

# The 2<sup>nd</sup> Texas A&M ENG-LIFE Workshop

At the Interface of Engineering and Life Sciences



Friday, April 24, 2015  
8:00 a.m. – 4:30 p.m.

Health Professions Education Bldg (HPEB) LL30

# **Welcome!**

We would like to welcome you to the 2<sup>nd</sup> Texas A&M University ENG-LIFE Workshop: At the Interface of Engineering and Life Sciences. The purpose of this workshop is to promote multidisciplinary interaction and scientific communication in the field of engineering and life sciences. This event will also offer a venue for graduate and undergraduate students to gain valuable experience by presenting their latest research results as well as interacting with fellow students and prominent researchers from Texas A&M University. We hope that you all enjoy this workshop.

Sincerely,

The 2015 Symposium Organizing Committee

# Acknowledgements

## Symposium Organizing Committee

Arum Han

*Associate Professor, Dept. Electrical and Computer Engineering*

Arul Jayaraman

*Ray Nesbitt Professor, Dept. Chemical Engineering*

Allison C. Rice-Ficht

*Interim Vice President for Research, Texas A&M Health Science Center*

*Regents Professor, Dept. Molecular and Cellular Medicine*

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# Program

Time	Activity	Speaker
8:00 - 8:30	Registration/Breakfast	
8:30 - 9:00	Opening Remarks	Dr. Brett P. Giroir: Executive Vice President and CEO, College of Medicine, Texas A&M Health Science Center Dr. M. Katherine Banks: Vice Chancellor for Engineering, Texas A&M University System; Dean, Dwight Look College of Engineering Dr. Eleanor M. Green: Dean, College of Veterinary Medicine & Biomedical Sciences
9:00 - 9:40	Opening Keynote	Convergence of Engineering and Biology to Generate Biotherapeutics of the Future Dr. Sally Ward (Molecular and Cellular Medicine) & Dr. Raimund J. Ober (Biomedical Engineering) Tag Team
9:40 - 9:55	Presentation 1	Applications for Imaging in Mycobacterium Tuberculosis Dr. Jeffrey Cirillo (Microbial Pathogenesis and Immunology) <i>Professor of Microbial Pathogenesis and Immunology</i>
9:55 - 10:10	Presentation 2	Artificial Sensory Augmentation via Skin Stretch Feedback and its Effect in Postural Control Dr. Pilwon Hur <i>Assistant Professor of Mechanical Engineering</i>
10:10 - 10:35	Coffee Break	
10:35 - 10:50	Presentation 3	Wrapping for Life: Neuroprotection and Myelin Regeneration in Demyelinating Diseases Dr. Jianrong Li <i>Associate Professor of Veterinary Integrative Biosciences</i>
10:50 - 11:05	Presentation 4	Synthetic Hydrogel Matrices for Stem Cell Delivery in Musculoskeletal Tissue Engineering Dr. Daniel L. Alge <i>Assistant Professor of Biomedical Engineering</i>
11:05 - 11:20	Presentation 5	Bacteriophage Antimicrobials Dr. Jason J. Gill <i>Associate Professor of Animal Science</i>
11:20 - 11:35	Presentation 6	MD Simulations and Free Energy Calculations in Protein Structure Prediction, Design and Biomaterials Dr. Phanourios Tamamis <i>Assistant Professor of Chemical Engineering</i>
11:35 - 11:50	Presentation 7	Potential Therapeutics for Fibrosing Diseases Dr. Richard Gomer <i>Professor of Biology</i>
11:50 - 1:00	Lunch	
1:00 - 1:45	Plenary Talk	Engineering Personalized Cancer Therapy Dr. Mauro Ferrari <i>President and CEO, Houston Methodist Research Institute</i>
1:45 - 2:00	Presentation 8	New Ultrasound Techniques for Musculoskeletal Applications Dr. Raffaella Righetti <i>Associate Professor of Electrical and Computer Engineering</i>
2:00 - 2:15	Presentation 9	Integrated Roles of Biology and Engineering in our Understanding of Lymphatic Function Dr. David C. Zawieja <i>Regent Professor and Interim Chair of Medical Physiology</i>
2:15 - 2:30	Presentation 10	Actin Remodeling by Nck Regulates Endothelial Lumen Formation and Promotes Angiogenesis Dr. Gonzalo M. Rivera <i>Assistant Professor of Veterinary Pathobiology</i>
2:30 - 2:40	Closing Remarks	Drs. Arum Han and Allison Rice-Ficht
2:40 - 4:30	Poster Session	

# Plenary Speaker



## **Mauro Ferrari, PH. D.**

President and CEO

Ernest Cockrell Jr. Presidential Distinguished Chair

### **Engineering Personalized Cancer Therapy**

Dr. Mauro Ferrari serves as President and CEO of Houston Methodist Research Institute (HMRI), where he holds the Ernest Cockrell Jr. Presidential Distinguished Chair. He is also Executive Vice President of Houston Methodist Hospital System and Director of the Houston Methodist Institute for Academic Medicine. He concurrently serves at Senior Associate Dean and Professor of Medicine at Weill Cornell Medical College, in New York, and holds Adjunct and Honorary Professorships at many universities around the world.

Dr. Ferrari's degrees are in Mathematics (Padova, 1985, Italy), and Mechanical Engineering (U.C. Berkeley, M.S. 1987, & Ph.D. 1989). He attended medical school at the Ohio State University (2002-03).

Dr. Mauro Ferrari is a founder of biomedical nano/micro-technology, especially in their applications to drug delivery, cell transplantation, implantable bioreactors, and other innovative therapeutic modalities. In these fields, he has published more than 250 peer-reviewed journal articles and six books. He is the inventor of more than 30 issued patents, with about thirty more pending in the US and internationally. He has received many prestigious honors, and research funding from NCI, NIH, DoD, NASA, NSF, DARPA, DoE, the State of Texas, and the State of Ohio, The Ohio State University, and several private enterprises. Dr. Ferrari served as Professor at Berkeley, Ohio State, and University of Texas, and served as Special Expert on Nanotechnology at the National Cancer Institute in 2003-2005, providing leadership into the formulation, refinement, and approval of the NCI's Alliance for Nanotechnology in Cancer, currently the world's largest program in medical nanotechnology. Dr. Ferrari is an academic-entrepreneur, with several companies that originated from his laboratory.

# Poster Session Abstracts

Health Professions Education Building (HPEB) Rm LL30

2:30 – 4:30 PM

(Arranged in alphabetical order by title)

## 1. Acoustofluidic Measurement of Cell Mechanical Property for Metastatic Cancer Diagnosis

H. Wang<sup>1</sup>, Z. Liu<sup>2</sup>, D.M. Shin<sup>3</sup>, G. Chen<sup>3</sup>, Y. Cho<sup>4</sup>, Y.-J. Kim<sup>2</sup>, and A. Han<sup>1</sup>

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Mechanical properties of cells such as compressibility are regarded to be different as cancer cells progress into metastatic state. Traditional methods for measuring mechanical properties of single cells such as AFM and micropipette aspiration, require labor-intensive procedures and can cause damage to cells due to direct contact, thus unsuitable for high-throughput measurement. Acoustophoretic force exerted on particles under acoustic-standing-waves depends on the particle and medium's vibro-acoustic properties. Thus, cells with different mechanical properties show different mobility under acoustic resonant field which can be analyzed to decipher the mechanical properties of cells. Here we present a high-throughput, single-cell-resolution, cell compressibility measurement approach based on acoustic-standing-wave-induced force, and the finding that breast cancer cells having different metastatic capacities show noticeable differences in compressibility. Trajectories of moving cells in the channel under acoustic standing wave excitation in the absence of flow are recorded. By using a microfluidic acoustophoretic model, the simulated trajectories of cells are calculated. Cells with highest metastatic capacity showed highest compressibility, consistent with previously reported clinical observations.

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## 2. Acute Phase Proteins and Arachidonic Acid Pathway Metabolites from Dogs with Spinal Cord Injuries

I. Reyes, A. Mondragon, B. Lee

Dept. of Veterinary Integrative Biosciences, Texas A&M University, USA, School of Veterinary Medicine and Biomedical Sciences, College Station, TX

One of the biggest challenges of today's world has been identifying the elusive cure for spinal cord injuries (SCI)<sup>1</sup>. The immune system's response to spinal cord injuries is to metabolize and release fatty acids such as arachidonic acid<sup>3</sup>. This metabolite undergoes the arachidonic acid metabolism pathway, which mediates the inflammatory response in humans and dogs to generate eicosanoids, signaling metabolites which play an important role in the body's immune system<sup>2</sup>. Previous studies mainly focused on mice and rats as animal models for spinal cord injuries have not proven to be very successful due to the difficulty in replicating symptoms. However, in our study, we have utilized *Canis familiaris* as our central animal model to test multiple samples of cerebral spinal fluid and plasma. We utilized enzyme-linked immunosorbent assays (ELISA) to quantify the concentration of multiple target metabolites, Prostaglandins E2 and Leukotriene C4. A precursor to the arachidonic acid pathway, Phospholipase A2, was also measured. In addition, Acute Phase Proteins present in *Canis familiaris* cerebral spinal fluid and plasma were quantified using enzyme-linked immunosorbent assays. The acute phase proteins under investigation were C-Reactive Protein, Haptoglobin,  $\alpha$ -1-glycoprotein, and Serum Amyloid A. Results in *Canis familiaris* showed that prostaglandin E2 concentrations were below the detection limit, Leukotriene C4 concentrations were measured to be high, and no correlation was detected between spinal cord injuries and Phospholipase A2. In addition, among the acute phase proteins, Haptoglobin had the highest concentration. P-values were adequate for all acute phase proteins measured excluding  $\alpha$ -1-glycoprotein.

## References:

- 1.) **Shearin AL, Ostrander EA.** 2010. Leading the way: canine models of genomics and disease. *Disease Models & Mechanisms.* **3**:27-34.
- 2.) **Sjölander, M et al.** 1999. Characterization of a leukotriene C4 export mechanism in human platelets: possible involvement of multidrug resistance-associated protein 1. *Journal of Lipid Research.* **40**:439-446.
- 3.) **Toborek M, Malecki A, Garrido R, Mattson MP, Hennig B, Young B.** 1999. Arachidonic Acid-Induced Oxidative Injury to Cultured Spinal Cord Neurons. *Journal of Neurochemistry.* **73**:684-692.

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## 3. Actin Remodeling by Nck Regulates Endothelial Lumen Formation and Promotes Angiogenesis

Sankar P. Chaki,<sup>1</sup> Rola Barhoumi,<sup>2</sup> Gonzalo M. Rivera<sup>1\*</sup>

<sup>1</sup>Department of Veterinary Pathobiology and <sup>2</sup>Department of Veterinary Integrative Biosciences, Texas A&M University

Multiple angiogenic cues modulate phosphotyrosine signaling to promote vasculogenesis and angiogenesis. Despite its functional and clinical importance, how vascular cells integrate phosphotyrosine-dependent signaling to elicit cytoskeletal changes required for endothelial morphogenesis remains poorly understood. The family of Nck adaptors couples phosphotyrosine signals with actin dynamics, and therefore, is well positioned to orchestrate cellular processes required in vascular formation and remodeling. Using a combination of endothelial cell culture in three-dimensional collagen matrices, molecular genetics, optical imaging, and biochemistry, we show that Nck-dependent actin remodeling promotes endothelial cell elongation and proper organization of VE-cadherin intercellular junctions. Major morphogenetic defects caused by abrogation of Nck signaling included loss of endothelial apical-basal polarity and impaired lumenization. Time-lapse imaging using a Förster resonance energy transfer biosensor, immunostaining with phospho-specific antibodies, and GST-pull down assays showed that Nck determines spatiotemporal patterns of Cdc42/aPKC activation during endothelial morphogenesis. Our results demonstrate that Nck acts as an important hub integrating phosphotyrosine signals with cytoskeletal changes that enable endothelial polarization and lumen formation. These findings point to Nck as an emergent target for effective antiangiogenic therapy.

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#### 4. Amorphous Silicon Oxynitride Thin Films Enhance Osteogenesis of Human Periosteal Cells and “Bone-like” Mineral Formation via Rapid Carbonate and Phosphate Functional Group Formation

Venu G. Varanasi,<sup>a,\*</sup> Azhar Ilyas,<sup>a</sup> Megen F. Velten,<sup>b</sup> Nickolay V. Lavrik,<sup>c</sup> Harry Meyer,<sup>c</sup> Karen More,<sup>c</sup> Harry K.W. Kim<sup>d,e</sup> and Pranesh B. Aswath<sup>b</sup>

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<sup>e</sup> Center for Excellence in Hip Disorders, Texas Scottish Rite Hospital, Dallas, TX 75219, USA

Structurally unstable fracture sites require metal fixative devices, which have long healing times due to their lack of osteoinductivity. Bioactive glass coatings lack in interfacial bonding, delaminate, and have reduced bioactivity due to the high temperatures used for their fabrication. Here, we test the hypothesis that low-temperature PECVD amorphous silica can enhance adhesion to the underlying metal surface and that N incorporation enhances osteogenesis and rapid biomineralization. A model Ti/TiO<sub>2</sub>-SiO<sub>x</sub> interface was formed by first coating Ti layers onto Si wafers, patterning, thermally annealing to form TiO<sub>2</sub>, and depositing SiO<sub>x</sub>/Si(ON)<sub>x</sub> overlays. TEM micrographs showed conformal SiO<sub>x</sub> layers on Ti/TiO<sub>2</sub> overlays while XPS data revealed the formation of an elemental Ti-O-Si interface. Nanoscratch testing verified strong SiO<sub>x</sub> bonding with the underlying TiO<sub>2</sub> layers. In-vitro studies showed that the surface chemistry changed from Si(ON)<sub>x</sub> to hydroxycarbonate apatite within 6 hours and Si(ON)<sub>x</sub> surface chemistry induced osteogenic gene expression of human periosteal cells and led to a rapid bone-like biomineral formation within 4 weeks. XANES data revealed that the incorporation of N increased the surface HA bioactivity by increasing the carbonate to phosphate ratio. In conclusion, silicon oxynitride overlays on bone implant systems enhance osteogenesis and biomineralization via surface nitrogen incorporation.

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## 5. Applications of 3D Printing in the Natural and Biomedical Sciences

Jonathan E. Bravo, Constance J. Woodman, Donald J. Brightsmith

Schubot Center, Dept. of Veterinary Pathobiology, Texas A&M University, USA

The increasing availability of 3D printing is changing the scope of manufacturing and research by allowing low cost, accurate production in the laboratory. We began experimenting with 3D printing only four months ago and in this time we have used it to facilitate work in multiple fields: field ornithology, behavioral research, animal welfare and university level education. In order to record wild macaw incubation behavior in Peru, we are using 3D printing to create artificial eggs to hold temperature and motion sensors. The resulting data will help us quantify wild bird incubation timing and movements and allow us to improve artificial incubation procedures in captivity. We have used 3D printing to create custom parts to design and build an automated enrichment robot capable of simultaneously improving captive animal welfare and recording data useful for behavioral research. We are also working with anatomy instructors in the College of Veterinary Medicine to print animal bones using medical imaging data. These bones are then used as teaching models and learning tools, reducing the need for real bones. Our work shows that with relatively little previous experience one can quickly use 3D printing to enhance a wide variety of research and teaching endeavors.

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## 6. A Biomimetic-computational approach to optimizing the quantum Efficiency Of Photovoltaics

A. Holzenburg<sup>1</sup>, L.M. Pérez<sup>2</sup>, and X. Qian<sup>3</sup>

<sup>1</sup>Microscopy & Imaging Center, Dept. Biology and Biochem. & Biophysics, Texas A&M University, USA

<sup>2</sup>Laboratory for Molecular Simulation, Dept. Chemistry, Texas A&M University, USA

<sup>3</sup>Materials Science & Engineering, Texas A&M University, USA

The most advanced low-cost organic photovoltaic cells have a quantum efficiency of ~10%. This is in stark contrast to plant/bacterial light-harvesting systems, which offer quantum efficiencies close to unity. The principal components of photosynthesis are: (i)  $h\nu$  is harvested and its energy transferred between pigment molecules. (ii) Energy transfer is optimized by funneling from lower to higher  $\lambda_{\max}$ , spectral overlap and quantum coherence, which constitutes the focus point of this project. (iii) The transferred energy leads to the quintessential charge separation ( $P \rightarrow P^* \rightarrow P^+ + e^-$ ) while (iv) recombination is avoided via the production of oxygen, reducing equivalents and membrane potential. Noting that quantum coherence is promoted by charged residues and local dielectrics, classical atomistic simulations and timedependent density functional theory (DFT) are used to identify charge/dielectric patterns and electronic coupling at exactly defined energy transfer interfaces. The calculations make use of structural information obtained on photosynthetic protein-pigment complexes while still in the native membrane making it possible to establish a link between supramolecular organization and quantum coherence in terms of what length scales enable fast energy transport and prevent quenching. Calculating energy transfer efficiencies between components based on different proximities will permit the search for patterns that enable defining material properties suitable for advanced photovoltaics.

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## 7. A Broadband MRI System for Simultaneous Multinuclear MR Spectroscopy and Imaging

J.C. Bosshard, S.E. Ogier, S.A. Blasczyk, and S.M. Wright

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Dept. of Electrical and Computer Engineering, Texas A&M University

The idea of combining magnetic resonance spectroscopic studies from multiple nuclei is quite compelling. A number of papers have been published dating all the way back to 1979. However, other than a few isolated studies, combined multinuclear studies are generally performed sequentially or interleaved. We are not aware of any systems that support simultaneous multi-coil, multinuclear studies. Motivation for revisiting simultaneous multinuclear spectroscopy and imaging comes from a number of sources. The development of hyperpolarized MRI and MRS, and the importance of magnetization management from the transient hyperpolarization may generate renewed interest in simultaneous multinuclear studies. Additionally, the increasing movement towards quantitative MRI and MRS provides motivation for truly simultaneous monitoring of the kinetics of some chemical reactions. This poster reports initial results in the Magnetic Resonance Systems lab in developing a wideband spectrometer capable of simultaneous excitation and acquisition of multiple nuclei and from multiple channel (array) receive coils. Results are shown from simultaneous  $^1\text{H}/^2\text{H}$  imaging on a 1.0T magnet, as well as simultaneous  $^1\text{H}/^{13}\text{C}$  spectroscopy on a 4.7T magnet, with both sets of results obtained from the same prototype broadband MR scanner.

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## 8. Calcium Measurement in Hippocampal Cultures with a Genetically-Encoded Indicator

A.H. Mahnke<sup>1</sup>, J. Wang<sup>1</sup>, U.H. Winzer-Serhan<sup>1</sup>

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Calcium plays a pivotal role within the cell as a second messenger, propagating signals from both external stimuli and intracellular events. Genetically-encoded calcium indicators (GECIs) are powerful tools for visualization of calcium dynamics. Unlike calcium-sensitive dyes, GECIs can be permanently expressed, allowing for longitudinal analysis of intracellular calcium changes. The GCaMP family of GECIs uses calcium-binding calmodulin conjugated to the green fluorescent protein (GFP). Elevation of intracellular calcium results in rapid binding of  $\text{Ca}^{2+}$  to calmodulin, inducing increases in fluorescence signals. Here we describe the use of the highly sensitive GCaMP6s to visualize spontaneous and evoked calcium signals in primary hippocampal cultures. After three days in vitro, cultures were transduced with an AAV5 viral vector containing GCaMP6s. Starting four days after transduction, confocal microscopy was used to record  $[\text{Ca}^{2+}]_i$  signals. The results revealed that both neurons and glia cells expressed functional GCaMP6s in the soma and far into cellular processes. Spontaneous calcium signals were detected in glia cells suggesting the presence of astrocytic calcium waves. In neurons, external stimuli that cause calcium influx, such as nicotine and KCl, transiently increased  $[\text{Ca}^{2+}]_i$  fluorescence signals in soma and dendrites. Subsequent fixation and antibody probing of the GFP moiety allowed for the analysis of GCaMP6s cellular localization as well as double immunocytochemistry for the characterization of cell types. The results confirmed the expression of GCaMP6s in soma and processes of neurons and glia cells. This methodology provides a novel approach of assaying calcium dynamics, allowing for new insight into cellular function.

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## 9. Characterization of ventral hernia repair using shear wave elastography

Chaudhry A<sup>1</sup>, J. S. Fernandez-Moure<sup>2</sup>, J. L. Van Eps, F. J. Cabrera<sup>2</sup>, S. Shajudeen<sup>1</sup>, E. Tasciotti<sup>2</sup>, B. K. Weiner<sup>2</sup>, M. Ferrari<sup>2</sup>, R. Righetti<sup>1</sup>

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In the United States, approximately 360,000 ventral hernia repairs (VHR) occur each year. In some cases hernia recurrence following repair may be as high as 30%. In order to assess the integrity of the repair imaging modalities such as computed tomography (CT) or ultrasound B-mode (US) are used. In both modalities, identification of the actual mesh is often difficult when mesh incorporation or breakdown has occurred. Also neither modality in its current embodiment has the capacity for simultaneously image the structure and stiffness of the mesh, which in many cases is the strongest indicator of recurrence of hernia. Shear Wave elastography (SWE) is a recently developed ultrasound technique that can be used to image the propagation of slow moving transverse waves through a tissue. The speed of the shear wave depends on the underlying tissue stiffness and can be mapped into Young's modulus by assuming the tissue to be incompressible single phase solid. In this study, we propose the use of 2D and 3D SWE to image ventral hernias repaired using polyester mesh. We expect that by using ultrasound shear wave elastography, we will be able to visualize both the hernia and the implanted mesh on the basis of their elastic modulus.

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## 10. Circadian Clock Regulation of the Three-Dimensional Chromatin Organization

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Virtually every mammalian cell harbors a molecular circadian clock that coordinates the rhythmic expression of 10-15% of the transcriptome and promotes the activation of key biochemical and metabolic pathways at the most appropriate time of day. Although rhythmically expressed genes have been identified, the mechanisms underlying their coordinated expression remain unclear. Recent studies have shown that chromatin looping generates long-range interactions between enhancers and genes' promoter that are critical for transcriptional regulation. Moreover, these chromatin rearrangements promote interactions between numerous genes and tissue-specific enhancers to synchronize the expression of many transcripts in a spatial and temporal manner. For these reasons, we hypothesized that clock-controlled long-range chromatin rearrangements underlie the harmonized expression of circadian transcripts in the mouse. To test our hypothesis, we applied the next-generation sequencing technique called Chromatin Interaction Analysis by Paired-End Tag sequencing (ChIA-PET) to mouse liver chromatin. This technique includes a RNA Polymerase II chromatin immunoprecipitation step that enables the detection of long-range chromatin interactions in transcriptionally active genes and the identification of interactions between enhancers and/or transcription start sites of genes that are directly engaged in transcription. Preliminary analysis of our mouse liver Pol II ChIA-PET datasets indicates that the technique is set up and readily detects specific long-range chromatin interactions of up to 200 kb. We will present a more complete analysis of our approaches, which are expected to reveal the importance of time-dependent genome reconfiguration for circadian transcription.

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## 11. A Complex-based Normal Mode Analysis for Conformational Changes Upon Protein Binding

Tomasz Oliwa<sup>1</sup> and Yang Shen<sup>1,2</sup>,

<sup>1</sup>Toyota Technological Institute at Chicago, USA, <sup>2</sup>Dept. Electrical and Computer Engineering, Texas A&M University, USA<sup>1</sup> Dept. Biology, Texas A&M University, USA, <sup>2</sup> Genetics Program, Texas A&M University

Protein motions and conformational changes are often associated with protein-protein interactions (PPIs), which is important to protein functions. However, it remains a fundamental challenge to understand and to anticipate those conformational changes. Models have been proposed to describe mechanisms of PPIs and resulting conformational changes. In particular, conformational selection theory considers protein motions as a pre-existing conformational ensemble, whereas induced-fit theory emphasizes binding effects.

Our study aims at modeling the contributions of both intrinsic flexibility and intermolecular interactions to protein conformational changes upon binding and applying the knowledge to protein docking. To that end, we extended the framework of anisotropic network model (ANM) to include both intra- and inter-molecular interactions with varying parameters to differentiate them, applied the models to a non-redundant set of encounter complexes approximated by rigidly-docked protein pairs, and assessed how well the resulting slowest normal modes could approximate conformational changes compared to those derived from traditional ANM that only accounts for intrinsic flexibility. Results indicated that the resulting Hessian matrices of the encounter complexes has a special structure related to those of individual components, and their lowest-eigenvalue eigenvectors (slowest normal modes) could approximate conformational changes better than traditional ones did, especially for cases with significant amounts of conformational changes (and thus with high levels of docking difficulty). Furthermore, our results reveals the importance of both intrinsic flexibility and intermolecular interactions on protein conformational changes and provides an efficient dimensionality-reduction approach for protein docking.

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## 12. Computational Identification of Genetic Subnetwork Modules Associated with Maize Defense Response to *Fusarium verticillioides*

Mansuck Kim<sup>1</sup>, Huan Zhang<sup>2</sup>, Charles Woloshuk<sup>3</sup>, Won-Bo Shim<sup>2</sup>, and Byung-Jun Yoon<sup>1,4</sup>

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<sup>4</sup>College of Science, Engineering, and Technology, Hamad bin Khalifa University (HBKU), Doha, Qatar.

Maize, a crop of global significance, is vulnerable to a variety of biotic stress leading to economic losses. *Fusarium verticillioides* is one of the key fungal pathogens of maize, causing ear rots and stalk rots. To better understand maize defense as well as *F. verticillioides* pathogenicity, a systematic investigation of the host-pathogen interactions is needed. The aim of this study was to computationally identify potential maize subnetwork modules associated with defense against *F. verticillioides*. We captured time-course RNA-seq reads from B73 maize inoculated with wild-type *F. verticillioides* and a loss-of-virulence mutant and subsequently performed a network-based comparative analysis. Specifically, we first analyzed the RNA-Seq data based on a cointegration-correlation-expression approach, where maize genes were jointly analyzed with known pathogenicity genes in *F. verticillioides*; (i) cointegration was used to analyze their expression trends over time, (ii) correlation was used to investigate the expression patterns across different replicates, and (iii) expression levels in all replicates were checked to focus on significantly expressed genes. We then searched the maize co-expression network to detect subnetwork modules that are differentially expressed with high significance when inoculated with two different fungal strains. Most of the candidate maize genes identified through the cointegration-correlation-expression approach were subjected to GO (gene ontology) term designation, thereby further selecting for the modules that are likely to be associated with maize defense response. In conclusion, we report potential maize defense response modules predicted by our proposed method as well as the interactions amongst the genes in the modules.

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### 13. Cytokines and Chemokines, Acute Phase Proteins, and Arachidonic Acid Pathway Metabolites from Dogs with Acute Spinal Cord Injuries

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Canine intervertebral disk herniation (IVDH) is a common, naturally-occurring form of spinal cord injury (SCI) which is increasingly being used in pre-clinical evaluations of therapies. Although IVDH bears critical similarities to human SCI with respect to lesion morphology, imaging features, and post-SCI treatment, limited data are available concerning secondary injury mechanisms. Here we characterized cerebrospinal fluid (CSF) cytokines and chemokines in dogs with acute, surgically treated, thoracolumbar, and healthy control dogs to investigate early inflammatory events following SCI. Additionally, we measured the levels of acute phase proteins (APP) and arachidonic acid pathway (AAP) metabolites in both sets. Interleukin (IL)-2, -6, -7, -8, -10, -15, and -18, granulocyte macrophage colony-stimulating factor (GM-CSF), interferon gamma (IFN-g), keratinocyte chemoattractant (KC)-like protein, IFN-g-inducible protein-10, monocyte chemotactic protein 1 (MCP-1), and tumor necrosis factor alpha (TNF- $\alpha$ ), were measured using a bioplex system. Enzyme-linked immunosorbent assay (ELISA) kits were used to measure the APP profile: c-reactive protein (CRP), serum-amyloid-A (SAA), haptoglobin,  $\alpha$ -1-acid glycoprotein, and AAP metabolites: leukotriene C4 (LT-C4), prostaglandin-E2 (PG-E2), phospholipase-A2 (PL-A2). The cytokine/chemokine profiles indicated that IL-8 and KC-like concentrations were significantly higher in dogs with SCI than the healthy control dogs. In the APP profile, haptoglobin levels were determined to be significantly higher in dogs with SCI. Finally, in the AAP profile, PG-E2 levels were significantly higher in dogs with SCI than the control dogs. These tests indicate that cytokines, chemokines, APP, and AAP metabolites, may play a role in indicating acute SCI in canine IVDH and also in the recovery process.

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### 14. Degradation of Shape Memory Polyurethanes and Foams

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Polyurethanes have been the material of choice for cardiovascular devices and blood-contacting applications for decades, however the degradation products of these materials in many cases are suspected or known carcinogens, mutagens, toxic, ect. Generally, these toxic products also only begin appearing months or years after implanting the medical device, requiring the use of accelerated testing methods to examine degradation; the degradation of the urethanes occurs spontaneously over time due to hydrolysis. Shape memory polyurethanes (SMPs) shows promise for medical devices, are thermally sensitive, and possess excellent bulk biocompatibility. We examine a series of thermoset SMPs and their foams for degradation. Variations in crosslink density, chemical composition, porosity, and hydrophobicity are considered for their role in altering degradation rates and products. Preliminary results indicate that substantial degradation occurs via oxidation, not hydrolysis. The hydrophobicity and chemical composition of the backbone of the polymer are shown to dramatically alter the rates of degradation over time. Correlations between previous studies and commercially available polyurethanes are made, showing how studied SMPs different behavior can be related to the examined properties and how this can be controlled in medical devices.

## 15. Development and Use of Biosensors to Measure Spatial and Temporal Concentration Profiles of Phosphate in Plants and Animals

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Phosphate (Pi) is an essential nutrient that is found in every compartment of a eukaryotic cell and serves key roles in energy conversion, signal transduction and metabolism. To fulfill these fundamental roles, plants and animals rely on a series of specialized transport processes to acquire and distribute Pi to all cells and subcellular compartments. The mechanisms of these transport processes, and how these are coordinated with environmental and metabolic conditions are poorly understood, largely due to the inability to monitor Pi with high spatial and temporal resolution in live organisms. To overcome this limitation we have developed a suite of genetically encoded FRET-based biosensors that permit live imaging of Pi in individual cells and subcellular compartments. These biosensors consist of a translational fusion of CFP, a cyanobacterial Pi binding protein, and a circularly permuted version of YFP (Venus). Live imaging of Pi biosensors in a plant, *Arabidopsis thaliana*, revealed cell-specific changes in cytosolic Pi concentrations in response to environmental supply, and confirmed the activity of a Pi transporter in an organelle. Live imaging experiments in which the same biosensors were expressed in the nematode *Caenorhabditis elegans* suggest functional conservation of Pi mobilization processes for organisms of different biological kingdoms.

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## 16. Development of the wild tomato *Solanum pennellii* as a novel and sustainable biofuel feedstock

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*Solanum pennellii* (Solanaceae), a wild relative of cultivated tomato (*S. lycopersicum*), is a potential new feedstock for biofuels. In order to reduce water loss in their native dry habitat in Peru, these plants secrete massive amounts of 2,3,4 tri-O-acylated glucose esters through their trichomes. Transesterification of the secreted compound yields three molecules of C4 to C12 fatty acid esters, which can be used as biogasoline. We are currently growing these plants on a large scale in order to produce enough fuel for testing. The biosynthetic pathway of the glucolipids involves only four or five enzymes, making it a good candidate for transfer to other plants. We successfully cloned cDNAs encoding the first two enzymes of the pathway (UDP:glucose glucosyltransferase and glucose acyltransferase). We used GFP reporter constructs to determine whether expression of these genes is restricted to trichomes. Future goals include discovering the remaining genes in the glucolipid biosynthetic pathway and transferring at least the first two genes into tobacco and other plants with large leaf surfaces to improve yield of this novel biofuel. We used next-generation DNA sequencing and analysis and assembly tools on the BioGalaxy server to construct a reference transcriptome to serve as a baseline for additional RNA-seq experiments between high- and low-glucolipid-producing accessions to reveal the genes associated with this pathway. Our reference transcriptome contains 68,864 unigenes (average length 740.07 ± 679.79 bp, N50 = 1151) and transcripts for 456 of 458 core eukaryotic proteins, indicating the high quality of this assembly.

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## 17. Effect of Customized Haptic Feedback on Navigation Characteristics and Performance

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This study presents an approach to customize haptic assistance based on subject's performance and performing-characteristics. Determining the level of haptic assistance, e.g., the magnitude and direction of haptic force, is a challenging problem. For instance, an excessive assistance level may degrade user's performance and cause discomfort, whereas a lack of enough assistance may yield task-failure. In our approach, expert's control strategy was utilized to provide novices with customized haptic guidance. First, subject's control strategy for given tasks was parameterized by inverse optimal control. The obtained parameters serve as metrics to customize haptic feedback for each subject by i) assigning a coach whose task-performing characteristics are the desired characteristics for the rest of the subjects, ii) defining a spine (guiding path) which guides subjects based on the coach's strategy, and iii) determining the level of assistance. Sixteen healthy young adults participated in the experiment consisting of baseline and evaluation sessions. Based on each subject's baseline data, subjects' control strategies were parameterized, and customized haptic feedback for each subject was determined. In the evaluation session, subjects' performances were evaluated while customized haptic guidance were provided. The results showed that task-completion time for slower subjects was enhanced and the variability of subjects' performance reduced when customized haptic guidance was provided. Interestingly, variability reduced prominently for those subjects whose task-performing characteristics differed from the coach's. The findings in this study can contribute to the development of powered wheelchair with customized haptic guidance system for persons with difficulties in navigation.

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## 18. Effect of Sensory Augmentation via Skin Stretch Feedback on Quiet Standing Balance

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Postural control and balance are two important factors to humans in performing activities of daily living (ADL). In the past two decades, studies have shown that light touch of fingers on fixed surfaces can reduce postural sway during quiet standing and walking for people with or without dysfunction of sensory systems. With the help of additional sensory information from biofeedback, individuals with neurological impairments may improve their balance in ADL. We developed a portable sensory augmentation device that can induce skin stretch feedback in response to signal postural sway to enhance balance. The efficacy of the developed device was evaluated and our results indicated that the presence of additional sensory input helped maintaining balance when both visual and vestibular inputs were removed while quietly standing. We expect that this system will have great impacts on the development of portable balance rehabilitation device to enhance postural control and to help reduce fall risk for neurologically impaired patients and the elderly and eventually lead to enhanced quality of their lives.

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## 19. Effects of isophorone diisocyanate on the thermal and mechanical properties of shape-memory polyurethane foams

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Previously developed shape-memory polymer foams display fast actuation in water due to plasticization of the polymer network. The actuation presents itself as a depression in the glass transition temperature when moved from dry to aqueous conditions; this effect limits the working time of the foam to ten minutes when used in a transcatheter embolic device. We have developed reproducible foams, by altering the chemical backbone, which can achieve working times of greater than twenty minutes. We accomplished this by incorporating isophorone diisocyanate into the foam, resulting in increased hydrophobicity, glass transitions, and actuation time. This delayed actuation, when compared to our previous systems, allows for more optimal working time (< 5 minutes) in clinical applications.

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## 20. Efficient Photothermal Therapy of Brain Cancer Through Functionalized Graphene Oxide

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Current clinical treatments including surgical resection, radiation therapy and chemotherapy for brain cancer result in high mortality due to complex structure of brain and aggressiveness of brain cancer. Recently, non-invasive photothermal therapy (PTT) with near infrared laser irradiation has been developed as an alternative emerging therapy for brain cancer. In this paper, a biocompatible porphyrin functionalized graphene oxide (PGO) with high absorbance at 808nm is synthesized as a photothermal platform for brain cancer therapy. Graphite flake was intercalated, exfoliated and then conjugated with porphyrin through  $\pi$ - $\pi$  interactions. This PGO is two times more stable than reduced graphene oxide (rGO) in aqueous solution. Most importantly, the efficiency of photothermal conversion of PGO is increased by 89% compared to GO, and 33% compared to rGO with 808nm laser irradiation, causing large number of brain cancer cells ablation in vitro. This PGO platform containing active functional groups allows specific targeting in PTT without harming healthy cells and tissues.

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## 21. Effects of Co-Planar Shielding of Array Elements for High Field MRI

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Shielding of MRI radiofrequency (RF) coils decreases radiative and dielectric losses and becomes particularly important with increases in magnetic field strength with the linear relationship to frequency of operation. Use of array coils is standard for increasing SNR and decreasing scan time in MRI, with typical clinical scanners equipped with at least eight receiver channels. Shielding the elements of an array coil that are overlapped for geometric decoupling becomes problematic however. This work investigates the efficacy of “coplanar shielding” of array elements, in which copper shields are oriented concentric and coplanar to the RF coils rather than implemented as a full ground plane behind them, allowing for ease of fabrication and overlapped, inter-element, geometric decoupling. Two 5-channel receive arrays were constructed on a cylindrical former in an overlapped, “Olympic ring” geometry with 18 AWG Cu wire: one with unshielded elements and one with coplanar shields with a geometry optimized from FDTD simulation. A 15-cm diameter container of saline (0.58 S/m) was imaged with both arrays on a whole-body 7T scanner (Achieva, Phillips Medical Systems). Average SNR increased by 13% for the shielded vs. unshielded case; inter-element isolation improved, showing reductions in noise-correlation between channels (average values: 3.3% vs. 4.6%); g-factor maps in accelerated imaging showed 12-15% reductions in average g-factors and 30-36% reductions in max g-factors, indicating an increased ability to provide acceleration with less expense to SNR. Coplanar shielding was shown to be effective at increasing imaging quality and acceleration performance while offering advantages in terms of array fabrication.

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## 22. Electrochemical biosensors based on polymer brushes and hybrid metallic nanostructures for real-time detection of foodborne pathogens

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Recent foodborne outbreaks have heightened public concern about food safety and created a greater impetus to improve methods for pathogen detection. Traditional analytical techniques for detecting pathogens are time-consuming, labor-intensive, and expensive. More rapid, sensitive, and reliable methods are needed for pathogen detection in food products to ensure public health. Electrochemical nanotechnology offers a unique opportunity to address some of these challenges using biopolymer-nanocomposite based electrochemical platforms as biosensors for the detection of these pathogens. In this work, we develop graphene-nanometal hybrid materials combined with stimuli-responsive biopolymer nanobrushes (chitosan and poly(N-isopropylacrylamide, PNIPAAm) functionalized with various capture probes (aptamers and lectins) for targeting foodborne pathogens (*Listeria monocytogenes*). We determined the best loading capacity for the different capture probes on the biopolymer nanobrushes and characterize their electrochemical properties. Furthermore, their actuation using pH and temperature variations was evaluated for capturing and detecting pathogens to determine the optimum configuration. Electrochemical impedance spectroscopy measurements in phosphate buffer and fresh produce homogenates demonstrate the effectiveness of the biosensors in detecting pathogens in a broad concentration range. Limit of detection, sensitivity, selectivity, linear range, response time, and durability for each biosensor platform were evaluated. Results show that the combination of stimuli-responsive polymer nanobrushes and hybrid metallic nanostructures enhanced capture of target bacteria and electrochemical acquisition signal with enhanced sensitivity and detection limit. Consequently, the developed platform demonstrated a rapid (within minutes) biosensor system that can be used for pathogen detection in food.

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### 23. Epigenome Method and its Application to Hematopoietic Stem Cells

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5mc and 5hmc can be quantitatively measured at base level by whole genome bisulfite sequencing. However, lack of accurate methods describing and utilizing digital methylation information from single base to region level, and lack of comprehensive analysis pipeline are two major challenges when WGBS is expensive and hence sequencing depth is low and replicate number is low. One good example is highly purified Hematopoietic Stem Cells(HSC), which are extremely low in number and expensive to sequence.

These two bioinformatics challenges were solved by MOABS, a comprehensive, accurate and efficient solution for DNA methylation data analysis. Especially MOABS is accurate even at low coverage and is aware of replicate reproducibility.

MOABS was applied to the first epigenome dataset of mouse HSCs. In order to investigate the HSC biology of cancer from epigenetic perspective, two strategies were used including aging and DNA methylome perturbation by knockout of DNMT3A and DNMT3B. The aging study described correlated epigenome and transcriptome changes, discovered ribosome up-regulation and aberrant methylation on MDS genes during aging process. The knockout studies discovered large under-methylated regions (Canyons) and revealed the specific role of DNMT3B.

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### 24. The FIT in fat: Mating behavior and metabolism are influenced by fat body FIT expression and secretion into hemolymph

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Vertebrate adipose tissue modulates central nervous system (CNS) activity via the secretion of hormonal factors known as adipokines. These factors can affect the development of the neural circuitry as well as the response of the CNS to environmental changes. Insect fat body functions analogously to vertebrate adipose and hepatic tissues. Fat body serves as a repository for lipid and glycogen stores, but it also is an endocrine gland that produces and releases factors into the hemolymph (insect plasma). Our lab's previous work indicated that sexual and social experience affect expression of a fat body-expressed gene, *female-specific independent of transformer (fit)*. Unpublished work suggests that *fit* modulates both courtship behavior and starvation responses. The predicted FIT protein has a signal sequence, indicating that the protein may be secreted by the fat body into hemolymph. We hypothesized that circulating FIT signals to the CNS to influence behavior and metabolism. To determine if FIT is secreted, we developed an anti-FIT antiserum and tested it in *D. melanogaster* via Western blotting. Since *fit* has female-biased expression, we first tested female tissues for FIT protein. We detected FIT in female whole bodies and hemolymph. *fit* transcripts previously were detected predominantly in head fat body and at much lower levels in the brain. Although we also detected FIT in heads, expression was lost when we compared FIT expression separately in brains and head carcasses (contain head fat body). One explanation for not detecting FIT in head carcasses is that the rate of protein translation does not match the rate of transcription for the gene. It could also be that FIT is quickly secreted into the hemolymph and is not present in fat body in detectable amounts. Determining where FIT is located can help in understanding which tissues are influenced by this fat-expressed and secreted protein that modulates mating and metabolism.

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## 25. Functional Materials: Functionalization of Ultrabithorax Materials with Vascular Endothelial Growth Factor Enhances Primary Human Endothelial Cell Survival and Activation

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Advances in tissue engineering will lead to new therapies that restore, maintain, or enhance function in damaged tissues. However, the most successful applications have been in thin (~2 mm) tissues, in which delivery of essential nutrients occurs by diffusion. Tissue engineering of more complex tissues cannot be supported by simple diffusion and will require vascular networks to support cellular function. In this study, we have tested the ability of materials derived from the *Drosophila melanogaster* Hox protein Ultrabithorax (Ubx) because they can be readily functionalized with full-length proteins by fusing DNA encompassing the functional protein to the *ubx* gene. Because Ubx self-assembles rapidly in mild aqueous buffers, proteins fused to Ubx are incorporated into the resulting materials in a native, fully functional material that is not toxic to human vascular cells and promotes sustained cellular interactions in culture. Our initial studies have focused on whether vascular endothelial growth factor (VEGF) fused to Ubx provides a functional scaffold that can influence endothelial cell survival, activation, and sprouting in *in vitro*, *ex vivo*, and *in vivo* experiments.

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## 26. Galectin-3 binding protein secreted by breast cancer cells inhibits fibrocyte differentiation

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To metastasize, tumor cells often need to migrate through a layer of collagen-containing scar tissue which encapsulates the tumor. A key component of scar tissue and fibrosing diseases is the fibrocyte, a collagen-secreting pro-fibrotic cell. To test the hypothesis that invasive tumor cells may block the formation of the fibrous sheath, we determined if tumor cells secrete factors that inhibit fibrocyte differentiation. We found that the human metastatic breast cancer cell line MDA-MB 231 secretes activity that inhibits human fibrocyte differentiation, while less aggressive breast cancer cell lines secrete less of this activity. Purification indicated that Galectin-3 Binding Protein (LGALS3BP) is the active factor. Recombinant LGALS3BP inhibits fibrocyte differentiation, and immunodepletion of LGALS3BP from MDA-MB 231 conditioned media removes the fibrocyte differentiation-inhibiting activity. LGALS3BP inhibits the differentiation of fibrocytes from wild-type mouse spleen cells, but not from SIGN-R1<sup>-/-</sup> mouse spleen cells, suggesting that CD209/SIGN-R1 is required for the LGALS3BP effect. Galectin-3 and galectin-1, binding partners of LGALS3BP, potentiate fibrocyte differentiation. In breast cancer biopsies, increased levels of tumor cell-associated LGALS3BP were observed in regions of the tumor that were invading the surrounding stroma. These findings suggest LGALS3BP and galectin-3 as new targets to treat metastatic cancer and fibrosing diseases.

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## 27. A High-Throughput Microfluidic Droplet-based Polymicrobial Assay for Antimicrobial Drug Discovery

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Antibiotic resistant and rapid continuously evolving microbes are becoming commonly prevalent in nature, proposing an enormous challenging to discover and develop drugs which exhibit antimicrobial properties. Conventional antimicrobial drug discovery approaches can be drastically improved, in terms of cost, throughput, and repeatability, by implementing a droplet microfluidics screening assay to identify and analyze antimicrobial products generated by microbes. The assay consists of two high-throughput droplet generators to continuously encapsulate a given number of microbes/media in fluorinated oil, followed by droplet synchronization using a railroad-like microfluidic structure to prepare droplets for one-to-one merging. Droplets are then merged using continuous high-throughput electric field based merging, followed by droplet storage and cultivation or co-cultivation. After co-cultivation, fluorescently labeled microbes of interested are detected using a compact dual-wavelength home built free optics-based detection platform. In this way, one can continuously and simultaneously detect the presence of two cells (i.e. GFP/RFP label cells) on the fly at a high-throughput to determine which droplets contain cells producing antimicrobial properties. The system is capable of providing antimicrobial property analysis of cells to a known pathogen of interest and isolating a single droplet from a million droplet assay. Droplets of interest, which contain microbes exhibiting antimicrobial properties, are sorted using a high-throughput electric field based droplet sorting method. Droplets exhibiting antimicrobial properties are then transferred back to a conventional culture and antimicrobial compounds synthesized by microbes are identified and characterized. This approach can drastically improve the identification of novel molecules that previously had unrecognizable mechanisms of microbial inhibition/attenuation.

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## 28. Identifying effective pathway markers for breast cancer prognosis through relative gene expression analysis using decision trees

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Identification of robust biomarkers for cancer classification is an important research problem in translational genomics that has received significant attention by many re- searchers. Several studies have shown that module-based markers tend to be more robust and lead to higher prediction accuracy than classic single-gene markers. These modular markers jointly analyze the gene expression activities of closely associated genes; for example, those that belong to a known pathway or genes whose protein products form a subnetwork module in a protein-protein interaction (PPI) network. Recently, decision trees have been shown to be very effective for logically interpreting the expression level of interacting genes, as they can capture a wide range of coordinated gene activities within a given module. Here, we propose a novel method for identifying robust pathway markers that can accurately predict breast cancer metastasis based on decision trees and their logic functions. In our method, we utilize relative expression levels obtained through pairwise comparison of genes, which have been previously shown to yield more reproducible results with better interpretability. Through extensive evaluations using multiple breast cancer gene expression datasets, we demonstrate that the proposed method can identify better pathway markers that can improve the overall performance of prognostic cancer classifiers.

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## 29. The Impact Of Obesity On Walking And Dual-Task Situations In Older Individuals: A Prefrontal fNIRS Study

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Recent evidence of obesity-related changes in the prefrontal cortex (PFC) during cognitive and seated motor activities have surfaced; however, the impact of obesity on PFC regulation during ambulation remains unclear. The purpose of this study was to determine obesity-specific differences in prefrontal cortex activation using functional near infrared spectroscopy (fNIRS) during single- and dual-task walking situations in older adults. Ten non-obese and ten obese individuals, 65 years and older, performed single (walking) tasks and dual (walking with: verbal, motor, precision gait, and decision-making) tasks. Maximum oxygenated hemoglobin (HbO<sub>2</sub>) was measured bilaterally using a portable fNIRS system. Gait speed and performance on the dual tasks were also obtained.

Dual-task conditions, namely precision gait and decision-making tasks, were associated with greater PFC activity compared to the single-task condition (both  $P < 0.0001$ ) despite the precision gait task being associated with slower gait speeds ( $P = 0.0001$ ). Obesity was marginally associated with greater activation in the PFC during the single- and dual-task conditions ( $P = 0.057$ ) despite both obesity groups having comparable gait speeds ( $P = 0.229$ ) and performances in the secondary tasks (all  $P > 0.232$ ). The study is one of the first to examine the dependence of gait on PFC function in non-obese and obese older adults during walking and dual-tasks using fNIRS. These preliminary findings have strong public health implications in identifying individuals who are at greater risks of falls, particularly when performing complex mobility tasks.

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## 30. Impact of Myosin5a Mutation in Neurodegeneration: a Rat Animal Model

Dr. George Stoica.

Myosin5a (Myo5a) is an actin-dependent motor protein that is highly expressed in the brain, and involved in vesicular/organelles transport and its absence leads to movement disorders in humans and animal species (Griscelli and Elejalde syndromes in humans), rodents (dilute lethal phenotype in mice, and dilute-opisthotonus of Wistar rats), and Arabian horses Lavender Foal Syndrome.

A spontaneous autosomal recessive rat model for neurodegeneration caused by a mutation in the Myo5a gene was developed in our laboratory. The pleiotropic effects of this mutation affect the coat color, central nervous and neuroendocrine systems.

Preliminary data from our model of Myo5a mutant Berlin-Druckrey (BD-IV) “shaker” rats demonstrated marked alterative changes involving the alpha-synuclein (a-syn) overexpression, decrease dopamine (DA) levels, alteration of DA metabolism, and overexpression of tau protein in specific anatomical area of brain in shaker rats compared with non-affected siblings. The mechanisms responsible for neurological phenotypes in the deficient Myo5a affected animals are less understood and suggest pleiotropic origins.

These neurological degenerative changes are common in human neurodegenerative diseases such as Alzheimer, Parkinson’s, and Lewis Body dementia, which make this animal model ideal for mechanistically investigating human diseases with potential development of novel therapy, which can lead to translational studies. The main challenge for the future will be to investigate the molecular mechanisms of Myo5a and interaction with other proteins underlying its functions.

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## 31. Integrin Signaling Regulates Actin Cytoskeleton Organization

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Vascular smooth muscle cells (VSMC) predominantly express integrins  $\alpha_5\beta_1$  and  $\alpha_v\beta_3$  which are involved in the regulation of contractile function. Mechanical stretch alters conformation of  $\beta$ -tails to provide binding sites for key focal adhesion (FA) proteins. This process is followed by actin cytoskeleton remodeling to alter VSMC function accordingly. Our study investigates the relationship between integrin distribution and function with respect to stress fibers morphology in VSMC. To determine the role of integrin activation in FA formation, VSMC co-expressing integrin  $\beta_3$ -EGFP variants (wild-type (wt), inside-out and outside-in constitutively active (CA), dominant negative (DN) mutants) with vinculin- or dSH2-mCherry have been imaged by total internal reflection fluorescence (TIRF) microscopy. Measurements of integrin area relative to cell area showed that expression of CA integrin mutants induced significant integrin clustering all over the basal cell surface. In contrast, lack of integrin localization at FA has been exhibited by expression of the DN-mutant. To determine the integrin functional involvement in stress fiber morphology, VSMC coexpressing actin-mRFP and integrin  $\beta_3$  variants were imaged by confocal microscopy. Cells expressing integrin  $\beta_3$  CA-mutations exhibited a significant increase in stress fiber formation compared to wild-type, while cells expressing DN-mutation presented thinner actin fibers. These results show that F-actin area and  $\alpha_v\beta_3$  integrin clustering at FA exhibit similar trends, suggesting that stress fiber formation may be related to integrin activation state.

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## 32. Laboratory for Optical Diagnosis and Imaging (LODI)

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The Laboratory for Optical Diagnosis and Imaging (LODI) was established within the Department of Biomedical Engineering at Texas A&M University in 2007. The overall mission of our laboratory is to develop technologies to further the understanding of the underlying physiology and biology in various pathological conditions, and to translate such technologies into the clinical arena. The focus of the laboratory is to develop optical spectroscopy and imaging instrumentation and related signal and image processing tools to nondestructively quantify the structure, molecular composition, and physiological state of biological tissues with both macroscopic and microscopic resolutions. Early tissue pathological transformations are accompanied by subtle changes in tissue microstructure, biochemical composition, and physiological regulations. Thus, our hope is that some of our developing technologies will help to clinically detect diseases during their early stages as well as to guide, monitor, and personalize clinical interventions.

Here, we will give an overview of our current efforts to develop optical instrumentation and computational methods for both pre-clinical and clinical applications. In particular, we will focus on Fluorescence Lifetime Imaging Microscopy (FLIM), which can be used to characterize tissue biochemical composition. We will discuss the application of this optical imaging technology as a clinical tool for oral cancer diagnosis and as a pre-clinical tool to study mechanisms of coronary atherosclerosis development.

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### **33. Lack of Efficacy of Benzodiazepines to Stop the progression of epilepsy development: A video-EEG analysis of DFP-treated rats**

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Presently, there is no cure for epilepsy. Acquired epilepsy is triggered by diverse cascading factors such as brain injury, stroke, infections, or prolonged seizures caused by chemical exposures. Organophosphate (OP) nerve agents and pesticides cause seizures, status epilepticus (SE), and brain damage due to neuroexcitotoxicity. The development of epilepsy is a major chronic neurological dysfunction in survivors of OP poisoning. In the present study, a video-EEG technology was utilized to determine the incidence of epilepsy and its modification by benzodiazepine treatment in rats exposed to the OP agent DFP. 24/7 video-EEG activity was recorded continuously for up to 24 days around 3 months post exposure to verify the occurrence of epilepsy with spontaneous recurrent seizures after DFP exposure. DFP exposure led to persistent acute seizure activity in rats. Among the control (untreated) rats, approximately 78% exhibited spontaneous seizures within 3 months after DFP exposure. Rats exhibited generalized seizures (stage 3 to 5) of 30–60 s duration with a frequency of 2 to 5 seizures per week. Sixty-seven percent of the rats treated with diazepam developed spontaneous seizures. The incidence spontaneous seizures were 64% in the midazolam-treated group. Neuronal degeneration evaluated through Nissl staining revealed damage in cortical and hippocampal areas. The data from chronic video-EEG recording confirm the development of epilepsy with spontaneous recurrent seizures in the DFP model. Benzodiazepines failed to stop the progression of the epilepsy development triggered by DFP exposure. New drugs are needed for preventing epilepsy in victims of chemical exposures. \*\* NIH Grant U01-NS083460 (to DSR)\*\*

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### **34. Lymphatic endothelial cell response to X-ray irradiation**

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The lymphatics play a key role in the redistribution of interstitial fluids, cytokines, immune cells, and macromolecules via formation of lymph. Impairment in lymphatic function causes chronic swelling and inflammation, characterized as lymphedema. Secondary lymphedema development has been a long-known issue in cancer patients receiving radiation therapy, with the underlying mechanisms for lymphedema development unknown. Furthermore astronauts develop edema while in space, being exposed to an environment that alters normal hydrostatic pressure gradients as well as exhibiting chronic, low-dose background radiation even with shielding. This is one of the first investigations looking at viability and cell morphological response in primary, rat mesenteric lymphatic endothelial cells in response to a broad range of X-ray doses at multiple time points. Briefly, we characterized cell viability by a XTT cell viability kit and identified cell structural changes via immunofluorescence for markers  $\beta$ -catenin, F-actin, and DRAQ7. Doses applied include 0.5, 1, 1.5, 2, 4, and 16 Gy, with measures being performed at 6, 24, 72, and 264 hours post-irradiation. Preliminary observations include acute responses (24-72h) in all irradiated groups, with decreased viability, loss of cell-to-cell adhesion, and increased cell surface area. Further protein markers for cell morphology will be conducted (VE-cadherin, VCAM), as well as functional measures of permeability. These experiments will build the framework for future investigations towards clinical and space-flight relevant radiation exposure response.

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### 35. Mechanical and *in vitro* Evaluation of an Experimental Canine Patent Ductus Arteriosus Closure Device

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Patent ductus arteriosus (PDA) is a congenital cardiovascular disease in which a connection between the aorta and pulmonary artery does not close shortly after birth. If this defect is not closed, it can lead to serious complications and even death. A prototype device, the nitinol foam cage (NFC), has been developed in an attempt to address the shortcomings of the current treatment methods. The NFC utilizes a nitinol frame and a shape memory polymer foam to promote embolization and tissue healing.

The NFC's mechanical properties were evaluated and compared to the Amplatz Canine Ductal Occluder's (ACDO) in multiple mechanical and *in vitro* experiments. The NFC exerted similar radial pressures to those of the ACDO for all but the two largest vessel diameters tested, but it required a much lower force to dislodge the device from its ideal position compared to the ACDO. The NFC exhibited minimal device migration, remained in the desired location in the *in vitro* models, and received positive clinician feedback, including that it offered less resistance and was easier to deliver in the same sized sheath as the ACDO. While the ACDO exhibited superior mechanical properties, the NFC performed well in the *in vitro* experiments, warranting further development of this design as an alternative method to treat PDA.

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### 36. Microfluidic Microalgae Screening Platforms for Growth and Oil Production Analysis

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Microalgae have been envisioned as a future source of renewable energy. However, significant improvements such as development of better performing microalgal strains, optimization of culture conditions, and better understanding of microalgal biology are required to obtain economically feasible microalgal biofuels. To resolve these limitations, massively parallel studies are needed, however, current microalgae culture systems lack high-throughput screening capabilities, and thus not suitable for the parallel studies. Here, we present series of microfluidic microalgae screening platforms, each of which addresses major bottlenecks towards commercial viability. The first platform, a microfluidic photobioreactor array has been developed to investigate the effect of different culture conditions on microalgal growth and oil production. This platform can provide up to 64 different culture conditions on-chip, such as combinations of different light intensities, light cycles, and culture media/chemical compositions. The second platform, a high-throughput droplet microfluidics-based microalgae screening platform has been developed to investigate/compare the growth and the oil production of microalgal libraries with much higher throughput. On-chip microalgal oil staining and high-throughput optical detection capabilities in the developed platform allows for large numbers of microalgae samples to be analyzed within a short period of time. The third platform utilizes Raman spectroscopy to perform non-destructive *in vivo* identification and quantification of microalgal biofuel production. We expect that these platforms will serve as a powerful tool to investigate better microalgal strains as well as optimum algae culture conditions at significantly lower costs and shorter times, which can dramatically enhance the development of renewable algal energy systems.

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### **37. miRNA-lncRNA interactions as a regulator of neural stem cell pluripotency and a target for ethanol teratogenesis**

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Fetal alcohol exposure is a leading non-genetic cause of neurodevelopmental disability. Neural stem cells (NSCs) that give rise to most neurons of the adult brain during the first and second trimester are particularly vulnerable. We previously found that ethanol exposure did not result in NSC death, but rather, the loss of NSCs due to premature maturation. Moreover, we found that a class of small non-protein-coding regulatory miRNAs was decreased following ethanol exposure. We recently found that the loss of miRNAs result in premature expression of a network of genes that support premature NSC maturation. However, the question that remains is whether ethanol also specifically prevents NSC renewal by interfering with miRNA-regulated processes. To address this question, we assessed the regulation of the homeobox transcription factor, Oct4/POU5F1, which is important for maintaining stem cell renewal and pluripotency. The Oct4 family includes a number of transcribed non-protein coding pseudogenes (lncRNA). We hypothesized that these lncRNAs serve as miRNA sponges. Immunoprecipitation studies with the miRNA binding protein, Ago-2, showed that several Oct4 pseudogenes bind miRNAs in both the nucleus and cytoplasm of NSCs, supporting their role as miRNA sponges. Ethanol exposure resulted in a decrease in expression of Ago2-binding pseudogene transcripts and also decreased expression of Oct4 mRNA and protein. These data suggest that lncRNA-miRNAs interaction may protect pluripotency factors and facilitate NSC renewal. These data also advance a novel mechanism for ethanol teratology in that ethanol exposure may disrupt lncRNA-mediated protection resulting in miRNA-mediated loss of renewal capacity in fetal NSCs

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### **38. Modeling and Inference of the MAP Kinase Cascade in Plant Defense Signaling**

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In order to compensate for their immobility, plants have developed multiple layers of defense to protect themselves from different types of infectious agents such as fungi, viruses, bacteria and oomycetes. But a number of these pathogens have co-evolved with plants and developed mechanisms to render plant defense systems ineffective. This has motivated the need for development of strategies for improving plant disease resistance to harmful pathogens. The Mitogen Activated Protein Kinase (MAPK) pathways are highly conserved regulators of growth, proliferation and stress response in all multi-cellular organisms. Recent plant biological studies have implicated the MAPK cascade as the point of convergence of various biotic and abiotic stress stimuli, and suggested the targeting of specific components of the cascade for manipulating plant defense responses. We utilize a Bayesian Network based approach which incorporates prior biological information for modeling and inference of the MAP Kinase cascade.

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## 39. Modeling Lymph Flow and Fluid Exchange with Blood Vessels in Lymph Nodes

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Lymph nodes (LNs) are positioned strategically throughout the body as critical mediators of lymph filtration and immune response. Lymph carries cytokines, antigens, and cells to the downstream LNs, and their effective delivery to the correct location within the LN directly impacts the quality and quantity of immune response. Despite the importance of this system, the flow patterns in LN have never been quantified, in part because experimental characterization is so difficult. To achieve a more quantitative knowledge base of LN flow, a computational flow model has been developed based on the mouse popliteal LN, allowing for a parameter sensitivity analysis to identify the important system characteristics. This model suggests that about 90% of the lymph takes a peripheral path via the subcapsular and medullary sinuses, while fluid perfusing deeper into the paracortex is sequestered by parenchymal blood vessels. Fluid absorption by these blood vessels under baseline conditions was driven mainly by oncotic pressure differences between lymph and blood, although the magnitude of fluid transfer is highly dependent on blood vessel surface area. We also predict that the hydraulic conductivity of the medulla, a parameter that has never been experimentally measured, should be at least three orders of magnitude larger than that of the paracortex to ensure physiologic pressures across the node. These results suggest that structural changes in the LN microenvironment, as well as changes in inflow/outflow conditions, dramatically alter the distribution of lymph, cytokines, antigen and cells within the LN, with great potential for modulating immune response.

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## 40. A multi-channel microfluidic system for modelling the blood-brain barrier (BBB)

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Blood–brain barrier (BBB) is an extraordinarily selective permeability barrier which impedes influx of most drug compounds from blood to the central nervous system (CNS). In order to develop effective CNS drugs, it is necessary to do the functionality study about the BBB to overcome the major obstacle such as not allowing large molecule drugs to pass through. Thus, adequate in vitro models need to be developed so that we can characterize the penetration properties of drug candidates into the CNS. Here, we present a multi-channel microchip capable of physically modelling the structure of the BBB consisting of endothelial cells, which can form tight junctions among cells with showing an extremely high electrical resistivity. This chip consists of 4 x 4 patterns of intersecting microfluidic channels, which are separately assembled with porous membrane as the central figure. Since the interface cultured with endothelial cells has the porous structure, various biomolecules in media can pass through the interface between intersecting microfluidic channels. However, when the endothelial cells form tight junctions, most large biomolecules cannot pass through it anymore. Due to the structure of multi-intersecting microfluidic channels, this chip enables to conduct perfusion culture mimicking blood vessels and investigate the permeability of drug compounds. Using the electrode arrays patterned on the substrate that each microfluidic channel layer is attached on, we can monitor the trans-endothelial permeability in real time by measuring trans-endothelial electrical resistance (TEER). In this regard, this microchip may serve as a powerful tool not only for investigating factors that control the BBB functions, but also for applying to develop drug compounds against brain diseases.

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#### **41. Next Generation Condom-are we there yet?**

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Several antioxidants are used as stimulants to increase sexual pleasure and intimacy in many countries. The constant search for flavonoid (natural plant based) antioxidants is still attractive because they are readily accessible, affordable, and less toxic. On the other hand if individuals only consume these products without using a condom (a frequent complaint with male condoms is the decreased pleasure), it might result in unintended pregnancy or the transmission of sexual transmitted infections (STIs). So the next logical question is “how should we deliver the antioxidant in the genital tissue in conjunction with condom?” We have developed a hydrogel which can be stretched up to over 30 times its length at low thickness. The hydrated and soft nature of these hydrogels ensures that a condom of this material would be naturally well lubricated. This would also simulate the vaginal environment in a superior manner as compared to latex condoms that cause a reduction of tactile sensation and attenuate heat transduction. Based on these scientific evidence and our preliminary data, we expect to create an antioxidant embedded hydropolymer as a next generation condom.

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#### **42. Obtain polyclonal antigen-specific chicken IgY within less than 2 weeks after a single immunization**

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For over five years, our group has worked on the development of *in vivo* antibody-guided vaccines against various chicken pathogens. The central tenet of this concept is the *in vivo* targeting of endocytotic receptors expressed on antigen-presenting cells (APCs; dendritic cells, macrophages and B-cells), such as CD40 and CD205, with the intention of enhancing immunogenicity of protective epitopes in various ways. For instance, a 17-mer peptide induced a significant antibody response within a week after administration. The most remarkable observation during these experiments was that both *IgY* (a chicken's IgG) and *sIgA* were produced *within a week* after s.c., oral, oculo-nasal, and cloacal administration, unlike what one would expect in a primary immune response. This can only be explained by accepting that ligation of the APC's CD40 receptor by the targeting antibody triggers downstream signaling resulting in surprisingly fast isotype switch from IgM/IgD to IgG and/or IgA. A corollary of this work offers an opportunity for any researcher who uses primary antibodies in his/her research to obtain substantial amounts of polyclonal IgY in less than two weeks, regardless of whether the antigen of interest is a peptide, protein, carbohydrate, nucleic acid or lipid. This avenue may result in substantial time gain, e.g. during “quick” generation of crucial preliminary data underpinning a proposal.

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### **43. Orally-delivered, tetrahydrobiopterin-containing nanoparticles targeting gastrointestinal lymphatics modulate nitric oxide synthesis by aortic endothelial cells in diabetic rats**

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A deficiency of tetrahydrobiopterin (BH4), a critical cofactor for endothelial cell (EC) nitric oxide (NO) synthase, may be a common basis for vascular dysfunction in many diseases. Our goal was to modulate BH4 levels in ECs to increase NO bioavailability and improve vascular function in diabetes. We previously showed that BH4 encapsulated in poly(D,L-lactide-co-glycolide) nanoparticles (NPs) and injected intravenously significantly improved vascular function in diabetic rats. In the current study, we synthesized solid lipid NPs (SLNPs) containing BH4. Our objective was to allow oral delivery of BH4 in a form that would show preferential uptake via the gastrointestinal lymphatic lacteals, bypassing the portal circulation and delivering BH4 directly to the peripheral circulation to increase circulating levels of this critical cofactor. SLNPs were administered via oral gavage once per day for two days (SLNPs) and endothelium-dependent vessel relaxation assessed 24 hours later by measuring the response to increasing concentrations of acetylcholine. Lymphatic uptake of orally delivered nanoparticles was verified by sampling mesenteric lymph. BH4 levels were determined using a reversed phase high-performance liquid chromatography method. Orally delivered BH4-loaded SLNPs significantly increased BH4 levels in coronary endothelial cells, compared to coronary endothelial cells taken from non-treated diabetic rats. Importantly, BH4-loaded SLNPs delivered by gavage increased vasodilatory responses of aortic rings taken from diabetic rats, compared to SLNPs loaded with water. These data support the feasibility of using oral NPs to deliver therapeutic agents to ECs in the general circulation via the lymphatic system to attenuate dysfunction associated with vascular disease.

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### **44. Quantitative Analysis of Peripheral membrane Protein Binding Kinetics**

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The attachment of peripheral membrane proteins (PMP) to cellular surfaces regulates most of the cell signaling processes and other essential cellular processes. Quantitative analysis of PMP retention on cellular membranes is essential to determine the rate of these biochemical reactions. We have developed a nanocube sensor coupled with complex reaction analysis to quantitatively explore the PMP binding kinetics. The nanocube sensor is surrounded by lipid bilayers that possess the same physical and chemical properties as cell membranes. This biomimetic surface then enables the label-free detection of PMP bindings by observing the absorption spectra shift of localized surface plasmon resonance (LSPR) peak. This biosensor works with standard laboratory plate reader for high-throughput binding kinetic analysis. The simple protocol ("mix-and-then-detect") allows any end users performing the analysis in their own laboratories. We have successfully explored many essential PMP binding events, including pleckstrin-homology (PH)-lipid and toxin-ganglioside interactions. Moreover, we have introduced complex reaction analysis technique to model the binding cooperativity among PMPs, ligands, and accessory molecules. This sophisticated model-fitting approach can analyze rich kinetic data acquired by the high-throughput nanocube sensor and quantitatively predict the association/dissociation rate of PMP on any desired membrane environment. This technique can also be used to study other biomolecule adhesion, including nucleic acid and pathogen adhesions.

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## 45. Querying networks using context-sensitive random walk

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Cellular functions are carried out by complicated interactions among numerous constituents in the cell rather than a single functional molecule. Recent studies have shown that protein complexes and signaling pathways involved in core cellular processes are often conserved across different species, and that querying networks to detect conserved molecular complexes or pathways can provide effective means of transferring knowledge between species. In this work, we propose a novel network querying algorithm that can effectively scan a large-scale PPI network to detect conserved protein complexes or pathways that are similar to a given query complex or pathway. To this aim, we adopt a context-sensitive random walk (CSRW) model to estimate the functional correspondence between proteins in the query and the target networks, in a way that sensibly integrates the molecular similarity between proteins and their interaction patterns in the respective networks. The functional correspondence scores estimated by the CSRW are used in subsequent stages to gradually zoom into regions in the target network that bear high similarity with the query network, and ultimately, to identify the best match for the query. Based on extensive simulations, we demonstrate that the proposed network querying algorithm can identify conserved protein complexes and pathways with high accuracy and at low computational cost, clearly outperforming existing methods.

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## 46. Regeneration of pulpodentin complex using biophysical cues

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Successful pulpodentin regeneration should include both dentin and pulp tissues in order to fully restore the biological and mechanical functions of the diseased tooth. To date, a rational scaffolding design to regenerate a complete tooth-like dentin-pulp complex has not been achieved. In this study, we aimed to use biophysical cues (scaffolding stiffness) to control dental pulp stem cell (DPSC) fate and regenerate a complete pulpodentin complex. We developed a facile method of integrating the nanofibrous gelatin matrices with two different rigidities into a single scaffold. Our results indicate that DPSCs differentiated into odontoblasts and formed mineralized tissue on high-stiffness structures, whereas pulp-like tissue was regenerated on low-stiffness structures. A complete pulpodentin complex similar to natural pulpodentin was successfully regenerated after subcutaneous implantation of the DPSC/scaffold in nude mice for 4 weeks. A quantitative analysis indicated that the regenerated dentin density was more than 58% of natural human dentin. Histological staining showed a significant amount of ECM formation in the newly formed pulpodentin complex, and a number of blood vessels were observed in the pulp tissue. Taken together, modulation of the scaffolding stiffness is a successful approach to regenerating a complete tooth-like pulpodentin complex.

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## 47. A Role for Serum Amyloid P Glycosylation in Regulating Innate Immunity

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Fibrosing diseases such as pulmonary fibrosis, congestive heart disease, and renal fibrosis result from inappropriate scar tissue formation in internal organs, and are associated with ~45% of deaths in the U.S. Fibrosis is caused by deregulated recruitment and activation of neutrophils, lymphocytes, and monocytes. Two closely related human serum proteins, Serum Amyloid P (SAP) and C-reactive protein (CRP), strongly affect fibrosis. In multiple animal models, and in Phase 1 and Phase 2 clinical trials, SAP suppresses several aspects of the innate immune system to reduce fibrosis, whereas CRP appears to potentiate fibrosis. However, SAP and CRP bind the same Fcγ receptors (FcγR) with similar affinities, and why SAP and CRP have opposing effects is unknown. Here we report that SAP but not CRP binds the receptor DC-SIGN (SIGN-R1) to affect the innate immune system, and that FcγR are not necessary for SAP function. A polycyclic aminothiazole DC-SIGN ligand and anti-DC-SIGN antibodies mimic SAP effects in vitro. In mice, the aminothiazole reduces neutrophil accumulation in a model of acute lung inflammation, and at 0.001 mg/kg alleviates pulmonary fibrosis by increasing levels of the immunosuppressant IL-10. DC-SIGN (SIGN-R1) is present on mouse lung epithelial cells, and SAP and the aminothiazole potentiate IL-10 production from these cells. Our data suggest that SAP activates DC-SIGN to regulate the innate immune system differently from CRP, and that DC-SIGN is a novel target for anti-fibrotics.

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## 48. The Role of NLRC5 Receptor in the Development of TMEV-induced Epilepsy

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Theiler's murine encephalomyelitis virus (TMEV) is a positive-sense single stranded RNA virus that belongs to the *Cardiovirus* genus of the *Picornaviridae* family. It is a natural pathogen of mice and is transmitted through the fecal-oral route. The BeAn strain of TMEV, when injected intracerebrally, in different strains of mice results in different neurological diseases that resemble human multiple sclerosis and epilepsy. In the C57BL/6 mouse strain, TMEV causes acute seizures 3-4 days post infection (dpi) that may later develop into epilepsy. Although, TMEV is cleared from the brains of these mice within 2 weeks post infection by both innate (NK cells, macrophages) and adaptive (cytotoxic T cells) immune cells but the resulting inflammation causes bystander hippocampal damage and increased pro-inflammatory cytokine (such as IL-6, TNF-α, IFN-γ, IFN-β etc.) levels in the brain.

The nucleotide-binding oligomerization domain receptors (or NLRs) are a family of intracellular pathogen sensors expressed by a variety of cell types in the body, including those of both the innate and adaptive immune systems, and activation of these receptors leads to inflammatory cascade. In the current study, we examined the role of the NLRC5 receptor in the development of TMEV-induced epilepsy. NLRC5 acts as a crucial transactivator for MHC class-I and its related genes (CITA). NLRC5 knockout mice (NLRC5 K.O.) were infected with TMEV via intracerebral injection and compared to wild-type C57BL/6 (W.T.) mice for clinical signs of epilepsy. The incidence of acute seizures was around 80% in W.T. C57BL/6 mice while no seizures were observed in the NLRC5 K.O. mice. Mice were sacrificed at 7 dpi and their brains were harvested to study hippocampal lesions using immunohistochemistry and measure cytokine expression using quantitative real time PCR. We found that the clinical signs of epilepsy correlated with the development of increased inflammation in the hippocampus.

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## 49. Sequence-specific incorporation of DNA into protein-based biomaterials

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Following straightforward chemical principles, both protein and DNA fold to unique structures with diverse, yet highly specific molecular functions. Protein and DNA can also be designed to self-associate in pre-defined patterns, creating ordered nanoscale to macroscale materials. Composite materials, composed of protein and DNA, expand the possibilities for molecular functionality and structural design. An ideal protein to interface with these materials would self-assemble and specifically direct DNA binding and orientation. We have previously produced biomaterials composed of a recombinant *Drosophila* transcription factor, Ultrabithorax (Ubx). The Ubx sequence includes a homeodomain, a region that binds its target DNA sequence with very high (picomolar) affinity. We find the homeodomain in Ubx fibers remains capable of sequence-specific DNA binding. Ubx fibers can bind linear, supercoiled, and minivector (strained) DNA. Although binding is reversible, DNA sequences with many binding sites can remain bound for many days. DNAs reorient on the surface of fibers to maximize the number of binding sites in contact with the fibers. Finally, fiber binding protects DNA from degradation. Ubx monomers, pre-bound to DNA, can assemble into fibers with DNA located throughout the fiber. This system provides the necessary level of control to design the bottom-up assembly of functional 3D composite biomaterials.

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## 50. Shared and task-specific muscle synergies during normal walking and slipping

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The CNS controls motor tasks using a low-dimensional modular organization of muscle activation (muscle synergies), and some synergies might be shared across different behaviors. Studying slip, as a leading cause of falling accident is of importance. In this study, EMG data was collected from eight muscles in lower extremity of eleven healthy subjects during normal walking and slip initiation, and four muscle synergies and their activation coefficients were extracted using a nonnegative optimization algorithm. The synergies and their activation coefficients for normal walking and slipping were then compared to each other using correlation coefficients and one sample *t*-test to study if some of the synergies are shared. One sample *t*test results showed that there is one shared synergy ( $r=0.82$ ,  $p=0.002$ ) and one marginally shared synergy ( $r=0.62$ ,  $p=0.024$ ) between these two motor tasks, while the remaining two synergies were considered to be task-specific. The activation coefficients were studied for the first 200 ms after heel contact (as the reaction time to slip is about 200 ms), and they showed a significant correlation for the first shared synergy ( $r=0.84$ ,  $p=0.004$ ) and the second shared synergy ( $r=0.59$ ,  $p=0.026$ ) but not for the task-specific synergies. Moreover, after 200 ms of slip, the activation levels were different, matching with known primary and secondary response of the CNS to slip. Future work will include investigating shared/task-specific synergies based on slip severity, which will help identify factors responsible for severe slips.

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## 51. Simple, Lithography-free Fabrication of Embedded Multi-scale Surface Features via Crystallographic Imprinting

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There is currently a need for straightforward methods to embed physical barrier structures for separations in miniaturized systems. Here we introduce a novel approach to address these needs by exploiting the sensitivity of enzymatic activity to a biodegradable substrate's crystallinity as a vehicle to imprint complex micro- and nano-scale surface features. This is achieved in an enzyme/substrate system involving proteinase K and poly(lactic acid) (PLA), where a strong etch rate selectivity to PLA crystallinity is observed. Scotch tape is used as a mask to localize subsequent enzymatic etching, which is straightforward and can be easily performed in any lab. The PLA crystalline morphology is governed by its thermal history and material properties, enabling the size and density of the imprinted features to be precisely manipulated.

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## 52. A smartphone based PCR assay platform enables rapid point of care diagnostics on a quadcopter drone

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The lack of affordable, rapid, and easy to use diagnostic technologies is one of the most critical issues confronting global public health. We describe new efforts to address this need by coupling an innovative thermocycling system that harnesses natural convection to perform rapid DNA amplification via the polymerase chain reaction (PCR). The unique isothermal design allows PCR to be executed via a single miniature heater, dramatically reducing electrical consumption to a level provided by ordinary 5 V USB sources that power consumer mobile devices (even via solar or hand crank action). The device also integrates a smartphone camera with a dedicated image analysis app for real time fluorescence detection of PCR products. Our instrument is incredibly robust and lightweight, enabling pinpoint deployment of gold standard nucleic acid-based diagnostics to remote field sites using commercially available quadcopter drones. Standard sample preparation is enabled by leveraging the drone's motors as centrifuges via 3D printed snap-on attachments. These advancements make it possible to build a complete DNA analysis system for under \$20 (\$US) that can deliver results in 10 – 15 min. We also demonstrate in-flight analysis, suggesting potential to obtain an unprecedented dynamic picture of outbreaks that can inform improved resource deployment strategies

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### 53. *Sxl* transgene system – to characterize and quantify the activity of a transiently acting promoter in *Drosophila*

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The primary determinant of sexual fate in *Drosophila* is the number of X-chromosomes. XX embryos develop as females, while XY embryos develop as males. The target of the X chromosome signal is *Sex-lethal (Sxl)* gene that lies at the top of the sex determination and dosage compensation. *Sxl* is stably expressed in females via autoregulatory mRNA splicing that occurs as a consequence a brief pulse of transcription from establishment promoter *Sxl<sub>pe</sub>*. Female-specific expression of *Sxl<sub>pe</sub>* requires a two X chromosome dose of the X-signal elements *sisA*, *sc*, *upd* and *runt*. Males fail to express *Sxl<sub>pe</sub>* because they carry on a single dose of the X-signal elements.

We are interested in understanding how this two-fold difference in XSE's concentration is interpreted at *Sxl<sub>pe</sub>* to give an all or nothing response. Understanding regulation of *Sxl<sub>pe</sub>* demands an advanced quantification tool to monitor *Sxl* activity in vivo. *Sxl*-swap allows the monitoring of endogenous *Sxl* transcription, both as nascent transcripts and as mature mRNA. The key feature of this system is that intron sequences are swapped between related species to allow allele-specific detection, by in situ hybridization, of expression from mutant and wild type transgenes side-by-side in every nucleus of the embryo. The transgene system is fully functional, so we can also exploit classical *Drosophila* genetics to monitor the biological effects of engineered *Sxl* mutations. We will exploit this powerful system to characterize the cis interactions of key regulators including the X-signal elements *Sc/Da*, *Upd*, and pioneering transcription factor, *Zelda*, to discover the means by which this sensitive promoter switch operates.

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### 54. Using Boolean Logic Modeling of Gene Regulatory Networks to Exploit the Links between Cancer and Diabetes for Therapeutic Purposes

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The uncontrolled cell proliferation that is characteristically associated with cancer is usually accompanied by alterations in the genome and cell metabolism. Indeed, the phenomenon of cancer cells metabolizing glucose using a less efficient anaerobic process even in the presence of normal oxygen levels, termed the Warburg effect, is currently considered to be one of the hallmarks of cancer. Diabetes, much like cancer, is defined by significant metabolic changes. Recent epidemiological studies have shown that diabetes patients treated with the antidiabetic drug Metformin, have significantly lowered risk of cancer as compared to patients treated with other anti-diabetic drugs. We utilize a Boolean logic model of the pathways commonly mutated in cancer to not only investigate the efficacy of Metformin for cancer therapeutic purposes but also demonstrate how Metformin in concert with other cancer drugs could provide better and less toxic clinical outcomes as compared to using cancer drugs alone.

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## 55. Noncanonical Neurotransmission at the neuromuscular junction

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Large-scale proteomic analyses of heterogeneous CNS preparations, such as isolated CNS synaptic vesicles, have provided a wealth of information related to synapse specific proteins, and synaptic signaling pathways. However, these studies under-represent, or lack entirely, many vesicle and neurotransmitter signaling pathways. In particular the neuromodulatory adrenergic and cholinergic pathways are often under-represented. In order to better understand the molecules and signaling pathways involved at cholinergic synapses, we have focused on the classic preparation of the electric lobe and electric organ of *Torpedo californica*. The abundant macromolecular material available from this preparation has contributed greatly to our understanding of the synthesis, storage, release and receptor activation of the neurotransmitter acetylcholine. Many lines of evidence support the idea that in addition to acetylcholine, additional neurotransmitters and/or neuromodulators are also released from cholinergic synapses. Utilizing a single-vesicle imaging approach, we have found that individual synaptic vesicles from the electric ray possess neurotransmitter transporters for glutamate, ATP, and acetylcholine. In addition to those transporters, cholinergic synaptic vesicles from the electric ray also possess an orphaned transporter from the bile acid transport family. We are now expanding our research to identify the molecules transported by the vesicular SLC10A4 transporter using multiple biochemical approaches and a mouse model.

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