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**Accelerating Monte Carlo Simulation of Protein-Protein Interaction**

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Protein-protein binding is an essential phenomenon in molecular-level biology including cell signaling and regulation of enzyme activity. The binding and unbinding of the proteins can be studied by Monte Carlo simulation. At each step of the simulation, one protein can either be translated or rotated relative to the other. The energy of the new configuration can then be compared to that of the previous configuration to determine whether the system should return to the previous configuration or stay in the new configuration. These energy calculations are computationally costly, but computing the energy of the system in a six-dimensional grid of configurations before the simulation is run can mitigate this cost. Monte Carlo simulation has now been run using a pre-computed clash table and a table of energies due to electrostatic interactions. These simulations illustrate the feasibility of accelerating Monte Carlo simulation using pre-computed tables.

3

**Biophysical characterization of a G-Quadruplex within p250GAP mRNA and of its interactions with the Fragile X Mental Retardation Protein**

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p250GAP is a messenger RNA encoding for Rho GTPase-activating protein 32 that affects the plasticity of dendrites in the nervous system. It is predicted that p250GAP mRNA is a target of the Fragile X mental retardation protein (FMRP), whose absence leads to fragile X syndrome, the most common form of inherited mental impairment. FMRP, an RNA binding protein, uses its arginine-glycine-glycine (RGG box) domain to bind with high specificity and affinity to secondary RNA structures named G-quadruplexes. We identified a guanine rich stretch in the p250GAP that has a high probability to form a G-quadruplex structure and used various biophysical methods such as nuclear magnetic resonance spectroscopy, circular dichroism spectroscopy, UV spectroscopy and native gel electrophoresis to prove the existence and characterize folding of a G-quadruplex structure within p250GAP mRNA. Additionally, we have analyzed the interactions of this G-quadruplex structure with the FMRP RGG box.

2

**Accuracy of Personal Care Provider's (PCP's) Self-Report on Screening Procedures of Adolescents on Substance Use and Mental Disorders**

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Recently, marijuana has increased drastically, seen heavily in people in their late teens and twenties. Prevention begins by regular screening from Primary Care Providers (PCPs) before experimentation occurs in youths. This study compares PCP self-reported screening procedures with self-reports of their adolescents receiving care. It is hypothesized that PCPs overrate their screening implementation, especially for marijuana use, but those at high risk for drug abuse, will be more likely to be screened. Practitioners recruited through PittNet were surveyed on their Screening Brief Intervention Referral For Treatment (SBIRT) practices. Participating adolescents were assessed for substance abuse risk with Computer Assisted Decision Support (CADS) tablets and were given post-visit assessments to verify screens fulfilled during visit. This project is currently taking place; collected data shows Mental Disorders being screened at early ages as PCPs reported, but substance screenings given to adolescents older ( by 2-3 years) than reported.

4

**The Investigation of the Presence of Organic GSR on SEM Stubs**

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A Scanning Electron Microscope (SEM) stub was looked over to identify both inorganic and organic gunshot residue (GSR) to ensure that the stub contained GSR and not particles found in the environment. To find the inorganic GSR particles, the SEM was used. The Liquid Chromatography- Mass Spectrometer (LC-MS/MS) was used to find organic GSR. The limit of detection of the components of GSR- Akardite II solid, Methyl Centralite, Ethyl Centralite, and Single Based Gun Surveillance – was detected using the LC-MS/MS. After finding the limit of detection, an extraction method was developed to extract organic GSR from an SEM stub. The presence of organic GSR was tested for on a cloth with GSR particles found on it, an SEM stub with GSR particles on it, and an SEM stub with GSR, which was scanned on the SEM. Findings from the cloth, SEM stub, and the scanned SEM stub were compared.

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**Characterization of Steroid Sulfatase in *Xenopus laevis*.**

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Steroid sulfatase (STS) removes the sulfate group from inactive steroids, activating the hormone. Amphibians have sulfated steroids; however, no reports exist on STS in amphibians. We sought to determine if STS exists in an amphibian, *Xenopus laevis*. Tissue homogenates were assayed for STS activity, using an  $^3\text{H-E}_1\text{S}$  conversion assay. STS activity was present in all tissues, with testes and intestine having the highest levels. Microsomes and cytosol were prepared from female liver. An  $^3\text{H-E}_1\text{S}$  conversion assay of microsomes and cytosol was run in the presence and absence of STS inhibitors, EMATE and Coumate. STS activities of both were eliminated by the inhibitors, indicating the activity measured is STS. STS activity was temperature dependent, with lower activity at 4°C. Lineweaver-Burke analysis of microsomes indicated the Km for *Xenopus laevis* STS to be higher than mammals. Our data indicate that *Xenopus laevis* tissues possess STS and that this enzyme varies between tissues.

7

**Expression, Purification, and Preliminary Kinetic Study of the Periplasmic Nitrate Reductase from *Campylobacter jejuni***

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The reduction of nitrate has been shown to play an important role in the infectious cycle of the microaerophilic bacterium, *Campylobacter jejuni*. *C. jejuni* serves as the leading cause of gastroenteritis in the United States and can, in rare cases, lead to Guillain-Barre Syndrome. While other organisms have a variety of nitrate reducing enzymes, to date, only a single nitrate reductase has been identified in *C. jejuni*. Nap, the periplasmic nitrate reductase, is a dissimilatory nitrate reductase that catalyzes the reduction of nitrate to nitrite via a molybdopterin based cofactor (known as the molybdenum cofactor or MoCo). In this work, we have successfully overexpressed the *C. jejuni* catalytic subunit (NapA) in *Escherichia coli* and purified the enzyme via Immobilized Metal Affinity Chromatography. Furthermore, preliminary kinetic studies have been attempted in order to determine the functionality and properties of the produced enzyme.

6

**Protein-small molecule interactions between a Vitamin D agonist (2MD) and the Vitamin D receptor by using alanine scanning computational methods**

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Identifying the importance of residues at protein-protein interfaces and within a binding pocket is a challenge experimentally and computationally. A method known as alanine scanning has been used to analyze protein-protein interactions and protein-small molecule interactions. The method works by mutating selected residues from the wild-type to alanine and then observing the impact. The interactions between 2MD, a molecule similar to Vitamin D, and the Vitamin D receptor (VDR) have been experimentally studied. The thermodynamic cycle – perturbation method is used to simulate alanine scanning and computationally determine the important residues in the Vitamin D receptor binding pocket. The results from these calculations will be presented in this poster.

8

**Correlation between band gap and electronegativity of substituted atoms in the TiO<sub>2</sub> crystalline structure**

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The electronic structure of TiO<sub>2</sub> has been extensively studied through a variety of experimental and computational methods. Its properties range from thin film photovoltaic cells to optics. TiO<sub>2</sub> provides an excellent model to study computationally due to the wealth of experimental data and its inexpensive computational cost. Our hypothesis is that the electronegativity of a substituent changes the band gap of crystalline TiO<sub>2</sub>. Atoms of different electronegativities were selected for substitution into the three polymorphic forms of TiO<sub>2</sub>, which are rutile, anatase, and brookite. Our computational approach utilizes the linearized-augmented plane-wave approach of density functional theory in the WIEN2k computational software, and includes the incorporation of the modified Becke-Johnson potential, to determine the band gap and density of states for each case. Initial results showed that fluorine substitution in a 2x2x2 rutile supercell resulted in a slight decrease in the band gap.

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**Conformational Changes of the Human Serotonin Transporter**

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Duquesne University

One important ability of human cells is to regulate the passage molecules and ions across the cell membrane. While there are hundreds of transporters in the body, this research focuses on the human serotonin transporter or hSERT. The hSERT is a highly allosteric protein, involved in the re-uptake of serotonin. A better understanding of the factors underlying the transporter's allostery could lead to advancements in drugs that affect the hSERT. Three plasmids were chosen for experimentation: X8C, 109A, and 109A Bluescript. Each plasmid contained hSERT mutants void of some or all accessible cysteine. The plasmids were transformed into competent human cells, amplified, and extracted for purification of the DNA. The plasmids were then mutated to introduce the 109A cysteine. Once mutants are successfully created, the protein will be enriched in different conformational states from which structural data will be ascertained through the use of cross-linking and mass spectrometry.

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**Computational modeling of spatial heterogeneity in growing tumors**

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As tumors age they gain a large degree of genetic, metabolic, and other types of cellular heterogeneity. This heterogeneity raises the risk that one cell type will resist the effects of cancer therapies, leading to recurrence and high probability of patient death. Furthermore, subpopulations are often spatially segregated throughout tumors, resulting in incomplete biopsy analyses. And yet, the origin and development of such intratumor heterogeneity are not understood. We have constructed an avascular tumor growth model using the Cellular Potts Model to understand some of the factors that affect spatial patterns of heterogeneity in primary tumor growth. After confirming that our *in silico* tumors display the expected exponential growth at short times, we explore the effects of cellular growth and mutation rates on tumor heterogeneity. Our simulations illustrate that different spatial patterns and morphologies arise from the subtle interplay between adhesion energies and probability of cell type mutation.

10

**De Novo Synthesis of a Molybdopterin Analogue**

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Duquesne University

The molybdenum cofactor (Moco) forms the active site of many enzymes, including sulfite oxidase. It consists of a molybdenum center bound to one or two organic chelates, which are commonly known as molybdopterin. *De novo* synthesis of Moco is of significant interest due to the occurrence of Molybdenum Cofactor Deficiency (MoCD), a rare but fatal genetic disorder. Moco is formed by four conjoined cyclic units: a pyrimidine unit, a pyrazine, a pyran, and a dithiolene. This synthetic approach focuses on synthesis of Moco by first forming a precursor that includes the pyran and dithiolene units of the structure (6-hydroxy-4-(hydroxymethyl)-2-thioxo-4H-[1,3]dithiolo[4,5-c]pyran-7(6H)-one), then adding the pyrazine and pyrimidine units by reacting this precursor with 4-Hydroxy-2,5,6-triaminopyrimidine, a commercially available reagent, in a Gabriel-Isay condensation. Future research could potentially yield a biologically active form of Moco.

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**NR2B mRNA as a potential target of the Fragile X Mental Retardation Protein**

Underwood, Ayana; Stefanovic, Snezana; Katrancha, Sara; Zhang, Yang; Mihailescu, M. Rita  
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Duquesne University

Fragile X Syndrome, an inherited disorder caused by the expansion of CGG repeats within the fragile X mental retardation (*FMR1*) gene, is the most common form of mental impairment. The numerous CGG repeats silence the *FMR1* gene on the X chromosome, leading to the loss of the Fragile X Mental Retardation Protein (FMRP). NR2B mRNA, a potential target of FMRP, encodes for a subunit of the glutamate receptor NMDA, which is involved in synaptic plasticity and memory retention. In this study we postulated that NR2B mRNA adopts a G-quadruplex secondary structure that is recognized specifically by the RGG box of FMRP. Several biophysical techniques such as NMR spectroscopy, CD spectroscopy, UV spectroscopy and non-denaturing polyacrylamide gel electrophoresis were used to determine if a G-quadruplex is formed in the 3'-untranslated region of NR2B mRNA. An electromobility shift assay was employed to examine interaction of this region with the FMRP RGG box.

**13****Geometry optimization and vibrational analysis of a Rieske-type iron-sulfur cluster model**

Benjamin R. Jagger, Bao-Linh Nguyen, and Ralph A. Wheeler

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Duquesne University

The Rieske-type iron-sulfur (Fe-S) cluster is a catalytic subunit of the mitochondrial cytochrome bc<sub>1</sub> complex. The cluster accepts an electron and a proton when the mobile electron carrier, ubiquinol, is oxidized at the Qi site. One of the two histidine ligands of this cluster (His141 and His161) is believed to act as the acceptor, therefore, the mechanism of proton coupled electron transfer is dependent on the protonation states of these ligands. To better understand this electron transfer mechanism, a detailed description of the Fe-S cluster is necessary. Two models for the oxidized Rieske Fe-S cluster were created: one with both histidine ligands protonated and one with a single ligand deprotonated. The geometries of both models were optimized and vibrational frequencies were calculated. The vibrational spectra were then compared to experimental values as well as to previous computational work.

**15****Replication of Genetic Associations with Dental Caries**

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University of Pittsburgh

Dental caries is the most widespread disease in children and a major public health concern due to its increasing incidence, health/social co-morbidities, and socio-demographic disparities. We performed a follow-up study to verify/replicate previously nominated candidate genes affecting tooth decay in primary dentition. Affection status was defined as 1 or more teeth with evidence of decay based on intra-oral examination. Genes nominated were tested for association in children and adults. While no associations met strict criteria for genome-wide significance ( $p < 10E-7$ ), several loci (ACTN2, TFIP11, PLUNC, and EPHA7) with plausible biological roles in dental caries exhibited suggestive evidence for association. This study reinforces the complexity of dental caries, suggesting that numerous loci, mostly having small effects, are involved in cariogenesis. Further investigation into these suggestive loci may highlight biological mechanisms and/or pathways leading to a fuller understanding of the genetic risks for dental caries.

**14****Nanotoxicity, the Other Side of Nanotechnology**

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University of Pittsburgh

Functional nanoparticles have widespread use in consumer products such as cosmetics, clothing, and electronics. However, increasing evidence demonstrates that nanoparticles show elevated toxicity compared to their bulk analogs, thus calling for a reliable method to detect nanotoxicity. Silica nanoparticles are accepted as being non-toxic, prompting the hypothesis that embedding metal nanoparticles in silica could reduce toxicity while still providing accessibility of the metal nanoparticle, therefore maintaining the functionality of the embedded nanoparticle. Three different nickel embedded silica complex nanostructures are investigated in this study via *in vitro* human embryonic stem cells (hESCs) and fibroblast cultures. Fibroblasts are an established model for assessing nanotoxicity using cell viability, morphology and proliferation. hESCs are a new and emerging human developmental model and they also allow for assessment of differentiation. Our initial tests have been completed using nickel salt and all three nanoparticles using fibroblasts and more comprehensive tests with hESCs are in progress.

**16****Inflammatory Pain and the Central Amygdala: Immunohistochemical Analysis of Pain Markers**

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Duquesne University and Swarthmore College

The central nucleus of the amygdala (CeA) integrates information about pain and influences other pain centers in the brain. Phosphorylated extracellular signal-regulated kinase (pERK) is a signaling molecule activated in the right CeA three hours post formalin injection, a common model of inflammatory pain in mice. This activation of pERK is necessary for prolonged peripheral behavioral hypersensitivity after formalin injection. The goal of the present study was to investigate which molecules in the CeA co-localize with pERK, and therefore are somehow involved in inflammatory pain signaling in the brain. Specifically, immunohistochemistry was used to assess GABA, substance P, and mGluR5 (metabotropic glutamate receptor 5) localization three hours post-4% formalin paw injection. Results using epifluorescence and confocal microscopy will be presented at the symposium.

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**Signaling interactions in regulation of autophagic protein degradation**

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Abnormal muscle protein degradation producing muscle wasting and atrophy is seen in numerous disease states, starvation, or prolonged spaceflight. Low-level autophagic degradation maintains the dynamic balance of proteins in healthy muscle, but an increase in autophagic signaling may promote abnormal degradation of bulk proteins. Autophagic degradation is promoted by autocrine signaling through the Ras-Raf-MEK-MAP kinase cascade or deficient insulin signal. My research explores signaling interactions between proteins downstream of MAP kinase (MAPK), the protein kinases RSK and TOR. RSK activation is required for cytosolic protein degradation via autophagy, while TOR opposes it. It remains to be discovered if *both* increase in RSK signal and inhibition of TOR are required to allow autophagy or if either is sufficient. To differentiate between these interaction models, I used transgenic strains of *C. elegans* deficient in MAPK and RNAi knockdown of TOR, assessed by the loss of cytosolic LacZ reporter protein in muscle.

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**Molecular biological and Biochemical techniques used in investigating periplasmic nitrate reductase**

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The molybdoenzyme, periplasmic nitrate reductase (NapA), plays an important role in the vitality of the pathogenic bacterium, *Campylobacter jejuni*. *C. jejuni* is a microaerophilic bacterium that grows anaerobically by utilizing nitrate as an electron acceptor. Infection by *C. jejuni* is one of the most common causes of gastroenteritis in the United States. NapA cloned from *C. jejuni* has successfully been overexpressed in *E. coli*. Produced protein has also been successfully isolated and purified for further studies using a reduced methyl viologen assay. Other techniques, such as Immobilized Metal Affinity Chromatography (IMAC) and Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE), have been used to further study this enzyme. IMAC was performed to retain the protein in a column that contained immobilized nickel ions for the purification of the polyhistidine tag. SDS-PAGE was done to see if NapA was present in the protein sample.

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**Chemical Vapor Deposition Growth of Large Domain Graphene**

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Graphene is a single layer of graphite, a one atom thick honeycomb lattice of carbon atoms. Despite being only one atom thick, a sheet of graphene can cover many square centimeters and is strong enough to be manipulated and processed to make electronic and mechanical devices. The D'Urso group uses graphene to make nanoelectromechanical devices, which are micrometer-scale bridges of graphene which span across grooves cut in a substrate. This project is about improving the methods that the D'Urso group uses to grow graphene for use in their devices, since the quality of the graphene is critical for the performance of the devices. I am working on modeling the methane gas concentration gradient that is being used to grow the large domain graphene, and an apparatus to control the gradient.

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**The synthesis and characterization of nanocrystalline quaternary sulfide semiconductors**Simmons, Cheyenne; Brant, Jacilynn; Aitken, Jennifer  
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Duquesne University

The purpose of this project was to recreate the synthesis of  $\text{Cu}_2\text{CoSnS}_4$  nanoparticles, which was reported in a paper published by Changhua An, et al. The synthesis of new nanoparticles with other structurally and chemically related compounds which have not been previously prepared in nanoparticle form. After synthesizing the nanoparticles were analyzed using Scanning Electron Microscopy (SEM) along with the Energy Dispersive Spectroscopy (EDS). With the Scanning Electron Microscope, the size and shape of said nanoparticles were characterized. Energy Dispersive Spectroscopy was used to determine the composition and formula of the new nanoparticles to see if the intended elements were incorporated into the nanoparticles. To further characterize, X-ray powder diffraction was used to identify the structures of crystals made, and UV-Vis-NIR Spectroscopy to determine the band gaps.

**21****HPLC Method Development for the Separation of Niacin and Niacinamide**

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Niacin is a water soluble B vitamin commonly used as a dietary supplement in solid dosage forms. At high temperatures, in acidic or alkaline solutions, and *in vivo*, niacin undergoes hydrolysis to yield niacinamide. Because these two compounds differ functionally, developing a method to quantify the amounts of each compound in a sample is of interest. A high performance liquid chromatography (HPLC) method was developed to separate these two components. Mobile phase composition, flow rate, and detector wavelength were altered to achieve optimal peak resolution of both niacin and niacinamide. The experiments were carried out on a Waters 2790 apparatus using a C18 column. Maximum peak resolution was achieved with a mobile phase composition of 82 parts sodium 1-hexane-sulfonate in water, 10 parts methanol, 4 parts acetonitrile, and 4 parts glacial acetic acid with a flow rate of 0.50 mL/min and a detector wavelength of 260 nm.

**23****G-quadruplex Determination and Characterization of the low density lipoprotein receptor related 1 precursor**

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The pathogenesis of Alzheimer's disease, a frequently occurring disease among the growing elderly population, is related to the accumulation of a peptide (amyloid- $\beta$ ), which forms aggregates in the brain. The low-density lipoprotein receptor related 1 precursor (LRP1) protein has been implicated in the clearance of the amyloid- $\beta$ , as well as in the pathogenesis of tumor metastasis. Thus, the expression of the LRP1 protein has to be finely regulated. LRP1 mRNA contains in its 5'-untranslated region a G rich sequence which has a high probability to adopt a secondary structure called G-quadruplex, which has been shown to be involved in the regulation of translation. In this study we employed different biophysical methods to determine if LRP1 mRNA adopts a G-quadruplex structure and to characterize its fold.

**22****Structure and Function of Mutant Serotonin Transporters**

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Duquesne University

Serotonin is a neurotransmitter that is released in the brain to convey feelings of happiness and safety. The serotonin transporter (SERT) is a sodium dependent transporter that transports serotonin across biological membranes in neurons. This transporter has one external reactive cysteine residue at the 109 position. Mutant SERTs without cysteines were acquired as a template so that single point mutations could be made. Primers were employed in polymerase chain reaction to create point mutations that insert cysteine at 310 or 406 in SERT, followed by sequencing. Further studies will be conducted using thiol-reactive crosslinkers to label the cysteines. Photoactivation of the attached benzophenone then forms non-specific crosslinks to nearby residues in SERT. Analysis of the crosslinked sites will be conducted using mass spectrometry to observe crosslinker mass shifting. Knowing of the conformation of SERT would lead to more specific binding targets for future drugs for depression and other mood disorders.

**24****Drug Design to Reduce Inflammatory Response of Macrophages Aimed at Spi-1/DNA Interaction**

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The transcription factor Spi-1/PU.1 (Spi-1) is required for expression of the macrophage cytokine, IL-1 $\beta$ . This protein is a potent mediator of inflammation in health and disease. Spi-1 possesses a DNA binding domain containing a recognition  $\alpha$ -helix, which also serves as a NLS and protein-protein interaction element. We used molecular dynamics (MD) as a means of evaluating the Spi-1 x-ray structure for the potential of binding an inhibitory small molecule that could be a model for drug design. Structure preparation and solvation of the protein was conducted in Molecular Operating Environment (MOE) before the protein was relaxed via dynamics simulations in NAMD2.8. Analysis of the behavior of the protein and the flexibility of individual amino acid residues including Arg<sup>232</sup> and Arg<sup>235</sup>, which are exposed on the recognition  $\alpha$ -helix, suggested a starting point for optimal small molecule design. Further results of the MD and small molecule development are described on the poster.

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**Traveling Waves in a Wilson-Cowan Model of Cortex**

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Cortical slow oscillations play a significant role in activating subcortical structures and determining internal brain states. Recent investigations have characterized the spread of activity across and between the six layers of neocortex as a wave of neuronal activation, and have suggested that infragranular layer 5 is primarily responsible for initiating and maintaining widespread cortical activity whereas supragranular layers (layer 2/3) are subsidiary. We propose a model of interacting excitatory and inhibitory neural fields in layers 2/3 and 5 that illustrates the existence, stability, and properties of these waves. Our analysis demonstrates numerically and analytically that small amplitude traveling waves can be initiated in either cortical layer but require the contribution of layer 5. We consider the dynamics resulting from varying vertical and laminar connectivity parameters and find that the dominance of layer 5 can be attributed to increased local connectivity and stronger vertical projections originating in this layer.

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**Small molecule-protein interactions involving Substrate 4 of the Vitamin D receptor using computational alanine scanning**

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Vitamin D is an essential enzyme that assists in the mineralization of bone, bone growth and bone remodeling. Experiments have been done to investigate the allosteric effects of ligands in the function of nuclear receptors using a method known as alanine scanning. Alanine scanning involves the mutation of binding pocket amino acids to alanine so that the importance of that amino acid in the binding pocket is determined. The free energy perturbation method in conjunction with the thermodynamic cycle is used to perform computational alanine screening. Using Substrate 4 in the vitamin D receptor the computational alanine scanning method can be validated. Preliminary results of the computational alanine scanning method will be presented in the poster.

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**A Computational Approach for Identifying a Transcriptional Network Used in the Development of a Novel Appendage**Lynch<sup>1\*</sup>, Danny; Rebeiz<sup>2</sup>, Mark

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The posterior lobe, an appendage that protrudes from *Drosophila melanogaster's* male genitalia, is a recently derived novelty. Two cell-signaling pathways, JAK-STAT and Notch, have been discovered to underlie the development of this structure, suggesting that complex transcriptional networks have been co-opted to drive the evolution of this appendage. In order to identify the downstream members of this novel network, we modified the SCORE (Site Clustering Over Random Expectation) method that identifies transcriptional cis-regulatory modules. This computational method found the number of binding sites of the Suppressor of Hairless protein and JAK-STAT pathways in GAL4. We then created a new algorithm to parse through these potential enhancers to count the concentration of binding sites. Next, we screened several of our highest hits for expression in the developing male genitalia using the Gal4 – UAS system. This approach has the potential to assist in identifying any transcriptional network that is deployed during development. This research was funded by National Science Foundation and the Department of Defense Grant # 1263020

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**Efficient Employment of Novel Binuclear Copper Complexes in Atom Transfer Radical Addition (ATRA) via Catalyst Regeneration Technique**

Evan Perez, Aman Kaur and Tomislav Pintauer

Atom transfer radical addition (ATRA) is fundamental and versatile organic process for C-C bond formation via addition of various alkyl halides to double bond of alkene catalyzed by copper complexes. The usage of catalyst regeneration technique has dramatically decreased the amount of the catalyst from to 5-30 mol% to 0.003 mol% with the aid of reducing agents such as 2,2'-Azobis(2-methylpropionitrile) AIBN and ascorbic acid. Using this technique, two novel binuclear copper complexes have successfully employed  $[Cu^{II}(TPEN)X][X]$ , and  $[Cu^{II}(TPEN^*)X][X]$  (X=Cl,Br) (TPEN = Tetrakis(pyridin-3-ylmethyl)ethane-1,2-diamine; TPEN\* = Tetrakis((5-methoxy-4,6-dimethylpyridin-3-yl)methyl)ethane-1,2-diamine) on various alkenes (1-octene, acrylonitrile, methyl acrylate, methyl methacrylate) in the presence of ascorbic acid as reducing agent.

**29****Understanding the mechanics behind cell morphodynamics using model simulations**

Harris, Demetrius; Miller, Callie; Davidson, Lance  
Bioengineering  
University of Pittsburgh

The processes underlying the mechanics of cell and tissue morphodynamics are difficult to understand, because of this, we simulated cytoskeletal dynamics within a hexagon-shaped subunit of the cell cortex. Understanding how the cytoskeleton controls cell shape is essential in interpreting how small changes in molecular processes can result in developmental defects; e.g. a hole in the heart or cleft lip. Our simulations represent rotating and sliding F-actin filaments within a viscous cytoplasm. Filament movements and morphology depend on the density of actomyosin in the cell subunit, the span of the myosin II minifilament, and attachment/detachment rates of the myosin to F-actin filaments. At present, this model does not include fluid dynamics or vesicle/organelle transport, however, processes such as these may be added in a straightforward manner in future versions. Larger patches of the cortex or combining multiple subunits can simulate the mechanics of entire cells or multicellular arrays.

**31****Quantum dynamics of carboxyphosphate and associated stability**

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Center for Computational Sciences and Department of  
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Duquesne University

It is not possible to isolate or synthesize carboxyphosphate in aqueous solution. However, carboxyphosphate is hypothesized to be stable in nonpolar environments, such as enzymatic pockets. To study the effect of hydrogen bonding on carboxyphosphate, 15 waters were placed around a linear dianion conformation of carboxyphosphate to fulfill the possible hydrogen bond donor and acceptor sites. The M06-2X functional with the 6-31G(d), 6-31+G(d), and aug-cc-pv[D-Q]z basis sets have been used to identify the strength of individual hydrogen bond configurations. Shorter hydrogen bond lengths associated with linear carboxyphosphate indicate stronger interactions with solvent to unfold the pseudo-chair conformation. Quantum dynamics of carboxyphosphate have been carried out using TeraChem with ~100 waters for ~2 ps using the B3LYP/6-31G(d) functional for the pseudo-chair and linear forms of mono and dianionic carboxyphosphate. Elongation of the CO bond length suggests that linear carboxyphosphate will decompose into CO<sub>2</sub> and inorganic phosphate, which is consistent with experimental findings.

**30****Acceptance Studies for 4He(e,e'p)X Reaction up to High Missing Energies and Momenta**

Finton, Drew; Benmokhtar, Dr. Fatiha  
Department of Physics  
Duquesne University

Data collected from the Helium-4 target in Hall A at Thomas Jefferson National Accelerator Facility (TJNAF) in Newport News, Virginia, was analyzed using the object-oriented data analysis software ROOT and used to create Missing Energy Spectra for Missing Momenta ranging from 150 MeV/c to 755 MeV/c for 4He(e,e'p)X reaction channels. Jefferson Lab is a continuous electron beam accelerator facility, and Hall A contains two high resolution spectrometers as well as the cryogenic Helium-4 target. Acceptance cuts were made to six measured variables to remove background noise, and then applied to produce a Missing Energy Spectrum showing two- and three-body break up as well as pion electro-production energy threshold. The missing energy spectra are used to choose between theoretical models for one-, two-, and three-body interactions. Target density was also determined and virial corrections at low temperatures (17 to 20 K) were estimated.

**32****Design and Development of High School Science Laboratories Utilizing Remote-Access Scanning Electron Microscopy**

Janicki, Emily L.; Gault, Joe H.; Rosmus, Kimberly A.;  
Larry, Nolan R.; Aiken, Jennifer A.  
Department of Chemistry and Biochemistry  
Duquesne University

Four laboratory experiments utilizing a scanning electron microscope (SEM) were designed to be implemented into local high school science classrooms following the Pennsylvania State Standards and the Science Keystone Assessment Anchors. SEM is used to observe the surface morphology of a material and to measure sample features such as crystallite size. The students will be introduced to basic techniques of electron and light microscopy with an introductory lab that uses a copper penny. Observing differences between the morphology of salt and sugar (rock candy) as well as differentiating between NaCl and KCl will also be examined. The microscopy experiments were developed to include the following teaching resources: remote access guidelines, pre-lab presentation, teacher and student edition laboratory instructions, and SEM videos and micrographs. This project was funded by the National Science Foundation, Grant No. CHE-0923183.



**33****Search of Genetic Signaling Pathway with the Traveling Salesman Problem**Lorenzo, Eney<sup>1</sup>; Ropelewski, Alexander<sup>2</sup>; Cabrera, Mauricio<sup>1</sup>; Isaza, Clara<sup>3</sup><sup>1</sup>Industrial Engineering Department, University of Puerto Rico, Mayagüez Campus<sup>2</sup>Pittsburgh Supercomputing Center<sup>3</sup>The Autonomus University of Nuevo León

Establishing how a series of potentially important genes might relate to each other is relevant to understand the origin and evolution of illnesses such as cancer. High-throughput biological experiments have played a critical role in providing information in this regard. A special challenge, however, is that of trying to conciliate information from separate microarray experiments to build a potential genetic signaling path. We have developed a method that models this problem in a manner similar to a Traveling Salesman Problem. To keep the method tractable, a preselection of genes obtained using multiple criteria optimization is found first. The genes are laid out in a graph as nodes joined by arcs with linear correlation coefficients. The optimal solution corresponds to the most correlated path or tour. The preliminary results in independent databases show similarities at two pairs of neighboring genes. These are currently under analysis for biological interpretation.

**35****Effects of IFN- $\gamma$  on AMPA and NMDA receptor activation and cell survival**

Gray, Erica; Ganesan, Priya; Kulkarni, Apurva; O'Donnell, Lauren

Mylan School of Pharmacy  
Duquesne University

Interferon-gamma (IFN- $\gamma$ ), a proinflammatory cytokine, is elevated in anti-viral responses, many inflammatory conditions, and brain injury. IFN- $\gamma$  may cause neuroprotection or neurotoxicity through  $\alpha$ -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) and possibly N-methyl-D-aspartate (NMDA) receptors, depending upon which excitotoxins are expressed during inflammation. We hypothesize that IFN- $\gamma$  influences activation of these receptors and ultimately affects cell survival. To test this hypothesis, primary hippocampal neurons were isolated from embryonic day 16 mice and cultured for 5, 10, and 14 days *in vitro*. Neurons were treated with either an AMPA or NMDA receptor agonist and cultured with or without IFN- $\gamma$ . The expression level of active and inactive forms of each receptor was measured by western blot analysis. To evaluate neurotoxicity, cell viability was measured with Cell Titer Glo®. Current studies are addressing whether IFN- $\gamma$  affects AMPA or NMDA receptor activation and whether cell survival is compromised by IFN- $\gamma$  alone or a combination of IFN- $\gamma$  and AMPA/NMDA.

**34****Chemical Immobilization of Bone Morphogenetic Protein 2 on Ceramic Scaffolding Enhances Substrate Bioactivity**Farrell, Drew C.; Romeo, Jared D.; Gawalt, Ellen S.  
Department of Chemistry and Biochemistry  
Duquesne University

Bone morphogenetic protein 2 (BMP-2) is a member of the TGF- $\beta$  growth factor family that exhibits multiple functionalities in the process of bone remodeling. As such, surface modification of intrinsically bioactive ceramic scaffolds with BMP-2 could serve to further enhance the osteogenic effects of the scaffolds at a bone defect site. In this study, calcium aluminate and calcium phosphate scaffolds were modified with BMP-2 by direct surface adsorption or chemical immobilization utilizing an organic linker system. The effect of surface modification was evaluated by human osteoblast growth at Day 4 using scanning electron microscopy. Differentiation of adult mesenchymal stem cells was evaluated on the substrates using an alkaline phosphatase assay and a live/dead cytotoxicity assay. It was shown that the method of biomolecule attachment as well as scaffold composition influenced cell viability and differentiation and that chemically immobilized BMP-2 on HA scaffolds promoted cell viability to the highest degree.

**36****Thermodynamics of Binding Site Water Molecules**James Castiglione, David Koes Ph.D.  
Department of Computational and Systems Biology  
University of Pittsburgh

When a protein and ligand bind, the water molecules inside the protein's binding site are displaced into the bulk solvent. This displacement is thought to greatly contribute to the free energy change of binding. To predict this change, we implemented the grid inhomogeneous solvation theory (GIST) methods developed by Crystal Nguyen et al. These methods divide the active site into a grid of voxels, bin the water molecules from explicit solvent molecular dynamics simulations into the voxels, and use the distribution of the molecules' positions and orientations to calculate the energetic and entropic contributions of each voxel to the region's free energy. Using GIST, we analyzed the 2012 CSAR competition protein targets and our own target proteins, rationalizing the activity of ligands known to bind to them. We also looked to minimize the simulation time required to generate trajectories that produce results substantially equivalent to those from much longer simulations.

**37****Introduction Of Fluorous Lipophilic Diblock Constructs To Stabilize Triphasic Nanoemulsions**Mountain, Gregory<sup>1</sup>; Jelier, Benson<sup>2</sup>; Friesen, Chad<sup>2</sup>, Janjic, Jellena Ph.D.<sup>3\*</sup><sup>1</sup>Department of Chemistry, Saint Francis University, Loretto, Pennsylvania, USA 15940-9704<sup>2</sup> Department of Chemistry, Trinity Western University, Neufeld Science Centre, 7600 Glover Road, Langley, BC V2Y 1Y1, Canada<sup>3</sup> Graduate School of Pharmaceutical Sciences, Duquesne University, Pittsburgh, PA USA 15282-0001

Nano-emulsions have become a promising new formulation platform for drug delivery and imaging. Colloidal stability of these emulsions is important to their efficacy. Recently we designed a novel triphasic system: fluorocarbon (FC) in oil in water nanoemulsions. We report here the introduction of a fluorophilic-lipophilic diblock (FLD) into triphasic nanoemulsions. FLD was incorporated into these nanoemulsions to stabilize the hydrocarbon oil-fluorocarbon oil interphase. The FLD produced an increase in particle stability and a decrease in degradation over time. The ratio of FLD to perfluoropolyether (PFPE), used as the model FC, was optimized using sonication as the emulsification technique. Microfluidization was then used to scale up the optimized nanoemulsion. Using Dynamic Light Scattering Spectroscopy (DLS) nanoemulsion particulates were shown to have increased stability with the addition of the FLD. Temperature and serum stability testing results further support the claim that the addition of the FLD increases colloidal stability.

**39****Neuroprotective Effect of Picied in SH-SY5Y Cells**

Mazzoni, Jessi; Potdar, Sneha; Cavanaugh, Jane; Parmar, Mayur; Allen, Erika; Moonlish, Darlene. Department of Pharmacology, Duquesne University.

Resveratrol, a natural compound, has properties as an antioxidant, anticancer, anti-inflammatory, and neuroprotective agent. However, resveratrol has low bioavailability. Picied, an analog of resveratrol, is found in the same fruits but in higher concentrations than resveratrol. Our research looked into the protective effects of picied *in vitro*. After testing toxicity of high concentrations of dopamine in the neuronal dopamine-like SH-SY5Y cell line, we treated cells with varying concentrations of picied and exposed them to dopamine for 24 hours. Using cell titer glow to measure levels of ATP, which is an indicator of cell viability, we found that ATP levels in picied-treated cells were significantly higher than those exposed to dopamine only. To determine how picied may be involved in neuroprotection of SH-SY5Y cells, we used western blot to see if pro-survival and anti-apoptotic pathways ERK, Bcl-2, or DJ1 are activated in the cells by picied.

**38****Parallel Benchmarking and Performance Profiling of de novo Genome Assembly Algorithms Appropriate for (NGS) Data**

Jan Salomon (University of Puerto Rico Mayaguez) Alex Ropelewski (Pittsburgh Supercomputing Center) Dr. Bienvenido Velez (University of Puerto Rico Mayaguez) Electrical &amp; Computer Engineering Department, University of Puerto Rico Mayaguez Pittsburgh Supercomputing Center, Pittsburgh, PA

Next Generation Sequencers (NGS) provide high throughput by parallelizing the sequencing process, producing millions of sequences in a relatively short amount of time. Because NGS is still relatively new, the methods to assemble data have not been fully explored from an optimization perspective. Such an assembler is ALLPATHS-LG, whose optimization profiling is the focus of this poster. In order to carry out the profiling tasks, the CPU and memory usage of each step of the program was analyzed using profilers. The profiling process highlighted which steps were taking the most amount of time, and if possible, optimized each step accordingly. In order to maximize the efficiency and throughput of the program as a whole, steps with the highest amount of I/O, memory, and CPU time were given the most priority, in order to decrease the amount of time on sequence assembly.

**40****Design and Execution of Digital Signal Processing System for Materials Characterization**

Sunseri, Jocelyn Department of Physics and Astronomy University of Pittsburgh

A system was created to process analog input signals, reducing noise and rapidly relaying data to a computer for visualization. A combination of a bandpass input filter and an Arduino programmed to heterodyne and buffer the data were utilized. Among various applications, the system is relevant to mechanical assessment of graphene for the purposes of microstructural characterization. The signals are used to produce oscillations in nanoscale graphene "drums," and the detection of variations in the oscillations will be used to compare single-crystal and polycrystalline graphene grown by modified chemical vapor deposition techniques.

41

**Developing an infection-based bladder pain model in mice**

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 Research Consortium  
 Duquesne University, Pittsburgh, PA.

Painful bladder syndrome/interstitial cystitis (PBS/IC) is a chronic pain condition whose sufferers are commonly unresponsive to regular pain treatment. Current models of bladder pain fail to recapitulate many of its important features. To more accurately mimic PBS/IC, we sought to infect mice bladders with a uropathogenic *E. coli* strain thus mimicking the hypersensitized bladder of PSB/IC patients. Following infection, urinary bladder distension (UBD) was performed in conjunction with electromyogram (EMG) recording. The EMGs are quantified as a visceromotor response (VMR), and are representative of pain-like responses by the animal. Initial experiments showed no evidence of consistent sensitization with infection. It was discovered that body temperature may have been a confounding factor that skewed the VMR results. After adjusting for body temperature, additional 24-hour infection UBD experiments were performed. However bladder sensitization did not occur at this time point.

43

**Analyzing the Computational Demands of the Trinity RNA-Seq Assembler and Supported Downstream Analyses**

Strickland, Jonathan; Ropelewski, Alexander  
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 University of Arizona, affiliated with Pittsburgh  
 Supercomputing Center

With the affordability of next generation sequencers and the data they produce, RNA sequencing is becoming increasingly available to researchers. To assemble RNA sequence data, de novo de Bruijn based assemblers, such as Trinity, are becoming preferred over assemblers that map reads to a reference sequence. Unfortunately, de Bruijn assemblers come with high computational demands, both in terms of memory and time. In this poster, we quantify the computational demands of Trinity through the use of several different paired and unpaired RNA sequence data sets from NCBI's sequence read archive. We further illustrate with these datasets how computationally demanding the specific stages of Trinity are, as well as the Trinity supported downstream analyses.

42

**Characterization of the Arsenic Oxido/reductase from *Alkalilimnicola ehrlichii* strain MLHE-1**

Jeffrey L. Hunker<sup>1</sup> and John F. Stolz<sup>1</sup>  
<sup>1</sup> Duquesne University Department of Biological  
 Sciences, Pittsburgh, PA

The haloalkaliphilic gammaproteobacterium *Alkalilimnicola ehrlichii* strain MLHE-1 uses the unique arsenite oxido-reductase *Arx* to oxidize arsenite as an electron acceptor. As this enzyme represents a new clade in the DMSO reductase class of mononuclear molybdoenzymes, further characterization of its activity and functionality was warranted. Large volumes of cells were grown using arsenite as the electron donor and nitrate as the electron acceptor under anaerobic conditions. Cells were harvested, lysed through sonication and separated into soluble (periplasm and cytoplasm) and particulate (membranes and particulates) fractions through centrifugation. Activity assays were done to determine the localization of active enzyme. The greatest amount of activity was found in the membrane fraction with little solubilized with 2% CHAPS treatment. Further investigation has prompted experimentation with alternative non-ionic detergents such as Triton X. Upon obtaining a soluble fraction, ion exchange chromatography with DEAE Toyopearl was employed to obtain a highly enriched fraction. Activity assays were used to determine specific activity and confirm the bifunctionality (e.g., arsenite oxidase/arsenate reductase) of *Arx*.

44

**Novel and Selective Ligand Discovery for the Dopamine D<sub>3</sub> Receptor via Computational Methods.**

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 Department of Chemistry and Biochemistry  
 Center for Computational Sciences  
 Duquesne University  
 Pittsburgh, PA 15282

Dopamine receptors have been widely targeted in the treatment of schizophrenia, psychosis and drug addiction. Current studies have focused on the dopamine D<sub>3</sub> receptor because of a reduction in extrapyramidal side effects as opposed to the effects experienced at other dopamine receptors. The development of small drug-like molecules that bind selectively to the D<sub>3</sub> receptor can be accomplished through computational methods. Using AutoGrow 3.0 and MedChem Transformations found in Molecular Operating Environment, novel antagonistic ligands that are selective for the D<sub>3</sub> receptor are created. AutoGrow 3.0 has been used to generate 20 such ligands whereas MedChem Transformation in conjunction with pharmacophore filtering has been used to generate 45 potential ligands.

45

**Small molecule-protein interactions involving Vitamin D of the Vitamin D receptor using computational alanine scanning**

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Center for Computational Sciences

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Vitamin D is an important enzyme in cell proliferation; it is vital in the immune system and key in turning on or shutting off certain functions. Deficiencies in Vitamin D can result in; bone deformities, osteoporosis, muscle weakness, increased risk of fracture, and some cancers. Alanine scanning experiments have identified important residues involved in the binding of agonists to Vitamin D. Applying the Thermodynamic cycle--Perturbation method on the Vitamin D receptor with the bound substrate Vitamin D the computational alanine scanning method can be validated. Preliminary results of the computational alanine scanning method will be presented in the poster.

47

**Measurement of Rate of Activation Constants ( $k_{act}$ ) of Copper Catalysts Utilized in Atom Transfer Radical Addition Reactions**

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Department of Chemistry and Biochemistry

Duquesne University

Copper catalyzed atom transfer radical addition (ATRA) reactions are the fundamental organic transformation used to add C—X across a double bond of an alkene. The reaction involves the formation of a radical, called activation, and has a rate constant of activation  $k_{act}$ . Using a UV-Vis spectrophotometer and pseudo-first order kinetics, we calculated  $k_{act}$  for different catalysts using several alkyl halides. The reactions were catalyzed by different copper complexes,  $[Cu^I(TREN-R)Cl]$  ( $R=H, CH_3, Cl, CF_3$ ). Through the kinetic studies, we were able to show that the rate constant for the  $CH_3$  derivative is the highest, the  $Cl$  and  $CF_3$  the lowest. The electron donating effects of the methyl group increase the rate constant and the electron-withdrawing effects of  $Cl$  and  $CF_3$  decrease the rate constant. Therefore, it can be concluded that the electronic environment around the copper center has an effect on how fast the reaction occurs.

46

**Antioxidant Mechanisms of Nitrite-Mediated Neuroprotection after Asphyxial Cardiac Arrest (ACA)**

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Safar Center for Resuscitation Research

University of Pittsburgh

Cardiac arrest carries  $\geq 90\%$  mortality with 75% of patients initially resuscitated dying in the hospital from neurologic injury; post-resuscitation therapies are therefore urgently needed. We have shown that nitrite protects the heart and brain in experimental ACA. Nitrite therapy to achieve relatively low blood levels results in significant neuroprotection in animal models with little adverse effects in human safety studies. The extent and mechanisms by which this occurs need to be studied to optimize this promising therapy. We hypothesize that nitrite acts as an antioxidant reducing brain free radical generation during reperfusion providing neuroprotection. Preliminary quantification of free radical injury, free radical generation, and brain mitochondrial function appear to support this hypothesis. Our results show that following ACA, brain mitochondria from animals treated with nitrite have reduced hydrogen peroxide production, enhanced ATP-linked function and efficiency in complex I and II, and greater ATP production, compared to placebo-treated counterparts.

48

**Mechanistic Investigation of Stem Cell Differentiation Pathway Triggered by Nanoparticles**Padgaonkar, Kimaya<sup>1</sup>; Richardson, Tommy<sup>1</sup>; Candiello, Joseph<sup>2</sup> (PhD); Banerjee, Ipsita<sup>1,2</sup> (PhD)Department of Chemical Engineering<sup>1</sup>Department of Bioengineering<sup>2</sup>

Swanson School of Engineering, University of Pittsburgh

The ability to transplant human embryonic stem cell (hESC) derived islet-like cells could potentially change the lives of the almost 300 million people suffering from Diabetes. There is a critical need to develop a mechanistic understanding of the process of differentiation to design an appropriate microenvironment niche for maturation. When hESCs were co-encapsulated with chitosan nanoparticles (CNP) in calcium alginate, the cell-nanoparticle interaction was found to induce endoderm specific differentiation without directed growth factor induction. The focus of the current project is to characterize the CNP induced differentiation of the hESCs by incorporation of CNPs within embryoid bodies (EBs) using common EB forming techniques: hanging drop, stirred suspension, and microwells. Of the three methods of forming EBs, those created in microwells were found to have the best results and will further be analyzed through gene and protein expression to gain a better understanding of the mechanism.

49

**Further Development of RAE: To Enable Patient Follow-Up Presentations**

Wai-lun Lin; Cynthia Martincic, PhD, MSIS; Mandy Raab, PhD, MSIS; Mark Sangimino, MD\*

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\*Department of Orthopedic Surgery, Allegheny General Hospital, Pittsburgh, PA

RAE is a biomedical information system used by the doctor to aid parents and their children with orthopedic disorders to understand their children's condition and treatment plan during an educational session. RAE's primary function is to create customized presentations that are tailored based on the patient's age, diagnosis, gender, and other personal information. After evaluating the use of the software in a clinical setting, we improved the program's flexibility by adding a number of modifications. I provided the doctor with the option to upload multiple x-ray or MRI images of each patient. In order to assist with the clinical trial of the RAE system, as well as to expedite "continuity of care" visits, I updated the software to record the pages discussed with the patient during his/her educational session. This allows the doctor to review pages discussed with each patient in the redesigned dropdown box and the new report page.

51

**Further Development of RAE: To Assemble RAE Data to Analyze Patient Understanding**

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The RAE program is a web-based application used by pediatric orthopedic doctors to further educate patients about their disorders. The program generates dynamic presentations specific to each patient, based upon their diagnosis and several other factors. Since the generated presentations are to be used in a flexible education session between the patient and the doctor, not all of the pages of a presentation need to be covered. The focus of my internship was to create a centralized report page that displays the pages viewed for each patient presentation. A clinical trial utilizing pre- and post-surveys is also ongoing to determine the effectiveness of RAE at teaching patients and their families about their disorders and treatment plans. My centralized report page also allows the doctor to review the patient's pre-and post-survey answers to determine how well they understood the information in their dynamic presentation.

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**Further Development of RAE Clinical Trial: To Improve the Efficacy of an Interactive Biomedical Information System**

Jennifer Klimek; Mandy Raab, PhD, MSIS; Cynthia Martincic, PhD, MSIS; Mark Sangimino, MD\*

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\*Department of Orthopedic Surgery, Allegheny General Hospital, Pittsburgh, PA

Jennifer Klimek

RAE is an interactive tool used to further facilitate the educational process that occurs between patient and doctor. Previous work by Dr. Sangimino showed that supplemental instruction improved the patient's ability to manage their musculoskeletal disorders. RAE software was created to automatically create personalized presentations that allows for a standard of knowledge to be achieved by the patients. A clinical trial was developed to test the efficacy of the software. The first version of the clinical trial was a worthy beta test of the experimental setup and revealed some roadblocks that needed to be cleared before continuing the clinical trial. After observing the Learning Center during the educational presentations and the pre/post testing of the patients, a second version of the clinical trial was created to increase user friendliness and to smooth the process. This second version was redesigned with a new information flow, control panels, user interface and functionality.

52

**The Effect of Stress on the Ability to Infect Salamanders with Probiotic Bacteria.**

Shober, Lauren; Woodley, Dr. Sarah

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Amphibian populations are declining all over the world, in part due to infection by a lethal chytrid fungus. There is a probiotic bacterium, *Janthinobacterium lividum*, residing on salamander skin that may provide resistance to infection by the chytrid fungus. A conservation strategy is to inoculate susceptible amphibian species with *J.lividum*. We asked whether chronic stress, known to suppress the immune system, affects susceptibility of amphibians to inoculation with *J.lividum*. We exposed two species of salamanders to *J.lividum* and predicted that exposure to chronic stress prior to inoculation would lead to greater infection compared to unstressed control animals. Weekly following inoculation all salamanders were swabbed for the probiotic bacterium. Body weight and feeding were also indications of infection. All animals regardless of treatment or species were infected. Therefore, we were unable to evaluate whether stress affects infection rate. This demonstrated that *J.lividum* holds potential for inoculating multiple species of amphibians.

**53**

Depression Modulation through CNS Active Compounds from a Marine Cyanobacterium

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Comorbid pain and depression directly and indirectly affect millions of Americans. Most pharmaceuticals treat either pain or depression, but not both. Those suffering from comorbidity must take multiple drugs, which can elicit negative side effects. This project focuses on the anti-depressive capabilities of novel compounds from a marine cyanobacterium. In this project, we extracted and purified compounds from a marine cyanobacterium using chromatography (silica and HPLC) and analyzed the fractions using NMR and HPLC-MS. Fractions were pre-screened for activity of the CNS receptors. Fractions 2064e and 2064h were found to bind to 5-HT<sub>2B</sub> and 5-HT<sub>2C</sub> receptors, respectively. They were tested *in vivo* by administration to the CNS utilizing intracerebroventricular (ICV) cannula injections in C57BL/6 mice. After delivery, we performed the tail suspension test (TST), a widely used depression assay, to test the anti-depressant potentials of these compounds. Biological results to date will be presented.

**55**

**The Effect of Voltage-Gated Calcium Channel Organization on Function in the Mouse Neuromuscular Junction**

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University of Pittsburgh  
Pittsburgh Supercomputing Center, Carnegie Mellon  
University

Through the use of computer simulations, we can study the structure and function of the mouse neuromuscular junction (NMJ) at a microscopic level inaccessible through experimental techniques. We utilized MCell, a stochastic reaction-diffusion simulator, to conduct our computational study. Our MCell model of the mouse NMJ contained a realistic 3D representation of an active zone, voltage-gated calcium channels (VGCCs), calcium ions, synaptic vesicles, calcium ion sensors on vesicles, and calcium buffer molecules. Based on experimentally known putative locations of VGCCs in the mouse NMJ, we investigated the impact of channel location on synaptic vesicle release. We found that vesicle release was most prominent with closely associated VGCCs and paired-pulse facilitation was greatest at intermediate distances. We also observed that with more distant VGCCs it took longer for vesicles to release from the active zone. Our results will help identify likely organization of VGCCs within active zones.

**54**

**Effects of Stress on Rate of Infection by A Deadly Amphibian Pathogen in Salamanders**

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Duquesne University, Pittsburgh, PA

Amidst recent amphibian declines, it is important to understand what is offsetting it and how to prevent it. Chronic stress exposure may negatively impact skin microbiota, suppress immune response to pathogenic stressors, and increase susceptibility to a chytrid fungus pathogen, *Batrachochytrium dendrobatidis*. I tested whether exposure to the stress hormone, corticosterone (CORT), increases susceptibility to infection and development of chytridiomycosis in red-legged salamanders. Daily exposure to CORT or vehicle for 10 days was followed by inoculation with chytrid ( $6.9 \times 10^7$  zoospores/animal) or synthetic spring water. Subjects were monitored for changes in body weight, skin sloughing, and behavior. Subjects exposed to the chytrid pathogen showed signs of disease, but initial results found no effect of prior exposure to CORT. Future work includes the use PCR to quantify chytrid zoospore loads on infected subjects to better understand the effects of stress on the rate of infection and disease progression.

**56**

**A Study of the Presence of Gunshot Residue in Pittsburgh Police Stations**

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Due to the risk of secondary transfer of gunshot residue (GSR) by police vehicles or stations prior to GSR sampling, it was necessary to create a baseline for the amount of GSR present in police stations or in police vehicles. Samples taken from the back of a Ross Township police vehicle, jail cell, and interview room were automatically analyzed using scanning electron microscopy-energy-dispersive X-ray spectroscopy (SEM-EDS) techniques for the presence of particles containing at least ten percent by weight antimony, barium, and lead. These particles were classified as characteristic GSR, while particles containing various combinations of lead, antimony, barium, aluminum, titanium, and zinc were classified as consistent. For a sample to be considered as positive for GSR there must have been at least three characteristic GSR particles present. Three hundred and seven characteristic and no unique GSR particles were found among the samples analyzed; no sample was positive for GSR.

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**Effect of Varying Carbon Chain Length on SAMs on SS316L**

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 Department of Chemistry and Biochemistry, Duquesne University<sup>2</sup>

A self-assembled monolayer (SAM) is a two-dimensional molecular array that is spontaneously organized by adsorption of amphiphilic organic molecules on a solid inorganic surface. The head group of adsorbing molecules strongly binds with the metallic surface, which can provide highly ordered monolayers. Because of this, SAMs have been used to modify the properties of surfaces such as friction, lubricity, hydrophobicity, etc. In this study, SAMs of tetradecylphosphonic acids (TDPA) and octadecylphosphonic acids (ODPA), and mixed SAMs of each, were produced on the oxide surface of stainless steel (SS316L). A total of five samples, including 100%, 50%-50%, and 25%-75% ODPAs (18 carbons) and TDPA (14 carbons), were prepared using solution deposition and analyzed using diffuse reflectance infrared Fourier transform spectroscopy (DRIFT) and atomic force microscopy (AFM). DRIFT revealed that highly ordered SAMs of ODPAs and TDPA were formed. Interfacial friction was measured with AFM on the monolayers and mixed monolayers.

59

**Nitrite regulates the activation of mitochondrial protein kinase A (PKA) and modulates mitochondrial function in isolated rat heart mitochondria.**

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 University of Pittsburgh

Nitrite at a concentration of 25  $\mu$ M increased both protein kinase A (PKA) activity and maximal respiration in isolated mitochondria from rat heart. These effects were abolished in the presence of the PKA inhibitor, PKI. We have previously shown that treatment of rat cardiomyocytes with nitrite protects against ischemia/reperfusion (I/R) injury by increasing PKA activity and regulating mitochondrial function. Whether nitrite can directly modulate mitochondrial PKA activity to regulate mitochondrial function is unknown. In mitochondria isolated from rat heart, PKA activity was measured by an Enzyme-Linked Immuno-Absorbent (ELISA) assay. Maximal oxygen consumption, measured using a Clark electrode, increased with nitrite. The PKA-dependent increase in maximal oxygen consumption after nitrite treatment suggests that nitrite could regulate the activity of Complex IV, a known phosphorylation target of PKA.

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**Creating a Nitric Oxide leaving group through the use of 4-NBA SAMs**

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 Duquesne University

Nitric oxide (NO) is suggested to have a variety of therapeutic properties, one being the ability to cause dispersal of the antibiotic resistant *P. arginosa* biofilm within the Cystic Fibrosis lung. Upon the biofilm being broken down, the bacteria can be treated. Nitric oxide can be released from 4-nitrobenzoic acid (4-NBA). When exposed to a reactive oxide surface of SS316L, 4-NBA may form a self-assembled monolayer (SAM). On formation of a SAM, the molecule can be treated to release the desired NO. Basic deposition of a 4-NBA SAM onto SS316L was unsuccessful using either solution or aerosol deposition. Polished SS316L with an aerosol spray provided promising results. A secondary method was explored utilizing the formation of 12-aminododecanoic acid (12-ADA) SAMs as surface links along with a PFP/EDC coupling technique. All samples were examined through diffuse reflectance infrared Fourier transform spectroscopy (DRIFT).

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**Potassium Translocation Pathways in the Dopamine Transporter**

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Dopamine (DA) is a neurotransmitter that, when released into the synaptic cleft, causes a happy feeling. The dopamine transporter (DAT) is responsible for the reabsorption of DA into the pre-synaptic cell from the synaptic cleft. The translocation of DA involves an open-outward (OO) to open-inward (OI) conformational change. The reverse conformational change has not been thoroughly studied. Our hypothesis is that  $K^+$  is needed to drive this conformational change from OI to OO. We simulated this phenomenon using molecular dynamics—the propagation of atomic positions that follow Newton's second law of motion. The system simulated consisted of two bilayers, each containing one DAT. Between the bilayers is the extracellular space and on the other side, the intracellular space; each space contained the physiologically correct  $Na^+$  and  $K^+$  ion concentrations. Current results show a  $K^+$  ion 40% translocated through DAT. Further details of one analysis will be presented in this poster.

**61****Calculating Vibrational Frequencies and Modes of N-methylacetamide Using Principal Component Analysis**

McGovern, Marissa; Wheeler, Ralph A.; Boesch, Scott  
Department of Chemistry and Biochemistry  
Duquesne University

Vibrational frequencies and modes are crucial in confirming the synthesis of a compound, and distinguishing different conformations of proteins and polymers. Principal Component Analysis is a new method to determine the normal modes and vibrational frequencies of molecules in solvents, proteins or polymers melts, which are difficult to find using current methods. Quantum Mechanics/Molecular Mechanics (QM/MM) trajectories were calculated for N-methylacetamide in isolation and in water to be used in this new method. The Principal Component Analysis of the QM/MM results will be performed and these frequencies will be compared with the frequencies resulting from published experiments and from standard quantum chemical calculations.

**63****Predicting the Structures and Properties of Biochemical Electron Transfer Intermediates**

Fowkes, Melissa; Boesch, Scott; Wheeler, Ralph A.  
Department of Chemistry and Biochemistry  
Duquesne University

Quinones, a class of organic molecules, are important in electron transfer. The para-benzosemiquinone neutral radical is an intermediate between the fully oxidized quinone and the fully reduced quinol. A variety of quantum chemical methods will be used to calculate the optimized geometries, vibrational frequencies and modes, and other properties of this radical in the gas phase and with implicit solvent. The computed data will be compared to available experimental data and methods of detecting the radical in proteins will be suggested.

**62****Development of *Streptomyces coelicolor* spores for Vaccine Delivery**

Kocher, Matthew; Moore, Carrie Ann; Mrohs, Kevin;  
Fucich, Dan; Dr. McCormick, Joseph  
Department of Biological Sciences  
Duquesne University

Endospores from unicellular bacteria have been used for vaccine production, probiotics, and as biosensors. Exospores from filamentous bacteria have not been thoroughly investigated for use in similar applications. SapA, SapC, SapD, and SapE are spore-associated proteins of the filamentous sporulating soil bacterium *Streptomyces coelicolor*. The goal of this project was to fuse the gene encoding SapC to the gene encoding the B subunit of the heat labile toxin of *Escherichia coli* (LTB) or a truncated gene encoding a portion of the tetanus toxin of *Clostridium tetani* (TTFC) to determine if fusion proteins containing either antigen portion can be expressed and incorporated on the spore surface. The Sap-toxin fusion genes were inserted into the chromosome. SapC-LTB expression and localization was characterized by SDS-PAGE and Western Blot analyses. If antigens can be localized on the spore surface, it could allow for oral vaccines with an extended shelf life.

**64****Sequence Relationships of Universal Stress Proteins of *Clostridium* Species**

Thompson, Michael R.<sup>1</sup>; Nicholas, Hugh<sup>2</sup>; Isokpehi, Raphael<sup>1</sup>

<sup>1</sup>Center for Bioinformatics and Computational Biology,  
Jackson State University

<sup>2</sup>Pittsburgh Supercomputing Center

The United States is continually faced with cases of foodborne illnesses that are related to environmental pathogens such as the genus *Clostridium*. This provides a need to understand the inherent features that permit species from this genus to thrive in the harsh conditions that they are faced with whether it be man-made or ecological. Universal stress proteins (USP) play a vital role in *Clostridium* species allowing them to flourish in a variety of stressful environments due to the activation of various intracellular signaling pathways. The purpose of this research is to analyze the various species of *Clostridium* and understand the key aspects of sequences that are connected to USP. A suite of bioinformatic tools were used to aid in the understanding of significant data including multiple sequence alignments and motifs using multiple expectation maximization for motif elicitation (MEME).



**65****Modeling Embryonic Stem Cells: How does Cyclin D and p21 affect cell cycle transition?**Lawrence, Milani<sup>1,2</sup>; Task, Keith<sup>1</sup>; Banerjee, Ipsita<sup>1,3,4</sup><sup>1</sup>Chemical Engineering Department, University of Pittsburgh<sup>2</sup> Mathematics Department, Mount Holyoke College<sup>3</sup> Bioengineering Department, University of Pittsburgh<sup>4</sup>McGowan Institute for Regenerative Medicine, University of Pittsburgh

Embryonic Stem Cells (ESCs) have unique cell cycle characteristics compared to adult cells, such as fast proliferation and a short G1 phase which lengthens during differentiation. Understanding this unique cell behavior becomes difficult with a purely experimental approach. We have previously created a model that can explain and predict ESC cell cycle behavior during self-renewal. To explain the cell cycle transition from pluripotent to differentiated, in the current work we have developed a model to show how two cell cycle proteins affect the dynamics of the probability of G1-S transition during differentiation. The inclusion of the two proteins, cyclin D and p21, change the probability density function of transition, causing a lengthening of G1. Furthermore, this effect is dependent on the dynamics of the proteins' expression. Our model can capture and explain the G1-S transition variability and lengthening during differentiation, giving better insight into the specific behavior of ESCs.

**67****Glucose Transporter Glut8 Used In Next Generation Sequencing Of Chicken Transcriptome**Nelson, Rhia; Wang, Xiaofei<sup>1</sup>; Nicholas, Hugh<sup>2</sup>, Ropelewski, Alex<sup>3</sup>

Pittsburgh Supercomputing Center, Pittsburgh, PA

College of Agricultural, Human, and Natural Sciences, Tennessee State University, Nashville, TN

We are studying how to efficiently produce chicken for either meat or eggs by determining the factors effecting growth. Glucose transporter GLUT8 is used as an expression we will study with complete chicken transcriptomes at two and eight weeks of age. We are creating a curated dataset of GLUT8 transporters that will provide us with specific insights into its functions and properties. We have begun with a high quality multiple sequence alignment and prediction of the transmembrane helices followed by, a phylogeny of several GLUTn transporters, where n is 6, 8, 10, 12, and 13. We will also determine the individual amino acids responsible for the specific actions of the GLUT8 transporter within this family of closely related transporters as well as visualizing its 3 dimensional structure.

**66****Quantitative Analysis of Simulation Techniques Using a Rule-Based Model of JAK/STAT**

Essex, Morgan and Harris, Leonard; Lezon, Tim (mentors)

Computational and Systems Biology  
University of Pittsburgh

The STAT (signal transducer and activator of transcription) family of proteins has become a cancer therapy target of interest in the last decade. The STAT3 protein specifically is activated in a wide array of both blood and solid tumors, indicating it as a key player in general cancer development. When activated by IL-6 signaling via the JAK/STAT pathway, the STAT3 protein has two regulators: a tyrosine phosphatase (SHP2) and a suppressor of cytokine signaling (SOCS3). A rule-based model to look at the this pathway in a single cell with these regulators is a simplified approach with good potential to explain the behavior of STAT3. Simulation of the generated network can be done using ordinary differential equations or stochastically. Each method produces different trajectories, and quantifying the conditions under which ODE methods become unreliable will be useful for future modeling and simulation in a variety of cellular applications.

**68****RICH Detector for Jefferson Lab Experiments**

Lois, Robert

Dr. Fatiha Benmokhtar

Department of Physics

Duquesne University

The Ring-Imaging Cherenkov (RICH) detector is a device that allows exclusive particle identification and is currently being designed for the CLAS12 spectrometer at Thomas Jefferson National Accelerator Facility (TJNAF). The upgrade to the spectrometer will improve hadron identification from 3 GeV/c to momenta exceeding 8 GeV/c by replacing the low-threshold Cherenkov counter (LTCC) with the RICH detector. When a charged particle beam is travelling faster than the speed of light in the radiator material RICH Cherenkov radiation is emitted. The radiation is reflected by an optical mirror system and finally measured by photon detectors. Simulations of the CLAS12 detector with the GEant4 Monte-Carlo package are written and the RICH detector is currently being implemented to the simulation package.

69

**Characterization of Several Pharmaceutical Compounds and the Use of Carnuba Wax to Improve the Compressibility of Problematic Materials**

Erickson, Patrick W.; Katz, Jeffrey M.; and Buckner, Ira S.

Graduate School of Pharmaceutical Sciences

Duquesne University

Pharmaceutical materials have unique compressibility properties. These properties must be understood before attempting to manufacture new drugs into tablets. Nineteen pharmaceutical compounds exhibiting various characteristics were compressed, in order to obtain the compressibility data for each material. In-die solid fraction was measured during compression of powders on an Instron Universal Testing System. Out-of-die solid fraction was measured by recording tablet dimensions over several days using a digital caliper. In-die solid fraction data was used to predict each out-of-die compressibility profile by correcting for elastic and viscoelastic deformation that occurs during compression in-die. Additionally, chlorpropamide, calcium alginate, and phenytoin were mixed in varied proportions with carnauba wax in an attempt to improve tablet compressibility. Each material was successfully characterized based on the retrieved profiles from the data, most of the out-of-die data for the materials were correctly predicted, and the carnauba wax generally increased the compressibility for all three problematic materials.

71

**Site-Directed V489C and R564C Mutagenesis of hSERT**

Ayinde, Safiat; Fogl, Anthony; Veeramachanenin, Rathna; Dr. Cascio, Michael

Department of Chemistry and Biochemistry  
Duquesne University

The human serotonin transporter, hSERT, is responsible for re-uptake of serotonin after neurotransmitter release and is targeted by antidepressants. hSERT has many poorly characterized conformational structures. It is anticipated that mutagenesis of V489C or R564C in a mutated form of the receptor that lacks external cysteines can be used as targets for cross-linking of hSERT. PCR (polymerase chain reaction) mutagenesis of the hSERT expression vector was followed by transformation into competent cells to isolate the mutated expression vector. Plasmid containing SERT mutated cDNA will be transfected into mammalian cells to express the receptor on the cell surface. MTS-benzophenone ligand will form disulfide at the introduced cysteine that will be activated with UV light to form non-specific crosslinks. The sites cross-linked to the mutated cysteine will be identified through mass spectrometry where the binding sites can be characterized.

70

**Quantum Models of Methyl Phosphate Adsorption onto the Rutile (110) Surface**

Clifford, Sadie W.; Hebert, Sebastien P.; Srnec, Matt N.; Gwalt, Ellen S.; Evanseck, Jeffrey D.

Center for Computational Sciences and the Department of Chemistry and Biochemistry  
Duquesne University

Adsorption modes and strength of the bond(s) between the rutile (110) surface and methyl phosphate have been investigated using quantum chemistry. Model systems were developed to conserve the coordination geometry and valency of each atom within the periodic cell. Specifically, the octahedral geometry of titanium and trigonal planar geometry of oxygen were maintained, and the overall negative charge was neutralized with the appropriate number of terminating hydrogen atoms. A simple model,  $Ti_3O_{11}H_{10}$ , was partially optimized where only the hydrogen atoms were allowed to move and results were used for further calculations. The Crystal09 quantum software package was used to run calculations with the DFT functional Perdew-Burke-Erzerhof and the triple-zeta valence with polarization quality basis sets corresponding to each atom; Ti, O, H, P, and C. Results that describe the binding strengths and adsorption modes of the phosphate to the surface will be discussed.

72

**Design, Synthesis and Characterization of Non-redox Metal Complexes of Redox Active Ligands**

Ratvasky, Stephen; Mogesa, Benjamin; Basu, Partha

Department of Chemistry and Biochemistry

Duquesne University

Redox active dithiolene ligands can exhibit multiple well-defined redox states, but their coordination chemistry is primarily focused on the reduced form of the ligand with transition metal ions with multiple redox states. These complexes are important in understanding the metal-ligand redox interplay. Another side of this fascinating equation, where ligands are redox active but the metal is not, is described herein. For this purpose Diisopropylpiperazine-2,3-dithione ( $Pr_2Dt^0$ ) and N,N'-Dimethylpiperazine-2,3-dithione ( $Me_2Dt^0$ ) were used to represent the oxidized state of the ligands. The coordination chemistry of the non-redox active metal, zinc, was explored. The synthesis of these ligands and their zinc complexes, as well as a mixed redox complex with a reduced dithiolene co-ligand maleonitriledithiolate (mnt) Zn(II) complex are described. These compounds were characterized using  $^1H$  and  $^{13}C$  NMR, IR spectroscopy, UV-Vis spectrophotometry, and X-ray crystallography. The challenges and novelty of this system are described.

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**GeneDoc Migration**Reed, Tevin<sup>1</sup>; Ropelewski, Alexander<sup>2</sup>;<sup>1</sup>North Carolina Agricultural and Technical State University, Greensboro, NC;<sup>2</sup>Pittsburgh Supercomputing Center, Pittsburgh, PA

GeneDoc is a full featured multiple sequencing alignment visualization, editing, and analysis tool. Multiple Sequence Alignments provide a map of the evolution of a particular gene or protein family. GeneDoc highlighting facilities provide good guidance for experiments by indicating important residues in the gene or protein family. GeneDoc has an easy-to-use point and click user interface with extensive keyboard mapping for advanced users. Despite the plethora of tools GeneDoc provides to the bioinformatics world; GeneDoc is bound to Windows systems, because it makes use to Microsoft foundation classes (MFC). To combat this problem we elected to convert GeneDoc from MFC into wxWidgets. wxWidgets provides a single, easy to use API for writing GUI applications on multiple platforms that still utilize the native platform's controls and utilities. Moving GeneDoc to this new platform not only adds accessibility to more users but also adds potential to move GeneDoc on to tablets.

75

**Comparison of Transfection Efficiency and Cytotoxicity in Primary Stem Cells Using Non-viral Transfection Reagents**

Garbrecht, Grace; Shekhar, Sudhanshu; Kumta, Prashant

Department of Bioengineering  
University of Pittsburgh

Gene therapy has the potential to treat many diseases such as genetic disorders, infections, and cancer. However, additional research must be done to improve gene transfer efficiency and to enhance efficacy especially in primary cells, which, unlike immortalized cell lines, represent more realistically the in vivo application of gene delivery. Transfection efficiency and cytotoxicity were compared among several commercially available transfection kits as well as synthesized hydroxyapatite and silicon substituted hydroxyapatite nanoparticles. GFP-expressing plasmid DNA was used for the transfection of different cell types including human mesenchymal stem cells and human umbilical vein endothelial cells. Transfection efficiency and cytotoxicity were measured at day 1 and day 3 using flow cytometry and MTT assay. Although no results have been recorded as of yet, we hypothesize that the nanoparticles will exhibit higher transfection efficiency as well as lower cytotoxicity in comparison to the commercially available reagents.

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**Comparative Analysis of Polymer Additives in Carpet Fibers using Matrix Assisted Laser/Desorption Ionization Time of Flight (MALDI-TOF) and Electrospray**Cutia, Zachary; Wetzel, Dr. Stephanie J  
Department of Biochemistry  
Duquesne University

Polymers have an impact on all of our lives, because they are essentially everywhere. Manufacturers compound synthetic polymers with numerous additives, affecting their chemical and physical properties. Qualitative analysis of synthetic polymers can yield the identities of specific additives. The additive composition of the polymer may be useful in forensic fiber analysis. It is hypothesized that the additive composition can be used to identify the manufacturer of the fiber. Nylon, polyolefin, and polyester carpets from three different manufacturers were studied. Synthetic carpet fibers were either extracted with a 1:1 ratio of hexane: acetone or methylene chloride, and then sonicated and heated. Samples were dried with nitrogen gas, and reconstituted with isopropanol, methanol, or acetonitrile. MALDI-TOF-MS and Electrospray (ESI-TOF-MS) were used to obtain additive compositions of the fiber extracts. The resulting mass spectra were used to compare the two techniques for optimal solvent extraction, instrument method, and manufacturer identification.

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**Optimization of Fluorine content in a Hydrogel Designed for Multimodal Imaging and Drug Delivery**George, Simon; Domenech, Trystan; Tunney, Robert; Hitchens, Kevin; Velankar, Sachin; Janjic, Jelena  
Mylan School of Pharmacy  
Duquesne University

A novel hydrogel with drug delivery as well as multiple imaging capabilities such as photoacoustic, near infrared and <sup>19</sup>F magnetic resonance imaging (MRI) was optimized. More specifically, the fluorine content was optimized to obtain effective photoacoustic and <sup>19</sup>F MR imaging. This is the first reported case of a hydrogel containing this triple imaging capacity to date. Our preformulation strategies and methods are reported. The designed hydrogel contains four components: perfluoropolyether (PFPE) ester as the fluorine component, Pluronic® F127 as the block copolymer framework, branched Polyethyleneimine (PEI) to stabilize PFPE, and glycerol as a thickener. Optimization studies were performed by gradually increasing the PFPE and testing gel imaging capacity. Rheological studies and microscopy were utilized to evaluate the stability and structure. Imaging capacity was also demonstrated by the 3 different modalities. In conclusion, we report a novel hydrogel with triple imaging capacity and a broad number of possible applications.

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**Developing Tools for Analyzing Unique Cell Cycle Features of Embryonic Stem Cells**Barner, Sierra<sup>1</sup>; Task, Keith<sup>1</sup>; Banerjee, Ipsita<sup>(1,2,3)</sup>

1 Department of Chemical Engineering

2 Department of Bioengineering

3 McGowan Institute of Regenerative Medicine  
University of Pittsburgh

Embryonic Stem Cells (ESC) have unique cell cycle characteristics when compared to adult cells, exhibiting a brief G1 phase and high population proportion in the S phase. When ESC differentiate their cell cycle significantly changes, including a lengthening of G1. While these features are important for ESC, little is known about how the cell cycle fate is regulated during self-renewal and differentiation. In this work we investigate synchronized ESC to obtain a better understanding of these cell cycle changes. We have synchronized pluripotent and differentiated ESC in the G2 phase with nocodazole. Synchronized cells were analyzed by flow cytometry to determine population-level information on the cell cycle. The data obtained from flow cytometry was further analyzed by ModFit software to quantify the portions of cells in each phase. The tools developed in this work allow for more efficient ways to obtain a better understanding of cell cycle behavior of ESC.

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**The Baculoviral Expression of the Serotonin Transporter in Sf9 Insect Cells**Underwood, Soraya; Cascio, Michael  
Chemistry and Biochemistry Department  
Duquesne University

Research involving the human serotonin transporter (hSERT) can lead to the development of novel antidepressants. hSERT studies are limited by the very low expression of the transporter. Thus, the overall goal is to produce recombinant hSERT-baculovirus, infect Sf9 insect cells, which in turn will produce relatively large quantities of the serotonin transporter as infection is a very successful method of producing large quantities of or overexpressing a protein. This insect cell- baculovirus expression vector system (IC-BEVS) can express products of genes from many organisms and from any location within a cell. Initial studies excised a portion of the pcDNA3 plasmid containing hSERT that contains no free accessible cysteines, using specific restriction enzymes and ligated this fragment into pFASTBAC1. The expression vector is a recombinant baculovirus that encodes a protein of interest under transcriptional regulation of the strong polyhedrin promoter. The recombinant baculovirus will be made, amplified and tested for expression.

78

**Intermolecular effects and their influence on the <sup>13</sup>C chemical shift anisotropy in organic crystal lattices**Marsh, Zachary<sup>1</sup>; Madura, Jeffrey D.<sup>2</sup>; and Luliucci, Robbie J.<sup>1</sup><sup>1</sup>Department of Chemistry, Washington and Jefferson College, Washington, Pennsylvania 15301, USA<sup>2</sup>Department of Chemistry and Biochemistry, Center for Computational Sciences, Duquesne University, Pittsburgh, Pennsylvania 15282, USA

Anisotropic components of the chemical shift can be used to understand molecular structure and the impact of its surrounding environment. Accurate magnetic shielding calculations in solid phase systems require the lattice structure be included. Intermolecular effects such as hydrogen bonding have a significant influence on the molecule's crystal structure. Methyl-glucopyranoside, which crystalizes in the P<sub>212121</sub> space group, is an excellent test system due to extensive hydrogen bonding in the solid state as well as the availability of <sup>13</sup>C chemical shift anisotropy data. The <sup>13</sup>C magnetic shielding anisotropy values of methyl-glucopyranoside were calculated using the GIPAW method with Vanderbilt ultra-soft pseudo potentials in the framework of the Quantum Espresso program. Calculations, one of an isolated molecule, and one including the lattice structure, were compared to experimental <sup>13</sup>C anisotropic values. Intermolecular effects change the magnitude of the principal components by five percent. Additional details and results will be given in the poster.

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**Methods to Analyze Discrete Logical Models of Cell Signaling**Mendes Marina; Natasa Miskov-Zivanov; James Faeder  
Computational & Systems Biology  
University of Pittsburgh

When developing discrete logical models of cell signaling, the analysis of outcomes can be challenging. In this work, I am applying several analysis methods that are often used for studying biological models of other types (e.g., continuous, ODE models), but are not common for analysis of discrete logical models. Two of the methods I am using are Principal Components Analysis (PCA) and Boolean function sensitivity analysis. I am studying models of control circuitry of T cell differentiation and Malaria parasite infection in mosquito cells. PCA is used to compress a large amount of data obtained from simulation of these two models, while Boolean difference is applied to study sensitivity of Boolean functions in the models. The final goal of this work is to better uncover key regulatory components of these systems and predict how cells will respond to different stimuli.

**81****Pharmacokinetic-Pharmacodynamic Model of the Anti-Cancer Agent Topotecan**

Boley, Eric; Carcillo, Christine M.; Ho, Thang; Parker; Robert S.

Department of Chemical and Petroleum Engineering, University of Pittsburgh

Topotecan is a chemotherapy drug used to treat ovarian cancer, small cell lung cancer, and cervical cancer. Because chemotherapy kills healthy cells (undesired toxicities), as well as cancer cells (desired effect), design of the correct dose magnitude and schedule is crucial. A mathematical model for a chemotherapy drug can help to determine the patient-optimal dose by evaluating the patient's plasma drug concentration-vs-time (pharmacokinetics, PK), and the antitumor effect and toxicity of the drug dose (pharmacodynamics, PD). We fit a 2-compartment PK model that captures human plasma concentrations after intravenous administration at dose levels of 0.5-1.5mg/m<sup>2</sup>. This PK model drives a PD toxicity model originally derived in our lab to capture neutropenia after docetaxel chemotherapy; we refit the 3 drug-associated parameters to evaluate the model's ability to capture topotecan-induced neutropenia. These models can be incorporated in a dose scheduling design algorithm to aid clinical decision-making while managing toxicity for patient-specific chemotherapy.

**83****Dissipative Particle Dynamics: Modeling 3D-Vesicles**Rosscoe, Michael<sup>1,2</sup>; McCarthy, Joseph<sup>1</sup><sup>1</sup>Department of Chemical and Petroleum Engineering, University of Pittsburgh<sup>2</sup>Department of Chemical and Biochemical Engineering, University of Maryland, Baltimore County

The study of biological science is imperative to our understanding of biological systems, particularly through computational and experimental methods. Current theoretical models for dynamic particle simulation are limited to timescales in the micro- and nano- second time frame. Dissipative Particle Dynamics (DPD) is an innovative simulation technique where particles represent whole molecules or fluid regions rather than single atoms to bridge the hydrodynamic time gap and space scale versus experimental data. This study exemplifies the purpose of DPD by simulating a vesicle in a three-dimensional environment over 300 simulation seconds. The results show a direct correlation between spring forces and particle interaction resulting in various vesicle formations. Ultimately, the goal of this research is to better understand vesicle interactions within a biological system when varying solutes are introduced into the environment on larger scales than previously achieved using other theoretical methods.

**82****Exploration of Cement Reactions of Crystalline and Amorphous Trimagnesium Phosphate**

Delaney, Kayla; Ostrowski, Nicole; Lee, Boeun; Kumta, Prashant

Department of Bioengineering

University of Pittsburgh

Currently calcium phosphate based bone cements are commonly used in bone repair and replacement due to their high biocompatibility; however this solution still has its downfalls. An alternative may be magnesium phosphate based bone cements due to their higher initial strength and shorter resorption times. In this study, amorphous, semi-crystalline, and crystalline trimagnesium phosphate powders were reacted with an aqueous potassium phosphate solution at various powder to liquid ratios to form cements. The samples were characterized through x-ray diffraction, scanning electron microscopy, and set time measurement; revealing that semi-crystalline samples had the most clinically relevant set times. Testing of sample stability in phosphate buffer solution indicated that all but the amorphous samples remained intact. Cell viability was tested through multiple methods showing conflicting results. Despite these conflicting results further experimentation may confirm semi-crystalline trimagnesium phosphate as viable substitute for current bone repair methods.

**84****Tool to Analyze Protein Localization during Tissue Assembly**Wallace, Anathae<sup>1</sup>; Lynch, Holley<sup>2</sup>; Davidson, Lance<sup>2</sup><sup>1</sup>Systems Medicine REU, Department of Chemical and Petroleum Engineering<sup>2</sup>Department of Bioengineering, University of Pittsburgh

During morphogenesis, cells polarize and intercalate, developing the features and shape of the mature organism. These processes are prompted by proteins such as frizzled and disheveled moving to certain sites in the cells, producing planar cell polarity. To study these localizations, I designed a plugin in ImageJ to analyze the patterns produced by fluorescently labeled proteins. In particular, I studied the localization of yellow fluorescent protein labeled myosin regulatory light chain (YFP-MRLC) along cell boundaries identified with mCherry labeled membrane protein (mem-mCherry) within animal cap ectoderm cells of *Xenopus laevis*. The plugin measures the intensity of the fluorescing protein oriented along the cell boundary, and outputs graphical representations of this data for individual cells so that they can be compared regardless of differences in size and shape.

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**Blood Flow, Blood Shunting, and Edema:  
Modeling Physiological Responses to Inflammation  
in Critical Care**

Shreves, Caitlyn<sup>1,2</sup>; Pritchard-Bell, Ari<sup>1</sup>; Clermont, Gilles<sup>1,3</sup>; Parker, Robert S.<sup>1,3</sup>

<sup>1</sup>Department of Chemical and Petroleum Engineering, University of Pittsburgh

<sup>2</sup>Department of Biomedical Engineering, University of South Carolina

<sup>3</sup>CRSMA Laboratories, Department of Critical Care Medicine, University of Pittsburgh Medical Center

“Stress hyperglycemia” often occurs in patients post-surgically and during sepsis in critical care. This diabetes-like condition leads to elevated glucose concentrations, as well as increased morbidity and mortality. Additional side-effects of the inflammatory response to trauma are shunting of blood and edema – extravascular fluid pooling. The purpose of this study is to create a model that predicts inflammation-induced changes in blood flow and fluid pooling, and to couple this model to a model of glucose concentration and regulation in critical care. A physiologically-based model of inflammatory response, capturing inflammatory cytokines, blood flow, and tissue pooling was created in MATLAB (© 2013, The MathWorks, Natick, MA) our parameterization allows us to tune vascular/tissue equilibration rates for cytokines, cytokine-induced changes in shunting of blood, and the impact of net physiological fluid balance on edema. Future work will address changes in cardiac output and tissue-specific blood flow in response to inflammatory cytokine dynamics.

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**Constructing Bacterial Two-Hybrid Plasmids to  
Analyze Chromosome Segregation in Filamentous  
Sporulating Bacteria**

Fucich, Dan; Hasipek, Metis; Dr. McCormick, Joseph  
Department of Biological Sciences  
Duquesne University

*Streptomyces* are the major producers of antibiotics and other naturally occurring biological compounds. For *S. coelicolor*, the mechanism and proteins involved in DNA segregation are largely unknown. Important segregation proteins in the chromosome partitioning system such as ParA, ParB, and ParJ have been identified and characterized, however, other unknown proteins are believed to be involved. The ultimate goal of this project is to determine if protein-protein interaction partners for ScpA and ScpB can be identified. ScpA and ScpB are accessory proteins for the condensin protein structural maintenance of chromosome (SMC). To investigate protein-protein interaction, the genes that code for these proteins are being cloned into Bacterial Two-Hybrid (BTH) vectors pKT25 and pUT18. Using BTH, the *in vivo* protein-protein interactions result in expression of a reporter gene.

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**A Subcutaneous Insulin Absorption Model for  
Critically Ill patients**

Vilkhovoy, Michael<sup>1,2</sup>; Pritchard-Bell, Ari<sup>1</sup>; Clermont, Gilles<sup>1,3</sup>; Parker, Robert S.<sup>1,3</sup>

<sup>1</sup>Department of Chemical and Petroleum Engineering, University of Pittsburgh

<sup>2</sup>Department of Chemical Engineering, University of Massachusetts Amherst

<sup>3</sup>CRISMA Laboratories, Department of Critical Care Medicine, University of Pittsburgh Medical Center

Critically ill patients suffer from “stress hyperglycemia,” a diabetes-like condition of elevated glucose concentrations. Preventing these episodes can drastically reduce morbidity and mortality rates for patients. Currently, physicians maintain glucose levels via strategic administration of insulin, with the intent to return patients to a normoglycemic state. We look to automate this insulin delivery process to improve glucose control, without hypoglycemia, using mathematical model-based tools. Key to performance is a subcutaneous insulin delivery model. Beginning from [1], we add an extra compartment to capture the kinetics of both subcutaneous insulin infusion and bolus injections. The proposed subcutaneous model, in concert with our whole-body patient model, is able to capture data from type 1 diabetics as well as healthy patients. Future coupling of this model to a model-based control algorithm will facilitate clinical decision-making for glucose control and insulin delivery in critical care. [1] Wilinska, M.E. "Insulin kinetics in type-1 diabetes: continuous and bolus delivery of rapid acting insulin," *Biomedical Engineering*, vol.52, no.1, Jan. 2005

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**SLURP: Service Learning in an Undergraduate  
Research Program**

Shober, Lauren; Patel, Shreya; Woodley, Dr. Sarah  
Department of Biological Sciences  
Duquesne University

Service learning combines formal instruction with a related service in the community with the goal of promoting discipline-specific learning. This summer, we combined our undergraduate research experience with a service learning project. We partnered with the Hazelwood-based Center of Life to engage youth in STEM (science, technology, engineering and math) activities via a summer camp for 1-8<sup>th</sup> graders. While working on our individual research projects, we donated 20% of our time to preparing activities for the camp and taught over 80 campers STEM-related activities. Our goals were to increase our ability to problem solve, communicate science effectively, and promote social awareness while at the same time, improve the scientific literacy of campers and inspire them to consider science careers.

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**Intramolecular hydrogen bond (IMHB) strength in dianionic pseudo-chair carboxyphosphate**

Sarah E. Kochanek, Sebastien P. Hebert, Traci M. Clymer, Steven M. Firestine,<sup>1</sup> and Jeffrey D. Evanseck\*  
*Contribution from the Center for Computational Sciences and the Department of Chemistry and Biochemistry, Duquesne University, 600 Forbes Avenue, Pittsburgh, Pennsylvania 15282-1530. <sup>1</sup>Eugene Applebaum College of Pharmacy and Health Sciences, Wayne State University, Detroit, Michigan 48201.*

Dianionic carboxyphosphate is predicted to exist in a novel conformation known as pseudo-chair, stabilized by an intramolecular hydrogen bond (IMHB), which has significant mechanistic implications in understanding ATP-grasp carboxylases important in obesity, diabetes, and microbial infections. To quantify the IMHB strength, the open-closed method, the isodesmic approach, the related rotamers method, the rotational barrier method, and NBO have been employed using Truhlar's Minnesota M06-2X functional and Dunning's aug-cc-pVnZ ( $n=D, T, Q, 5$ ) basis sets. These estimation methods have been evaluated for their ability to provide quantitatively accurate results. We predict that the IMHB strength lies between 12 to 15 kcal/mol in vacuum and 6 to 8 kcal/mol in implicit water. It is noted that the IMHB strength decreases by 50% when in implicit water. The quantitative determination of IMHB strength supports the existence of dianionic pseudo-chair carboxyphosphate in nonpolar environments, such as that found in enzymatic active sites.

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**Improving Cancer Detection Performance of Quantitative Phase Microscopy through Image Registration**

Kita, Stephen; Uttam, Shikhar  
 Departments of Medicine and Bioengineering  
 University of Pittsburgh

An image registration algorithm using algebraic invariants of sequential corners [1] is implemented using MATLAB and applied to images of cells acquired via spatial-domain low-coherence quantitative phase microscopy. Previous screening efforts have shown variation in optical path length difference (OPD) of cell nuclei to be a potential optical biomarker for breast and colon cancer detection [2,3]. For pathological diagnosis the cell nuclei need to be identified using Hematoxylin and Eosin (H&E) staining, which interferes with OPD calculations [4]. In the presented approach, OPD information is taken from unstained slides while nuclei location is extracted afterwards from H&E stained slides and the two images are registered using the aforementioned algorithm. Synthetic and experimental results showing the efficacy of the registration algorithm are presented.

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**Examination of Deep Sequence Influenza Virus Data from a Ferret Transmission Study**

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As part of a larger study to investigate the mechanisms that drive influenza virus evolution and the emergence of new antigenic strains, virus sequence data generated on the Illumina HiSeq2000 system are being compared with that of data collected from the novel PACBIO RS System. The single molecule real time (SMRT) approach of the PACBIO RS provides a unique way to scaffold short HiSeq sequence reads, phase mutations found across the viral genomic segments, and detect genetic variation that might otherwise be overlooked by either methodology alone. Parsed PACBIO data largely confirm SNPs identified by HiSeq and link mutations into variant strains. The combined data from these two platforms will contribute to understanding the frequency at which genetic variation is created within viral hosts and the transmission of strain variants. The success of these goals has the potential to aid in developing better vaccines for influenza.

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**Conformations of key interactions between Acetylcholinesterase and its Inhibitors**

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Acetylcholinesterase (AChE) is a key enzyme in the regulation of the neurotransmitter acetylcholine in neuronal synapses. AChE is inhibited by organophosphate (OP) compounds that covalently bind to the enzymes active site. Reactivation of AChE inhibited by OPs can be facilitated using molecules containing an oxime functionality, but various oximes reactivate inhibited AChE differently and have differing levels of success. To understand why this is the case, we have investigated the motion of AChE alone and complexed with different oximes to see how both the overall structure and active site are affected in a solvated environment. Classical molecular dynamics simulations were used to investigate the stability of various AChE complexes and understand the interactions of oximes with residues in the protein.

**93****KCND2 mRNA formation of G quadruplex structure**Ciccariello, Lauren<sup>1</sup>; Katrancha, Sara<sup>2</sup>; Stefanovic, Snezana<sup>2</sup>; Zhang, Yang<sup>2</sup>; Mihailescu, M. Rita<sup>2</sup>  
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Fragile X Syndrome (FXS), the most common inherited form of mental impairment, is caused by the transcriptional silencing of Fragile X Mental Retardation Protein (FMRP) due to a CGG trinucleotide repeat expansion within the 5'- untranslated region of the *FMR1* gene. FMRP is a translation regulator, one of its targets being the KCND2 mRNA, which encodes for the KCND2 protein, a potassium voltage-gated channel that helps regulate synaptic plasticity. FMRP uses its arginine-glycine-glycine domain (RGG box) to recognize G quadruplex structures formed by a subset of its mRNA targets. G quadruplex structures are formed by stacked planar guanine quartets that are stabilized by potassium ions. Thus, we used the QGRS Mapper prediction software to determine if KCND2 has any regions with the potential to form G quadruplex structures as well as various biophysical methods to characterize G quadruplex formation in the KCND2 mRNA and examine their binding by the FMRP RGG box.

**95 The Effect of Shortening Muscle Contractions on Dystrophic Muscle Histopathology**

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Duchenne Muscular Dystrophy (DMD) is the most common lethal genetic disease in boys. The boy's muscles are very fragile and physical therapy and basic exercises (even riding a bike) are typically discouraged for fear of damaging the muscle, though there is little evidence to support this. The effect of muscle contraction type and intensity has not been examined previously. Our goal was to determine the effect of low intensity and high intensity shortening muscle contractions on muscle pathology in the mdx mouse model of DMD. Methods: mdx mice were randomized to 2 experimental groups and 1 control group for each time point. The experimental groups received either low intensity or high intensity stimulation of the sciatic nerve to produce muscle contraction, three times each week for 3 or 6 weeks. Muscle tissues were collected and H&E staining was performed for the currently ongoing microscopy analysis of tissue pathology.

**94****Machine Learning Algorithms for Mitosis Detection in Whole-Slide, Digital Histopathology Images**Prince, Lauren; Burger, Virginia; Chennubhotla, Chakra  
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A key factor in cancer prognosis is the mitotic index (MI), which is the ratio between the number of cells in mitosis and total number of cells visible in the tissue area. Manual evaluation of MI is tedious, error-prone and subjective. With technological advances in digital imaging, we now have access to digitized whole-slide histopathology images, and these can aid in the development of automated image analysis algorithms for mitosis detection. On a training set of 311 high power fields (HPFs), regions of interests (ROIs) containing mitotic nuclei were identified through Gaussian smoothing and other filtering methods. Principal component analysis was performed on the training ROIs in order to create basis images representing the variance of mitotic nuclei in the data set. Classifiers for detecting mitotic nuclei were then built from the basis images. The detection algorithm was then tested on a data set containing 295 HPFs.