

56th Metropolitan Association of College and University Biologists (MACUB) Conference November 4, 2023 University of Bridgeport

Author Name(s): Bah, Kadiatou; Sarosiek, Kristopher; Inde, Zintis Poster Presentation Title: Role of BAX and BAK in Apoptotic Priming Affiliations: Medgar Evers College Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Apoptosis is a type of cell death in which a series of tightly-controlled steps result in the controlled removal of cells undergoing stress or other pro-death signals. Apoptosis is essential for many normal biological functions, including neurodevelopment, and may contribute to a range of diseases, including cancer, autoimmune diseases and neurodegeneration. Apoptosis is involved in early brain development. It is used to eliminate unwanted cells. However, there is proof that neurons or other cell types in the brain undergo apoptosis during Alzheimer's Disease (AD) and other neurodegenerative diseases. BAX and BAK are two nuclear-encoded proteins present in higher eukaryotes that are able to pierce the mitochondrial outer membrane to mediate cell death by apoptosis. This change can be measured by BH3 profiling. Briefly, this is done by measuring the amount and type of pro-apoptotic signal a given cell requires to initiate apoptosis. BH3 profiling utilizes flow cytometry to measure the release of cytochrome c, a key event in the initiation of apoptosis, after treatment with pro-apoptotic peptides. Cells that require only a weak pro-apoptotic signal to release cytochrome c are considered "primed" for apoptosis; cells that require a stronger signal are considered "unprimed." In order to study the role of two critical proteins, BAK (B-cell-2 antagonist killer) and BAX (B-cell-2 associated x protein), in Alzheimer's disease and other disorders, we have generated cell lines lacking these proteins to analyze using BH3 profiling. Knockouts were made in HeLa cancer cells, allowing for easy culturing and experimental analysis. The cells are collected and prepared into a single-cell suspension, then added to a prepared 96 well plate containing BH3 peptides. The different genotypes of the HeLa cancer cells that were used are, HeLa WT, HeLa BAX -/- (BAX KO), HeLa BAK1 -/- (BAK KO), and HeLa BAX -/- BAK1 -/- (BAX and BAK double KO – DKO). These genotypes are analyzed by flow cytometry and priming is measured by the amount of cytochrome c that is released from the mitochondria. The goal is to compare the different genotypes and see how each cell line responds to the amount of BH3 peptide applied to the cell. We hypothesize that the more primed a cell gets, the more BAX/BAK get activated, which increases the likelihood of cytochrome c release and sensitivity to apoptosis. Using the BH3 profiling assay, we found the HeLa WT cells released more cytochrome c than the BAX KO and BAK KO. The HeLa DKO, which was missing both BAX and BAK, were completely resistant to pro-apoptotic peptides as indicated by the lack of cytochrome c release. These results will enable continued research to determine whether apoptosis in neurons and other cell types plays a role in Alzheimer's disease and to test novel drug candidates to block apoptosis.

Author Name(s): Zafar, Wajiha; Denton, Richard W. Poster Presentation Title: Synthesis of Novel Isoxazolidine-mimetics of DAG-lactones: In Search of New and Specific Protein Kinase C Agonist Affiliations: Medgar Evers College Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Diacylglycerol (DAG) is a prolific second messenger that activates proteins in various signaling cascades. They target a family of protein kinase C (PK-C) isoenzymes. These isoenzymes catalyze the Ophosphorylation of serine/threonine of proteins involved in the signaling pathways that regulate cell growth, differentiation, apoptosis, and the promotion of tumors. A more potent kinase C agonists are the DAG-lactones. The syntheses of these compounds are widely reported in the literature, and they promise therapeutic targets for cancer, dementia, HIV, AIDS, and multiple other disorders. In this project, the concept behind scaffold hopping is utilized to prepare isoxazolidine compounds, which are suitable mimetics of DAG-lactones. We hypothesize that compounds with a specific isoxazoline scaffold containing key functional groups such as a -CH2OH and ester functionalities are possible activators of protein Kinase C enzymes. Several methods were used to purify and characterize the products formed in this project: These are outlined below: 1) flash column chromatography is used to purify chemical mixtures. It is also known as flash purification. It uses a glass column packed with silica as the stationary phase and a solvent – either polar, non-polar or a mix of the two depending on the compounds to separate. This is the mobile phase and is initially added to cover the silica. A layer of sand can also be added to the top, bottom, or both. 2) thin layer chromatography (TLC) consists of a stationary phase as a thin adsorbent material layer, usually silica gel or aluminum oxide, coated onto an inert plate surface, typically glass, plastic, or aluminum; 3) nuclear magnetic resonance (NMR) was used to characterize the compounds isolated. It is a physical phenomenon in which nuclei in a strong constant magnetic field are perturbed by a weak oscillating magnetic field and respond by producing an electromagnetic signal with a frequency characteristic of the magnetic field at the nucleus, and 5) infra red (IR) spectroscopy was used to determine the functional groups present in the molecules based on their vibration mode when irradiated. The results showed that an isoxazolidine scaffold was prepared from the 3+2 cycloaddition reaction between 2-(((tert-butyldiphenylsilyl)oxy)methyl)allyl acetate, and the undecanal oxime was effectively prepared in our laboratory in less than 30% yield. This product is a key precursor to isoxazolidine mimetics of the potent DAG-lactones. The project was able to synthesize an isoxazolidine scaffold, a key compound to novel DAG-lactone mimetics that are possible activators of PKC enzymes. The work is pertinent to the preparation of therapeutics that can manipulate the signaling pathways that regulate cell growth, differentiation, apoptosis, and the promotion of tumors.

Author Name(s): Rumora, Ana; Hopkins, Liliana; Yim, Kayla; Baykus, Melissa Poster Presentation Title: Microbial Fuel Cells Affiliations: Bergen Community College Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Microbial fuel cells (MFCs) are bioelectrical devices powered by the oxidation of organic and inorganic compounds due to microbial activity. Twelve soils from Bergen Community College (BCC) or areas nearby were used to generate mud suspensions and MFCs with the anode buried with the mud, while the cathode rested on top. MFCs were incubated at 37°C with electrical output and numbers of electrogenic bacteria measured using an application developed for iPhones. The most productive MFC generated a maximum of 161 microwatts with 3.38 x 109 electrogenic bacteria after addition of cellulose. Addition of cellulose optimized electrical output and electrogenic bacterial with more than double the numbers compared to previous reported studies. Clones libraries of 16S rRNA genes showed the presence of different types of electrogenic bacteria in the anodes related to bacterial phyla such as uncultured members of Chloroflexi, Bacteroidetes, and Acidobacteria. Some bacteria did not match any known bacterial phylum. Bioelectrical devices such as MFCs provide sustainable and clean alternatives to future applications for electricity generation, waste treatment, and biosensors.

Author Name(s): Nagapurkar, Akash R.; Sun, Jonathan W.; Renfrew, P. Douglass; Montclare, Jin Kim Poster Presentation Title: Developing a Computational Workflow to Model and Target Viable Supercharged States of siRNA-delivering Coiled-coil Proteins Affiliations: New York University Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Short-interfering RNAs (siRNAs) have emerged as promising therapeutic modalities for targeting diseases by silencing specific genes; however, efficient intracellular delivery of these biomolecules remains a major challenge, limiting their clinical translation. This study presents a computational approach to design new positively supercharged variants of the Cartilage Oligomeric Matrix Protein's coiled-coil domain (COMPcc) (PDB: 3V2P) to serve as a non-viral proteinaceous siRNA carrier. New sequences with specific net charges were generated using the Rosetta macromolecular modeling suite and subsequently analyzed for thermodynamic stability and structural alignment with their COMPcc parent. Computationally generated Coiled-coiled Supercharged Protein (CSP) variants exhibit robust Rosetta scores between -900 and -800 energy units. Moreover, low root mean square deviation (RMSD) values that suggest CSP mutants maintain their alpha-helical secondary structure and align well with native COMPcc. Docking siRNA molecules (PDB: 1R9F) reveal more energetically-favorable binding states associated with CSP, indicating strong interactions with the siRNA backbone. Future studies will explore complementary negatively supercharged CSP species that are hypothesized to co-assemble with their positive counterparts into larger supramolecular complexes. Overall, this computational framework offers innovative prospects to rationally design unique sequences, structures, and eventually morphologies that could be leveraged for the next-generation of gene and drug delivery vehicles.

Author Name(s): Tariq, Hafiz; Spence, Nickayla; Felix, Rose; Johnson, Qiaxian; Chauhan, Bhanu P.; Chauhan, Moni; Ghoshal, Sarbani Poster Presentation Title: Synthesis and Anticancer Properties of Polyrhodanine Copper

Nanocomposites.

Affiliations: Queensborough Community College of CUNY

Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Rhodanine (derived from thiazolidine), a heterocyclic compound, plays an essential role in the biological system of humans. Its derivatives are present in drugs used in antibiotics, antiviruses, antidiabetics, and antifungals. We hypothesize that the shape-controlled synthesis of PolyRhodanine will provide an exciting perspective for diagnosing and treating diseases, including cancer. In our research, we investigate the synthesis of PolyRhodanine in a single-step oxidation-reduction reaction in the presence of transition metals in the microwave. Two morphologies for PolyRhodanine have been identified depending on the metal: core-shell and nanotubular. In the first step of the reaction, the rhodamine monomer forms a onedimensional complex with the metal ions (Copper (I) Acetate) due to coordinative interaction. In the second step, the oxidation of Rhodanine and the reduction of metal ions result in the polymerization of Rhodanine into core-shell nano-micro spheres with embedded metal nanoparticles. The product is analyzed via Infra-Red and UV-vis Spectroscopy, SEM (Scanning Electron Microscopy), EDX (Energy Dispersive X-ray spectroscopy), and TEM (Transmission Electron Microscopy). Subsequently, we tested our compound in a human lung cancer cell line, namely A549, to measure cancer cell viability by the colorimetric MTT (3- [4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide) assay. The MTT assay is used to measure cellular metabolic activity as an indicator of cell viability and cytotoxicity. The underlying principle is the ability of NADPH-dependent cellular oxidoreductase enzyme secreted by the mitochondria to convert the tetrazolium dye into insoluble formazan crystals. More formazan crystal formation indicates more viable cells. In our experiment, 20,000 cells were plated in each well of a 96well plate and treated with the compound for 48 hours to investigate the viability of lung cancer cells. Our data shows viability of A549 cells decreases in a dose dependent manner with treatment concentrations from 0.01µM to 1µM in comparison to cells in the DMSO control treatment group. Our present focus is to investigate the cell signaling pathways, by real-time PCR and Western blotting, that could be altered by our compound to identify expression of key genes and protein, that are known to be dysregulated in lung cancer. Future studies will focus on investigating effect in other cancer cell lines, including triple negative breast cancer cells. Acknowledgements: Queensborough Community College NIH Bridges to Baccalaureate Program 5R25GM065096-21 and Research Foundation, CUNY.

Author Name(s): Nidhi Chandra Kannekanti Poster Presentation Title: The protective effect of commercial Moringa Oleifera extract on Chinese Hamster Ovary cells when treated with Chromium. Affiliations: University of Bridgeport Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Moringa Oleifera is an Indian plant found in tropical and subtropical climates all over the world. Its most common names include "drumstick tree" and "horseradish tree. This plant has valuable nutrients, and every part of the tree is useful for either nutritional or economic uses because of its high nutritional value. Furthermore, this plant extracts have antioxidants that can neutralize free radicals preventing cell toxicity.

Chromium (VI) in its hexavalent oxidation state is hemotoxic, genotoxic, mutagenic, and carcinogenic. This metal is a very powerful oxidizing agent and can cause DNA damage.

This study was conducted to investigate the possible protective effect(s) of. M. Oleifera commercial extracts, against Cr (VI) in vitro cytotoxicity.

Chinese hamster ovary cells (CHO) were treated with various concentrations of Cr (VI) {0.25 uM, 0.5 uM and 1 uM)} alone and in combination with extracts of M. Oleifera (0.9 mg, 4mg, 9mg and 17.5 mg). Cell viability was assessed by the Trypan Blue assay. Our results indicate that cell viability decreases as the metal concentration was increased in a dose response fashion. For the combined treatments of the metal with M. Oleifera cell viability was increased at 0.5 μ M and 0.25 μ M Cr (VI) with 0.9 mg Moringa, indicating that M. Oleifera had a protective effect against Cr (VI) toxicity. However, at higher M. Oleifera treatments (4mg, 9mg and 17.5 mg) this extract indicated toxic effects in a dose depended on fashion. Since the M. oleifera plant extracts are widely used in many areas in the world, therefore, caution should be exercised when people using it as a nutritive supplement.

Author Name(s): Zhang, Yi; Tamari, Farshad Poster Presentation Title: DNA Extraction in Hydrangea macrophylla using the Edwards Buffer Method: DNA Extraction of Small Tissue Quantities using Affiliations: Kingsborough Community College Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Hydrangea macrophylla is an economically valuable ornamental crop with many potential medicinal applications. However, molecular research using H. macrophylla and other plants often requires access to the plant's DNA, which can be challenging to extract due to the secondary metabolites and flavonoids present within the plants. Previous work by the Tamari lab has demonstrated that the Edwards buffer method of DNA extraction is superior to the commonly used CTAB method, resulting in higher quality and greater DNA yields (Tamari et al., 2013). This study aimed to investigate whether the Edwards buffer method could be applied to other angiosperm species with comparable DNA yields and, if so, whether the technique is also appropriate for DNA extractions using larger quantities of tissue samples. We hypothesized that 1. The Edwards buffer is suitable for DNA extraction from small tissue samples for another angiosperm species. There will be a linear relationship between the amount of tissues used and the quantity and quality of DNA yield.

A slightly modified protocol from Tamari et al. (2013) was used for the DNA extractions using H. macrophylla. Tissue amounts used were 10, 50, 100, 150 and 200 mg. All samples were prepared in triplicates. DNA concentration was quantified using a spectrophotometer (Lambda XLS, BIORAD). Initial data manipulation was performed on Microsoft Excel, and statistical analysis were carried out using SigmaPlot. Our results show that the DNA quantities obtained for H. macrophylla are comparable to those for Petunia hybrida, supporting our first hypothesis. A linear increase in DNA concentration was observed from 10 to 100 mg, but not beyond that, partially supporting our second hypothesis. The statistical analysis supports this trend. The reasons for the observed plateau are discussed.

Author Name(s): Munn, Laura; O'Connor, Michael; Multani, Harpreet; Scanze, Nina Poster Presentation Title: Leptin exacerbates inflammatory cytokine production within a 3D mouse model of osteoarthritis Affiliations: Molloy University Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Osteoarthritis, a painful chronic disease in which joints begin to deteriorate, affects approximately 500 million people worldwide. Obese individuals are at high risk of developing osteoarthritis due to increased pressure exerted on the joints. Leptin, a hormone that is produced by adipocytes, is elevated in obesity and has been shown to enhance the effects of cartilage degrading matrix metalloproteinases. Due to the inflammatory characteristics of obesity and osteoarthritis, it was hypothesized that high levels of leptin would exacerbate the production of inflammatory cytokines within a 3D model of inflammatory osteoarthritis. 3D chondrogenic cultures were derived from the differentiation of mouse mesenchymal stem cells, which were then maintained within six different environments to determine the impact of leptin on the progression of inflammatory osteoarthritis. These environments included an untreated control (UC), a leptin concentration within normal range (NL, 5ng/mL), an elevated range of leptin (HL, 25ng/mL), an inflammatory control (IC), an inflammatory environment with leptin concentration within normal range (INL), and an inflammatory environment with elevated range of leptin (IHL). The inflammatory environment was created through the use of activated macrophage medium. The cytokine profiles of the conditioned medium from the experimental groups were analyzed via a Proteome Profiler[™] Mouse XL Cytokine Array Kit, which determined the levels of 111 different cytokines within the array. The inflammation-inducing and tissue damaging cytokines, CCL2/JE/MCP-1, CCL3/CCL4/MIP-1 (α/β), and TNF- α , were found in higher concentrations within the IHL group, indicating that high levels of leptin could exacerbate osteoarthritic conditions. Understanding the relationship between leptin and osteoarthritis is critical in determining whether therapies targeting the leptin pathway could be used as future treatments for obesity related osteoarthritis.

Author Name(s): Talmadge, Ta'Meir; Yoh, Andrea; Rivera, Nilka Z. ; Wallace, Connie L.; Marquez, Felipe; Asare, Belinda; Zhou, Chun
Poster Presentation Title: Mechanistic study of the impact of designed mutagenesis on bacterial β-Glucosidase B
Affiliations: Mercy University
Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

β-Glucosidase B (BglB) is an enzyme continuously researched for its potential application in producing alternatives for fossil fuels. BglB catalyzes the hydrolysis of terminal, non-reducing beta-D-glucosyl residues with the release of beta-D-glucose. It is involved in producing liquid fuel (glucose) from the hydrolysis of carbohydrate polymers, such as cellulose. Previous studies have shown that certain mutations on this enzyme can either improve or reduce the enzymatic activity or stability. The present research is aimed to understand the mechanisms by which certain mutations can change enzyme activities. Specifically, we focused on two amino acids, leucine at position 171 (L171) and asparagine at position 220 (N220). Polypeptide segments of BglB, which form the active sites, were identified using Foldit Standalone computational software which runs the Rosetta protein modeling software. Using this software, we're able to create point mutations of L171 by replacing leucine with phenylalanine and tryptophan to create the mutations L171F and L171W. We also made mutations of N220 by replacing asparagine with threonine and arginine, N220T and N220R. We hypothesized that changing leucine to aromatic residues or modifying the hydrogen bond at position 220 might affect the enzymatic activity. Utilizing Kunkel Mutagenesis, we generated the L171F, L171W, N220T, and N220R mutations. We confirmed the mutations via DNA sequencing. The mutant proteins were then purified from bacteria along with the wild-type protein using Ni-NTA affinity chromatography. To test the enzymatic activity of these mutations, standard colorimetric assays were performed. While mutations N220T, N220R, and L171W did not show measurable activity, L171F showed robust enzymatic activity. Thus, three independent kinetic assays were completed, followed by two independent assays for thermal stability. Compared to the wild-type enzyme, L171F exhibited a slight increase in the binding affinity during enzymatic assays but had the same thermal stability. Further testing of L171F will need to be done to fully confirm the thermal stability of this mutation. The results of the kinetic reading for L171W, N220T, and N220R might be due to a loss of enzymatic activity during the purification or storage processes, which needs to be clarified through additional experiments. The results support our hypothesis that the L171F mutation presents higher enzymatic activity as a result of the aromatic biochemical property of phenylalanine. The findings of the present study and future studies will benefit ourselves and others in further understanding the mechanisms that mutations can change enzyme activity or thermal stability.

Author Name(s): Elizabeth Bahar; I. Alexandra Amaro; Mariana Wolfner
Poster Presentation Title: Seminal Fluid Proteins and Post-Mating Responses: Exploring Protein
Binding and Pathways in Aedes aegypti
Affiliations: Cornell University
Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Aedes aegypti mosquitoes transmit diseases such as dengue and Zika, posing a substantial threat to public health. One way to control the spread of these serious diseases is to control the prevalence of the mosquitoes that transmit them. As a step to do so, we would like to characterize the reproductive molecules in this insect, in particular the seminal fluid proteins (SFP) derived from males' accessory glands that modulate their mates' post-mating responses (PMR) and thus fertility. The Wolfner/Harrington labs have identified several SFPs that their data suggest may regulate females' PMR. By investigating the binding partners for these SFPs, our research aims to uncover the underlying reasons for their impact on PMR, as our results can identify how SFPs interact with the reproductive system of Ae. aegypti. To achieve this, we will use a new high-throughput screening method called BASEHIT, in which a protein of interest is tested for binding to a library of Ae. aegypti proteins. This study's main objective is to generate query proteins in a mammalian cell culture system. The gene sequence encoding the query protein was obtained and Gibson assembly was used to insert it into a plasmid designed for the expression of cloned candidate SFPs in tissue culture cells. After verifying the plasmids, we will introduce them into tissue culture cells and then purify their expressed protein that will be used to screen the BASEHIT mosquito protein library. I have successfully cloned 10 target seminal fluid proteins including, a metalloprotease and C type lectin to name a few which are known to have reproductive roles in insects. This work, followed by screens of the BASEHIT library, will contribute to our deeper understanding of the intricate molecular mechanisms involved in reproductive biology and sexual interactions specifically in Ae. aegypti.

Author Name(s): Ishwarya Krishna, Jessica Wong, Jacob Kronenberg, Maria Kulapurathazhe, Dustin Britton, Nada Haq-Siddiqi, and Jin Kim Montclare Poster Presentation Title: Protein Engineered Hydrogels for Wet Adhesives Affiliations: New York University Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Wound healing involves a stage of inflammation, which can be prolonged in patients with diseases such as diabetes, leading to discomfort and poor patient outcomes. Hydrogels have the potential to promote wound recovery, serving as depots for sustained drug release and scaffolds for tissue repair. In particular, wet adhesion is important for wound healing because it ensures that the material will be held in place in the presence of bodily fluids such as blood. Here we describe the design and synthesis of a triblock protein polymer bearing the non-canonical amino acid, dihydroxyphenylalanine (DOPA). We conduct physicochemical characterization involving circular dichroism, UV-Vis spectroscopy, Fourier-transformed infrared spectroscopy, and mechanical testing on the protein engineered hydrogel bearing DOPA. The characterization of a non-DOPA containing negative control and our triblock polymer with DOPA confirmed the adhesive properties of DOPA. The results confirm the feasibility of our DOPA-containing triblock polymer as a material for a wet-adhesive hydrogel drug delivery system.

Author Name(s): Hintelmann, Thomas; Enny, Olivia; Schievelbein, Mika; Tilton, James Tilton; Ouellet, Jonathan
Poster Presentation Title: Creating a Cost Efficient Method to Determine Theophylline Riboswitch Activation
Affiliations: Monmouth University
Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

A riboswitch is a small piece of RNA in structure that binds a molecule to itself to turn on

or turn off a gene. As a result, riboswitch regulation provides an opportunity to develop targeted therapies for various diseases. Currently one of the only ways to determine activation of a riboswitch in an organism is to perform Fluorescent Activated Cell Sorting (FACS). This machine requires a large amount of upkeep and a trained technician to use. Most smaller research universities do not have the resources to be able to use this type of machine which greatly limits the amount of research that can be performed on riboswitches. This project is built based on 3 different cloning methods; Gibson assembly, Golden Gate assembly, and PCR assembly. To test this, the lab is currently taking plasmids and performing these assemblies using fluorescent genes, GFP-UV and mCherry, as well as the sequence for the theophylline riboswitch. Once these genes are cloned, the hope is to view under UV light which of the fluorescent genes are expressing. The amount of expression will be quantified using ratio-metric fluorescence. If GFP-UV and mCherry express, this will tell that the theophylline riboswitch is active. If only mCherry is active this will tell theophylline is riboswitch is inactive. These measurements can be used to measure activation of other riboswitches as well Author Name(s): Baiju, Rabina
Poster Presentation Title: Effect of mineral sunscreen in protecting plasmid DNA damage induced by UV light
Affiliations: University of Bridgeport
Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Ultraviolet radiation (UV) from the sun is a well-known environmental factor which induces substantial DNA damage leading to various skin conditions and increased risk of skin cancer, both in human and environmental DNA, such as plasmid. Sunscreens, particularly those containing mineral filters such as zinc oxide and titanium dioxide, are widely used to protect the skin from UV-induced harm. In this study we compare the mineral sunscreen efficacy related to prevention of UV induced plasmid DNA damage with a focus on understanding the potential application in environmental protection and sunblock development.

The experimental approach involved subjecting a plasmid DNA sample to a range of high controlled UV light with 8- watt bulb, machine (wavelength 312nm) for 5, 10, 20 and 30 minutes in the presence and absence of three different brands of commercial mineral sunscreen: Cera Ve 30 SPF, Eucerin 30 SPF and Cetaphil 50 SPF. More specifically plasmid within a tube was covered outside with sunscreens and then the tubes were irradiated with UV light. DNA damage induced by UV was assessed using qPCR technique. Our results indicated that UV radiation significantly damages plasmid DNA leading to strand breaks and alteration in its structural integrity. Damage increased with increasing time of UV exposure. Exposure for 30 minutes indicated higher DNA damage (92.742%) while for 5 minutes treatments being the lesser damage (50.3%). However, the application of mineral sunscreen provided a substantial protective effect, reducing DNA damage as evidenced by low quantification cycle (Cq) value. Plasmid DNA without any sunscreen application showed high Cq value suggesting less amplifiable DNA. Sun Protection Factor (SPF), which is thought to be the primary determinant of sunscreen effectiveness, however did not demonstrate any appreciable difference in the level of protection between 30 SPF and 50 SPF. Eucerin 30 was found to be the most effective of all. The protective strength of Cetaphil 50 was found to be the most consistent while Cera Ve 30 and Eucerin 30 depicting changes in their protection level throughout the exposure despite the Cq value for Cetaphil exposure was high in compared to other two sunscreens. All the treatment reached more or less a plateau after certain time interval suggesting either a complete damage or effective sunscreen application. This protective effect was attributed to the ability of the sunscreen's mineral components to act as shield and scatter UV rays, preventing their penetration into the DNA strands.

These findings indicate that mineral sunscreen formulated with zinc oxide and titanium dioxide effectively protect plasmid DNA from damage induced by UV radiation. Further research is necessary to explore the broader applications of mineral sunscreen in protecting genomic DNA.

Keywords: mineral sunscreen, UV exposure, plasmid DNA damage, qPCR, DNA protection, zinc oxide, titanium dioxide, skin cancer prevention.

Author Name(s): Pellerito, Madison; Ouellet, Jonathan Poster Presentation Title: Catalysis Chemistry of the I-R3 DNA Enzyme through Ion-Promoted Cleavage Affiliations: Monmouth University Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

DNA enzymes are synthetically engineered deoxyribozymes that function in hydrolyzing DNA, breaking the phosphodiester bonds that are important to the long term storage of genetic information. In the I-R3 DNA enzyme, there is a catalytic core consisting of 17 nucleotides that form an asymmetrical loop when the DNA enzyme is annealed to the DNA substrate. When this structure is in the presence of zinc chloride under neutral pH, the substrate strand will be cleaved between two adenosines in positions A15 and A16, resulting in the creation of a 5' product and a 3' product.

This project is designed to study the cleavage behaviors of the I-R3 DNA enzyme through the introduction of different metal ions in place of the known zinc ions that promote cleavage. Metal ions were chosen based on their atomic radii size and their overall similarities to the atomic radius size of zinc. The current hypothesis is that metal ions with similar atomic radii to the zinc ions may promote cleavage of the DNA substrate when bound to the I-R3 DNA enzyme. At the discovery of the I-R3 DNA enzyme, Zn2+ allowed cleavage, while Cd2+, Co2+, Cu2+, Ni2+, Mn2+, Ca2+, and Mg2+ did not allow cleavage.

The chosen metal ions will be added to 100 pmol DNA enzyme, 10 pmol DNA substrate, 50 mM HEPES pH 7.05, and 0.1M NaCl after an initial heating of 97°C and slow cooling to 37°C. Then after 5 minutes the sample will be quenched with a denaturing formamide solution, will be run on a 20% acrylamide gel, and then stained with SYBR gold. The gel will then be analyzed by photodensitometry to find the percent of cleavage.

Understanding the correlations between metal ions and cleavage of the I-R3 DNA enzyme would have a significant impact on the understanding of deoxyribozymes and their overall catalytic function. This research could also have further biological and medicinal applications with targeting and cutting single stranded viral DNA, such as parvovirus most commonly seen in dogs.

Author Name(s): Moparthi,Naveen; Mudu,Pavan;Muttineni,Keerthan;Kandi,Sindhu Poster Presentation Title: Development and Porosity Analysis of PLA Polymer Scaffold for Tissue Engineering Applications Affiliations: University of Bridgeport Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Poly(lactic acid) (PLA), a biocompatible and biodegradable polymer, has gained prominence for its potential in scaffold design. Its biocompatibility and tunable porosity make it a promising candidate for various tissue engineering applications, including bone regeneration, cartilage repair, and wound healing. The scaffold's porosity is tailored to promote effective nutrient transport, cell infiltration, and tissue regeneration. The development of this controllable porosity porous PLA scaffold represents a significant advancement and opportunity in the field of tissue engineering. Therefore, it becomes particularly important to optimize the scaffold's mechanical properties and functionalization for specific tissue types, enhancing its potential applications in the field of regenerative medicine. In this poster, we present a study on the development of a scaffold using PLA polymer and the assessment of its porosity. By evaluating its biocompatibility through cell compatibility testing, we can determine whether the PLA polymer is suitable for tissue regeneration and its potential applications.

Author Name(s): Houseknecht, Alexa; Rosen, Maxwell; Jose, Davis Poster Presentation Title: Investigating local conformational changes from ligands on G-quadruplex complexes using fluorescent base analogues Affiliations: Monmouth University Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

DNA sequences rich in guanines readily fold to form quadruplex structures, which are bound by Hoogsten-type hydrogen bonding of four guanine nucleotides (G4). These G-quadruplexes (GQs) make up many aspects of structural DNA, including the telomeres which function as a protective barrier to uphold the integrity of the chromosome. GQs are important structural components involved in many physiological functions including limiting telomerase activity which is seen in 85-90% of human tumor cells. Telomerase activity can be influenced by introducing small molecules that can interact with GQs. This interaction of small molecules can alter the stability and local conformations of the GQ at the guanine tetrad level which in turn can affect the telomerase activity and cancer progression.

To identify changes in the local conformations of the telomeric sequence upon interaction with small organic molecules, we incorporated 6-methylisoxanthopterine (6MI), a circular dichroism (CD)-active fluorescent base analogue of guanine in place of guanine at distinct positions in the human telomeric GQ sequence. Three variations of DNA sequences were used, 22AG, 22AG6MI, and 22AG6MIup to monitor the conformational changes at different locations of the GQ structure. Past studies investigated binding of (5,10,15,20-Tetrakis-(N-methyl-4-pyridyl)porphine (TmPyP4), a telomerase inhibiting ligand, to the GQ but only addressed their interaction in a global conformational perspective. In this study, we used the fluorescent base analogue to track the local conformation at individual G-tetrad levels using various spectroscopic methods.

The results demonstrated an initial stabilization followed by destabilization of 22AG DNA sequence with increasing ratios of TmPyP4. However, with 22AG6MI and 22AG6MIup strands stabilization was observed at all concentrations with the sequence 22AG6MIup showing a greater stabilization than 22AG6MI. The results suggest by using site-specific fluorescent probes we can monitor the structural and stability changes in GQs.

This method of studying ligand interactions with fluorescently active base pairs can be used to understand small molecule interaction at individual G-tetrad levels. We hypothesize that this new site-specific approach might help to identify more targeted drugs to treat cancer and other telomere-related diseases.

Author Name(s): Farhath Ayesha Poster Presentation Title: The Interaction of Glutathione with Nickel, and Chromium in the Induction of Cytotoxicity in Chinese Hamster Ovary Cells Affiliations: University of Bridgeport Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Glutathione is a well-known antioxidant, present in all mammalian cells as the most abundant non-protein thiol defending against oxidative stress, as a result it can protect DNA damage caused by reactive oxygen species.

In This study we investigated the possible protective effect(s) of Glutathione against Nickel (Ni II) and Chromium (Cr VI) toxicity. Chinese Hamster Ovary cells (CHO) were treated with various concentrations of Ni (II) {1uM, 5uM, 10uM, 25uM} and Cr (VI) {0.5uM, 1uM, 2.5uM, 5uM}, alone. Furthermore, we treated cells with Ni (II) {1uM,5uM,10uM,25uM} combined with glutathione {0.006M, 0.01M,0.02M} as well as, Cr (VI) {0.5uM, 1uM, 2.5uM, 5uM} with glutathione {0.006M, 0.01M,0.02M}. The Cell viability was assessed by the Trypan blue assay.

Our results indicate that cell viability decreases as the metal concentration was increased. Specifically, as the concentration of Ni (II) and Cr (VI) increases, the cell viability for both metals decreased in a dose dependent fashion. For the combined treatments of both the metals with glutathione, we observed that cell viability increased when the cells were treated with 5uM, 10uM, 25uM of Ni (II) and 0.01M glutathione as well as 1uM,2.5uM, 5uM of Cr (VI) and 0.01M glutathione, indicating that glutathione had a protective effect against Ni (II) or Cr (VI) toxicity.

No protective effect was seen at lower concentration (0.006M) of glutathione treatments. However, at higher levels of glutathione (0.02M) the cell viability decreased, indicating a toxic effect of glutathione.

This study provides information about the interplay between glutathione, Ni, and Cr treatments and their effects on cell viability. Furthermore, our results indicate the importance of maintaining an optimal concentration of glutathione in protecting heavy metal-induced toxicity and caution should be exercised when glutathione is used as a dietary supplement.

Author Name(s): Eleanor Parks-Orr and Jonathan Ouellet Poster Presentation Title: Spinach Aptamers Affiliations: Monmouth University Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Baby Spinach RNA aptamer that has been designed by SELEX to make an RNA fluorescent similar to GFP. It has been shown by X-Ray crystallography structure that the spinach contains contains a G Quadruplex capped at the end by triple base pairs in a triangular plane. The G Quadruplex is a structure of DNA or RNA that has coiled up to the point of forming layers. Four Guanines (G) all exist on the same physical plane from each other forming hydrogen bonds, this forms a tetrad. Three tetrads are stacked on top of each other. G Quadruplexes are found at the ends of DNA, as DNA replicates over time the G Quadruplexes are lost and DNA shorterns. This causes aging. Telomerase is an enzyme that rebuilds the ends of the DNA and slows the process of aging–however–too much telomerase activity is a symptom of cancer cells.

Baby spinach aptamer's melting temperature is unreported in the scientific community, as it is a fairly new model to work with. The spinach aptamer is also a short RNA sequence, containing 51 nucleotides. For these reasons it was chosen to be used to study G Quadruplexes stability.

For the spinach aptamer to be fluorescent, it needs a dye. DFHBI is one of the several fluorescent dyes used for spinach. Its structure allows the molecule to rotate between two central carbons in the molecule and vibrate when free in solution. It's energy dissipates when it vibrates, without its energy the dye will not glow. DFHBI will slot itself between the top tetrad of a G Quadruplex and the three base pair gap. These boundaries will stop DFHBI from rotating and expelling energy, causing the molecule to glow green. If DFHBI is glowing in the presence of a G Quadruplex, it proves that the G Quadruplex is properly folded.

Finding the melting point of G Quadruplex is the same thing as finding the temperature in which the G Quadruplex breaks. By using a fluorescence instrument to scan the levels of fluorescence in a solution of spinach aptamer at different temperatures, the melting point can be found.

A small chemical compound (ligand) called TMPyP4 has been found to stabilize the G Quadruplex. If TPMyP4 (as well as organic compounds made by an organic chemist collaborator) is added to a solution of RNA spinach aptamer and DFHBI, the melting temperature will rise.

Because the G Quadruplex is folded up DNA or RNA, it is bulky and large. Ribosomes are slowed by its bulk, as it is hard to read, and slows the synthesis of proteins in translation regulation. By understanding the melting temp of the G Quadruplex with ligands through the usage of DFHBI, it can provide better understanding in translation regulation of messenger RNAs and viral RNAs and the structure of G Quadruplex as a whole.

Author Name(s): Deirdre Campbell; Jonathan Ouellet Poster Presentation Title: Isolation of an Aptamer Selective to Glucose Affiliations: Monmouth University Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Diabetes is a disease that hundreds of million people live with daily throughout the world. Although this is disease is typically not fatal, it can be if not treated properly. The day to day life of a person with diabetes consists of blood sugar monitoring by finger pricks, insulin injections and strict diet. The research for a glucose aptamer would be the first step to eliminate the need for all of this. This project uses Systematic Evolution of Ligands by Exponential Enrichment, or SELEX, to select RNA that binds specifically glucose. The process is a cycle beginning with a PCR from a pool of millions and billions different DNA sequences, then transcription to RNA, negative selection, positive selection, and reverse transcription back to DNA. The conclusion of the reverse transcription is the beginning of the next generation where each generation becomes more selective to glucose. Eventually the RNA would be sequenced and converted to a riboswitch. A riboswitch is a sequence of untranslated mRNA that can bind a specific ligand, in this case glucose, and transmit a signal to the expression platform to start the reaction to make a protein. For this project the riboswitch would begin the production of insulin only in the presence of glucose. By making insulin outside of the pancreas, diabetes patients would no longer need insulin injections or constantly monitor their blood sugar levels. The project is currently on its 25th generation and is continuing to move forward. Once we obtain a high ratio of positive over negative cleavage percentages we will begin the process to clone DNA and individually test sequences to find an aptamer that cleaves only in the presence of glucose.

Author Name(s): Oliver, Crystal; Ghoshal, Sarbani; Sullivan, Regina; Rajapakse, Harsha Poster Presentation Title: Extraction of Low Molecular Weight Proteins Derived From Murraya koenigii (L.) for their Bioactivity Affiliations: CUNY - Medgar Evers College Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Murraya koenigii (curry tree) plants are a therapeutic staple in South Asian cultures. The plant has a broad spectrum of properties to treat ailments, including antioxidative, anti-inflammatory, antimicrobial, anthelminthic, and wound healing. Additionally, one specific study showed that curry leaf extracts could induce cell death in two human breast carcinoma cell lines, MCF7 and MDA-MB 231 by inhibiting proteasome activity. For all these purposes, the curry tree plant has been a mainstay in South Asian traditional medicine and can potentially be used as an anticancer agent. Previous studies have isolated phytochemicals from different structures in the M. koenigii plant as well as proteins weighing more than 30 kDa in the leaves; however, there are no studies researching the functions of the plant's low molecular weight proteins. Low molecular weight proteins have emerged as a molecule of exploration in recent years. Studies focusing on low molecular weight proteins in mulberry (Bombyx mori (L.)) leaves and neem (Azadirachta indica) seeds have been shown to possess antioxidative, antimicrobial, anthelminthic, and antineoplastic properties once isolated. In this study, we isolated low molecular weight proteins of M. koenigii from freeze dried leaves using pH 7.4 Tris-HCl and precipitating with TCA/acetone. Our present study is focused on treating these small molecular weight proteins isolated from curry leaves in a lung cancer cell line, A549 to understand its effect on cell death.

Author Name(s): Abdur-Rehman Hussain; Taimoor Chaudhry; Jeleeta Jolly; Azza Gener; Muntaha Ahmad; & Dr. Jacqueline Keighron
Poster Presentation Title: Enhancing the stability and activity of glucose oxidase through interaction with highly curved gold nanoparticles
Affiliations: New York Institute of Technology
Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Interactions with gold nanoparticles have been shown to enhance the activity of enzymes, such as glucose oxidase (GOx), due to the stabilizing effects of the highly curved surface. The enhanced enzyme activity of glucose oxidase can be applied to the area of biosensors. Glucose oxidase is used here as a low-cost model for comparison with glutamate oxidase. By measuring the activity of glucose oxidase-gold nanoparticle conjugates, we optimized the interaction between the enzyme and the nanoparticle to enhance the design and function of enzymatic biosensors for glutamate, an important neurotransmitter. Overall, our results indicate that when glucose oxidase is adsorbed onto highly curved nanoparticles in a single, densely packed layer, its activity is enhanced compared to that of the enzyme free in solution.

Author Name(s): DuBois, Cora; Burdowski, Allen; Kita, Katsuhiro Poster Presentation Title: Combination of nano-hydroxyapatite/gelatin cryogels and 3D printing to manufacture scaffolds mimicking bone microenvironment Affiliations: St. Francis College Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

The field of regenerative medicine relies heavily on biomaterials, which are materials obtained from living creatures or engineered to interact with biological systems. An era of medical interventions targeted at restoring biological function and improving patient outcomes has been brought in by this interdisciplinary area, which utilizes biomaterials to help regenerate, repair, or replace damaged tissues and organs. Regenerative medicine, which attempts to replace or repair damaged human tissues and organs, heavily relies on biomaterials. In this sector, the selection of biomaterials is crucial because they offer a scaffold or support structure that allows cells to proliferate, differentiate, and eventually form functional tissues. Bone fractures are quite common in traumatic injuries, which account for nearly 80,000 deaths of adults younger than 45 in the United States alone. This number is generally higher than death tolls by infectious diseases (except the COVID-19 pandemic). Thus, the development of biomaterials that help bone and spinal cord regeneration is very important to improve the aftermath of traumatic injuries. Compared to wound coverages, the development of bone-mimicking scaffolds using biomaterials requires more studies, partly because biomaterials are mechanically weaker than synthetic materials. In addition, very commonly used types of materials, such as hydrogels, do not really recapitulate the microenvironment inside of the bones. Cryogels, in contrast, can provide a porous-like structure that mimics the bone microenvironment. In addition, our ongoing study has shown the mechanical properties of gelatin-based cryogels can be improved by combined with other materials. Therefore, we attempted to combine nano-hydroxyapatite (nHA) and gelatin to prepare cryogels as potential biomaterials to manufacture bone-mimicking scaffolds. To synthesize nHA, CaHPO4 2H2O and CaCO3 were mixed in a 2.5M NaOH solution and then reacted at 75°C for one hour. The resulting nHA powder was then collected by centrifugation (6,000×g, 10 minutes). Then nHA was washed with water four times. Finally, nHA slurry was dried at 70°C for 24 hours to obtain nHA powder. nHA-gelatin cryogel preparation is based on a recent report (K.T. Shalumon et al. (2019) Mater. Sci. C, 104, 109855). An equal volume of 10% gelatin solution (containing 0.02M glutaraldehyde) and 5% nHA solution were mixed. After mixing (final concentration: 5% gelatin, 2.5% nHA, and 0.01M glutaraldehyde), the mixture was poured into an in-house-made, disposable syringe-based mold. Then the mold was left at room temperature for 30 minutes before freezing at -20°C for 16 hours. After thawing, nHA-gelatin cryogels were ejected from the mold by gently pressing the piston, followed by a quick rinsing and quenching of the remaining aldehyde with NaBH4. The ultimate objective is to create a cast that resembles the shape of teeth and bones using a 3D printer. We obtained the shape design from the National Institute of Allergy and Infectious Diseases/NIH 3D resource. The image or edited image is used to print the prototype of the mold using Grab CAB software. We are currently designing the teeth- and bone-shaped, nHA-gelatin cryogels as the platform to grow human mesenchymal stem cells to differentiate them into osteogenic lineages.

Author Name(s): Trinh, Anh; Awawdeh, Abdelrahman; Cardenas, Samanta; Vetterlein, Amanda; Thompson, Robert
Poster Presentation Title: Spatial and Temporal Targeting of Cre/LoxP Recombination Using Photocaged 4-Hydroxytamoxifen
Affiliations: Mercy University
Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Cre/LoxP is a of site-specific recombination that can be employed in vivo to initiate deletions, insertions, and translocations of genes. In this system, Cre recombinase catalyzes the recombination of genes that are flanked by LoxP sequences. CreER recombinase is a fusion protein of Cre recombinase and the estrogen receptor (ER). This ligand-dependent Cre recombinase can be activated upon binding to the synthetic estrogen receptor ligand 4-hydroxytamoxifen. Photocleavable protecting groups are tools that allow for the controlled release of biologically active molecules in vivo using UV light. Photocaged 4-hydroxytamoxifen would allow for the spatial and temporal release of 4-hydroxytamoxifen using UV light. This, in turn, could create localized knockout models in transgenic organisms containing CreER/LoxP constructs. Here we report on our progress in synthesis of a photocaged 4-hydroxytamoxifen derivative. The photocleavable group (2-(4-methanesulfonyl-2-methoxy-5-nitrophenoxy)-N-(2-(N-tertbutoxycarbonylamino)ethylamine)acetamide) is ligated to 4-hydroxytamoifen to form photocaged 4-hydroxytamoxifen. Our progress on the synthesis and characterization of photocaged 4-hydroxytamoxifen is exemplified using infrared and nuclear magnetic resonance spectroscopy data.

Author Name(s): Pathak, Samya; Ghoshal, Sarbani Poster Presentation Title: Identification of Genetically Modified Plant Products Using PCR Affiliations: Queensborough Community College of The City University of New York, Bayside, NY Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

For centuries, human beings have used selective and conventional breeding techniques to enhance crops' yield, as well as desirable qualities in plants. Conventional breeding techniques may yield undesirable characteristics, and can also be time consuming. Modern biotechnology techniques have accelerated the process, where scientists can directly manipulate a DNA sequence to generate desirable traits, in addition to protecting plants from insects, and making products more nutritious. New genes can be inserted, deleted, or mutated by recombinant DNA technology, thereby giving rise to genetically modified (GM) organisms. Since there have been a lot of controversies regarding GM, the US government agencies maintain a strict vigil about the safety of such GM products. In the present work, we have determined whether certain commonly available food products have been genetically engineered. We isolated DNA from such plant products, and analyzed whether those were GM by Polymerase Chain Reaction (PCR), using three sets of primers directed at chloroplast gene, CAMV promoter and cry1f gene. Presence of a chloroplast band at 500bp confirmed that our DNA isolation, and PCR worked well. A band at 200bp for CAMV promoter denoted that the plant product was genetically modified, unless the product is a crucifer. CAMV promoter is usually used to introduce a new gene into the plant. Similarly, a band for cry1f gene, which provides insecticidal properties to the plant, denotes the product is genetically modified, as cry1f gene is not naturally found in plants. Importantly, our experiment identified a multitude of food products, especially a few that were marketed as organic, to be GM. Since a major limitation of our experiment is the number of primers used to identify a transgene, it is possible that a product we identified as non-GM, could still be a GM and has other genes that were not investigated in our experimental set up. In summary, we developed and utilized a robust and easily scalable PCR method for identification of genetically modified food products. Such methodology could prove to be highly useful in GM classification of plants produced from various origins.

The work is supported by a research grant provided to Dr. Ghoshal, from the Professional Staff Congress (PSC) CUNY.

Author Name(s): Itani, Riad ; Gonzalez, Zenovia ; Matthews, Janae ; Mahase, Syann ; Alokam, Yacoub ; Atiq, Daniyal ; Frias, Maria A.
Poster Presentation Title: Discovery of PLD Genomic Alterations in Human Cancer
Affiliations: St. Francis College
Category: Clinical (C)

Abstract

The major hallmarks of cancer are uncontrolled cell growth, division and survival, sustained by altered metabolism. Phospholipase D (PLD) controls all these cellular functions, therefore we hypothesized that PLD genomic alteration leads to cancer development.

To test our hypothesis, we searched for PLD1/2 genomic alterations in The Cancer Genome Atlas (TCGA) dataset. TCGA was a joint effort between The National Human Genome Institute and The National Cancer Institute (both NIH) between 2005 and 2018. The dataset of TCGA has genome sequences of 20,000 human samples (cancer and matching normal tissue), including 32 different types of cancer.

In this study, we found that the alteration landscape of PLD1 and PLD2 genes is very different. PLD1 is altered in 8% of TCGA cancer patients, while PLD2 is altered in 2%. The main PLD1 genomic alteration is amplification (gene duplication), while the main PLD2 alteration is single-base mutation. Crucially, PLD1 gene duplication is present in 35% of lung squamous cell carcinomas and reduces patient median survival by 20 months (almost 2 years). PLD2 had no impact on the clinical outcome survival.

This study provides the first attempt at determining the prevalence of PLD genomic alteration in human cancer. Because PLD controls cellular physiology as part of signaling pathways, future studies will evaluate the relationship between PLD and other gene alterations in the same pathway and how that contributes to cancer development.

Author Name(s): Steve Joseph Kuppili; R Santosh Kumar; Prabir K. Patra ; Peiqiao Wu Poster Presentation Title: Design, Optimization and Evaluation of Rifampicin Fast Dissolving Tablets Employing Starch Tannate by Direct Compression Method Affiliations: University of Bridgeport Category: Clinical (C)

Abstract

Fast-dissolving tablets, also known as fast-dissolving orodispersible tablets (ODTs), are solid dosage forms that disintegrate or dissolve rapidly (usually within a few seconds) in the oral cavity without the need for additional water. They offer an effective and convenient way of administering medication, particularly for patients who have difficulty swallowing traditional tablets or capsules. Various technologies can be used to formulate mouth-dissolving tablets. Among them, direct compression is one technique that involves incorporating super-disintegrants or highly water-soluble excipients in the formulation to achieve rapid tablet disintegration. Direct compression does not require the use of water or heat during the formulation procedure and is the ideal method for moisture and heat-liable medication. This work aims to formulate and characterize the fast-dissolving tablets of rifampicin by employing optimization techniques for rapid drug dissolution and absorption. In the present investigation, starch tannate, a new polymer was prepared to develop mouth-dissolving tablets of rifampicin to enhance its oral bioavailability, and it is evaluated for the application as a super disintegrant in fast-dissolving tablets.

Author Name(s): Solis, Valeria; Morkos, Celine; Vilchis-Ibarra, Cindy; Kita, Katsuhiro Poster Presentation Title: FLG mutations as a potential biomarker candidate to predict the risk of pancreatic cancer Affiliations: St. Francis College Category: Clinical (C)

Abstract

This year, there has been a total of 1.9 million new cancer cases and there is an estimate of 1,670 deaths per day. According to The American Cancer Society, there are 64,050 people diagnosed with pancreatic cancer in 2023 Out of the 64,050 around 33,130 are males and 30,920 are females. The mortality rate is extremely high with the expectance of at least 50,550 people dying from this disease. Pancreatic cancer is accounted for 3% of all cancers and 7% of all the cancer deaths in the United States. It is a malignant disease where cancer cells form in the tissues of the pancreas. It is usually fatal as it is typically diagnosed at the late stages. In addition, there is few treatments available and the five-year survival rate after the diagnosis is only 12% (glioblastoma: 6.9%). Thus, besides early detection, the development of useful biomarkers would be an urgent task. As the incidence of pancreatic cancer varies among races, it may make sense to compare the overlapping genes that are most frequently altered genes in both TCGA (The Cancer Genome Atlas) and ICGC (International Cancer Genome Consortium) datasets. There are 15 commonly mutated genes among TCGA and ICGC data (TTN, TP53, MUC16, ZFHX4, FLG, LRP1B, SYNE1, RYR2, CSMD3, USH2A, MUC17, DNAH5, FAM135B, COL11A1, RYR3) in lung cancer. One of them, FLG, is very frequently mutated (20%) in lung cancer data from cBioportal, however, it is hard to tell if FLG mutation can be a biomarker in lung cancer. When we take a look at FLG mutation in pancreatic cancer, 60 FLG mutations (missense and nonsense) were found. 88.3% were missense mutation and 11.7% were nonsense mutations. FLG, also known as filaggrin, encodes a protein that collects keratin mesenchymal structures in the human epidermis. Although apparently, the FLG gene is unlikely to be related to pancreatic cancer, our finding connects with one recent report (M. Cotterchio et al. (2015) PLoS One, 10, e0125273) pointing out the correlation between atopy-related genes, including FLG, and a higher risk of pancreatic cancer. Hence, our findings may shed a light on FLG in pancreatic cancer as a potential prognostic biomarker. When we focus on the patient data with known overall survival status, 63.33% of patients with FLG mutations were deceased, compared to 36.4% of living status. The percentage for deceased is higher as there is lack of treatment options. FLG mutation can potentially be used to find a high risk population in pancreatic cancer.

Author Name(s): Marin, Daisy; Hinkley, Craig S.; Nielsen, Lilja Poster Presentation Title: The FOXL2 Gene is Present in the Eastern Oyster (Crassostrea virginica) Affiliations: Kingsborough Community College Category: Developmental Biology and Genetics (DBG)

Abstract

Crassostrea virginica, also known as the Eastern oyster, can be found along the East coast of North America. The Eastern oyster plays an important role both environmentally and economically, thus it is essential to develop a clearer understanding of oyster population maintenance. In bolstering oyster populations and understanding their life cycle (especially breeding), it is of note that the Eastern oyster is able to change sex throughout its life. Generally ovsters begin life as males and develop into females, with the timing of this transition set to maximize the reproductive potential of both sexes. It is not fully understood what causes this change but it is known that there are environmental and genetic components. The FOXL2 gene is known to play an important role in ovary development and maintenance in other organisms. We theorize that the FOXL2 gene plays an essential role in sex determination in the Eastern Oyster, and therefore a first step is to detect whether the FOXL2 gene is present in C. virginica. Our hypothesis is that the FOXL2 gene is present in C. virginica. To determine if the FOXL2 gene is present, we conducted a BLAST search of the C. virginica genome using the H. sapiens Foxl2 protein. The results show that there was a protein that was similar to the H. sapiens Foxl2 protein with an E-value of 2x10 -56. Further examination showed that these 2 proteins were 90% identical in their Forkhead Box. The Forkhead Box is a DNA binding domain that is highly conserved in members of the FOXL2 gene family. An alignment of Foxl2 proteins from various vertebrates and invertebrates showed that the Forkhead Box was highly conserved from sponges to humans (80-99% identical). Based on these results we accept our hypothesis that the FOXL2 gene is present in C. virginica. Further analysis using a phylogenetic tree demonstrated that these organisms could be grouped using the C-terminus of the Foxl2 protein. Our future goal is to determine the role of FOXL2 in sex determination in the Eastern oyster.

Author Name(s): Elise Visser Poster Presentation Title: The Influence of St8Sia1 During the Embryonic Development of Zebrafish Affiliations: Manhattan College Category: Developmental Biology and Genetics (DBG)

Abstract

Zebrafish undergo a complex set of life stages that bring them from a fertilized egg to a fully functioning mature individual. As is the case for all vertebrates, these life stages unfold within the intricate framework of the extracellular matrix (ECM), a complex network of proteins and sugars that provide both structural support as well as govern molecular and cellular interactions that are vital for proper functioning of development. Within the ECM, sialic acids are terminal carbohydrates on proteoglycans and glycolipids that act as molecular messengers. To investigate the developmental requirement of sialic acids, the biosynthetic enzyme St8Sia1 was manipulated. St8Sia1 is a sialyltransferase required for the transfer of terminal sialic acids. The experimental design followed a three-step approach: real-time PCR to ascertain the temporal expression of St8Sia1, in-situ hybridization to localize expression, and morpholino inhibition to evaluate its function within development. Findings indicated that St8Sia1 expression was most prominent at 36 hours post fertilization (hpf) suggesting a role within neurogenesis/organogenesis. Through the in-situ hybridization it was determined that St8Sia1 expression was localized within the posterior ectoderm of the embryo. Functional analysis done through morpholino inhibition revealed truncation of the injected embryos as well as reduced tissue in the posterior ectoderm. These findings reveal that the cooperation between timing, location and function in the context of St8Sia1 provide compelling preliminary evidence to support a role in vital morphogenetic processes for sialic acids.

Author Name(s): Fitzgerald, Madelyn Poster Presentation Title: Kv2.1 and FAK: Protein Colocalization Visualized in The Embryonic Hindbrain of Zebrafish Affiliations: Manhattan College Category: Developmental Biology and Genetics (DBG)

Abstract

We are interested in the molecules that regulate the cellular processes within invertebrate organisms undergoing embryonic development. To this end, our lab has knocked-down the expression of Kv2.1, a voltage gated potassium channel, and shown that it results in abnormal morphological movements within the embryo. Interestingly this also phenocopies the independent loss of the FAK protein. We hypothesize that Kv2.1 and FAK, two proteins that are not typically seen working together, are genetically interacting within the same molecular complex to regulate embryonic development. In order to support this hypothesis, attempts were made to confirm the colocalization between Kv2.1 and FAK during embryonic development. Zebrafish embryos were treated with two different methods, thin section histological immunohistochemistry and whole mount immunohistochemistry. Using our antibodies, histological immunohistochemistry resulted in the disruption of the target epitopes making fluorescence colocalization with immunohistochemistry impossible. We therefore switched our approach to whole mount immunohistochemistry. Utilizing our two antibody assay, Kv2.1 and p-FAK were shown to colocalize as evidenced by the yellow fluorescent color. We further showed that the co-localization data was specific to the targeted epitopes by a series of negative control reactions. This data suggests that there is in fact colocalization between Kv2.1 and p-FAK during zebrafish embryonic development, suggesting a complex molecular mechanism between both of these proteins. Continued research will aim to elucidate how this molecular mechanism works.

Author Name(s): Christopher Colavito Poster Presentation Title: Investigating the Requirement of Sialic Acid During Zebrafish Neurodevelopment Affiliations: Manhattan College Category: Developmental Biology and Genetics (DBG)

Abstract

Sialic acid is a carbohydrate predominantly found in the nervous system of vertebrates, where it is localized to the terminal ends of polysaccharides. The purpose of this study is to determine if there is a role for sialic acids during the embryonic development of vertebrates. There is a family of enzymes that covalently attach sialic acid to the terminal ends of polysaccharides called sialyltransferases. Since it is difficult to specifically manipulate carbohydrates directly, one of the zebrafish genes that produces the sialyltransferase was investigated in this study, St6Gal1a. To understand the role of St6Gal1a, a Real-Time Polymerase Chain Reaction (RT-PCR) was performed to see when in development St6Gal1a is expressed. An in situ hybridization (ISH) was then performed to determine where in the zebrafish embryo St6Gal1a is expressed. To understand the ramifications of knocking down expression, morpholino oligonucleotide (MO) injections were performed. It was found that there were spikes in expression during specific organogenesis stages, and that St6Gal1a expression is localized in the ventricular region of the hindbrain. Following MO injection speckling was observed in the hindbrain during mid-organogenesis. An acridine orange stain showed that there was increased proliferation and apoptosis in the knockdown embryos when compared to controls. Based on this data, there is evidence to suggest that sialic acids play a role in the neural development of zebrafish.

Author Name(s): Perrillo-Sullivan, Stephen; Nicolas, Antoine Poster Presentation Title: Mitochondrial Genomics of Apiaceae Subfamily Azorelloideae Affiliations: Manhattan College Category: Developmental Biology and Genetics (DBG)

Abstract

Angiosperm mitochondrial DNA is difficult to use for phylogenetic analysis due to the complexity and size of the genome. With improved genomic-level sequencing technology, plant mitogenomes have become more popular markers to use in evolutionary studies. To study its structure and assess its phylogenetic utility, shotgun sequencing and de novo assemblies were conducted on 42 species of the Apiaceae subfamily Azorelloidae. The analyses showed high variations in gene size and order, even among very closely related species. For some groups, it was not possible to retrieve large assembled pieces due to the presence of a very high number of repeats that lead to conflicting gene order in cells of the same individual. The extracted dataset of coding region resulted in a phylogeny that was well supported and very similar to that based on the chloroplast genome, showing that some regions of the mitogenome are of high phylogenetic utility.

Author Name(s): Santospirito, Julianne; Nicolas, Antoine Poster Presentation Title: Comparison of Three Plant DNA Extraction Methods Using Parsley and Largeleaf Marsh Pennywort Affiliations: Manhattan College Category: Developmental Biology and Genetics (DBG)

Abstract

We compared three methods of plant DNA extraction, one using silica spin column, another using DNA magnetic beads, and a third using a propietary resin that traps polysaccharides. For each method, 12 silica-dried samples were used from each of two species Petroselinum crispum and Hydrocotyle bowlesioides. DNA quantity and purity was assessed with NanoDrop and Qubit. All methods resulted in high molecular weight, pure DNA, with the silica and bead methods being cheaper and less time consuming. The proprietary resin method retrieved the highest amount of DNA per mg of tissue, but with higher RNA and salt contaminants.

Author Name(s): Gunaratnam, Kughan; Nicolas, Antoine Poster Presentation Title: Comparing Three Approaches to Breaking Down Cell Walls for DNA Extraction from Historical Plant Samples Affiliations: Manhattan College Category: Developmental Biology and Genetics (DBG)

Abstract

We used 12 different plant samples varying of tissue types, and all dated around the 1891 and 1934, to test which method of cell wall lysis worked better. The three methods we used were bead mill, prolonged cold treatment at -80°C followed by manual grinding, and an enzyme mix to digest cell walls. We then used gel electrophoresis, NanoDrop, and Qubit to test the quantity and quality of each sample by measuring double stranded and single stranded DNA amounts, as well as RNA, protein, and salt contamination. We found that the method using enzymes was the least efficient and gave the lowest amounts of DNA. Bead mill and extreme cold treatment did not give significantly different amounts, but the type of tissue (e.g., thin or brittle vs. thick and flexible) was important to determining the better method.

Author Name(s): Arora, Bhavya; Shahzad, Zainab, Ahmad, Haziq; Akter, Tahamina; Bellantoni, Samantha; Hanson, Guevanie; Hussain, Syed; Koretz, Anna; Muralidhar, AJ; Smith, Steven; Traficante, Mirza, Waris; Miura; Wala, Zhakai; Nissen, Jillian
Poster Presentation Title: Investigating gene functions in the F1 cluster phage Akhila
Affiliations: SUNY Old Westbury
Category: Developmental Biology and Genetics (DBG)

Abstract

SUNY Old Westbury is a part of the 10th Cohort of the HHMI SEA-PHAGES program, and recently joined the 3rd cohort of the SEA-GENES project in 2021. The phage Akhila was isolated in Fall 2019 as part of the phage discovery component that was integrated into the honors section of Basic Biosciences I Laboratory (BS2401). As this section was switched to a remote format in Fall 2020, students in this course instead annotated Akhila's genome as a part of the SEA-PHAGES Bioinformatics project. Akhila has a Siphoviridae morphotype with a temperate life cycle. It was sequenced at the University of Pittsburgh Bacteriophage Institute using Illumina sequencing with a shotgun coverage of 1484. It has 56,251 base pairs and 62.1% GC content. It belongs to the F1 cluster of Mycobacterium smegmatis mc2155 phages. The final annotation of this phage identified 99 genes, 37 with assigned functions, and 62 with no known function. As the GENES program serves to investigate the function of these phage genes, 78 have been amplified from phage DNA, 37 of which have assigned functions, and 41 with no known function. Of these, 54 were chemically transformed into E. coli and confirmed as the correct gene through clone verification PCR. In future semesters, we intend to perform cytotoxicity and defense assays in M. smegmatis to determine the role of these genes in the phage life cycle.

Author Name(s): Tramell, Abigail ; Kennell, Jennifer Poster Presentation Title: Characterization of the CG5577 gene in Drosophila melanogaster Affiliations: Vassar College Category: Developmental Biology and Genetics (DBG)

Abstract

Currently, many of the genes in Drosophila melanogaster remain uncharacterized, especially genes involved in regulating metabolism. This summer, we focused on characterizing the CG5577 gene. Sequence similarity suggests this gene encodes a likely member of the haloacid dehydrogenase (HAD) domain superfamily of non-protein phosphatases, in particular, a protein similar to human pyridoxal phosphate phosphatase (PDXP). One function of PDXP is to dephosphorylate pyridoxal 5'-phosphate (PLP), the co-enzymatically active form of vitamin B6. Vitamin B6 is a crucial nutrient that maintains not only our muscular and skeletal structure but also our cognitive abilities. Our goal for this project is to determine if the CG5577 gene is the possible ortholog to PDXP by knocking out the CG5577 gene in Drosophila and determining if our mutants show similar phenotypes as reported in previous studies in other species. These phenotypes in mutant mice include muscle degeneration, neurodegeneration, and improper locomotion behavior. So far, we have seen that our mutants have decreased flight and climbing performance compared to our controls, as well as increased sensitivity to longer exposure times to carbon dioxide. Mutant females also have possible fertility defects. These results tell us this gene possibly affects muscle function and other aspects of physiology. Understanding more about how Drosophila regulates vitamin B6 levels may give us a better understanding of how it is regulated in humans. This can help develop treatments for diseases where vitamin B6 is unregulated, such as epilepsy, depression, certain cancers, and other neurological disorders.

Author Name(s): Rocenovic, Danilo; Wydner, Katherine Poster Presentation Title: Common Yellowthroat Song Variability and the Effects of Anthropogenic Noise Affiliations: Saint Peter's University

Category: Developmental Biology and Genetics (DBG)

Abstract

Bird songs are an essential part of a bird's life. Bird vocalizations are complex systems of bird communication which vary in regards to the purposes each song or call serves. These purposes include courtship, mate attraction, predator alarm, group cohesion, and territorial defense. Anthropogenic noise is a widespread pollutant in urban and suburban areas that impedes communication between birds of the same species. Interference with communication among birds can impact fitness and survival. Many species of birds, including song sparrows, alter their song frequencies in response to anthropogenic noise. The objective of this study is to investigate song variability of common yellowthroats (COYE) (Geothlypis trichas) and possible correlations between their song characteristics and levels of urban noise in different New Jersey areas. Song recordings of COYE birds were made in summers of 2020 and 2021. Selection of sites was based on the different levels of anthropogenic noise across New Jersey; a sound meter was used to record the ambient background noise respectively with each recording. The experiment focuses on several sites, including the Saint Peter's University campus (SP), East Brunswick Baseball Field (EBB), Heavenly Farms (HF), and Lincoln Park West (LPW). Spectrograms and measurements were analyzed using Raven Pro 1.6 software. We will present our preliminary findings regarding the following COYE song parameters: approximate minimum and maximum frequency, bandwidth, and duration as well as automated measures called frequency 95%, frequency 5%, bandwidth 90%, duration 90%, and peak frequency. This research is crucial in our ongoing efforts to assess the negative effects of anthropogenic pollution on bird populations and their overall well-being in their natural habitats.

Author Name(s): Gubitz-Hess, Eva; Kennell, Jennifer Poster Presentation Title: Characterization of CG17294 in Drosophila melanogaster through mutagenesis and analysis of its conservation across Drosophila species Affiliations: Vassar College Category: Developmental Biology and Genetics (DBG)

Abstract

Phosphoglycolate phosphatase (PGP) is a highly conserved protein in the haloacid dehydrogenase (HAD) domain superfamily of non-protein phosphatases, which is responsible for breaking down toxic side products of glycolysis and DNA repair as well as converting 3-glycerol phosphate to glycerol. Due to the important function of PGP in metabolism, our lab is interested in identifying possible orthologs of PGP in Drosophila melanogaster (D. mel). The goal of my project is to characterize the D. mel CG17294 gene, a predicted ortholog of PGP based on sequence similarity. CG17294 is located on the left arm of the second chromosome and is composed of 4 exons (~1.17 kb). CG17294 also belongs to the HAD family of non-protein phosphatases and across adult cell types, is most highly expressed in male germline cells.

My project involves the mutagenesis of CG17294 and its annotation in seven Drosophila species to explore both its function and conservation across species. I used CRISPR/Cas9 gene editing to induce random indels in my target gene, using a transgenic based approach. The fly progeny will be screened for indels, and any outcomes will be reported in my presentation. For the gene annotation portion of my project, I used databases such as the GEP UCSC Genome Browser Gateway, Basic Local Alignment Search Tool (BLAST), the Gene Record Finder, and the Gene Model Checker to create gene models for putative CG17294 orthologs in various Drosophila species. Overall, D. erecta was found to have the highest sequence identity (86%) and its genomic neighborhood shared complete synteny with D. mel. D. yakuba, D. ananassae, D. persimilis, and D. mojavensis also had high identity scores (~70-80%) and complete synteny. D. willistoni had a slightly lower identity (67%) and shared only partial synteny. Lastly, D. virilis had the lowest identity score (57%) and shared partial synteny with multiple CG17294 isoforms. These results show that CG17294 and its genomic neighborhood appears to be more highly conserved in species more closely related to D. mel.

Author Name(s): Morkos, Celine; Vilchis-Ibarra, Cindy; Solis, Valeria Poster Presentation Title: Y Chromosome gene, KDM6A a potential biomarker gene in various types of Cancer Affiliations: St. Francis College

Category: Developmental Biology and Genetics (DBG)

Abstract

Cancer is a complex group of diseases characterized by uncontrolled cell growth and metastasis to other parts of the body. It can affect any tissue or organ, leading to a wide range of symptoms and outcomes depending on the type and stage of the disease. The treatment of certain types of cancers has made significant progress in the past decades-as represented by the treatment of chronic myelogenous leukemia with imatinib, which improved the five-year survival rate of patients to 70% in 2016. The current human genome (version GRCh38) basically provides complete coverage for annotated protein-coded as well as non-coding RNAs and pseudogenes. There are important genes associated with cancer on sex chromosomes as better mapped on the X chromosome (A. Spatz et al. (2004) Nature Rev. Cancer, 4, 617-629) such as XIST non-coding RNA the androgen receptor gene in prostate cancer. Surprisingly, the human Y chromosome has been challenging to sequence due to its complex structure. Over half of it was missing from the GRCh38 reference sequence. Finally, the sequencing of unmapped regions of the Y chromosome was completed this summer (A. Rhie, et al. (2023) Nature, 621, 344-354). A recent study showed the loss of the Y chromosome is associated with multiple cancer types including 10%-40% bladder cancer. This prompted us to check if Y chromosome gene mutations associated with the increase risks of cancers. When we checked protein-coding genes located on the Y chromosome, the mutations rate of KDM6A was slightly higher than others. The Y chromosome plays a crucial role in determining male characteristics and development. Certain genes on the Y chromosome may have implications for predicting cancer risk and understanding the molecular basis of various diseases, including pancreatic, prostate, lung, and brain cancer. Recent studies have explored the potential role of Y chromosome genes in poor prognosis of bladder cancer (H.A. Abdel-Hafiz et al. (2023) Nature, 619, 624-631). Therefore, we hypothesized that the data mined in cBioportal might be able to find similar trends, or possibly novel biomarkers that can potentially predict high-risk patient populations in cancer. Among the majority of protein-coding, Y chromosome genes that we checked, lysine demethylase 6A (gene symbol: KDM6A) mutations appeared to be slightly higher than other protein-coding genes on the Y chromosome.Based on our analysis, there were 36.7% deceased and 6.7% living out of a total of 30 patient who show KDM6A alternations (splicing variants and frame shift mutations) in pancreatic cancer. Out of the 30 patients diagnosed with pancreatic cancer 17 patients' status were unknown. Recently, loss of KDM6A was reported to cause super-enhancer activation regulating DNp63, MYC, and RUNX3 (J. Andricovich et al. (2018) Cancer Cell, 33, 512-526), inducing metastatic pancreatic cancer. In our analysis, KDM6A splicing variants in pancreatic cancer showed xy% mortality. Thus, KDM6A alternations might be a useful biomarker to predict high-risk populations in pancreatic cancer.

Author Name(s): Vilchis-Ibarra, Cindy ; Solis, Valeria ; Morkos, Celine ; Kita, Katsuhiro Poster Presentation Title: Discovery of a new mutation hot spot in a microtubule plus-end binding protein, CLIP-170 Affiliations: St. Francis College Category: Developmental Biology and Genetics (DBG)

Abstract

The cytoskeleton is an interlinking protein polymer that forms in the cytoplasm of filamentous structures in a cell. Proper regulation of cytoskeletal machinery is essential for development. Among cytoskeletal proteins, microtubules (MTs) also play a very important role in directional cell migration, establishment of cell polarity, intracellular transport, and chromosome segregation during mitosis. Due to these important roles, MT-targeting drugs are one of the successful chemotherapeutic agents in clinical oncology. We recently reported a novel CLIP-170 variant that is associated with drug resistance in gastric cancer (P. Thakker et al. (2021) Dev. Cell, 56, 3264-3275). This prompted us to explore other variants of CLIP-170 in cancer. CLIP-170 is the first MT plus-end binding protein (+TIP) that tracks the growing MT-end in a cell and is shown to play an important role in MT plus-end dynamics, which is essential for cell migration. CLIP-170 also interacts with other +TIPs, including one of the well-known tumor suppressors (and a +TIP), adenomatous polyposis coli (APC) protein. Therefore, mutations that disrupt the normal function of CLIP-170 could affect a variety of cellular functions, including APC. When we conducted a comprehensive search in a variety of cancers through cBioportal, we found a mutation hot spot in lung, bladder and skin cancer: E1012K. All three types of cancer (2.1%, 5%, and 5%, respectively) share a higher percentage of CLIP-170 mutations compared to other types of cancers. We also found that skin cancer had a high rate of frameshift mutations specifically S1018fs mutations. These locations are close to the end of the long stretch of the coiled-coil region of CLIP-170, which is necessary for the dimerization of CLIP-170 molecule to function properly. In addition, the C-terminal portion of CLIP-170 zinc knuckle domain which is essential for CLIP-170 interaction with p150Glued, one of the dynactin complex proteins involved in retrograde vesicle transport. We suspect that this mutation may affect cancer cell migration and might be metastasis.

Author Name(s): Asare, Belina; Canger, Anthony
Poster Presentation Title: Expression of Actin Regulatory Proteins Paxillin, Zyxin, and Moesin during Neuronal Differentiation
Affiliations: Mercy University
Category: Developmental Biology and Genetics (DBG)

Abstract

In this research the expression of several proteins that are known to regulate actin were studied using chick primary midbrain cultures. A goal of using this model system is to better understand how neurons develop elaborate dendrites and grow axons that form synaptic connections. The expression of paxillin, zyxin, and moesin was studied at various at different stages of neuronal differentiation as well as astrocytes. Phase contrast microscopy was used to document the morphology and characteristics of the neurons in the cultures. Embryonic day six cultures grown at low density were plated on poly-Dlysine/laminin coated and immunofluorescence microscopy was done to study the expression pattern of these proteins. The expression of all three proteins was evaluated in neurons from 1 day in culture to 10 days in culture. Actin organization was studied in conjunction with the distribution of zyxin, paxillin, and moesin. Antibodies to identify growth cones, glial cells, focal adhesions, and axons were used along with antibodies specific for chick paxillin, moesin, and zyxin. Vinculin antibodies were used to evaluate the development of focal adhesions in the cultures. All these proteins were observed in neurites in day 7 cultures. The proteins vary in the level and distribution but all three are observed in a punctate distribution in both axons and dendrites. The results show that these proteins are focal adhesion components that localize with actin. Zyxin is expressed in neuronal growth cones in embryonic day 1 and 2 cultures. Zyxin was also observed at synapses between neurons and astrocytes in day 7 cultures. In neuronal process including filopodia, these proteins were observed in a punctate distribution which overlapped with microfilaments. The results show these proteins are expressed during various stages of neuronal differentiation.

Author Name(s): Edwards, Kristin; Kattela, Shalini; Belfiore, Natalia Poster Presentation Title: Determining the Genetic Diversity and Relatedness among North American River Otter (Lontra canadensis) Populations Affiliations: University of Bridgeport Category: Developmental Biology and Genetics (DBG)

Abstract

The North American river otter (Lontra canadensis) is a semiaquatic mammal in the Family Mustelidae which historically ranged throughout North America. The primary habitat requirements are a reliable food source and easy access to bodies of water. The wide diet of North American river otters includes fish, frogs, crayfish, turtles, insects, a few other small animals as well as seasonal fruit. They may hunt on land and in the water, and they are known to hunt in pairs or alone. The North American river otter feeds on aquatic and semi-aquatic species. The river otter's eating habits and preferred prey are mostly influenced by the prey species' vulnerability and seasonal availability.

Although North American river otters are among the more social members of the Mustelidae, the breeding season is limited and typically occurs between December and April. The actual gestation period for female otters lasts around 61–63 days, with the heat period lasting about 42–46 days. The time between copulation and giving birth can be 10–12 months due to the otters' distinctive reproductive technique of delayed implantation. Delayed implantation is defined by the blastocysts' failure to adhere to the uterine wall. Instead, the blastocysts remain freely in the uterine lumen for a considerable amount of time, extending the gestation period. The duration of the delay in all North American river otters is thought to vary among different geographical locations. The ability to delay implantation allows North American river otters the ability to adapt their reproductive cycles in response to changing environmental conditions.

These student projects are part of a larger study to determine genetic patterns across the range of the North American river otter, correlating them to local climate and seasonality. Specifically, the data obtained from these experiments will inform us about the relatedness or differences among the otters and the genetic diversity in each population. The differences or similarities in each sequence will then be examined to understand patterns of gene flow among river otter populations from each region.

Microsatellites, also known as short tandem repeats (STRs), are differentiated by short, repetitive sequence patterns that are usually 1-6 base pairs long. Microsatellites are significant in the field of conservation genetics and molecular ecology due to their high level of genetic variation. These variability in the amount of tandem repeats in different individuals is used to estimate genetic differentiation or similarities between different populations. This can then be used to understand population genetics, genetic diversity, and gene flow, which helps to compare the status and connectedness of different populations.

The mitochondrial control region, also known as the D-loop, is an easily accessible DNA region that usually contains sufficient variability to use for phylogeographic analysis. The variable portions of river otter control region will be sequenced and use these to assemble a phylogeographic hypothesis for relatedness among North American river otter populations.

Data from both of these genetic surveys will support one of several hypotheses about the adaptive evolution of river otters under variable environmental conditions.

Author Name(s): Apuango, Kiara; Jain, Chandna; Bradley-Ortiz, RJ; Mujica-Urzua, Patricio Poster Presentation Title: Isolation and Culture of Cardia Cells from the Chicken Embryo Affiliations: Mercy University Category: Developmental Biology and Genetics (DBG)

Abstract

Cardiomyocytes are specialized cells that make up the majority of the heart muscle. They are responsible for the heart's contraction and relaxation, allowing it to pump blood throughout the body. Now nitric oxide is a gas hormone that is an important regulator of cardiovascular function. In blood vessels, it causes vasodilation - low blood pressure.

Cardiac cells possess two NO sources, Neuronal nitric oxide synthase (nNOS) and Endothelial NOS (eNOS). Our project goal is to determine whether nitric oxide modulates the actions of acetylcholine and epinephrine in cardiomyocytes. Our research consisted of us incubating fertilized eggs @ 37 °C; 65% H2O + candling them. We extracted the embryos euthanized by decapitation, and then we performed mini surgery, extracting the hearts and isolating ventricles. Through tissue digestion we used enzymatic isolation (trypsin) + centrifugation, lastly, we set up the cell culture and incubated for 72-120 hrs. The plan was to Inhibit NOS with L-NMMA for Δ h then apply Epinephrine/ Acetocholyine/8CPT and record the rate of contraction.

We successfully isolated cardiac cells from E13 embryos and confirmed the presence of cardiomyocytes in our cultures using immunocytochemistry. The culture conditions increased the proliferation of fibroblasts therefore we were not able to enrich our cultures in cardiomyocytes.

Author Name(s): Liliana Hopkins; Kayla Yim; Ana Rumora; Melissa Baykus, Luisa Martinez Poster Presentation Title: Genetic Mapping of a Forest Microbiome Affiliations: Bergen Community College Category: Environmental Biology and Ecology (EBE)

Abstract

This study conducted a metagenomic analysis of trees and their rhizosphere microbial communities. We developed an accurate and replicable DNA barcoding method for tree species identification. We evaluated the ability of genetic barcodes primers for taxonomic classification of four trees: the first internal transcribed spacer (ITS1) region of the nuclear ribosomal cistron, it's second subunit, (ITS2), and chloroplast trnL (UAA) intron. The primers had a 50% success rate.

The metagenomic analysis of the trees' rhizosphere microbiomes revealed their relative bacteria compositions. All communities showed similar demographics at the phyla, class, and genus levels. Proteobacteria was the most abundant phyla in 75% of the samples. Alphaproteobacteria was the most abundant class in 100% of the samples. At the genus level, most species were unidentified. The first identified genus in 100% samples was Rhodoplanes.

These results call for replicated studies with larger sample sizes.

Author Name(s): Liang Xiao, Fanny; Colon, Christina P. Poster Presentation Title: Eastern Coyote (Canis latrans x lycaon) in NY: Will This New Predator Impact Bird Vocalizations on Long Island? Affiliations: Kingsborough Community College Category: Environmental Biology and Ecology (EBE)

Abstract

The eastern coyote (Canis latrans x lycaon) is a hybrid canine native to the Great Lakes region that, beginning in the 1970s, has spread to different areas of the East. In NYC, it was not until 2012 that coyotes were seen breeding in parks in the Bronx, however, Long Island remained the only territory in the US without a breeding population of covotes until 2016. With the arrival of this new predator, it was hypothesized that birds on Long Island would modify their vocal behavior in the presence of vocal coyotes. A decrease in bird diversity could be due to bird consumption by the predator, or an increase could be attributed to higher nest success if breeding is favored by the predator's consumption of mesopredators. Other acoustic studies elsewhere have also observed shifts in the bird call length and complexity as a result of embedding additional information about new predators. As part of a pilot study, a total of 10 Audio Moth acoustic devices were deployed opportunistically at three locations with coyotes present on Long Island. This collaborative project is part of a larger camera-trapping effort to track and study these urban covotes in and around the New York metropolitan area. The use of acoustic devices is to record shifts in the natural soundscape resulting from the covote's arrival. The purpose of this project is to quantify the impact that coyotes could have on the acoustic behavior of birds. Acoustic files were reviewed using Kaleidoscope software and the Merlin app was used to identify bird calls. A total of 39 bird species were recorded between December 2022 and May 2023. Bird species and frequency of calls were found to vary between locations, with Sands Point Preserve having the greatest bird diversity. However, a larger amount of data exclusively from Sands Point Preserve likely affected the results. A total of 38 presumed covote calls were recorded at Sands Point Preserve, the only location where vocalizations were detected. Coyotes rarely overlapped with the singing of birds and there was no evidence that howls elicited alarm calls from birds. While we cannot yet give a definitive answer to the proposed hypotheses, acoustic devices demonstrated the ability to provide answers in the future. They also captured the complexity and variety of calls that can be used by future students to analyze potential trophic effects of coyotes, including determining the correlation between the acoustic behavior of birds and coyotes. Other research on vocal shifts among the many frogs, crickets and other insects can also be examined by future cohorts as all data will reside on a dedicated server at the American Museum of Natural History.

Author Name(s): Cuevas, Zitlali Poster Presentation Title: Lichen and Air Pollution Affiliations: Mercy University Category: Environmental Biology and Ecology (EBE)

Abstract

Lichens and air pollution, the correlation between vehicle pollution and lichens.

This research aims to see if vehicle pollution will decrease the diversity and abundance of lichen. This study compares the lichen in two different locations of the Westchester Community College campus, the main road/bus route and near the science building where no car traffic is present. Surveys were done of the diversity and abundance of lichen living on the Honey locust trees at these two sites on the campus. Using apps, like iNaturalist, and lichen identification keys, the different types of lichen were recorded as well as their tolerance to pollution, either sensitive or tolerant. All data was put onto charts to display the differences between the two locations, showing clearly the difference in diversity and abundance of each lichen found. The difference in the diversity of lichen in each of the two locations was not large and can rule out that vehicle pollution has an effect on lichen diversity. On the other hand, the difference in the abundance of lichen in each of the two locations was large enough to support that vehicle pollution has an effect on lichen abundance. Although vehicle pollution does have an effect on lichen diversity there are different factors effecting its abundance and diversity such as weather, altitude, and tree species.

Author Name(s): Lutfur, Lutfur; Vasquez, Claudia; Eldesouky, Amal; Ortiz, Adam; Cardenas, Irma; Tessler, Michael; Herstoff, Emily Poster Presentation Title: Zooplankton and Microplastics in Brooklyn Bridge Park Affiliations: St. Francis College Category: Environmental Biology and Ecology (EBE)

Abstract

Waters around the world are contaminated with microplastics. Microplastics are small pieces of plastic (<5 mm), which are largely produced by the breakdown of larger pieces of plastic. Microplastics are prevalent in environments around the world, including our oceans. Organisms like zooplankton accidentally consume microplastics, which accumulate within their bodies and can pass up the food chain into larger consumers. While the prevalence of microplastics has been documented around the world, we are unaware of research about microplastics in the waters around New York harbor. In our study, we traveled to Pier 4 at Brooklyn Bridge Park, which is one of the two piers in the park where people can directly access the East River. Our research aimed to assess the levels of microplastics and zooplankton found in Pier 4 through water and sampling, comparing them during dry and wet periods of the summer. We also assessed the diversity and abundance of zooplankton communities in the East River at these same sampling periods. Fibers were the most abundant type of microplastic both in water and sand samples, and were more abundant during the dry sampling period. Within the zooplankton, copepods made up the largest portion of arthropods sampled, but there was little change in their proportional abundance through time. The non-arthropod zooplankton were less abundant and their proportions did not change through time. Because zooplankton are the base of the marine food web, and can act as the vector by which microplastics travel to higher trophic levels, it is important to continue to study this topic.

Author Name(s): Cardenas, Irma J.; Jules, Jurneal; Luna, Paula; Abdelhalem, Ali M.; Cunningham, Seth W.; David, Felix J.; Herstoff, Emily M.; Tessler, Michael
Poster Presentation Title: Suburban Soil Conservation with Backyard Meadows
Affiliations: Medgar Evers College CUNY & St. Francis College
Category: Environmental Biology and Ecology (EBE)

Abstract

Lawns cover 2% of the continental United States. Unfortunately, lawn monocultures decrease biodiversity, and lawn maintenance increases CO2, water scarcity, and chemical runoff of fertilizers and pesticides. A recent conservation solution is to convert lawns into meadows, which increase species diversity and can be aesthetically pleasing. However, most research on how converting lawns to meadows can benefit conservation has focused on pollinators, with little consideration of the impacts on soil microbial diversity and chemistry, which underpin many aspects of healthy habitats. In this study, we examined (1) soil microbial diversity, and (2) soil physical and chemical characteristics within suburban lawns and meadows in Cranbury, New Jersey. We found that species composition differed for microbes based on metabarcoding (bacterial 16S and fungal ITS2). Bacteria were also more diverse in meadows. Distance from the edge of the meadow also made a difference for bacteria, but the largest difference was simply the habitat type. Soil chemistry similarly differed between lawns and meadows, and was more optimal for plant growth in meadows (e.g. less acidic soils and more potassium). Our findings suggest that differences between lawn and meadow soils were driven by the organisms living in these habitats. Overall, the beneficial shifts in meadow restoration efforts can benefit ecosystem health and biodiversity.

Author Name(s): Eldesouky, Amal; Cardenas, Irma; Lutfur, Lutfur; Vasquez, Claudia; Ortiz, Adam; Richards, Eva; Tessler, Michael; Herstoff, Emily Poster Presentation Title: Leaf galls in Green-Wood Cemetery, Brooklyn NY Affiliations: St. Francis College Category: Environmental Biology and Ecology (EBE)

Abstract

A number of arthropods cause plants to form galls — abnormal growths in plant tissue that allow the animals to carry out their lifecycle. Although gall-forming arthropods are common, few studies have examined them specifically in urban environments, and no studies that we are aware of have studied this phenomenon in New York City. Here we examined gall-forming arthropods in Green-Wood Cemetery, one of the major green spaces in Brooklyn. First, we provide an overview of the diversity of gall-forming arthropods found at Green-Wood based on our observations coupled with iNaturalist records. We then compared differential gall prevalence on two species of Tilia trees at Green-Wood. While these Tilia tree species are planted within close proximity and are of similar sizes, they differ in their origin: one species is native (Tilia americana), while the other is European (Tilia cordata). Overall, our results begin to show some of the diversity and ecology of galls, an oft-overlooked form of biodiversity in urban environments.

Author Name(s): Rocenovic, Danilo; Quisbert, Helberth; Rodriguez, Katherine; Bautista, Isabelle; Wydner, Katherine
Poster Presentation Title: Project FeederWatch, Habitat Restoration, and Native Species: If You Build It, Will They Come?
Affiliations: Saint Peter's University
Category: Environmental Biology and Ecology (EBE)

Abstract

Despite the success of habitat restoration efforts in restoring native flora, they do not always result in the return of endemic wildlife. Starting in 2018, we renovated a patch of neglected landscaping on the Saint Peter's campus by removing foreign, invasive, ornamental plants and replanted it with native plants that are of value to birds and pollinating insects. After analyzing eight years of Project FeederWatch (PFW) data – from the four years before and after the creation of the native plant garden – we concluded that there was a significant increase in the number of avian species that winter on our campus from 2018-2019 and onward. However, despite the return of many native bird species, the dominant species on our campus remains Passer domesticus, the House Sparrow, a nonnative and sometimes aggressive species. In the 2022-2023 PFW season, we purposely changed the seed mix at our feeders to a combination of seeds that may be less preferred by House Sparrows. Compared to previous seasons, we did see somewhat reduced House Sparrow numbers along with numerous unexpected sightings of Black-capped Chickadees, Tufted Titmice, and White-breasted Nuthatches. These native species are associated with forested environments in suburban and rural areas, and have only been rarely (or never, in the case of titmice) reported for previous PFW seasons on campus. Our PFW results are presented along with our ongoing plans to encourage the return of native bird species.

Author Name(s): Roman, Isiah; Nicolas, Antoine Poster Presentation Title: Plastome Phylogenomics of the Pennywort Genus Hydrocotyle Affiliations: Manhattan College Category: Environmental Biology and Ecology (EBE)

Abstract

Hydrocotyle, the pennywort genus, includes over 170 species with centers of diversity in Australia and the Andes. To study the phylogenetics and biogeography of Hydrocotyle, we used whole genome sequencing to assemble, annotate, and analyze whole chloroplast genomes from 56 samples that represent its full geographic distribution. We recovered a well-supported phylogeny that showed distinct major clades, with geographic distinction. Successive early lineages are endemic to Australia, making it the center of origin of the genus, followed by diversification into New Zealand, Asia, and then the Americas. The American species were divided into four different groups: Andes , Brazil, the Caribbean, and North America. There is clear evidence of dispersal from New Zealand into Patagonian Argentina and Chile through a subantarctic route. The diversification into the Americas is less clear but most likely from Asia/Europe into North America and then the rest of the Americas.

Author Name(s): Mingone, Mario; Nicolas, Antoine Poster Presentation Title: Chloroplast Phylogenomics Of Apiaceae Subfamily Azorelloideae Affiliations: Manhattan College Category: Environmental Biology and Ecology (EBE)

Abstract

Azorelloideae is a subfamily of the carrot family Apiaceae that includes over 130 species in three major clades: Azorella, Asteriscium, and Bowlesia. The subfamily also includes two small genera: Diposis and Klotzschia. We sampled 42 species from all major groups of Azorelloideae for shotgun genomic sequencing and assembled, annotated, and analyzed their whole chloroplast sequences. Maximum likelihood phylogenetic trees showed well-supported relationships among the three main clades and full support of the placement of Diposis as sister to the Asteriscium clade. Chronograms generated in BEAST showed the age for the three major clades to be estimated at 65 million years, with an origin in Antarctica. Each of the three main clades originated in South America during the Eocene. Dispersals into Australia and New Zealand happened at least once in each of clades during the Miocene.

Author Name(s): Diaz-Oblitaz Esquivel, Abigail ; Carfagno, Gerardo Poster Presentation Title: Documenting ecologically driven differences in salamander morphology Affiliations: Manhattan College Category: Environmental Biology and Ecology (EBE)

Abstract

The red-backed salamander (Plethodon cinereus) can be found in relatively high densities in forests across eastern North America, and serves as an important link in these food webs. The species can even be found in highly urbanized locations, occurring within some New York City public parks. Given the species' relatively broad geographic distribution, P. cinereus is expected to experience a range of environmental conditions. It has been recently shown that morphological variation in salamander head shape may result from significant interactions between the species and environmental pressures. Head morphology is important for the species as mouth gape will determine the type of prey that this predator can consume. Therefore, my goal was to document head morphology as it might vary with changing environmental conditions. My hypothesis is that if different environmental conditions exert different selective pressures on populations, we should expect to see differences in environmentally relevant morphologies within the same species. During the summer of 2023, I searched for red-backed salamanders from two locations within Van Cortlandt Park, in the Bronx. Once a salamander was found it was weighed, its snout-to-vent length was recorded, and I took photographs of the animal from multiple angles for later analysis. The soil temperature at which the salamander was found and a random soil temperature nearby were also recorded. I also collected similar morphology data and photographs from fluid-preserved specimens at the American Museum of Natural History Department of Herpetology. All P. cinereus sampled at the museum were collected from the Bronx in 1967. For both living and preserved specimens, side and top-view digital photographs of the anterior end were analyzed using a software package (ImageJ) to measure distances between obvious landmarks on the heads of the animals. Salamanders were found in significantly cooler soil temperatures compared to the randomly sampled soil around them. I found no significant differences in the morphology of living salamanders found at the two locations in Van Cortlandt Park. However, there were some differences observed in the head morphology of living salamanders and those preserved at the museum. When controlled for body size, two of the nine head measurements were found to be significantly different. Both the relative snout to neck measurement and the head width of salamanders collected in 1967 were significantly larger than those from salamanders found in 2023. One possible explanation for these results is that the urban environment may be selecting for reduced head size, driven by a reduction in larger prey items. Future work should collect similar data of P. cinereus from additional locations throughout their natural range. It would be interesting to compare populations from different ecological environments to determine if there is any further evidence of morphological adaptations to local conditions.

Author Name(s): Osowiecki, Sabrina; Keena, Melody Poster Presentation Title: Determining the Effects of Prolonged Starvation on the Survival, Growth, and Development of Lymantria dispar Affiliations: Amity Regional High School Category: Environmental Biology and Ecology (EBE)

Abstract

Asian spongy moths (Lymantria dispar) are an invasive insect that can defoliate forests and cause health issues. They can withstand periods of starvation, which allows them to be transported, often by boats or trains, and consequently establish new populations. The purpose of the project is to determine if a period of starvation causes measurable differences in survival, growth, and development for Asian spongy moths. The hypothesis is that Asian spongy moths starved for the longest periods of time will have the lowest survival rate, the slowest development, and the lowest weight. The independent variable is the amount of time the insects were kept away from food, and the dependent variables are the length of time they survive, the speed at which they molt, and their weight gain. First, Asian spongy moths (RM strain) were weighed and placed into three groups of 100 each: a control group that received food for 21 days, a group that was starved for 4 days and then given food for 21 days, and a group that was starved for 8 days and then given food for 21 days, the amount of insects surviving, what instar each insect molted to, and approximately how much weight each insect gained since hatching were determined. Data partially supports the hypothesis. This project can help determine the threats posed in the long run by Asian spongy moths that are transported.

Author Name(s): Sekyere, Michael; Kang, Seokyoung; Dweck, M K, Hany Poster Presentation Title: Acid Taste Sensing in Spotted Wing Drosophila Affiliations: University of Bridgeport Category: Environmental Biology and Ecology (EBE)

Abstract

A major agricultural threat has recently emerged in North America and Europe. Drosophila suzukii (also known as spotted wing Drosophila) has begun to ravage fruit crops across these continents. In this study, we aimed to test the hypothesis that the D. suzkukii and its close relatives, Drosophila melanogaster, exhibit distinct behavioral responses to acids. We postulated that these differences might be linked to the varying levels of organic acids present in their preferred breeding sites: ripe fruits for SWD and overripe fruits for D. melanogaster. Using two egg-laying assays, we found that SWD exhibited a clear preference for laying eggs on certain acids found in ripe fruits. In contrast, D. melanogaster displayed either avoidance or neutral responses to these same acids. These results provided compelling evidence supporting the notion that the perception and response to acids are indeed critical factors contributing to the divergent egg-laying preferences between SWD and D. melanogaster.

Author Name(s): Aramis Medina Poster Presentation Title: The impact of rare recruitment events & the analysis of genetic variation among sub-populations in Cenchritis muricatus Affiliations: Manhattan College Category: Environmental Biology and Ecology (EBE)

Abstract

Metapopulations have varying degrees of isolation and connectivity. Species that freely move between populations tend to have a more homogenized genetic structure, whereas isolated populations show a more individualized genetic structure. In this study, we used a long-lived snail (Cenchritus muricatus) that only reproduces during rare events such as a hurricane, producing larvae that can disperse to other locations. Sequencing the ITS region of the genome, we utilized a fixation index (Fst) to analyze the population genetics of this species. Using this locus, we found no differences in genetic structure between six subpopulations in the U.S. Virgin Islands. Further sequence analyses may provide more insight into how rare dispersal events impact the population genetics of this species.

Author Name(s): Ali, Ashley; Medor, Gabrielle; Rene, Dorothee; Rivera, Noemi Poster Presentation Title: Water Quality Testing of Enterococcus Levels Along the East River Affiliations: St. Francis College Category: Environmental Biology and Ecology (EBE)

Abstract

Through the Billion Oyster Project's 2023 Community Water Quality Testing season, St. Francis College served as a lab site for testing water samples for enterococci at Piers two and four along the Easter River in Brooklyn, New York. The IDEXX Enterolert Test Kit was utilized for enterococci detection within 24 hours after incubation using a Defined Substrate Technology (DST) nutrient indicator that fluoresces when metabolized by enterococci. Water samples that are positive for this bacteria may suggest the presence of feces in the water, which would be harmful to the public that has utilized Dumbo Cover, Piers 2 and 4 at Brooklyn Bridge Park for kayaking and seining activities. Given that the public has direct access to the East River at Dumbo Cove and Pier 4, it was hypothesized that these locations would have higher levels of enterococcus levels. After reviewing the data from the twenty week sampling season, it was determined that overall Pier 4 beach had the highest levels of enterococcus levels when compared to Dumbo Cove, and that Pier 2 had acceptable levels at less than 35MPN for most of the season. The environmental conditions of rain, pollution, and ducks at these sites were shown to affect a higher level of enterococcus, particularly at Pier four.

Author Name(s): Diamond-Huey, Tosha Poster Presentation Title: Streamlining germination of natives for rewilding Affiliations: Borough of Manhattan Community College/CUNY Category: Environmental Biology and Ecology (EBE)

Abstract

Rewilding, the reintroduction of native species and restoration of natural self-regulating diverse ecosystems is at the core of improving resilience in a changing climate. As a "think global act local" approach, rewilding can be effective in small patches of mini-meadows in urban and suburban areas. Obstacles to rewilding a lawn or a weedy patch can be the germination and establishment of native species. By making science-based methods accessible to local gardeners, biodiversity islands can become reality. The start species here is Asclepias incarnata, swamp Milkweed, food and habitat for Monarch butterflies and not easy to germinate. Germination of 1500 fresh A. incarnata seeds was attempted with periods of cold stratification from 5 to 30 days, with and without scarification. Freezing and water floating were also attempted. The only treatments to result in some germinations were those involving heavy scarification. Apparently, for good reason, fresh A. incarnata is not meant to germinate. Possibly because establishment soon after seed production would prove difficult.

Author Name(s): Beas Romero, Agustin; Cabail, Zulema Poster Presentation Title: Meta-inflammation Increases the Migration and Invasiveness Potential of Prostate Cancer Cells Affiliations: SUNY at Old Westbruy Category: Microbiology and Immunology (MI)

Abstract

Obesity-induced chronic low-grade systemic inflammation, or meta-inflammation, is characterized by the accumulation of immune cells, mainly macrophages, in the adipose tissue (AT) and the deregulation of AT hormones and cytokines. Several studies have suggested that obesity is associated with a higher risk of fatal prostate cancer. Prostate cancer (PCa) represents one of the most common types of cancers diagnosed in American men and the third most common cause of cancer related-deaths, mainly due to incurable metastatic disease. Within the tumor microenvironment, there are several factors that play an important role in the proliferation and metastasis of cancer. Such factors include inflammatory chemokines and cytokines associated with infiltrating macrophages into the tumor environment. These infiltrating macrophages (also known as tumor associated macrophages) are a key component of inflammation during prostate cancer tumorigenesis and metastasis. The underlying mechanisms linking obesity with PCa are not completely understood. The goal of this project is to elucidate the molecular and cellular mechanisms underlying the association of meta-inflammation with prostate cancer migration and invasiveness. We hypothesize that a hypercaloric environment will promote the migration and invasion of PCa cells. To addresses this hypothesis we used U937 human macrophage-like cells and exposed them to a lipid-rich microenvironment to mimic the cellular metabolic inflammation observed in obese AT. After 16-18 hours of stimulation with 300 μ M Sodium Palmitate (inflammatory conditions), we collected the conditioned media which was used to stimulate DU145 human prostate cancer cells. The invasive cellular behavior was assessed by transwell migration, Boyden chamber invasion, and wound healing assays and we found that stimulation with conditioned media from inflammatory conditions promoted an increased in cell migration and invasion when compare with unstimulated DU145 cells. Taken together, our findings suggest that meta-inflammation promotes the progression of prostate cancer towards an invasive state.

Author Name(s): Pinheiro Machado, Adriana; Louis, Jerry; Bendaoud, Meriem Poster Presentation Title: Antibiofilm and Antimicrobial Properties of Schinus terebinthifolia Fruit Extract Affiliations: New Jersey City University

Category: Microbiology and Immunology (MI)

Abstract

In the past few decades, the misuse and overuse of antimicrobials has led to an increase in the number of antimicrobial resistant infections, which according to the World Health Organization, has become a major worldwide concern and a threat to public health. The need to develop new therapeutic alternatives to address the ineffectiveness of conventional antimicrobial treatments is crucial. The rising field of phytotherapy offers an efficient approach to addressing this global health crisis. In this study, we investigate the Schinus terebinthifolia plant fruit extract as a potential low-cost alternative to antibiotics. This highly available plant is widely used in gastronomy and known in folk medicine for its wound healing and health-promoting properties. In this study, we evaluate the plant's fruit extract antimicrobial and antibiofilm properties against 22 different strains of bacteria and fungi using the broth microdilution, biofilm, and spot assays in microtiter plates. The results show that the fruit extract has a significant antibacterial effect on several gram-positive and gram-negative pathogenic bacteria including Staphylococcus aureus, Enterococcus faecalis, Pseudomonas aeruginosa, Acinetobacter baumannii, Bacillus subtilis, Bacillus cereus, Escherichia coli, and Staphylococcus epidermidis. In addition, the plant extract displays varying degrees of antibiofilm properties at different concentrations against bacteria and fungi. These findings suggest that the S. terebinthifolia fruit extract has the potential to be used as a novel antimicrobial alternative in the treatment of infectious diseases. Future studies will focus on further characterization of the fruit extract.

Author Name(s): Djedji, Chloe; Gonzalez, Norma; Jimenez, Raul; Hana, Morris; Aqeel, Abdul Poster Presentation Title: Determination Antibiotic Resistance Pattern in Human Normal Microbiota Post-COVID-19 Pandemic Affiliations: Bergen Community College Category: Microbiology and Immunology (MI)

Abstract

In this study, seven hundred and twenty-two subsamples cultured on TSA and MSA, and EMB selective media. About one-third of the cultures showed resistance to antibiotics. Some of the bacteria were resistant to broad-spectrum antibiotics. Staphylococcus aureus was found among some cultures after being tested on Mannitol Salt Agar (MSA). This result demonstrates an increase of S. aureus among the collected population samples.

Author Name(s): Colares, Isabella; Atiq, Daniyal; Reece, Brianna; Frias, Maria A. Poster Presentation Title: Water Quality of the New York City East River Affiliations: St. Francis College Category: Microbiology and Immunology (MI)

Abstract

Water quality can be assessed by detection of the levels of bacterial indicator organisms such as coliforms and fecal coliforms. In this study, we tested water quality in a major urban water environment: the East River of New York City. We employed standard qualitative water analysis, which includes presumptive tests, confirmed tests and completed tests. We observed very high levels of total coliforms (920 per 100ml), and we isolated and identified several fecal coliforms. Our data indicates that the East River of New York City should not be used as recreational water.

Author Name(s): Williams, Z'Dhanne; Hernandez, Edwin; Evans, Jodi F Poster Presentation Title: Optimizing an Imaging Protocol to Quantify Mitochondrial Transfer Affiliations: Molloy University Category: Microbiology and Immunology (MI)

Abstract

Mesenchymal stem cells (MSC) play a critical role as connective tissue precursor cells and also have potent immunomodulatory properties. For instance, MSC can increase macrophage phagocytosis when in direct contact. The mechanisms through which they modulate this activity is under investigation and includes mitochondrial transfer. The long-term goal of the project is to develop a protocol to image the mitochondrial transfer between the MSC and macrophage cells (M Φ) and quantify the downstream changes of M Φ phagocytic activity. Intercellular transfer of mitochondria was observed by utilizing longterm static, dynamic, and fluorescence imaging. The MSC mitochondria were stained with fluorescent indicator, mitotracker red, and co-cultured with unlabeled macrophage cells. An initial baseline fluorescence image was taken to distinguish between the MSC and the M Φ . This was followed by time lapse imaging under phase contrast to reduce photobleaching. Images were collected every 30 seconds over 1-2 hours. A final static fluorescent image was taken to analyze mitochondrial transfer. In order to mark changes in phagocytic activity, the imaging protocol was optimized by decreasing the magnification to observe a wider field and greater number of cells. pH-rodo conjugated zymosan particles were added to the cultures after the second static image. Another time-lapse video capture was conducted to measure the $M\Phi$ phagocytic activity levels followed by a final static fluorescent image. Data analysis will be performed using MATLAB to quantify mitochondrial transfer and the resulting changes in phagocytic activity. This protocol, designed to retrieve quantitative and qualitative data, will further our understanding of mesenchymal stem cell regulation of innate immunity. The molecular pathways that contribute to the mitochondrial transfer from mesenchymal stem cells to macrophages are yet to be clearly defined. This information can be used to develop off-the-shelf immunomodulatory therapeutic treatment for a broad range of diseases including metabolic disorders and cancer.

Author Name(s): Gonzalez, Zenovia; Mlynarczyk, Coraline Poster Presentation Title: Generation of pre-clinical models of BTG1 mutated diffuse large B cell lymphomas to help develop rational targeted therapies Affiliations: St. Francis College Category: Microbiology and Immunology (MI)

Abstract

Diffuse large B cell lymphomas (DLBCLs) represent the most prevalent and aggressive form of lymphoid cancer. One recurrently mutated gene in DLBCL is BTG1 (B cell translocation gene 1), with the most frequent missense mutation [Glutamine $36 \rightarrow$ Histidine (Q36H)]. BTG1 mutations associate with worse clinical outcome. Our research aims to develop improved models for these BTG1 mutant tumors, to help advance treatment options, particularly for patients who exhibit incurable forms of DLBCL. BTG1 mutations favor rapid expression of the MYC protein, a master regulator of cell growth in most cells of our body. To test potential therapeutic options for treating tumors with BTG1 mutation, we decided to generate a new pre-clinical tumor model that would recapitulate key features of aggressive B cell lymphoma in patients. We crossed three mouse lines to get a specific genetic configuration. First, the Btg1Q36H conditional knockin mouse line was used to express the mutant form of BTG1. Next, the Myc conditional knockin allele was introduced to express an extra copy of the Myc gene. Finally, the Cgamma1Cre line was used to selectively activate the above two alleles in germinal center-derived B cells. We obtained murine tumor models from a combination of in vitro and in vivo approaches. Specifically, we removed tumors from old mice (greater than one year and two months) that had the additional copy of the Myc gene, either with or without expression of the BTG1 mutant form. We observed the presence of tumors in the mesenteric lymph node in two out of two Myc+Btg1Q36H mice, while the Myc-only mice only showed signs of enlarged spleens in two out of two mice. We purified cells from these tumors and enlarged spleens and re-injected them into two types of recipient mice: Rag1KO (no immune system and therefore no rejection of foreign tumor cells) and wild-type (in case the tumors would be able to engraft despite recipients being immune-competent). We also placed these cells in culture, for intro modelling. In vivo imaging of live recipient mice showed efficient engraftment of Myc+Btg1Q36H cells in Rag1KO, but not WT mice, after five months. We will collect again these tumors for serial transplantation into new recipients, until they grow rapidly (less than a month) and consistently in Rag1KO and WT mice. In vitro, through longitudinal imaging of the cells in culture, we observed that MycCKI+Btg1Q36H, but not Myc-only cells formed dense cellular clumps, reflecting efficient growth in vitro. These cultures will be used for high throughput screening of drugs or drug combinations prior to testing them in our mouse tumor models in vivo. In conclusion, the combination of Btg1Q36H and Myc expression allowed us to create new in vivo and in vitro models of aggressive B cell lymphoma forms, when compared to Myc alone. These models will be valuable for pre-clinical drug testing, particularly for drugs that target pathways expected to be highly activated in Myc+Btg1 mutant tumors.

Author Name(s): Wang, Jessica; Makedonska, Anna; Reynisdóttir, Natalía; Gibb, Bryan Poster Presentation Title: Isolation and Characterization of Two Citrobacter Bacteriophage Affiliations: New York Institute of Technology Category: Microbiology and Immunology (MI)

Abstract

Antibiotic resistance is a growing public health crisis as bacterial infections fail to respond to the treatments that we have relied on for the past eighty years. Citrobacter freundii is a member of the Enterobacteriaceae family, a group comprised of Gram-negative facultative anaerobes that inhabit a wide variety of environments. C. freundii is a treatable opportunistic pathogen, but antibiotic-resistant strains are making infections more difficult to treat and dangerous, especially in hospitalized patients. Bacteriophages are viruses of bacteria that contain the ability to replicate in bacteria they infect and ultimately lyse bacteria. There is growing interest in the therapeutic application for treating bacterial infections, especially those that pose a greater risk due to antimicrobial resistance. We isolated two bacteriophages that infect C. freundii from wastewater. The phages have myovirus morphology and are extremely lytic against the host bacteria. The genome of one of the phages was refractive to restriction enzyme digestion, which may be due to the presence of DNA-modifying enzymes in the genome. The genomes of both phages were sequenced and found to be approximately 180 kb long. A comparative genomic investigation reveals that both phages are related, but other more closely related phages have been found, and additional investigation of the genome appears to show that these bacteriophages are reasonable candidates for phage therapy.

Author Name(s): Kim, Min Joo; Leger, Margueritte Poster Presentation Title: Defense gene NPR1 against oomycete pathogens; plants diseased or resistant? Affiliations: Mercy University Category: Microbiology and Immunology (MI)

Abstract

Plants play a vital role in our ecosystem and are essential for many organisms that depend on them for survival. Like most things in the ecosystem, plants can be infected and become diseased. Plant pathogenic microorganisms like fungi and oomycetes are one of the greatest threats to agronomically important crops such as potato and soybean. They can infect their host by secreting effector proteins into the interior of the host cell and sabotaging the host defense machinery. The plants try to fight off these infectious pathogens and their effector proteins by using their elaborate primary and secondary defense systems. Our research investigates the oomycete Hyaloperonospora arabidopsidis (Hpa), and how it affects a plant's host defense gene and its host defense pathway. We hypothesize that RxL23, an effector protein from Hpa, will suppress the host defense gene NPR1 in wild type soybean (Glycine max) plants. NPR1 gene in host plants are involved in the autophagy pathway. NPR1 exists in normal conditions in the cytoplasm, but once infected with a pathogen, there is an accumulation of plant hormone Salicylic Acid or SA, causing NPR1 to migrate into the nucleus due to post-translational modification, also known as phosphorylation. Autophagy plays a role in promoting and inhibiting pathogens; host genes can induce or inhibit plant autophagy during pathogen infections which resist pathogen infection. This pathway is important for plant growth, development, maintenance of cell homeostasis, and immune response. NPR1 in the autophagy pathway is phosphorylated and transferred from the cytoplasm to the nucleus, where it forms a protein complex with CDK8 and EDS1, promoting the expression of the PR1 gene, ultimately leading to Systemic Acquired Resistance (SAR) and PCD (Programmed cell death or localized immunity). Our current experiments are conducted using a simulation PlantSimLab software. We used this software to create models of the host defense pathways and run virtual knockdown experiments. On running virtual simulation experiments we observed that the knockdown of either one or multiple genes led to the plant losing its resistance or becoming diseased. We also observed that effector proteins can reduce plant resistance by suppressing essential host defense genes. Future experiments will involve host defense gene suppression assay to determine how RxL23 suppresses NPR1 in Glycine max using RT-qPCR.

Author Name(s): Narchal, Gurvin; Oddman, Kevin Poster Presentation Title: Investigating the effects of consumption of a high sugar content diet on the diversity of the oral microbiome Affiliations: SUNY Old Westbury Category: Microbiology and Immunology (MI)

Abstract

The oral microbiome is a complex community of microorganisms that inhabit the human oral cavity. These microorganisms span microbes from all the domains of life, e.g. Bacteria, Viruses, and Fungi. They play a crucial role in maintaining oral health and overall well-being. Dietary habits have a significant impact on the composition of the oral microbiome and may determine the state of health of an individual. We know that consumption of high sugary foods leads to tooth cavities (caries). We were interested in determining the effect of a high sugar content diet on the diversity of the oral microbiome. We expect that individuals who consume a high sugar content diet will have a less diverse microbiome. We collected oral swabs of 68 subjects and we extracted total DNA from the swabs using a Qiagen DNEasy Kit. All 16S rRNA illumina-tag PCR reactions were performed on the DNA extracts per the Earth Microbiome Project's protocol (Walters et al. 2016). The 16S rRNA gene was sequenced by Wright Labs (Huntingdon, PA, USA) using an Illumina MiSeq v2 chemistry with paired-end 250 base pair reads. The resulting sequences were processed using the DNA subway purple bioinformatics pipeline. A survey was also given to the same subjects with questions about their dietary habits. The results of this experiment showed that the alpha diversity indices, Faith's Phylogenetic Diversity, showed significance(p=0.017), while Pielous's Evenness had no significance. Beta diversity indices show no significant clustering of samples based on sugar food consumption. When looking at soda consumption, clustering appears in the Bray-Curtis Principal Coordinates analysis graph for subjects who rarely consume soda. Some clustering appears in the Jaccard from soda consumers and the Weighted Unifrac shows some clustering as well. The most prevalent bacterial phyla in the oral microbiome for all subjects are Fusobacteria, Bacteroidetes, Actinobacteria, Firmicutes, and Proteobacteria in ascending order of abundance. Fusobacteria constitute a lower proportion of the microbiome in subjects that do not consume sugar. Cyanobacteria are only present in subjects who consume higher sugar content diets. At the class level Clostridia are more abundant in subjects who consume high sugar diets. Cyanobacteria have been found to correlate to disease states in humans. It's important to note that individual responses to dietary habits can vary, and genetics and oral hygiene also play a significant role in determining oral health. Maintaining good oral hygiene practices, such as brushing and flossing regularly and visiting the dentist for check-ups, is essential for keeping the oral microbiome in balance. Additionally, making informed food choices that support oral health can help prevent oral diseases and promote overall well-being.

Author Name(s): Goyvaerts, Noor; Cepeda, Myah; Piotrowski, Natalia Poster Presentation Title: Effects of Quorum Sensing Molecules on the Adhesion Properties of Newly Isolated Yeast Strains Affiliations: Molloy University Category: Microbiology and Immunology (MI)

Abstract

Most microorganisms, including fungi, grow in aggregates to form multicellular communities called biofilms that adhere to inert surfaces in the environment as well as tissue surface in humans. Pathogens within the biofilm are more resistant to antimicrobial therapies. Quorum sensing (QS) is a cell- cell communication mechanism that coordinates density-dependent social behaviors in microbes. Biofilm and QS are interconnected processes in that QS regulates genes involved in biofilm formation. The effects of inter- and intra-species QS molecules on the adhesion properties of two laboratory strains as well as four newly isolated wild yeast strains were tested at different concentrations and time points. In the simulated co-living conditions with other microbes, different quorum sensing molecules exhibited various regulatory effects on yeast adhesion. Our data support that QS has emerged as a potential alternative for antimicrobials due to its capability to interfere with the social behaviors of microbes, such as adhesive protein-based biofilm development.

Author Name(s): Kaczmarski, Michael; Oommen, Nigel Poster Presentation Title: Comparative Analysis of Illumina MiSeq and Oxford Nanopore Sequencing for Bacteriophage Genomes: Assessing Accuracy and Efficacy Affiliations: New York Institute of Technology Category: Microbiology and Immunology (MI)

Abstract

Next-generation sequencing technologies such as the Illumina MiSeq sequencer enable the rapid and costeffective sequencing of genomes, while also providing very accurate genomes. Illumina sequencers prove to have many benefits to other sequencing technologies but only become a cost-effective process when performed at a high-volume scale of bases analyzed. Oxford Nanopore developed a different sequencing technology that is cheaper to purchase and operate, making it attractive for undergraduate research projects. However, there are questions and concerns that emerge about the accuracy of Nanopore sequencing and whether this data is of sufficiently high quality to be used as the sole source of sequencing data when publishing bacteriophage genomes. We present a case study of bacteriophage Argan, isolated in Arthrobacter globiformis NRRL B-2880 from soil on Long Island New York at New York Tech. The genome of Argan was determined by Illumina MiSeq at UPitt and found to be 55 kilobases with ninety putative genes. In addition to Argan, 15 other phage genomes were sequenced in an effort to explore the efficacy of Oxford Nanopore technology by using a MinION and version R9 flowcells at New York Tech. Our results are consistent with other reports that the R9 flowcells from Nanopore has systematic issues with homopolymer regions on DNA where the resulting sequence has an insertion or deletion of a nucleotide. While these errors are rare, they have a significant impact resulting annotated genomes. Newer flowcells chemistries from Oxford Nanopore promise improvements, but we feel the R9 chemistry is not accurate enough to be used as the sole source of sequencing data for publishing finished genomes into repositories like Genbank.

Author Name(s): Gadula, Srinidhi; Patel, Yamini Bhaveshbhai; Qiu, Jerry; Hwang, Alex; Nagarwala, Hamza
Poster Presentation Title: The Isolation, Characterization, and Optimization of Bacteriophages Targeting Clinical Isolates of Staphylococcus Aureus.
Affiliations: New York Institute of Technology

Category: Microbiology and Immunology (MI)

Abstract

Staphylococcus aureus, a prevalent strain of pathogenic bacteria, has been recognized as the second greatest contributor to global deaths from bacterial pathogens with approximately 900,000 associated deaths and 178,000 deaths attributed to antibiotic-resistant S. aureus. Staphylococcus is one of the most prevalent bacterial infections in the world which is commonly found in the environment and in the nose and skin of humans. Antibiotic-resistant forms of S. aureus such as methicillin-resistant S. aureus (MRSA) and vancomycin resistant S. aureus (VRSA) are widespread in community-acquired and hospital-acquired infections, which are challenging to treat with traditional antibiotics. With the growth of antibiotic-resistant strains of Staphylococcus aureus, it is vital to find other forms of future treatment for infection. Bacteriophages are viruses that infect bacteria and are often found in areas inhabited by the host bacterial species. As a result, bacteriophages offer a promising alternative treatment for treating challenging infections caused by S. aureus. We are in the process of creating a library of bacteriophages capable of infecting a strain of MRSA that will apply to a mouse infection model system to study phage therapy. The first phage we isolated came from a commercial phage cocktail which was prepared for phage therapy. The phage infected well at 30 °C, but poorly at 37 °C. Using sequential culturing at 37 °C, we isolated a mutant phage that infected the strain of MRSA efficiently at 37 °C. We recently isolated two additional phages from human nares and another eight bacteriophages are in the process of being isolated. As we continue to further characterize these isolated phages the most optimal will be used in a library of phages to conduct experiments to evaluate the therapeutic efficacy of bacteriophages in a mouse infection model system.

Author Name(s): Palermo, Samantha; Kloc, Anna Poster Presentation Title: Analysis of Parvovirus B19 mutations in human heart tissue layers Affiliations: University of New Haven Category: Microbiology and Immunology (MI)

Abstract

Myocarditis is the inflammation of the heart muscle (myocardium). The heart's capacity to pump blood may be affected by such inflammation. Chest pain, breathing difficulty, and irregular heartbeat, known as arrhythmia, can all be symptoms of myocarditis. The cardiac muscle may be inflamed as a result of bacterial, viral or parasitic infection, although viruses are thought to be the most common cause. Previously, Parvovirus B19 has been shown to cause acute cases of heart inflammation. This single-stranded DNA virus is associated with the fifth disease, a common childhood illness that causes a number of symptoms including fever, joint pain, rash and anemia. Even though the virus does not directly infect cardiomyocytes, the cardiac damage is thought to occur via infection of endothelial cell of the myocardial vessels and activation of inflammatory cells. The persistence of Parvovirus B19 in cardiac tissue after the initial infection is a well-documented phenomenon that has been associated with atypical angina pectoris and mitochondrial impairment. Because Parvovirus B19 has also been found in the hearts of healthy individuals, it is not known if viral persistence can damage the cardiac tissue over time.

In this study, frozen human heart tissue samples were obtained from one patient diagnosed with viral cardiomyopathy (Gill Heart & Vascular Institute-University of Kentucky). These samples contained three different heart layers: epicardium, endocardium and myocardium. The patient's EKG was designated as abnormal, and showed normal sinus rhythm, T wave abnormality, and prolonged QT segment. This patient received a heart transplant due to this condition. In the lab, a homogenizer was used to dissociate the heart tissue and extract DNA and RNA. Multiple sets of Parvovirus B19 specific primers that spanned the coding region of the viral genome were used to amplify said genome from the extracted DNA. The PCR products were visualized using gel electrophoresis, followed by gel extraction and purification. After the appropriate DNA sizes were obtained, Sanger Sequencing was used to generate sequences of the Parvovirus B19 coding region for each of the three layers of the heart. Full sequences were aligned with the reference genome, as well as with each other to identify viral mutations. Ongoing studies are focused on determining the significance of these mutations in the Parvovirus B19 genome in each heart layer. In the future, the effect of viral mutations on the resulting proteins will be studied to reveal a potential correlation between the persistence of Parvovirus B19 and heart disease.

Author Name(s): Mansfield, Kera; Acheampong, Joana; Foster, Tia, Carroll, Margaret A.; Catapane, Edward J.
Poster Presentation Title: Western Blot Study of the Different Neurotoxic Effects of Manganism and Parkinson's Disease
Affiliations: Medgar Evers College
Category: Physiology and Neuroscience (PN)

Abstract

Manganism, which is caused by high brain manganese (Mn) levels, is a human neurodegenerative disease with symptoms similar to Parkinson's Disease (PD), but with a different cause and neurotoxic action. Both conditions interfere with the dopamine (DA) system that originates in the substantia nigra. The therapeutic treatment for PD does not benefit people with Manganism. Manganism is involved with impairment of DA postsynaptic signal transduction pathway, while PD destroys DA neurons. Impairment of postsynaptic signal transduction pathways decreases response to stimuli. Neuronal degradation can result in postsynaptic denervation supersensitivity. In this study we contrasted the neurotoxic actions of Mn and 6-Hydroxydopamine (6-OHDA), which destroys DA neurons, on the presence of DA D2R receptors in gill tissue of Crassostrea virginica. Our lab uses the bivalve C. virginica as a model to study both diseases. C. virginica has a cilio-inhibitory DA innervation from its ganglia to the ciliated gill lateral cells (GLC). Upon ganglionic stimulation, the effectiveness of these cilio-inhibitory DA neurons to slow down GLC cilia beating rates is decreased by both Mn and 6-OHDA treatments. However direct application of DA to GLC will reduce cilia beating rates even after 6-OHDA treatments, but not after Mn treatments. We hypothesize Mn treatment will decrease the presence or sensitivity of gill D2R receptors, while 6-OHDA treatment will increase their presence or sensitivity. To test this, we used PAGE and Western Blotting (WB) to view and quantify gill D2R of animals treated 5 days with Mn (500 µg) or 6-OHDA (500 µg). Control animals were similarly treated without Mn or 6-OHDA. Gills were homogenized in detergent and centrifuged. Protein concentration of the supernatant was determined by Bradford. Aliquots were treated with Laemmli followed by PAGE (20-40 µg protein/well). After electrophoresis the gels were sandwiched for transfer onto nitrocellulose membranes. After WB the membranes were blocked, then incubated with D2R-HRP conjugated antibodies for 24 hours in TBST and 2% blocker at 4°C. Blots were viewed and analyzed with an iBright Fl1500 image analyzer. The blots showed D2R bands for controls and treated animals. The intensity of the bands for 6-OHDA treatments were slightly more intense than controls (+110%); whereas intensity of the bands for Mn treated were significantly less intense (-80%). The results are consistent with physiological data we observed measuring GLC cilia beating rates in similar experiments. The study shows the 2 neurotoxins have distinct and different mechanisms of action. This can be helpful in designing more appropriate therapeutic treatments for these similar neurological disorders. It also shows this preparation is a useful model to study regulatory mechanisms of ciliary activity as well as the pharmacology of drugs affecting biogenic amines in nervous systems.

The work was supported by grants 2R25GM06003 of the Bridge Program of NIGMS, 0537231071 of the CSTEP program of the NYSDOE, P120A210054 of the MSEIP Program of the DoEd, and K12GM093854 of the NIH IRACDA Program of Rutgers University.

Author Name(s): Cayemitte, Laurent; Saqib, Mahnoor; Carroll, Margaret A.; Catapane, Edward J. Poster Presentation Title: 6-Hydroxydopamine Treatment Causes Supersensitivity of Dopamine D2R Receptors in Gill Lateral Cells of Crassostrea virginica Affiliations: Medgar Evers College Category: Physiology and Neuroscience (PN)

Abstract

Gill lateral cell (GLC) cilia of the bivalve molluses Crassostrea virginica and Mytilus edulis are controlled by a dopaminergic-serotonergic innervation originating from their ganglia. Dopamine (DA) decreases, while serotonin increases, cilia beating rates. 6-Hydroxydopamine (6-OHDA) is a neurotoxin that selectively destroys DA neurons and is used to induce Parkinson's Disease in animal models. Previous work of our lab demonstrated that in both C. virginica and M. edulis, 6-OHDA treatments decreased DA levels in the visceral ganglia (VG) and decreased the cilio-inhibitory actions of DA when it was applied to the VG. A common phenomenon observed after nerve cell damage or denervation of an innervated organ is denervation supersensitivity, which results in an increase in postsynaptic receptor sensitivity to neurotransmitter application. Manganese toxicity causes the neurodegenerative disease Manganism, which has similar symptoms to Parkinson's Disease, but affects the DA postsynaptic signal transduction pathway. Previous work of our lab found that manganese toxicity in C. virginica and M. edulis decreases the cilio-inhibitory effect of DA on GLC when applied to the VG or directly to the gill. In this study we hypothesize that treating C. virginica with 6-OHDA would cause a supersensitivity response when DA was applied directly to the GLCs. To test this, we first injected 500 µg of 6-OHDA into the animals' posterior adductor muscle. Controls were similarly treated with vehicle injections. Both control and treated animals were then placed in individual containers of aerated artificial sea water (ASW) for 5 days after which dose responses of DA on GLC cilia beating rates were conducted. GLC beating rates were observed by stroboscopic microscopy. Animals treated with 6-OHDA demonstrated supersensitivity, as compared to controls, when DA was superfused to gill. The DA dose response curve of the 6-OHDA treated animals was shifted 1 log dose to the left, compared to controls. In contrast. earlier work with Mn treatments reduced DA potency, shifting the dose response curve to the right. The study shows that 6-OHDA produces a supersensitive response of DA postsynaptic receptors present in GLC. This simple animal preparation serves as a useful model to study regulatory mechanisms of ciliary activity as well as the pharmacology of drugs affecting biogenic amines in nervous systems.

The work was supported by grants 2R25GM06003 of the Bridge Program of NIGMS, 0537231071 of the CSTEP program of the NYSDOE, P120A210054 of the MSEIP Program of the DoEd, and K12GM093854 of the NIH IRACDA Program of Rutgers University.

Author Name(s): Celey-Okogun, Osemenga; Wang, Ping Poster Presentation Title: Ovarian Tissue Cryopreservation and Transplantation into Brown Adipose Affiliations: Medgar Evers College Category: Physiology and Neuroscience (PN)

Abstract

A cutting-edge technique known as ovarian tissue cryopreservation and transplantation is being used more frequently to assist maintaining fertility after gonadotoxic therapies, particularly in cancer patients. With the aid of in-vitro fertilization, about 30% of patients who have undergone auto-transplantation are able to give birth to living children. For women who are having trouble getting pregnant or who are receiving chemotherapy or radiation therapy, which can harm the ovaries, ovarian tissue transplantation is a procedure with a lot of promise. A part of the donor's ovarian tissue is first taken out during a laparoscopic procedure. After being vitrified or slowly frozen, the cryopreserved tissue is then thawed and transplanted. In this study, we used female mice and transplanted ovarian tissue into the brown adipose tissue (BAT) of the same mouse to determine if the transplants survived and the follicle numbers remained the same. We hypothesize that the transplanted tissues would survive and remain functional, providing positive hope for treatments to restore fertilization ability in cancer survivors. Our results showed that we were able to transplant ovarian tissue from donor mice to microscopically view and analyze using histological techniques. Using hemoxylin and eosin staining we were able to visualize secondary follicles in the transplanted ovarian tissue. The next phase of the study would be to use Optical Coherence Tomography (OCT) to view the ovarian grafts and determine the survival rates of the follicles and the function of the ovaries. OCT is an emerging technology for performing high-resolution crosssectional images of tissue structure on the micron scale in situ and in real time. Ovarian tissue transplantation, as a whole, is an exciting new area of reproductive medicine, offering hope for regaining fertility and enhancing the lives of women who are struggling with infertility. The process will likely continue to be improved, and its range of potential applications will increase over time as a result of ongoing studies and breakthroughs in the field.

Author Name(s): Myrbel, Nedjee; Obianke, Victory; Carroll, Margaret A., Catapane, Edward J. Poster Presentation Title: A Neurophysiology Role of Glutamate in Ganglia of the Bivalve Crassostrea virginica Affiliations: Medgar Evers College

Category: Physiology and Neuroscience (PN)

Abstract

Glutamate (Glu) neurons are major excitatory neurons in mammalian brains and various invertebrate ganglia. Disfunction of Glu neurons are associated with a number of human disorders including Parkinson's disease, Alzheimer's disease, Huntington's disease, autism, depression and schizophrenia. In higher animals, Glu receptors are classified as ionotropic NMDA, AMPA and kainate types; or metabotropic group I, II, and III types. Glu neurons have not been reported present or have a physiological function in the adult bivalve oyster Crassotrea virginica. However, NMDA type Glu receptors recently have been found reported to be involved in regulating bivalve metamorphosis in C. gigas, Mercenaria mercenaria and Mya arenaria. GABA is synthesized from Glu by Glu decarboxylase. In C. virginica and other studied bivalves, gills are innervated by serotonin and dopamine nerves from their visceral ganglia (VG). Serotonin is cilio-excitatory, while dopamine is cilio-inhibitory to gill lateral cell (GLC). Recently our lab detected GABA in C. virginica, and showed it has a neurophysiological function as a ganglionic neurotransmitter that inhibits serotonin neurons. Since GABA neurons are present in C. virginica, we hypothesize that Glu neurons also are present and that Glu serves as an excitatory neurotransmitter in VG. To test this, we used whole animal preparations, which have their neural connections to the ganglia and organs intact, and examined effects of Glu application to VG on cilia beating rates of GLC. Shells were removed and preparations placed into chambers with a barrier so drugs could be discretely applied to the VG or to the gill. Beating rates of GLC cilia were measured by stroboscopic microscopy. Applying Glu to the VG caused a dose-dependent (10-5 - 10-3M) increase in GLC cilia beating rates from about 5 to 20 beats/sec. The dose responses were repeated in the presence of dextromethorphan hydrobromide (DMT), a NMDA receptor antagonist. Applying DMT (10-3M) to the VG by itself reduced cilia beating rates from about 7 to 0 beats/sec. Applying Glu (10-5 - 10-3M) after DMT did not increase the beating rates. The study thus far revealed a physiological role for Glu as an excitatory neurotransmitter in the VG, most likely exciting serotonin neurons to cause an increase in GLC cilia beating rates. The results of the antagonist DMT are inconclusive at this point and need to be further investigated, as DMT itself caused a decrease in beating. DMT is reported to have several side effects, including decreasing reuptake of catecholamines. If it is decreasing reuptake of dopamine in the VG, then that could cause the deceased beating rates observed, as the cilio-inhibitory actions of dopamine, a catecholamine, would be increased. The bivalve mollusc gill is a useful model to study regulatory mechanisms of ciliary activity as well as the pharmacology of drugs affecting biogenic amines in nervous systems.

The work was supported by grants 2R25GM06003 of the Bridge Program of NIGMS, 0537231071 of the CSTEP program of the NYSDOE, P120A210054 of the MSEIP Program of the DoEd, and K12GM093854 of the NIH IRACDA Program of Rutgers University.

Author Name(s): Wallach, Rosanne; Pierre, Kandy; Foster, Tia; Catapane, Edward J., Carroll, Margaret A.
Poster Presentation Title: Presence of Glutamate Neurons and Glutamate Receptors in Ganglia of the Bivalve Crassostrea virginica
Affiliations: Medgar Evers College

Category: Physiology and Neuroscience (PN)

Abstract

Mammals and various invertebrates have glutamate (Glu) neurons serving as excitatory neurons in their nervous systems. Neurodegenerative diseases associated with Glu dysfunction include Parkinson's, Alzheimer's, Huntington's, autism, depression and schizophrenia. Glu is synthesized into GABA by Glu decarboxylase. Recently, our lab found GABA neurons present in Crassostrea virginica and Mytilus edulis, and showed they have a neurophysiological function as ganglionic neurons inhibiting serotonin neurons. The Glu NMDA receptor recently was found in and reported to be involved in regulating bivalve metamorphosis in C. gigas, Mercenaria mercenaria and Mya arenaria. Based upon this and the presence of GABA neurons in C. virginica, we hypothesize that Glu neurons and receptors are present in the ganglia of C. virginica. To test this, we used immunohistofluorescence (IHF) microscopy to view Glu neurons and the Glu GluR-1 receptor in the visceral ganglia (VG) of C. virginica, along with PAGE and Western Blotting (WB) to detect GluR-1 receptors. VG and posterior adductor muscle (PAM) were dissected and prepared for IHF or for PAGE and WB. Briefly for IHF, VG and PAM were snap frozen, cryostat sectioned, fixed with EDAC (N-Ethyl-N'-(3-dimethyl-aminopropyl) carbodiimide hydrochloride), treated with blockers, and incubated with GluR-1 FITC conjugated antibodies. Sections were viewed on a Leica epilume fluorescence microscope with a Leica DFC400 camera, 50 watt mercury lamps and FITC excitation/emission filters. All sections were photographed with the same camera setting at 200X and 400X. For PAGE and WB, PAM and 10 pair of VG were homogenized in detergent and centrifuged. Protein concentrations of the supernatant were determined by Bradford. Aliquots were treated with Laemmli followed by PAGE (20-40 µg protein/well). After electrophoresis the gels were sandwiched for transfer onto nitrocellulose membranes. After WB the membranes were blocked, then incubated with GluR-1 HRP conjugated antibodies for 24 hours in TBST and 2% blocker at 4°C. Blots were viewed and analyzed with an iBright Fl1500 image analyzer. The IHF showed the presence of Glu nerves and GluR-1 receptors in the cortex, and nerve fibers in the neuropile of the VG. The PAGE and WB showed good bands of GluR-1 receptor proteins in VG and to a lesser extent in the PAM. The study shows Glu neurons and GluR-1 type receptor present in the VG of C. virginica. This study complements other work of our lab showing a neurophysiology function of Glu in the VG as an excitatory neurotransmitter. Together they demonstrate that C. virginica is a useful model to study neurophysiology as well as the pharmacology of drugs affecting the nervous systems.

The work was supported by grants 2R25GM06003 of the Bridge Program of NIGMS, 0537231071 of the CSTEP program of the NYSDOE, P120A210054 of the MSEIP Program of the DoEd, and K12GM093854 of the NIH IRACDA Program of Rutgers University.

Author Name(s): Small, Shatema; Wilson, Bellavia; Joseph, Kinida; Carroll, Margaret A.; Catapane, Edward J.
Poster Presentation Title: Comparison of the Neurotoxic Actions of 6-Hydroxydopamine and Manganese on Gill Lateral Cell Dopamine D2R Receptors of Crassostrea virginica
Affiliations: Medgar Evers College
Category: Physiology and Neuroscience (PN)

Abstract

Parkinson's Disease and Manganism are two human neurodegenerative diseases with similar symptoms but different neurophysiological causes. Parkinson's causes degeneration of dopamine (DA) neurons in the substantia nigra. Manganism, which is due to elevated manganese (Mn) levels in the brain, does not degenerate DA neurons; rather the postsynaptic signal transduction pathway is impaired. Neuronal degradation can cause denervation supersensitivity at the innervated cells, whereas postsynaptic signal pathway impairment causes decreased response to stimulation. 6-Hydroxydopamine (6-OHDA), a neurotoxin that selectively destroys DA neurons, induces Parkinson's Disease in animal models. Our lab showed the bivalve Crassostrea virginica is a useful model to study both diseases. In C. virginica, treatments with either 6-OHDA or Mn reduce the ability of cilio-inhibitory DA neurons to slow down the beating rates of gill lateral cell (GLC) cilia. The present study aims to contrast the neurotoxic actions of 6-OHDA vs Mn at the D2R of GLC by using immunohistofluorescence (IHF) microscopy to determine changes in D2R fluorescence intensity. We hypothesize that animals treated with 6-OHDA will have increased D2R fluorescence intensity at the GLC, while animals treated with Mn will demonstrate decreased D2R fluorescence intensity. To test this, we used IHF microscopy to view and quantify D2R fluorescence intensity of GLC of animals treated for 5 days with 500 µg of 6-OHDA or Mn. Control animals were similarly treated without 6-OHDA or Mn. After 5 days, gill sections were processed with D2R/FITC conjugated antibodies and viewed on a Leica fluorescence microscope with epilume illumination using a 50 watt HBO mercury excitation lamp fitted with FITC excitation/emission filters. Photomicrographs were taken with a Leica DFC400 camera at 100, 200 and 400X. All sections were photographed with the same camera settings and GLC fluorescence intensity was measured using ImageJ from NIH. Statistical significance was determined by ANOVA. The IHF results showed 6-OHDA treatments increased D2R fluorescence by 40%, while Mn treatments decrease it by 35% compared to the controls (p< 0.001 for comparison of 6-OHDA and Mn treated to controls). These results are consistent with the physiological data we observed when measuring GLC cilia beating rates in similar experiments. The study shows the 2 neurotoxins have distinct and different mechanisms of action that can be helpful in differentiating the cause and designing the appropriate potential therapeutic treatments for these neurological disorders. It also shows this preparation is a useful model to study regulatory mechanisms of ciliary activity as well as the pharmacology of drugs affecting biogenic amines in nervous systems.

The work was supported by grants 2R25GM06003 of the Bridge Program of NIGMS, 0537231071 and 0537231091of the CSTEP program of the NYSDOE, P120A210054 of the MSEIP Program of the DoEd, and K12GM093854 of the NIH IRACDA Program of Rutgers University.

Author Name(s): Miura Traficante, Saheed Lawal, Sung-Hoon Kim, Matthew Dickinson, Keith Yeung, Andrew Gallagher, Olivia Tabeka, Lauren Senia, Prerana Shrestha
Poster Presentation Title: Investigating Dysregulated Emotional Behaviors in Tuberous Sclerosis
Complex with Behavior Battery and Calcium Photometry
Affiliations: Stonybrook University
Category: Physiology and Neuroscience (PN)

Abstract

The present research seeks to better understand Tuberous Sclerosis Complex (TSC), a disorder with high neurological sequelae disorder comorbidity and the mechanism behind anxiety symptomologies that commonly exist within the TSC disease model archetype. More specifically, we seek to understand how TSC relates to emotional regulation with respect to emotional memory consolidation as well as the learned versus innate affective behaviors. This experiment was conducted in two segments. First, heterozygous mice with a knockout of a TSC2 allele were divided into single-housed and group-housed mice. Mice then performed a behavior battery that included the open field test, the marble burying test (MBT) and the 3-Chamber Social Interaction (3-CSI) test to analyze anxiety levels in TSC. Additionally, memory and learning was assessed in an avoidance paradigm using signal active avoidance (sAA), a test that utilizes Pavlovian cued fear conditioning to pair a foot shock with a threat cue (tone). The second segment of this experiment utilized fiber photometry to analyze calcium transients specifically in the medial prefrontal cortex (mPFC) during Pavlovian fear conditioning training, using genetically encoded Calcium sensor (CaMP). CaMP was expressed pan-cellularly in the mPFC. Anxiety-like behaviors were seen in the marble burying tests with significantly more marbles buried in cohorts who were singlehoused in comparison to group housed mice. For cohort 3, single housed mice spent less time in the middle and more time in the corners, suggesting higher levels of anxiety. During the 3-CSI deficits in social novelty were observed. Fiber photometry revealed a calcium transient pattern in mPFC of control wild-type mice that spiked during the shock but not during the tone presentation during the training session whereas there were also robust transients to the tone presentations in the memory retrieval test indicating that learning likely occurred. Interestingly, during consolidation, a similar pattern of cellular calcium activity was observed. Future research will investigate if deficits and dysfunction exist in calcium transient levels in the TSC2 heterozygous mouse model.

Author Name(s): Ethan Valle; Mohamed Noor; Emma Sarinick; Michael Coyle; Christopher Bishop Poster Presentation Title: The effects of the serotonin drugs Vortioxetine and Vilazodone on Apomorphine-induced dyskinesia in a rat model of Parkinson's disease Affiliations: Westchester Community College/Binghamton University Category: Physiology and Neuroscience (PN)

Abstract

Parkinson's disease (PD) is a movement illness that results from the death of nigrostriatal dopamine neurons and is characterized by bradykinesia, stiffness, tremors, and postural instability. L-DOPA, a dopamine precursor, is the most frequently prescribed medication for Parkinson's disease. However, L-DOPA-induced dyskinesia (LID), or abnormal involuntary movements, is brought on by long-term L-DOPA use. Vortioxetine and vilazodone, two FDA-approved serotonin receptor medications, have been shown in prior research to lower LID while maintaining the therapeutic effects of L-DOPA, although their exact mechanisms of action are yet unknown. We aimed to understand further the effects of vortioxetine and vilazodone, given previous research suggesting that 5-HT chemicals may operate at pre- and postsynaptic locations in the brain. In order to do this, adult male and female Sprague-Dawley rats were first made hemiparkinsonian by unilateral injection of 6-hydroxydopamine into the medial forebrain bundle and then made dyskinetic by prolonged L-DOPA administration. Before giving dyskinetic animals the post-synaptically acting dopamine receptor agonist apomorphine (APO), the rats were given a vehicle, a low, and a high dose of either vortioxetine or vilazodone. The findings indicate that our medications' effectiveness is probably mediated by presynaptic regulation of L-DOPA because they have very little postsynaptic effects.

Author Name(s): Tamayo, Sara; Shikapwashya, Gabriella Poster Presentation Title: Potential Role of LRRC8 in Supplying Cysteine to Neurons Via GSH Release Affiliations: Mercy University and Westchester Community College Category: Physiology and Neuroscience (PN)

Abstract

Astrocytes are a major source of the antioxidant glutathione (GSH) in the brain. Understanding the central role that astrocytes play in enhancing neuronal GSH levels is integral for developing therapeutics aimed at treating neurodegenerative diseases, such as Alzheimer's and Parkinson's, and acute disorders such as hepatic encephalopathy and stroke. The volume regulated anion channel (VRAC), composed of leucine rich repeat containing (LRCC8) subunits, releases chloride, GSH and other organic osmolytes in response to cell swelling. With prominent astrocyte swelling found in neurodegeneration and acute brain disorders, we hypothesize that LRRC8 represents a prominent pathway for GSH release and contributes to the antioxidant defense of the brain. While astrocytes release GSH, most evidence suggests that neurons do not directly take up GSH, but rather rely on the uptake of the GSH-rate limiting precursor cysteine. GSH, once released from astrocytes, becomes the source of cysteine for neurons and facilitates neuronal GSH synthesis. In the current study, we confirmed that GSH released from astrocytes cultured from chick embryonic optic tecta and swollen via either hypoosmotic or ammonia treatments to activate LRRC8, contributes to increased cysteine (reduced form) or cystine (oxidized form) levels in the extracellular space. Future experiments are underway to determine if this swelling-dependent release indeed occurs via LRRC8 and whether it subsequently contributes to neuronal GSH synthesis.

Author Name(s): Bozor, Katoucha; Austin, Mark Poster Presentation Title: Volume Regulated Anion Channels Are Essential for GSH Release in Astrocytes Affiliations: Mercy University Category: Physiology and Neuroscience (PN)

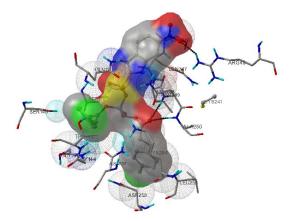
Abstract

Glutathione (GSH) is a major antioxidant that protects the brain from neurodegeneration and acute brain disorders, including Alzheimer's, Parkinson's, stroke and hepatic encephalopathy. Astrocytes produce higher levels of GSH than neurons and are central to supplying GSH precursors to neurons, thus facilitating neuronal GSH production. While the multi-drug resistant protein is considered the canonical GSH release pathway in astrocytes, we hypothesize that the volume regulated anion channel (VRAC) represents an equally important pathway for astrocytic-GSH release and subsequent neuronal GSH synthesis. VRACs, which activate in response to cell swelling and are composed of leucine rich repeat containing (LRCC8) subunits, have previously been found to release GSH. However, a connection between astrocytic VRAC and neuronal GSH synthesis has yet to be firmly established. As a first step, we characterized VRAC-dependent release in our model system, primary astrocyte cultures prepared from chick embryonic optic tecta. Astrocytes were swollen with hypoosmotic media or ammonia, which led to increased GSH levels in the extracellular space. Experiments with VRAC inhibitors are being performed to verify the role of VRAC in this swelling-induced GSH release. Future studies will establish if the levels of GSH released via this manner are sufficient to enhance neuronal GSH levels by supplying the rate-limiting precursor cysteine.

Author Name(s): Ronelle Robinson, Rochaiu Daye and Abu Gafar Hossion* Poster Presentation Title: Virtual Screening of Novel Benzimidazole Sugar Analogue with Microtubule Protein as Potent Chemotherapeutic Candidate Affiliations: University of Bridgeport Category: Biochemistry, Biophysics and Biotechnology (BBB)

Abstract

Cancer is essentially a disease of mitosis; consequently, owing to its essential role in mitosis, microtubule has been a key target in anticancer therapeutics.1 Microtubule binding agents (MTA) such as Colchicine "bind to β -tubulin in the α - β heterodimer and suppress microtubule dynamics. Colchicine binds strongly to the β -tubulin portion of the heterodimer at the CYS241 residue and sterically hinders the α -tubulin portion. This study utilized the computational aid, AutoDock 4.0, to conduct a virtual screening of the small molecule as inhibitors that potently inhibits the polymerization of tubulin. An aim to assist the development of novel microtubule inhibitors (MTI), a new class small molecules (TL-2) were designed. The TL-2 is an analogue of our recently developed ligand of T1, 2-thio- β -D-2',-3',4.'5'-tetra-Oacetylglucosyl-5- nitrobenzimidazole2. In an interest to develop novel Microtubule binding (MTI), TL-2 successfully designed with added side chain in the sugar moiety at 2' and 3' position of T1. The study revealed strong electrostatic interactions of TL-2 with the $\alpha\beta$ -heterodimer. Data were analyzed with respect to conformational binding energy and clustering of binding conformations. TL-2 demonstrated a greater number of electrostatic interactions than Colchicine. The strongest interaction observed for the ligand-protein complex was -9.24 kcal/mol. The observed intermolecular forces of the TL-2- protein complex showed 3 H-bonds with the β -chain residues ARG-48, ASN-249 and ALA-250. ASN249 and ALA-250 are H-bonded to the carbonyl group of the sugar moiety on TL2. A 4th H-bond between TL-2 and the GLN-11 residue of the α -chain, further stabilizes the complex in this conformation, as shown in Figure below.



References: 1. Bioorganic & Medicinal Chemistry, **2000**, 8, 2417. 2. Int. J. Phys. Chem. Biophys. **2022**, 12.