



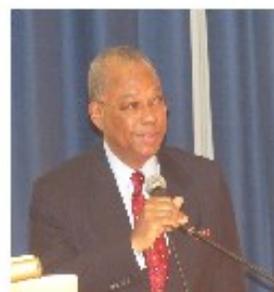
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Formation of Structures Resembling Early Embryonic Neural Plate in Traumatized Adult *Danio rerio* Optic Tectum Maintained in Organotypic Culture: a Morphological Study

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Abstract

Organotypic cultures of adult zebrafish brain pieces were used as an experimental system to study neurodegenerative and neuroregenerative processes happening in traumatized adult brain. We found that in these conditions, the surviving tissue pieces of the optic tectum of adult zebrafish brain show hallmarks of tissue degeneration leading to the classical spongiform appearance. However, some cell populations survive and form organized structures which resemble embryonic neuroepithelial tissue appearing similar to that seen in the neural plate and/or neural tube of vertebral embryos. These *restorative structures* or rudiments of a new brain formed in the traumatized, degenerating brain tissue could provide further avenues to analyze cell-specific events in a complex multicellular environment following brain trauma.

Keywords: traumatic brain injury (TBI), organotypic culture, zebrafish, optic tectum, degeneration, brain regeneration, embryoid brain rudiment

Introduction

Traumatic brain injury, frequently referred to as a silent epidemic, is a serious medical condition. Globally, it affects millions of people and it burdens not only the patients and their immediate families, but also the society as a whole^{1,4}. Although scientific evidence supports that in the adult mammalian brain, regeneration may occur and lead to some degree of functional recovery if the conditions are correct or treatment is delivered within the appropriate short window of opportunity, these events are limited because they are complicated by the intricate cellular relationships in the mammalian brain⁴. In the contrary, the more basic or simpler teleost fish brain shows high regenerative capacity of many

vital organs⁵⁻⁷ including the brain, even into late adulthood⁸⁻¹³. As such, they are good model animals to analyze biological phenomena leading to brain repair in adulthood following brain trauma, and may give some insight into what has changed in evolution of the more complex brain leading to reduced regenerative capacity. In this study, we selected a small teleost, the adult zebrafish (*Danio rerio*) brain to investigate posttraumatic cellular events.

Zebrafish lately have become a well-studied laboratory vertebrate animal in the fields of genetics, developmental biology, physiology and neurobiology to mention a few fields^{14,15}. Based on published data suggesting that adult zebrafish whole brain can survive in organotypic culture conditions for a considerable period of

time¹⁶, we hypothesized that chunks of adult brain would also survive in similar culture conditions. The process of removing the brain from the skull of adult zebrafish and chopping it up into pieces is, in itself, an extremely robust TBI and we anticipated that keeping these traumatized adult brain pieces in organotypic culture would help to display cellular events that may be complementary to those occurring in the *in vivo* brain and as such may provide a new model to study TBI.

Our objective was to find out whether dissected pieces of adult brain tissue survive in culture, and if they do, how long could they be kept viable and what would be the tissue characteristics at different time points of the cultivation period. We selected the optic tectum of the adult zebrafish (Fig. 1) as the region to be used because it is a well-studied, layered cortical structure with radially oriented cells interconnecting the ventricular (ependymal) and pial surfaces (Fig. 1B,C), a structure, the destruction of which as well as its reorganization, (regeneration) could be easily recognized¹⁷. The small size of the zebrafish brain makes it advantageous for easy visualization of an entire brain region under the light microscope (Fig. 1), therefore it takes considerably fewer images to assemble a high resolution montage from the analyzed region than of the mouse or rat brain, for example¹⁷.

Materials and Methods

Animals

Twenty seven fish were used in this study. The animals were maintained together in a large tank inside the lab on the lab's normal light cycle and fed twice a day. The experimental animals were

maintained individually, according to the Zebrafish Book until the surgery was performed to start the culture¹⁵.

This study was carried out in strict accordance with the recommendations in the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health. The protocol was approved by the Committee on the Ethics of Animal Experiments of the New York State Institute for Basic Research in Developmental Disabilities (Permit Number: 359). All surgery was performed under 0.04% tricaine solution, and all efforts were made to minimize the suffering of the subjects.

Surgical procedure

The surgical techniques were performed in aseptic conditions. This was important in order for the pieces to not carry over bacterial or fungal contamination into the culture. Surgery was carried out with micro-surgical tools and was done in a timely fashion, not exceeding five minutes to ensure that the cells did not die before the culture could be established.

In short, the skull was opened by cutting at the brain stem, severing the spinal cord and pulling forward. The brain was cut just below the medulla, lifted slightly and was then removed by grabbing and tearing the optic nerve, which is the main point of anchor of the brain to the skull. The brain was placed in a Petri dish with the pre-warmed culture medium to give the cells immediate nutrition and osmotic conditions and were first cut in the midsagittal plane for two symmetrical hemisphere pieces which then were cut in coronal plane at the border of the forebrain/optic tectum and again at the border of the optic tectum and cerebellum. The forebrain and

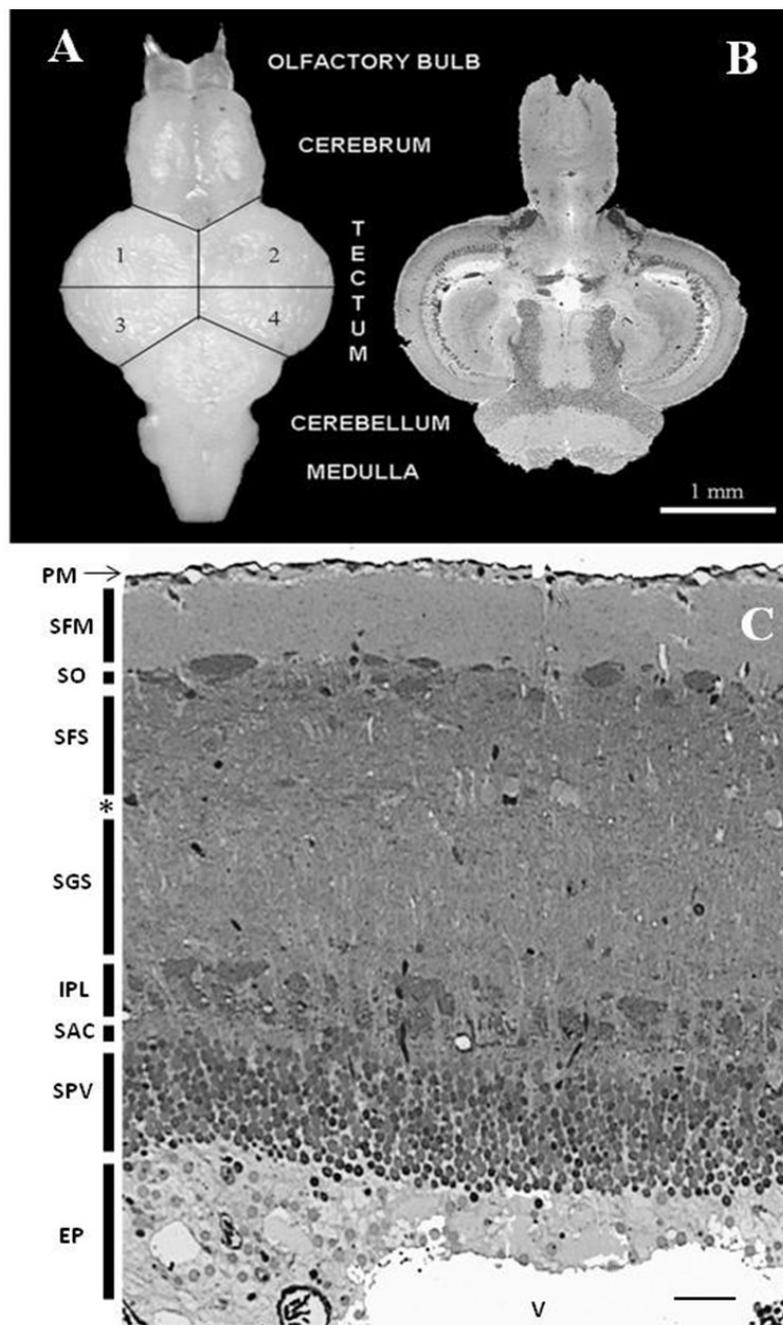


Figure 1 – General overview of the adult zebrafish brain and the basic structure of the optic tectum. Image A depicts the dorsal stereomicroscopic image of the adult zebrafish brain with demarcating lines and numbers indicating the pieces of the optic tectum used for organotypic culture. Image B depicts a montage of light microscopic images taken of a horizontal section of the whole adult zebrafish brain, using the 10x objective. Image C depicts a montage of light microscopic images of the cortical region of the right optic tectal hemisphere, taken using the 100x oil immersion objective. The layers of the optic tectum are: **PM** - Pia mater, **SFM**— Stratum Fibrosum Marginale, **SO**—Stratum Opticum, **SFS**—Stratum Fibrosum Superficiale **Asterisk**- Pyramidal-like cell layer **SGS**—Stratum Fibrosum Griseum **IPL**—Internal Plexiform Layer **SAC**—Stratum Album Centrale **SPV**—Stratum Periventriculare **EP**—Ependyma, **V**—Tectal Ventricle, For additional information concerning the morphological details of the optic tectum, refer to Corbo, 2012. Scale bar = 50 μ m

cerebellum were discarded. The two optic tectum hemispheres were cut in half in the coronal plane ending up with four pieces of optic tectum per brain (Fig. 1A). Each of the pieces was placed into its own chamber of a six well dish and were then transported to the tissue culture room.

Preparation and Maintenance of the Organotypic Culture

In a Laminar flow hood, the pieces of tissue were removed from the six well chambers and placed onto sterile Millicell tissue culture inserts (cat# PICM03050). The Millicell insert helped to keep a thin layer of medium over the tissue which was constantly changing due to capillary action. The medium recipe was adopted from Tomizawa *et. al.*¹⁶. All pieces were placed on the same insert in 6ml Petri dish. Exactly two milliliters of warm medium was added to the Petri dish. The cultures were kept in a carbon dioxide (5%) water jacketed incubator at 32°C for the appropriate time according to the time course of the research design. Samples were fixed at 0, 2, 4, 6, 12, 24, 48, 72, 96 hours and 7 and 14 days.

Histotechniques

At the selected time-point, the pieces were immersed into a picric acid fixative, containing 0.25% glutaraldehyde and 4% paraformaldehyde buffered to a pH of 7.2, for one hour followed by 1% osmium tetroxide post-fixation for another hour. The picric acid fixative was chosen due to its low concentration of glutaraldehyde. This fixative is necessary for electron microscopy analysis but did not destroyed antigens within the tissue allowing also for future immune-electron microscopy to be carried out.

The tissues were dehydrated through increasing concentrations of ethanol and then embedded into Durcupan (Electron Microscopy Sciences), which was infiltrated into the tissue using propylene oxide. The samples were polymerized in Durcupan overnight in a 70°C oven. Semithin-sections were cut on Sorvall MT-2B ultramicrotome using freshly prepared glass knives. The 1 micrometer thick, so called semithin-sections were stained with 1% toluidine blue containing 0.5% tetraborate.

Light Microscopy and Image Analysis

The semithin sections were analyzed on the BX-40 Olympus light microscope outfitted with a AmScope 10 mp digital camera. Photographs were captured on high magnification with ToupView capture software and are presented as montages, which were assembled using Adobe Photoshop.

Results

We analyzed the organotypic culture pieces on a time course, fixing four tissue samples from three animals each at 0, 2, 4, 6, 12, 24, 48, 72, 96 hours; 7 and 14 days. Our study clearly demonstrates that samples of adult zebrafish optic tectum were able to survive in organotypic culture for as long as we studied.

The basic histological structure of the optic tectum can be detected up to six hours of cultivation. Around 12 and 24 hours of cultivation, the tissue chunks obtain a flattened, up and down convex shape with a rounded footprint. Signs of spongiform degeneration start to appear as early as 12 hours after the beginning of the organotypic culture. While the expansion of the spongiform degeneration progresses, around 48 hours we begin to

see the appearance and accumulation of rounded, healthy looking cells at the periphery of the tissue sample (Fig. 2 A-C). Around 96 hours of survival, in certain regions, among the mixture of surviving spongiform degenerative tissue and regenerating regions, the formation of different, structural units can be also detected (Fig. 3A,B). Other regions of the surviving tissue show signs of spongiform degeneration (Fig. 3D), which are associated with large numbers of highly granulated and large cells (Fig 3C), which may be mast cells since they show metachromasia with toluidine blue.

Other formations show definite structural architectures some of which are resembling embryoid neural plate or even neural tube (Fig. 4-7). Although similar structures could be found in nearly every sample older than and including 96 hours of cultivation, their structural advancement differs between samples and depends on how progressed they are in development at the time of fixation (Fig. 4-6). While in this study we analyzed a large number of samples during the entire time course using plastic embedded semithin sections, different histo-technical methods and electron microscopy, in this paper we publish only our light microscopic (LM) morphological findings of the general composition of surviving samples and focusing on the neural-epithelial-like "new brain" rudiments, or as we call them, "restorative structures" presenting samples from 24, 96 hours as well as 7 and 14 days survival times.

These neural plate/tube-like structures always appear within the regions of spongiform degeneration and are surrounded by numerous, large, granulated cells. Metachromasia in our samples either appears reddish, if the tissue is not postfixed with osmium tetroxide, or green in osmified tissue

samples. Signs of degranulation of these cells were also readily detectable (Figs. 3B,D, 4, 5). While some surviving tissue components could be recognized throughout the entire specimen, they were easily distinguishable from the newly formed tissue components even with light microscopy.

An overall light microscopic image of a neural plate-like structure found in one of our 96 hour sample is depicted in figure 4. This particular image is a montage of around 50 individual micrographs captured using 100x, oil immersion objective. Due to the limitation of the print publication, the final montage is reduced to 15% of its original size. This rudiment is a horseshoe-shaped cellular band, consisting from neuroepithelial-like elongated cells with a pial-like tissue (external limiting membrane) at its convex surface and a ventricle-like region at its concave edge. Many small capillaries can be detected across the entire length of the pial surface while some large, vein-like, vessels are also present and filled with fresh-looking red blood cells as signs of neovascularization and hematopoiesis. The ventricle-like space at its concave edge is lined with a row of cells resembling early ependymal layer and is filled with a homogeneous, colloidal appearing fluid the upper boundary of which is studded with a row of vacuoles associated with the ventricular end-feet of the neuroepithelial layer cells. Surrounding this neural plate-like structure, newly-formed cell masses show further signs of neovascularization and "rosette" formation. Rosettes, typical components of the periventricular grey zone in a healthy, adult zebrafish optic tectum are described in our earlier publication Corbo and others (2012). At the lower boundary of the ventricle-like space (V), a row of cuboidal cells

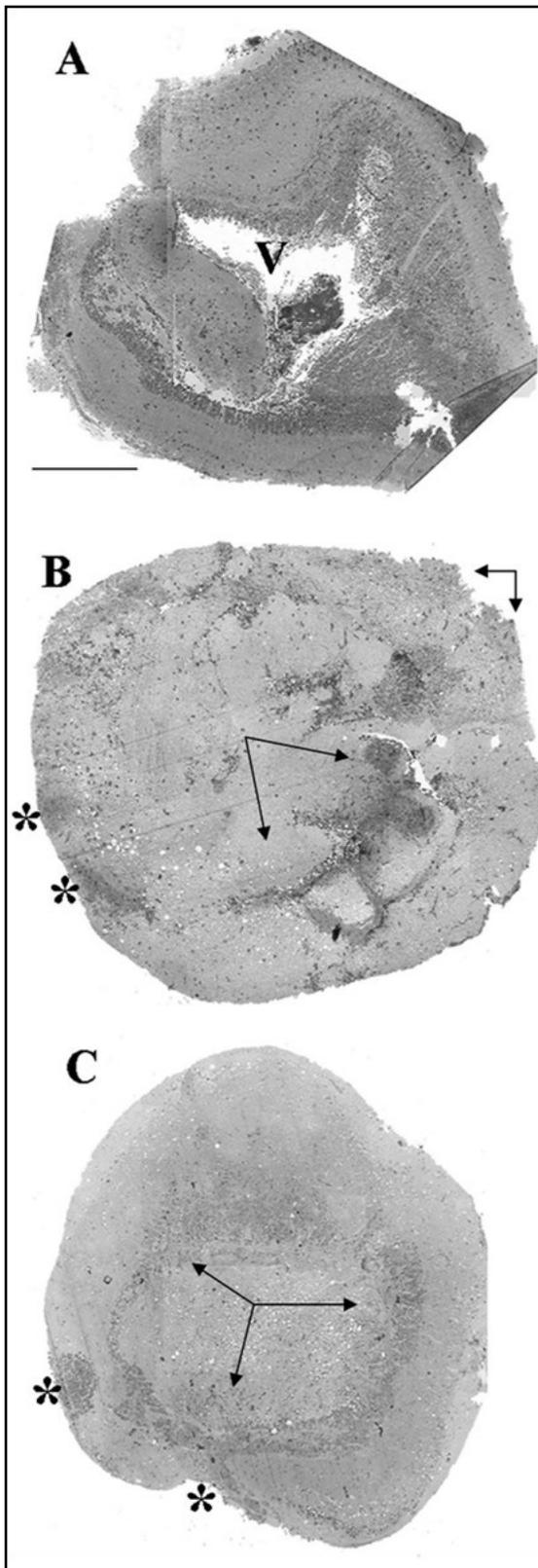


Figure 2 - General morphology of surviving tissue samples. Plastic embedded semithin sections stained with toluidine blue captured at high magnification and assembled into a montage. Images A, B, and C present a general overview of the cultivated optic tectum pieces at 24 hours, 96 hours, and 14 days, respectively. Note that at 24 hours survival time (A), the basic layered structural characteristics of the optic tectum together with the remnant of the lateral ventricle (V) can still easily be recognized. At 96 hours of cultivation (B) as well as at 14 days of cultivation (C), the samples have flattened and obtained a more or less rounded footprint while the cellular components are redistributed. Many large, rounded cells (shown at higher magnification in Figure 6A & B) accumulate around the periphery of the samples (asterisks or peripheral arrows). Some of these “simple appearing” cells are rounded with pale cytoplasm, centrally located, large nucleus with one nucleolus resembling undifferentiated neuroblasts while others are smaller with dense chromatin and a small cytoplasm. Some other cells form organized structures within the tissue chunks (internal arrows in images B and C) and form more complex cellular aggregates among regions with signs of spongiform degeneration (image D in Figure 6). Usually these structures are associated with large numbers of highly granulated and large cells depicted in a higher magnification in image C on Figure 3. Scale bars: A, B, and C - 500 μm .

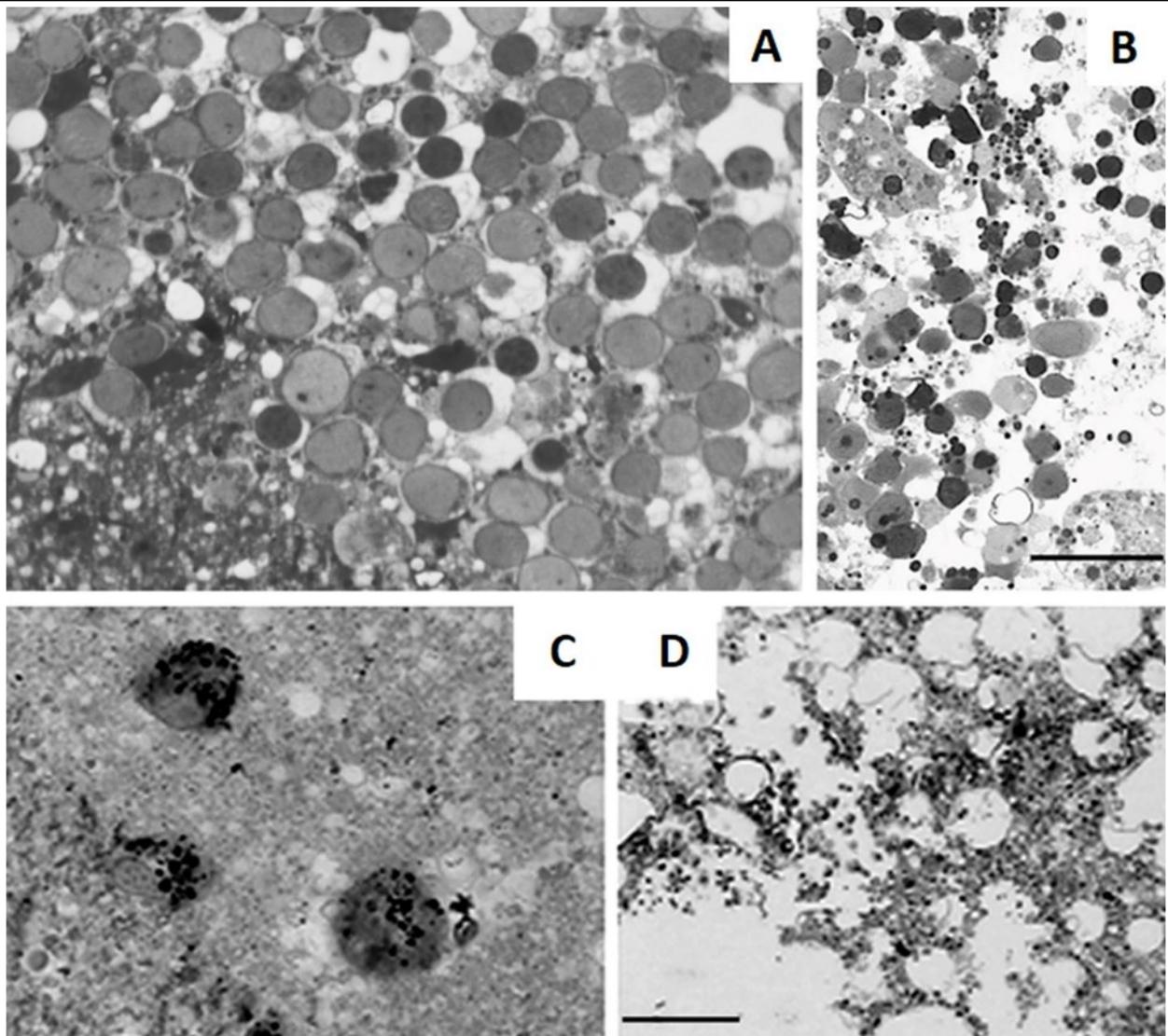


Figure 3 - Large, rounded cells accumulating around the periphery of the samples. (A & B) One type of these cells show pale cytoplasm, centrally located, large nucleus with one nucleolus resembling undifferentiated neuroblasts while other type represent smaller cells with dense chromatin and a small cytoplasm. Some other regions still show signs of spongiform degeneration (D). Usually these regions are associated with large numbers of highly granulated and large cells (C), which may be mast cells since they show metachromasia with toluidine blue. Scale bars: A, C & D - 75 μm ; B - 150 μm

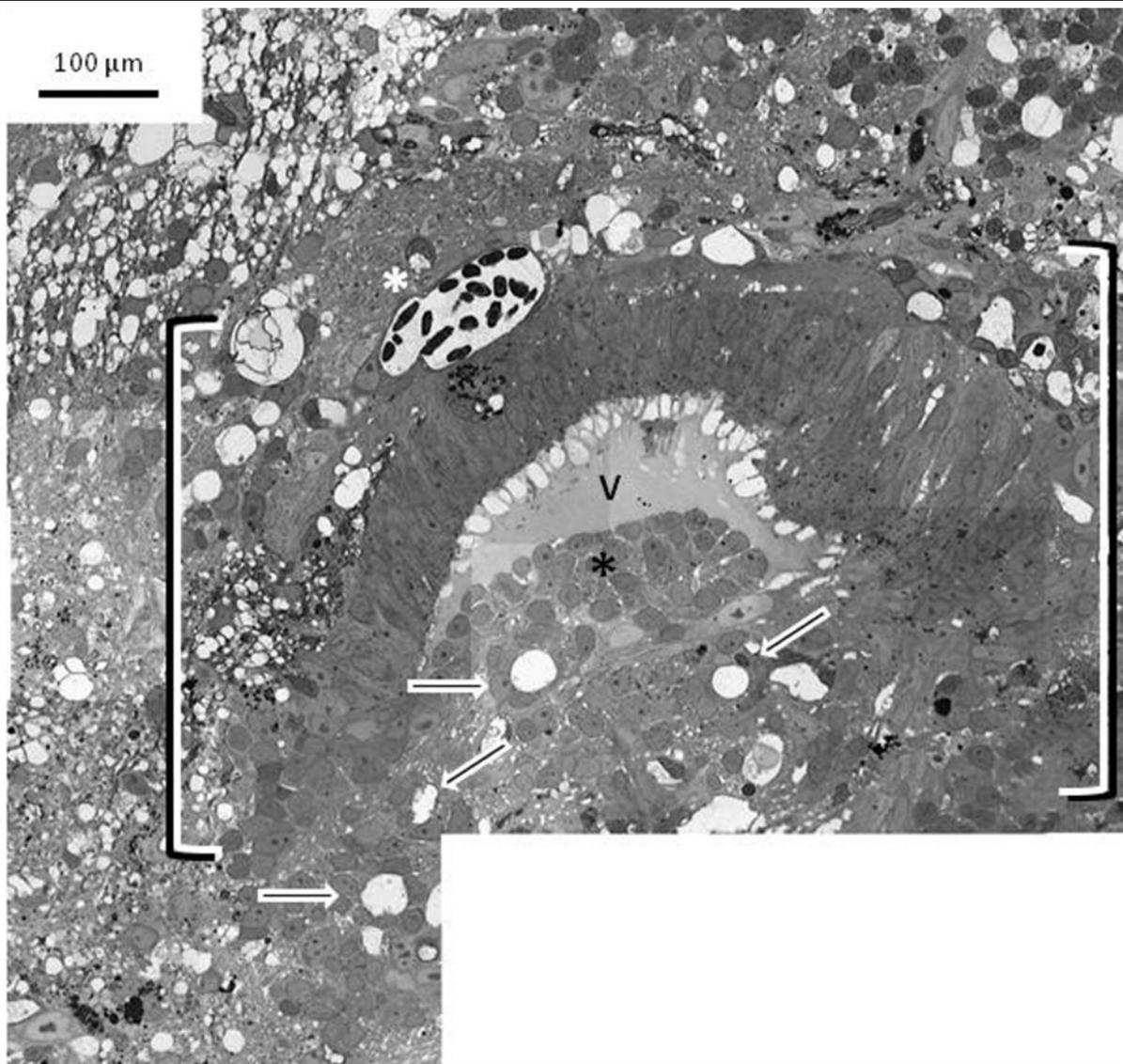


Figure 4 – An overall light microscopic image of neural plate-like structure in a 96 hours surviving organotypic culture of the adult zebrafish optic tectum. The image is a montage of 36 individual frames captured using the 100x oil immersion objective. The horseshoe-shaped structure located in the center of the image is framed by two large, black and white brackets on either side. At its convex upper surface, equivalent to the pia mater, a large, newly formed blood vessel (white asterisk) with fresh erythrocytes and at least one leukocyte can be recognized. Similarly, smaller capillary formation can be detected across the entire length of the pial surface. In the lower, concave area, a ventricle-like space (V) can be recognized filled with a homogeneous, colloidal-looking fluid the upper boundary of which is laced with a row of uniform, white vacuoles. Below the ventricle-like space, a newly-formed cell mass (black asterisk), which includes signs of neovascularization and “rosette” formation (white & black arrows). Surrounding the neural plate-like structure, a mixture of surviving spongiform degenerative tissue and regenerating regions can be found.

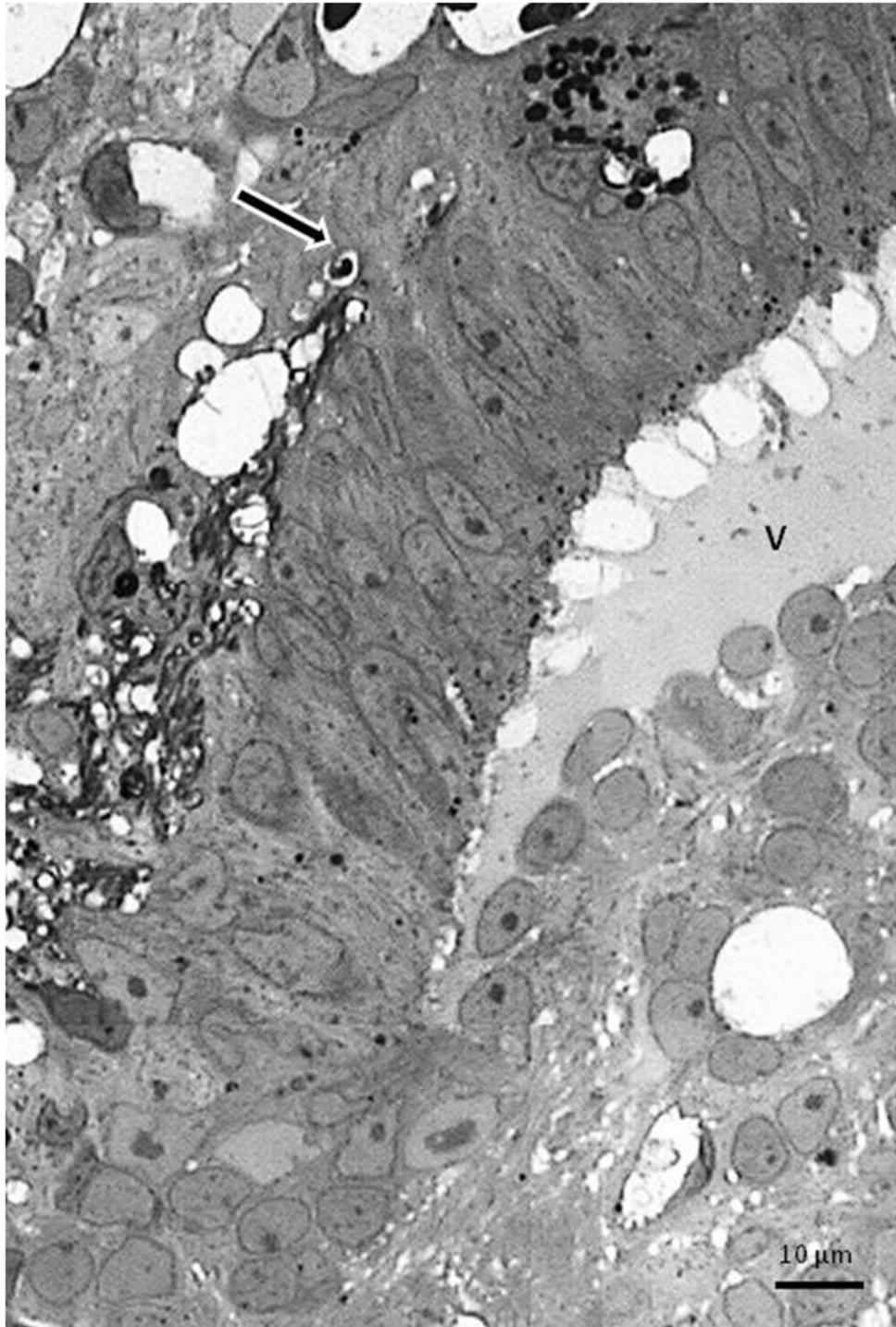


Figure 5 – An enlarged region of the embryoid body presented in figure 4. This figure highlights the similarity of this neural plate-like structure with embryonic neural tube neuroepithelial appearance with cells that are believed to be attached both at the ventricular and the pial surface and having peculiar, irregularly shaped nuclei (Sauer, 1935a; b). The atypical nuclei are radially oriented, constricted and scatter in several strata and give the impression of pseudostratified columnar epithelium. Many of the nuclei are bipartite and are lobulated. All of these features are typical of the neural tube. Among these cells, as is seen in the right lower corner of the image, large, granulated mast cells are present. At the pial-like surface (black and white arrow) of the neuroepithelium-like layer, formation of myelinated axonal bundles (cross- section) can be recognized. At the lower boundary of the ventricle-like space (V), a row of cuboidal cells resembling ependyma has been formed. Below it, a rosette-like arrangement of newly formed cells can be found around a white appearing cisternal space. At its left side, a capillary is formed with a clearly recognizable endothelial cell. Scale bars: - 500 μm

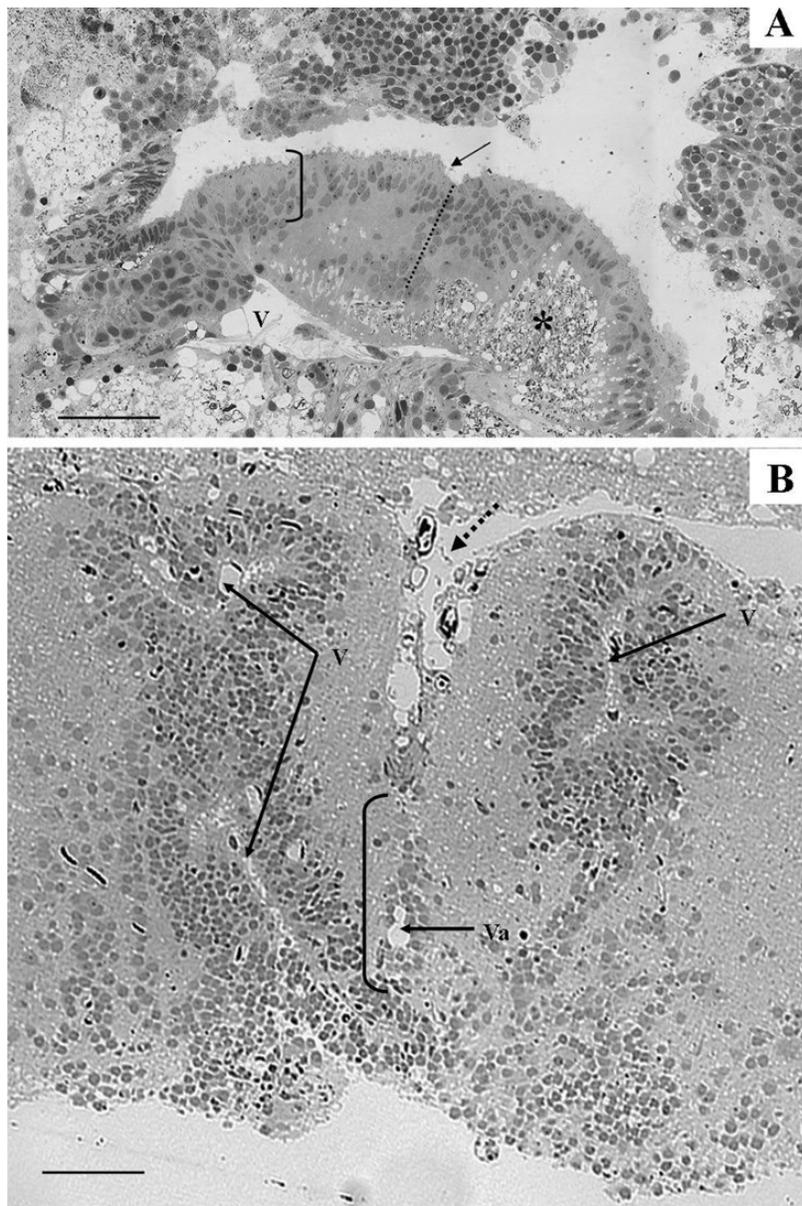


Figure 6 - Newly formed organized structures resembling embryonic neural plate and/or neural tube in a 7 & 14 days surviving samples. Plastic embedded semithin sections stained with toluidine blue Image A depicts a newly formed well organized structure resembling embryonic neural plate among the degenerative tissue. This structure was found in a 7 day surviving tissue sample montaged under the 40x objective. This neural plate-like structure appears to have bilateral symmetry with an indentation where the superior sagittal sulcus is to be formed (arrow) and a recognizable raphe-line. On the upper part of this structure several, newly formed layers resembling the mantle layer (bracket) can be seen. The plate is about to turn into a tube surrounding a distorted shaped space which may turn into the ventricular space (V) bordering a large piece of this structure is filled with degenerated, spongy looking tissue mass (asterisk). Image B depicts a more advanced neural epithelial structure found at 14 days survival time. It resembles a more reformed, two hemisphere-like formation of the early neural tube. The arrow at the top of the image points toward the superior sagittal sulcus filled with tissues appearing like forming pia mater. The well-developed two neural plate tubes surround recognizable lateral ventricular spaces (arrows with V) while underneath the raphe (bracket), between the two tubes, a space resembling the third ventricle (arrow Va) can be seen. Note that the two bilateral symmetrical tubes show a typical plexiform layer occupying the periphery of the tube and is dotted with some nuclei. Scale bar - 1 mm

resembling ependymal is being formed. Below it, a rosette-like arrangement of newly formed cells can be found around a white appearing cisternal space below which, a capillary is formed with a clearly recognizable endothelial cell.

Figure 5 is an enlarged print of the marked region in Figure 4 where the neural epithelial cell nuclei can be more clearly analyzed. This figure highlights the similarity of the nuclear forms of these cells with those of embryonic neural tube neuroepithelial cell nuclei¹⁸⁻²⁰ as they appear as pseudostratified, columnar epithelium and are believed to be attached both at the ventricular and the pial surface. In our sample as well, the cells are having peculiar, irregularly shaped nuclei, while the rows of atypical nuclei are similarly radially oriented and scattered in several strata. Many of the nuclei are constricted, bipartite or lobulated. At the pial-like surface of this structure (shown with a black and white arrow), sections of myelinated axonal bundles can be recognized. As seen in the right upper corner of the image, a large, granulated cell is also present.

In Figure 6, we demonstrate two, more advanced forms of these brain rudiments (A,B) found in samples at 7 and 14 days, respectively. Figure 6A shows a well-organized embryonic neural plate-like form among the degenerating tissue. It appears to be have bilateral symmetry with an indentation where the superior sagittal sulcus is to be formed (arrow) and a recognizable raphe-line. On the upper part of this structure, several, newly formed layers resembling the mantle layer (bracket) can also be seen. The plate is about to turn into a tube, surrounding a distorted shaped space which may turn into the ventricular space (V). A large piece of this structure is bordered with degenerated and spongy appearing tissue

masses (asterisk). Above and next to the featured structure (in the right upper corner of the figure), many newly formed cells are assembled into a tumor-like mass lacking any typical neural organization. Figure 6B represents a more advanced, neural tube-like level of organization of the rudiment, which resembles the early formation of the embryonic optic tectum viewed in a frontal plain²¹. The two hemispheres separated by a sagittal sulcus-like space (dotted arrow) can be easily recognized with the well-formed cellular layer made from neuroepithelial cells or already neuroblasts surrounding a potential lateral ventricular space (arrows labeled with V) while under the sagittal sulcus, the formation of the third ventricle (arrow Va) could be recognized surrounded by raphe cells (bracket). One can easily recognize the pial region with the formation of the pia mater itself and its blood vessels resembling superior sagittal sinus.

Discussion

This study identifies that the first events a few hours following the trauma of the adult zebrafish brain are inflammation followed by spongiform degeneration. In short, among the surviving cells, many of the original structural elements lose their typical morphology or disappear altogether and large numbers of vacuoles appear, giving a spongiform, degenerated appearance to the tissue. During the first two days of cultivation, the tissue follows its degenerative path paralleled with the appearance of round-shaped, simplistic cells, which start to accumulate at the periphery of the tissue chunk. At the same time, large, highly granulated cells begin to appear and increase in number throughout the sample. Some of these

cells show metachromasia with toluidine blue suggesting that they might be mast cells²². In some regions, the increase of these cells is paralleled with the appearance of healthy-looking differentiating new cells and formation of new capillaries, sometimes filled with cells resembling erythrocytes and leukocytes. Around 96 hours, organized spots resembling embryonic "pseudostratified" columnar neuroepithelium begin to appear in a close relationship with the newly formed blood vessels and the large, granulated and degranulated cells. While the size and cellular groupings of these restorative structures varies, most of them are well organized, and show similarities with the germinative layers of the developing cortical structures in vertebrates¹⁸⁻²⁰. It has been demonstrated previously that these neuroepithelial cells give rise to both neuronal and glial elements and are characterized as highly proliferative cells. The other interesting feature of these cells is the shape and arrangement of their nuclei as well as how they behave during their cell cycle, preceding mitosis. It was recognized as early as in 1935 that the radially oriented, frequently bipartite, oval nuclei proceed to oscillate (first move toward the external limiting membrane and then turn back and move to the periventricular zone) before they undergo mitosis^{19,20}. It was also shown in postnatal rat cerebellum that the radial neuroepithelium (Bergmann-glia) undergo morphologically asymmetric mitotic division when one of the newly formed daughter cell maintains the full length of its mother cell's radial fiber while the other obtains, undifferentiated, and rounded shape^{23,24}. The nuclear oscillation was later reinforced by several other laboratories and now is a well-accepted theory^{25,26}. Nuclear oscillation as a biological phenomenon was found in

other cell types maintained in cell culture when environmental conditions forced the cells to acquire an elongated shape with a long cytoplasmic process and is now in the center of renewed interest for research²⁷.

Based on the facts that radial glia is a typical organizer of embryonic neurodevelopment^{12,28,29}, and that it is being present in the adult zebrafish optic tectum^{30,31}, and also because the periventricular grey zone of the adult zebrafish optic tectum is a highly proliferative center containing neuronal stem cells and progenitor cells^{32,33}, we hypothesize that the first possible candidates for formation of such restorative rudimentary structures, as we see in our samples, are the radial glial components of the adult optic tectum. Anthony et al.²⁸ demonstrated that radial glia lead to the formation of all cells of cerebral cortex in mice using Brain Lipid Binding Protein as a marker. However, there is now also a bulk of experimental evidence indicating the possible involvement in adult brain regeneration of nearly all types of cells existing in the brain parenchyma. All other glial cell types, mast cells, endothelial cells, pericytes, fibroblasts, and smooth muscle cells all are involved in the inflammation of the brain tissue induced by the trauma³⁴⁻³⁶. Kyritsis report that injury induced inflammation in the zebrafish brain can activate, what they call, "executioner cells" of the immune system which work as switches to turn any other cells be involved in neurogenesis³⁷. Indeed, in our sample, observed signs of neovascularization, possible hematopoiesis and the formation of these rudiments are paralleled with an increasing number of large, highly granulated cells resembling mast cells, which may function as "executioner

cells³⁷. Mast cells are long known to become activated with injury³⁸, but new research provides evidence that their behavior on the molecular signaling level strongly depend on the severity of the injury and whether it is acute or chronic. With severe and chronic inflammation these cells have been shown to switch to neurogenesis³⁹⁻⁴². Our case represents a severe and long-lasting injury.

Conclusion

This study suggests that surviving organotypic culture of adult zebrafish optic tectum presents a complex interactive cellular microenvironment where epigenetic factors can change cellular behavior and activity. This approach may serve as a useful model system to study degenerative and regenerative processes induced by severe trauma in the adult brain, which is paralleled with cell survival and ontogenic events. In addition, the cultured optic tectum turned out to also be a model system to study intrinsic embryonic neuro-developmental processes in both an adult and degenerative brain tissue environments. The fact that well-structured brain rudiments do form as early as 96 hours, makes this model not only very useful to study the above mentioned embryonic neuro-developmental processes, but allows to focus on the very early inductive mechanisms. The fact that the number of surviving tissue pieces, cultivated at the same time, could be set by the investigator makes this model also appropriate to study, in relatively a short time, a larger sample size or a larger total tissue mass. This model can also help to investigate conditions that are different in the lower vertebrates' brain that still allows regeneration even into adulthood,

which cannot occur in higher vertebrates like mammals. This model also opens up several avenues to study the stem cells, mast cells, mechanism of neovascularization, for example, and of the use of other techniques (like immunohistochemistry) to further analyze tissue physiology and immunologically active, non-neuronal cells. It could become a good model for testing different neuroprotective agents, such as different growth factors, cytokines and hormones. Progesterone, for example, has become an extremely important hormone in brain restoration after TBI^{43,44}. Other avenues could be studying agents that block regeneration such as, for example, stress, temperature, and/or radiation. Additionally, the organotypic culture could be used to study neuronal glial relationships over time after TBI and how these interactions could affect plasticity and structural repair, among many other approaches. The use of zebrafish makes the entire process also relatively inexpensive.

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Clinical Reasoning Preparation: Gross Anatomy Case Studies in Occupational Therapy and Athletic Training

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Abstract

The purposes of this study was to determine if exposure to clinically-derived components in Gross Anatomy case studies were useful for preparation for clinical reasoning in Occupational Therapy and Athletic Training; if Occupational Therapists and Athletic Trainers continued to use paper case studies in their current professions, and what were the settings for this practice. A Qualtrics questionnaire, with a Likert scale to determine usefulness of case study components towards preparation for clinical reasoning and questions of case study use during practice, was distributed electronically to students that graduated AY 2005-2010. Descriptive statistics and correlations supported the usefulness of early exposure to concrete and cognitive elements towards preparation for clinical reasoning. It was found that the majority of respondents continue to use case studies of all types in a rehabilitative clinical setting, and that early introduction of all types of essential clinical reasoning components in Gross Anatomy will reinforce the clinical reasoning framework synthesized from other courses in the Occupational Therapy and Athletic Training curriculum, and in turn, will improve practice.

Introduction

What is clinical reasoning?

Every profession defines it differently based on the discipline, but in general, it is regarded as the decisions made regarding the prevention, diagnosis or treatment of a clinical problem associated with a particular patient. In Athletic Training (AT), clinical reasoning is described as a procedural process that encompasses knowledge, cognition and metacognition (thinking about thinking and or connecting knowledge to cognition)¹. Clinical reasoning within Occupational Therapy (OT) involves the search for the answers to three questions: What is the current state in occupational

therapy performance? What can be done to improve performance? What can be done to improve occupational competence²? Both of these views of clinical reasoning have a foundation within the scientific method of hypotheses testing, or Hypothetico-Deductive Reasoning (HDR): several hypotheses based on the clinician's knowledge/experience are proposed and then are either proved or disproved after conducting a series of evaluations/tests/experiments³. HDR is also a form of theoretical reasoning (applying theory to practice). Whether it is used to predict a specific case or in the broader context, theoretical knowledge of the anatomical sciences teaches that distinctive types of physical damage/diseases/conditions

results in unique deficits, signs, symptoms that can be applied universally to all patients even if we don't know the specifics of that patient's case⁴. The common denominator for all allied health professions is that clinical reasoning is linked with the initial evaluation, the clinical practice most aligned with a diagnosis⁴. Scientific reasoning utilizing HDR enhances the systematic process of both AT and OT practice and in turn "professionalizes" both of these allied health careers⁵. HDR is the initial form of clinical reasoning that students use in both fields of practice, but eventually other forms of clinical decision making are incorporated into practice as the Athletic Trainer or Occupational Therapist gains more clinical experience.

Practical Reasoning, as defined by Mattingly⁴, is a more advanced mode of clinical reasoning, differing from theoretical reasoning in that theoretical reasoning deals with what is possible and practical reasoning applies actual specific knowledge of the patient. Clinical reasoning is directed toward action, but determining how to act is more than theoretical knowledge, it involves critical thinking about the appropriate action for this unique case with this unique client/patient/athlete at this specific time and place. Other biological factors (current injury/ illness/condition, history of injury/ illness/condition), cognitive factors (motives, ethics, beliefs), and environmental factors (condition of rehab space, support system) influence how the injury, disease or condition becomes unique to the client/patient/athlete, therefore, a treatment plan requires more than anatomical/physiological knowledge⁴.

Narrative Reasoning, specific to OT practice, is rooted in the reality that Occupational Therapists must involve

their patients/clients actively in the therapeutic process, and therefore, a patient's perspective (motives, ethics, beliefs) towards the problem and the resulting therapy is the main clinical reasoning exercise⁴. How is that injury/disease/condition experienced by that particular client/patient? The Case Pattern Recognition (CPR) technique develops as experienced Athletic Trainers recognize the typical signs/symptoms, nature of injury, and physical presentation from past cases and does not rely solely on reasoning to produce diagnoses and plans of treatment⁶.

Practical reasoning, narrative reasoning and case pattern recognition are more advanced forms of clinical reasoning, reasoning skills that the novice Athletic Trainer and Occupational Therapist will eventually acquire with experience and time. HDR is the precursor clinical reasoning technique for allied health professions, and as reported by Heinerichs *et al.*⁷ can be learned through didactic courses that utilize inquiry-based instruction methods such as case studies, problem-based learning, peer coaching and role play.

Hypothetico-Deductive Reasoning

According to Geisler and Lazenby⁸, HDR is a better method for evaluating injury and illness during the beginning/intermediate curricular stages for three reasons. First, HDR reinforces the linear and organized relationship between the easily learnable concrete components of clinical evaluations such as signs and symptoms, history of illness/injury with a potential diagnosis. Secondly, HDR is an advanced form of clinical thinking that will improve upon practice as it requires more cognition than the early stages of Bloom's Taxonomy of Cognitive Development

(recognition, recall, comprehension), advancing into the higher stages of application, analysis and ultimately evaluation and synthesis. Thirdly, HDR allows the clinical instructor (CI) to monitor, question, and provide feedback and remediation, if necessary, to a student as he/she progresses through modes of clinical thinking. CI mentoring engages inexperienced clinicians through a process that improves their clinical skill sets. As a result, the HDR technique should be conducted as frequently as possible in all coursework. Students should expect and be comfortable with the fact that HDR will be their initial mode of clinical reasoning in the classroom and the clinic, and that other advanced forms of clinical reasoning will develop with more experience⁸.

In addition, results from the American Occupational Therapy Association/AOT Foundation Clinical Reasoning Study determined that the expected dysfunction-medical diagnosis relationship was the most important factor influencing how the Occupational Therapist assessed their patients, including the development of the treatment plan⁹. Knowledge of pathology or trauma leads to predictions of physiological and anatomical dysfunction, which then prescribes interventions that would improve dysfunction.

SNAPPS

A learner-centered HDR procedure (originating in medical schools) that links clinical reasoning (thinking) with a patient's examination results in a linear, timely manner goes by the mnemonic SNAPPS: **S**ummarize the history, signs and symptoms and nature of injury; **N**arrow the differential (diagnosis) from several possible diagnoses; **A**nalyze the differential; **P**robe the instructor for

uncertainties; **P**lan management of treatment and rehabilitation; and **S**elect an issue for self-directed learning^{10,11}. Within a study of SNAPPS on AT⁷, descriptors used to evaluate their case presentations (utilizing the SNAPPS technique) match the components of the case studies in my Human Gross Anatomy (HGA) and Basic Gross Anatomy (BGA) courses. The summary section of my gross anatomy case studies could include the chief concern, mechanism of injury, injury history, signs/symptoms of injury or illness, history of illnesses related to chief complaint, and abnormal physical presentation. These case studies also include details of and inquiries into a physical examination to aid participants in the process of narrowing the diagnosis differential: range of motion examinations, manual muscle/strength testing, dermatomal testing, cranial nerve examination and testing of extremity reflexes. Students then analyze the results of the examinations and summary information collected in the case study, and with this evidence, confirm why the diagnosis is included in the differential. Throughout the open discussion of these first three steps of the process, students are encouraged to probe the instructor/facilitator's knowledge via questions to help clarify any areas of confusion and raise concerns about testing, or the results of testing, that could be viewed as vague or ambiguous. As eloquently stated by Heinerichs *et al.*⁷, questions from the facilitator directed towards students during the initial steps of the SNAPPS process will help the instructor to determine student comprehension of these diagnostic components, provide a forum for students to express doubts about the narrowing of the differential diagnosis and allow students to vent uncertainties they may

feel about their own abilities. Because HGA and BGA are didactic courses in the OT and AT programs, the remaining steps of plan management and selecting a subject for self-directed learning are not emphasized as much as the previous steps.

The purpose of this clinical reasoning study is to collect and analyze quantitative data from a Qualtrics survey that inquired whether exposure to the SNAPPS components in my paper case studies was useful towards their preparation for clinical practice in their respective professions of OT and AT. The survey also inquired whether participants utilize case studies in their current profession and the setting for this practice. Responses will reflect what my previous students felt about this pedagogical method as a conveyor of precursor clinical reasoning knowledge. All data from this survey will ultimately be used for the assessment of case study practice in the gross anatomy classroom/laboratory as a tool for conveying elements of clinical reasoning in a didactic course.

Materials and Methods

A Qualtrics survey was electronically distributed in the summer of 2015 to students that had taken either my HGA or BGA course within the OT graduate program (HGA for graduate students) or the AT undergraduate program (BGA for undergraduates) that have graduated between AY 2005-2010. Case studies were presented identically by the instructor in both courses, regardless of the academic level. This particular time period was selected in order to survey students that have graduated from Kean and been employed in their current profession: Occupational Therapists (M.S. in Occupational Therapy) that have

worked in their field since graduation; and Athletic Trainers (B.S. in Athletic Training) either employed after undergraduate graduation or after earning a M.S. in AT and then employment. The content of the Qualtrics survey was created with the assistance and cooperation of the Assistant Director from the Office of Research and Sponsored Programs at Kean University (in charge of pedagogical research, Qualtrics program), Occupational Therapists in the OT program and Athletic Trainers in the AT program at Kean University. The Qualtrics survey, consent form, debriefing form, and research application was approved by the Kean Institutional Research Board committee (IRB00005689). Qualtrics software was used to process the data gathered on general information (about the participants) and questions related to the participant's current use of case studies in their profession. The software used for analysis of the Likert Scale quantitative data was IBM SPSS Statistics version 21. Descriptive statistics obtained via the SPSS software included mean, minimum value, maximum value and standard deviation. Correlations between components of the case studies were obtained via a bivariate Pearson Correlation two-tailed t-test, whereas $p < 0.05$ was significant.

The questions presented in the survey belong to two different categories: questions concerning general information about the participants and their current use of case studies in their professions; and questions related to use of specific components of paper case studies, presented in my gross anatomy courses, towards the preparation for actual cases in their clinical practice/profession (Fig. 1a,b). General questions included the year graduated, degree conferred, field of current clinical profession, use of case

studies within the clinical profession, and the setting the participants utilized case studies. Query of the usefulness of specific components of paper case studies towards their clinical practice/profession required participants to rank components of the case study using a Likert scale of 1-4: 1, "not useful"; 2, "somewhat useful"; 3, "useful"; and 4, "very useful." Survey participants ranked twelve components of case studies that were presented, discussed and analyzed in my gross anatomy courses: history of the events preceding illness/injury, physical symptoms/deficits, cognitive symptoms, physical presentation of the client/athlete as they enter the evaluation space, manual muscle testing including the results of range of motion and strength testing, dermatomal/sensory testing, gait analysis, specific testing including cranial nerve assessment and extremity reflexes, ability to diagnose or working with a diagnosis (specifically OT), designing a rehabilitation program or intervention, current mental status of the client/patient/athlete towards the prognosis, and preparation for the anomalous/atypical client/patient.

The risk for participation in this study was minimal as the surveys contained profession-related questions, without any questions of a personal nature, and because there were no unique identifiers (except geographical location) for participants, confidentiality was secured.

Results

Twenty-eight participants, 18 graduates that identified as obtaining a M.S. in OT and 9 former students that identified as graduating with a B.S. in AT, responded to the survey, providing all the general information requested and responses to queries that applied to their

current professions. The majority of participants identified themselves as graduating in 2010 and 2009, 48% and 24% respectively. Masters degrees of OT were conferred to 64% (18/28) of the survey participants and 32 % (9/28) of the responders identified as AT graduates. However, when a comparison is drawn between the number of survey takers versus the total number of individuals that received surveys in each group, the AT group had a 33 % participation rate (9/27) and the OT population had a 16% participation rate (19/117). The same number of participants that identified as having graduated with an OT degree also reported that OT was their current profession (64%, 18/28), whereas, only 5 of the 9 AT degree holders were currently practicing Athletic Trainers (18%, 5/28). Of the three remaining AT degree holders, one is a Physical Therapist (PT), one is a Doctor of Osteopathic Medicine (D.O.) and the other is a Physician Assistant (PA). Only 2 of the participants reported that they were not currently in a clinically-related profession.

When asked if they used case studies in their current profession, 61% (17/28) of the Kean alumni reported that they use paper and non-paper types of case studies. Of the participants that use paper case studies, 71% (12/17) were practicing Occupational Therapists and 29 % (5/17) were practicing Athletic Trainers. The majority of those professionals, 59% (10/17), use paper case studies in a rehabilitative clinical environment, whereas, 24% (4/17) practice with case studies in a non-clinical rehabilitative setting, and 17 % (3/17) present case studies in the classroom.

Participants were asked to rank 12 components included in case studies for usefulness as it pertains to preparation for clinical reasoning, and 11/12 components

Figure 1a. Survey: Clinical reasoning assessment: case study practice in the gross anatomy classroom/laboratory

In what year did you graduate from Kean University?

2005 2006 2007 2008 2009 2010

What degree did you graduate with from Kean University?

B.S. in Athletic Training B.A. in Psychology/ M.S. in Occupational Therapy

BA. in Biology/ M.S. in Occupational Therapy

BA. in Therapeutic Recreation (Rec Admin)/ M.S. in Occupational Therapy

B.A. (other)/ M.S. in Occupational Therapy M.S. in Occupational Therapy

B.A. or B.S. (other)

What is the field of your current clinical profession?

athletic training occupational therapy physical therapy

medicinal (MD/DO, physician assistant, nursing) non-clinical/ other profession

Do you use any form of paper clinical case studies in your current profession?

yes No

If so, in what setting?

instruction in a rehabilitation clinical setting instruction in a non- rehabilitation clinical setting

instruction in a classroom/school setting instruction in other setting

Do you make reference to clinical case studies, non-paper format (i.e. video), in your current profession?

yes No

If so, in what setting?

in a rehabilitation clinical setting in a non- rehabilitation clinical setting

on the athletic field in a classroom setting in other setting

acquired favorable means between 3 (“useful”) and 4 (“very useful”) (Table 1). Participants ranked the early introduction to observing physical symptoms/deficits as the most useful component (mean 3.79, SD .415), while the process of working with a diagnosis and manual muscle testing were both classified as the next most useful component (mean 3.68, SD 0.557).

The statistical data revealed several strongly significant correlations ($p < 0.001$) and numerous significant correlations ($p < 0.05$) between components in the gross anatomy case studies (Table 2). The five strongest correlations ($p < 0.001$) included connections between initial symptoms, testing, diagnosis, and incorporating mental status into the prognosis: 1) history

of the injury/illness/condition and gait analysis; 2) physical signs/symptoms, sensory testing and the ability to either hypothesize a diagnosis or work with a diagnosis; 3) initial cognitive symptoms of the client/patient/athlete and its subsequent influence on the mental status of the individual during the prognosis (i.e. treatment, rehabilitation); 4) results of manual muscle and sensory testing and the design of the rehabilitation plan; and 5) the inclusion of the mental status of the individual as a factor when designing the rehabilitation plan. With regards to approaching an anomalous presentation, there were 9 correlations between anomalous situations and the remaining 11 elements of case studies, statistically

Figure 1b. This question is for professions that involve clinical treatment only. Please rate how useful was the introduction of each component of the paper case study towards preparing you for actual clinical cases (clinical reasoning), using the Likert Scale: "very useful (4)," "useful (3)," "somewhat useful (2)," and "not useful (1)."

	Very Useful (4)	Useful (3)	Somewhat useful (2)	Not useful (1)
history (events before injury, onset of condition)				
physical symptoms/deficits				
cognitive symptoms/mental dysfunction				
physical presentation				
manual muscle testing (range of motion, strength)				
sensory/dermatome testing (modalities, receptive field)				
gait analysis				
specific diagnostic testing (i.e. cranial nerve assessment, reflexes)				
diagnosis or working with a diagnosis				
designing a rehabilitation program				
incorporating the mental status into the prognosis				
anomalous (atypical presentation, symptoms, diagnosis etc.)				

proving that exposure to the building blocks of clinical reasoning were useful when confronted with the atypical case in their profession.

Discussion

Twenty-eight participants, 18 graduates that identified as obtaining a M.A. in OT (all practicing Occupational Therapists) and 9 former students that identified as graduating with a B.S. in AT, responded to the survey, and as reported,

the majority of participants identified themselves as graduating in 2010 and 2009. This was expected because the surveys were distributed through Alumni Relations at Kean (to preserve confidentiality) and because the majority of the e-mail addresses on file were from the Kean server, the recently graduated students were more apt to check their alumni e-mail.

The majority of the survey respondents continue to use case studies in their current professions, and the same

Table 1: Ranking of components of paper case studies towards preparation for clinical reasoning

Component of case study	Very Useful (4)	Useful (3)	Somewhat useful (2)	Not useful (1)	Total	Mean ± sd
history (events before injury, onset of condition)	16	8	0	1	25	3.56 ± 0.712
physical symptoms/deficits	20	5	0	0	25	3.79 ± 0.415
cognitive symptoms/mental dysfunction	12	6	6	1	25	3.16 ± 0.943
physical presentation	17	5	3	0	25	3.56 ± 0.712
manual muscle testing (range of motion, strength)	18	6	1	0	25	3.68 ± 0.557
sensory/dermatome testing (modalities, receptive field)	16	7	1	0	24	3.62 ± 0.576
gait analysis	9	7	8	1	25	2.96 ± 0.935
specific diagnostic testing (i.e. cranial nerve assessment, reflexes)	11	11	3	0	25	3.32 ± 0.690
diagnosis or working with a diagnosis	18	6	1	0	25	3.68 ± 0.557
designing a rehabilitation program	14	10	1	0	25	3.52 ± 0.586
incorporating the mental status into the prognosis	10	8	7	0	25	3.12 ± 0.833
anomalous (atypical presentation, symptoms, diagnosis etc.)	11	9	5	0	25	3.24 ± 0.779

number of participants that indicated that they used paper case studies in their current professions also used non-paper types (i.e. video presentation). This result indicated that these professionals felt that practicing this inquiry-based pedagogical method was a valuable tool for teaching clinical concepts, which in turn, strengthens the development of clinical reasoning skills. Utilizing case studies for the purpose of acquiring clinical reasoning ability reinforces research that supports the use of case studies as an effective way to learn HDR, the precursor clinical reasoning technique⁷.

Correlations

History and gait analysis

A correlation between history and gait analysis ($p < 0.001$) points to an association between previous injuries/illnesses/conditions

affecting the musculature, peripheral nerves and skeletal system of the lower extremity (LE), and the resulting abnormal presentation during locomotion: muscle damage in the LE resulting from injury or pathology affects contraction of muscles in the hip, knee, ankle and digital joints limiting range of motion (ROM) and strength; neuromuscular junctions of LE musculature affected by nerve disorders present as either paralysis (complete loss of muscle function) or paresis (partial paralysis); and damage to bones, cartilage, and ligaments impacts ROM in lower extremity joint movement.

In gross anatomy case studies, the history of the protagonist could include injuries, illnesses or conditions related to the chief complaint, while gait analysis would fall under the physical signs/symptoms for the LE case study. According to the SNAPPS model for clinical reasoning in medically-related fields of study, summarizing the history, signs and

	H	PS	CS	P	MMT	ST	G	SDT	D	R	MP	A
History (H)			.015*				**	.004*				
Physical symptoms (PS)			.008*	.005*					.001**			
Cognitive symptoms (CS)	.015*	.008*		.005*	.037*	.019*	.015*	.012*		.025*	**	**
Presentation (P)		.005*	.005*		.017*		.017*			.003*	.026*	.003*
Manual muscle testing (MMT)			.037*	.017*		**			.020*	**	.006*	.003*
Sensory testing (ST)			.019*		**				**	.001**	.002*	.004*
Gait analysis (G)	**		.015*	.017*		**		.006*		.037*	.013*	.002*
Specific diagnostic testing (SDT)	.004*		.012*				.006*				.009*	.002*
Diagnosis, working with diagnosis (D)		.001*			.020*	**				.006*		
Rehab Plan design (R)			.025*	.003*	**	.001**	.037*		.006*		**	.026*
Mental status in prognosis (MP)		.027*	**	.026*	.006*	.002*	.013*	.009*		**		**
Anaomalous presentation (A)		.004*	**	.003*	.003*	.004*	.002*	.01*		.026*	**	
*p < 0.05, ** p < 0.001												

symptoms and nature of injury is the first step in the process^{10,11}. The strong connections between summary components observed in this study reinforces and complies with the first step of the SNAPPS model of clinical reasoning.

Physical symptoms, manual muscle testing, sensory testing and diagnosis/working with a diagnosis

As described by Mattingly⁴, combining the physical signs/symptoms of the client/patient/athlete and the results of the subsequent testing is essential for the initial evaluation, the clinical process that is closest to the expected dysfunction- medical diagnosis relationship. Correlations between the diagnosis and physical signs/symptoms, manual muscle testing, and sensory testing (p<0.001) reveal a logical clinical reasoning relationship: possible diagnoses hypothesized from initial physical signs and symptoms are narrowed down to the

differential diagnosis through further motor and sensory testing, which will either support or disprove the differential. These case studies included or discussed (via case study questions) details of tests used in physical examinations, derived in part by physical signs/symptoms, to aid participants in the process of narrowing the diagnosis differential: range of motion examinations, manual muscle/strength testing, dermatomal testing, cranial nerve examination and testing of extremity reflexes. Narrowing the differential (diagnosis) from several possible diagnoses is the second step in the SNAPPS model^{10,11}.

Manual muscle testing, sensory testing, and designing a rehab plan

Being a didactic course, the technical requirements for developing a rehabilitation plan for the protagonists was never discussed in the case studies, as this was beyond the scope of

the expertise. However, since students would be involved in designing rehab/treatment plans in their respective professions, the testing procedures cogent to the comprehension of concepts in gross anatomy to demonstrate the expected dysfunction-medical diagnosis relationship, establishes a relationship which aids development of the treatment plan⁹. For example, when a protagonist in a case study has damage to a specific nerve/plexus, it was discussed how the results of using manual muscle testing techniques like ROM and strength testing on the paralyzed/weakened muscles would be incorporated into a rehab plan to increase ROM and strength to normal levels.

As mentioned earlier, narrowing the differential (diagnosis) from several possible diagnoses via further testing complies with the second step of the SNAPPS model^{10,11}, but it also prescribes a preliminary treatment plan to restore function in AT/OT. Although treatment plans were not discussed in detail, the brief discourse regarding further testing alludes to rehab plan/therapeutic treatment management, the fifth step of the SNAPPS model of clinical reasoning^{10,11}.

Cognitive symptoms and incorporating mental status into the prognosis

The present cognitive state of the client/patient/athlete was viewed as being significantly linked ($p < 0.001$) to the future mental state and its influence on the prognosis of the individual. Within the scenarios of the gross anatomy case studies presented to these AT/OT students, the protagonists often injured themselves as a result of poor decision making when they misjudged the extent of their abilities or the environment in which

they carried out their activity. Cognitive factors such as the motives and beliefs of the protagonist and the environmental factors surrounding the injury/illness make the case unique and must be considered when determining the prognosis of the protagonist. Consideration of these elements when making decisions about the treatment describes an advanced type of clinical reasoning known as practical reasoning⁴. After the differential diagnosis was determined, a subsequent prognosis was proposed and the basics of the rehab plan were discussed, students were asked this simple question: "What kind of patient do you think this person will be during their treatment?" The question was elaborated upon by citing the mental state of the protagonist that led up to the trauma/illness and then participants were probed as to the influence that cognitive state would have during the protagonist's rehab program/treatment plan. For example, how will the motives and beliefs of the competitive athlete affect their ability to continue therapy or follow instructions in therapy? Negative outcomes could include decreased participation in therapy (belief system that leads to depression, anger, frustration) or the opposite, increasing their therapeutic exercises/activities beyond what is prescribed to regain lost abilities faster (motives), risking further injury.

With regards to clinical reasoning, citing and discussing how the cognitive state of the protagonist influences the level of cooperation during the evaluation and the subsequent prognosis is an important part of the post-diagnostic analysis. Narrative reasoning requires the professional to consider the beliefs, motives and ethics of the client when determining the prognosis and creating the treatment plan⁴. In order to ensure the best possible prognosis, the clinician

needs to have an idea of how cooperative the client/patient/athlete will be during treatment/rehab. At the same time, discussions between students and instructor pertaining to the protagonist's mental state allows the student to propose possible treatment outcomes to the instructor and then receive feedback about the theoretical case. This procedure gives the student the opportunity to probe the mind of the instructor for details of possible prognoses that they could encounter in their profession. This process of utilizing the protagonist's mental state to influence the prognosis, probing the instructor about possible outcomes based on the protagonist's mental state, and managing the treatment plan complies with the third, fourth and fifth steps of the SNAPPS model of clinical reasoning^{10,11}.

Designing a rehab plan and incorporating mental status into the prognosis

After the case study questions concerning the initial evaluation were thoroughly examined and explained, the management of treatment/rehab plans was theoretically discussed. Within this theoretical treatment plan, consideration of the influence of the protagonist's current mental status, identification of emotional factors (motives, beliefs) that could allude to the type of patient the protagonist would be during the ensuing therapy is vital⁴. Adapting the protagonist's present mental state into the prognosis must be considered as the success of the therapy depends on the cooperation of the client/patient/athlete to participate in the prescribed therapeutic process external to the clinical visits. If an Occupational Therapist or Athletic Trainer believes that the individual they are treating is not going to cooperate within the parameters set

forth for the treatment, then the professional has to take this into consideration when designing the rehabilitation/therapeutic plan. This correlation would also coincide with the plan management section of SNAPPS^{10,11}.

Anomalous presentation, cognitive symptoms and incorporating mental status into the prognosis

Correlations were found between the anomalous presentation piece and the cognitive elements ($p < 0.001$). During the open discussion of gross anatomy case studies, an atypical finding would often be substituted into the scenario, such as an abnormal or missing symptom, presentation or testing result and then students were asked "What would you do when you discover this unusual finding during your evaluation?" Initially, students are very uncomfortable discussing anomalies as they were saddled with the demanding tasks of learning the typical signs/symptoms, presentation and testing results associated with a particular injury/illness/condition. It was then explained to them that there are many variations in our internal anatomy (peripheral circulation, for example) and the possibility of coming across an anomalous case is very likely. That they will be responsible for the treatment of that patient/client/athlete as much as a typical case, using what they know from the typical cases to hypothesize the possible causes for the atypical case. Statistical analysis points to a strong connection between the present and future mental state and the atypical case. Indeed, the cognitive state of the client/patient/athlete can make the case unique, but it is surprising that the physical qualities were not more highly correlated, as they are typically the signs/symptoms of an evaluation that describes the anomalous

case. Because there were significant correlations between the anomalous case and eleven of the other case study components, professionals clearly felt that this early exposure to theoretical anomalous conditions has helped when treating the actual anomalous case.

Conclusions

The majority of the survey respondents continue to use paper case studies in their current profession. This result indicates that these clinicians use this inquiry-based, pedagogical method as a tool for teaching/reinforcing clinical concepts, which in turn, strengthens the reason to use case studies as a method of learning clinical knowledge, leading to clinical reasoning ability⁸. Utilizing case studies for the purpose of acquiring clinical reasoning ability reinforces prior research that supports the use of case studies as an effective way to learn HDR, the precursor clinical reasoning technique⁷.

Participants ranked the more demonstrative physical components of case studies as the most useful in their current professions: physical signs/symptoms, presentation and specific diagnostic testing such as cranial nerve examination and reflex testing. Summarizing physical deficits, the resulting physical presentation of those deficits and the results from further testing are important steps within the initial evaluation and subsequent diagnosis, complying with the SNAPPS regimen for developing clinical reasoning skills^{10,11}. With regards to the significantly stronger correlation between diagnosis/working with a diagnosis and designing a rehabilitation plan uncovered in this study, the connection between case study elements that narrow the diagnosis

differential and the development of the treatment plan, the second and fourth steps of the SNAPPS model^{10,11}, strengthens the link between practicing with the didactic based case studies and the development of clinical reasoning skills.

A future direction would be to continue to survey students in this manner, emphasizing even more the connections between components of clinical case studies and the steps of clinical reasoning. As theoretical clinical reasoning knowledge in the classroom is refined and strengthened, hopefully even more possibilities concerning clinical reasoning could be unlocked and shared with the students, and the ensuing effect from the increased exposure observed. Although preparatory clinical reasoning ability utilizing case studies was demonstrated and discussed with students for years, formal introduction of the concepts of HDR and Case Pattern Recognition to the students was done during the fall 2015 semester.

The results from this study, including the data/trends cogent to the HDR and Case Pattern Recognition styles of clinical reasoning, will be shared with the students and how the introduction of this information manifests in future professional behavior will be observed. Hopefully, this early exposure to clinical reasoning techniques will increase clinical competence within the OT and AT population that I have had the pleasure to teach in my Gross Anatomy courses.

Acknowledgments

I would like to thank the OT and AT students at Kean University that graduated from AY 2005-2010, the Assistant Director from the Office of

Research and Sponsored Programs at Kean University, and Occupational Therapists and Athletic Trainers at Kean University.

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MACUB 2016 Conference

Poster Presentation Award Winners

COMMUNITY COLLEGE

Biochemistry, Biophysics and Biotechnology

First Place

Novel Microwave Prepared Single-Walled Carbon Nanotubes-Ether Ionic Liquids Mixtures as Electrolytes for Dye-Sensitized Solar Cell Applications
Rawlric Sumner, Sharon Lall-Ramnarine and Tirandai Hemraj-Benny
Queensborough Community College, Bayside, N.Y

Second Place

Synthesis of Ruthenium Nanoparticle-Single-Walled Carbon Nanotube Composites for the Degradation of Congo Red Dye
Nicholas Carrero Rawlric Sumner and Tirandai Hemraj-Benny
Queensborough Community College, Bayside, NY

Third Place (tie)

Effect of Single Walled Carbon Nanotubes on Breast Cancer Cell Migration. Belkis Rodriguez, Jonathan Edouard, Mathiu Perez, Sunil Dehipawala, Tirandai Hemraj-Benny and Regina Sullivan, Queensborough Community College, Bayside, NY

Analysis of ETS2 Degradation through Exposure of Cyclin Dependent Kinase CDK10 Variants
Destinee B. Semidey, Valentina Schmidt and Luis Martinez

Developmental Biology and Genetics

First Place

Using CO1 vs. 16S rRNA DNA Barcoding for Taxon Clustering and Phylogenetic Resolution in Invertebrates
Harpreet Singh, Thomas Smith, Julian Stark and Nidhi Gadura
Queensborough Community College, Bayside, NY

Second Place (tie)

Common Periwinkles (*Littorina littorea*) from Woods Hole, Massachusetts, Plum Beach in Jamaica Bay, New York, and Fort Wadsworth in the Lower Bay of New York are Not Separate Populations
Mohamed Eid, Gary Sarinsky and Craig S. Hinkley
Kingsborough Community College, Brooklyn, NY

The Role of *Drosophila* SOCS36E in Linker Histone H1-mediated Heterochromatin Formation and Tumor Suppression

Ki Sum Fan, Andrea Mejia, Meghan Pfau, Nathan Doran, Amber Crockett¹, Arthur I. Skoultchi² and Na Xu¹
¹LaGuardia Community College, Long Island City, NY, and ²Albert Einstein School of Medicine, Bronx, NY

Environmental Biology and Ecology

First Place

**Pesticides and Alzheimer's Disease; High-mobility Group Box1 Protein(HMGB1)Pathway
and Microglis Dysfunction**

**Erick Subillaga¹, Maria Entezari² and Mohammad Javdan¹
¹Queensborough Community College, Bayside, NY
and ²LaGuardia Community College, Long Island City, NY**

Second Place (tie)

Identification of Plankton Species in the Hempstead Harbor Region Using DNA Barcoding

**Oscar J. Zagalo and Nidhi Gadura
Queensborough Community College, Bayside, NY**

Third Place

Isolation and Identification of Antibiotic-Resistant Bacteria From New York City Soil Samples

**Adolfo Coyotl and Joan Petersen
Queensborough Community College, Bayside, NY**

The Role of Social Learning on Ethanol Preference and Courtship Behavior in *Drosophila melanogaster*

**Erica Taveras, Matthew Romero, Arifa Altaf and Preethi Radhakrishnan
LaGuardia Community College, Long Island City, NY**

Microbiology and Immunology

First Place

Effects of Alcohol-Based Hand Sanitizer on the Diversity of Skin Microflora

**Johnessa S. Clarke, Elizabeth A. Mulligan and Grace Axler-Diperte
Kingsborough Community College, Brooklyn, NY**

Second Place

**Comparison of the Immunomodulation of Macrophages by Lipopolysaccharides (LPS) From Three
Different *E. coli* Serotypes. Ayinuer Aierken**

**A. Lucia Fuentes and Maria Entezari
LaGuardia Community College, Long Island City, NY**

Third Place

Antimicrobial Characteristics of Streptomyces

**Catherine Immordino, Brenda Torres and Mangala Tawde
Queensborough Community College, Bayside, NY**

Physiology, Neuroscience and Clinical

First Place

HMGB1 Inhibits Phagocytosis and Migration in LPS-stimulated Microglial Cell
Jordy Sepulveda, Maria Entezari and Lucia Fuentes
LaGuardia Community College, Long Island City, NY

Second Place

Comparison of N-methyl-d-aspartate NR2B Subunit Proportions Between Control, Gonadectomized, and Hormone Replaced Male Rats
Catherine Hernandez¹ and Mary F. Kritzer²
¹Nassau Community College and ²Stony Brook University, NY

Third Place (tie)

Memory Impairments During Cognitive Tasks in a Streptozotocin Induced Rat Model of Sporadic Alzheimer's disease
Raquel Romaine¹, Rashida R. Farokhi¹, Farrah Hussain¹ and Francisco Villegas²
¹Queensborough Community College, Bayside, NY and ²York College, Jamaica, NY

Neuronal Cell Death and Fluoro-Jade C Staining in Rat Model of Alzheimer Disease
Rashida R. Farokhi¹, Raquel Romaine¹, Sameh Hanna² and Francisco Villegas²
¹Queensborough Community College, Bayside, NY and ²York College, Jamaica, NY

MACUB 2016 Conference

Poster Presentation Award Winners

SENIOR COLLEGE

Biochemistry, Biophysics and Biotechnology

First Place (tie)

HDAC9 Linked to Lysine Acetylation Levels: a Potential Mechanism in Diabetic Nephropathy
Simran Alexandria Polce and Maria Alica Carrillo Sepulveda
NYIT-COM Old Westbury NY

Use of a Hollow Fiber Bioreactor for Analysis of Antiviral Compounds on the Cryptosporidium virus
Gabrel Samantha, Mary Morada, Mark Jenkins and Nigel Yarlett
Pace University, New York, NY

Second Place

Preferential Killing of v-Ras-transformed NIH 3T3 Cells by Inhibiting
Activated Cdc42-Dependent Kinase (ACK)
Ruth Opoku, Nadia Burgos, Ijaz Ahmed, Raj Rajnarayanan and Alam Nur-E-Kamal
Medgar Evers College, Brooklyn, NY

Third Place (tie)

Promoter Hypermethylation of SOX9 in Ovarian Cancer Correlates with Platinum-based Chemoresistance
Ryan Frank, Kim Doyle, Maria Samartino and Noelle Cutter
Molloy College, Rockville Centre, NY

Synthesis of Mini-Reporter Construct to Test Gene Transfer of RNA Therapeutics
Kerianne Fuoco, Koushik Muralidharan, Arbaz Khan, Hemangi Patel and Martin J. Hicks
Monmouth University, West Long Branch, NJ

Developmental Biology and Genetics

First Place (tie)

The Effects of Bisphenol A and Bisphenol F on the Reproductive System Using *Caenorhabditis Elegans*
Suyapa Penalva-Lopez, Merna Sawaged, Katherine Leonor
Saint Peter's University Jersey City, NJ

Juvenile Hormone Sensitivity of an Enhancer Region on the Foraging Gene. Marium Sarder and Rebecca Spokony, Baruch College, New York, NY.

Migration of Pc3 Cells is Regulated by Hexim 1
Awa Sow and Manya Mascareno
SUNY Old Westbury, Old Westbury, NY

Unraveling the CHD5 Interactome to Identify Anticancer Drug Targets. Ramish Nasir and Shilpi Paul, SUNY Old Westbury, Old Westbury, NY.

Production of Melanotic Tumors in *Drosophila* as a Result of Juvenile Hormone Mimic Treatment. Raquel Calero and Rebecca Spokony, Lehman College, Bronx, NY

Second Place (tie)

Impact of Combined Sewer Overflows on Water Quality and Aquatic Biodiversity at Selected Lower Hudson Raritan Estuary Sites. Carol Ellameh, Merna Bishai, Erick Gonzalez and Meriem Bendaoud, New Jersey City University, Jersey City, NJ

Low-level Copper Exposure Causes Cardiac and Neurodevelopmental Defects in the Embryonic Zebrafish.
K. Figueroa, J. Pagnotta, I. Mazaharul and A.L. Dell, St. Francis College, Brooklyn, NY

Environmental Biology and Ecology

First Place

New Perspective in the Treatment of Lung Cancer
David Donatucci, Michael Galarraga, Alix Duarte, Christina Faltas, Jan Osea and Natalia Coleman
New Jersey City University, Jersey City, NJ

Microbiology and Immunology

First Place

Direct PCR Detection, Cloning and Characterization of Mold Populations from Soils and Compost
Joy Bochis, Matthew Gardner, Theranda Jashari, Victoria Ellman, Jenifer Vasquez, Stephanie Zapata,
Victorya Ramos, Tina Choe, and Luis Jimenez
Bergen Community College, Paramus, NJ

Second Place (tie)

Bacteria for Inhibition of Biofilm Formation of Pathogenic Bacterial
Erick Gonzalez, Carol Ellameh and Meriem Bendaoud
New Jersey City University, Jersey City, NJ

Evaluation of Cytokines in Autoimmune Retinopathy
Pamela D. Marte Perez^{1,4}, Kelly Baloga², Doreen N. Nguyen², Christopher D. Heaney¹, Sapna Gangaputra³,
Nida Sen³, John Hooks³ and Barbara Detrick¹
¹The Johns Hopkins University, ²The Johns Hopkins Hospital, ³National Eye Institute, NIH and
⁴Caldwell University, Caldwell, NJ

**One Plus One is Greater than Two: Optimizing Synergistic Antibacterial Effect
Between Tea Polyphenols and Two Antibiotics**
Jonathan Valsechi-Diaz and Tinchun Chu
Seton Hall University, South Orange, NJ

**Anterior Cerebral Artery Stroke: A Case Study Created to Understand the Clinical Dysfunctions Related to
the Ischemic Brain Regions**
D. Colgan, W. Mirza, L. Lorentzen and K. Reilly
Kean University, Union NJ

Direct PCR Detection, Cloning, and Characterization of Bacterial RubisCO Genes from New Jersey Soils
Stephanie Zapata, Anna Gonzalez, Margarita Kulko, Ryan Kim, Theranda Jashari,
Aidan Holwerda, Tina Choe and Luis Jimenez
Bergen Community College, Paramus, NJ

Third Place (tie)

Nesprin-1 Regulation of Organelle Positioning During Endothelial Cell Migration
Nicole Williams, Zayna King and Monn Monn Myat
Medgar Evers College- CUNY, Brooklyn, NY

**Trans-splicing of Interleukin-13 Receptor Alpha Variant 2 into the EGFR Transcript to Block Growth and
Reactivate Immunogenic Potential of GBM Cells**
Sarah C. Falotico and Martin J. Hicks
Monmouth University, West Long Branch, NJ

**CD105 Deficiency in Mouse Aorta-derived Mesenchymal Stem Cells Promotes An Enhanced Inflammatory
Response to Lipopolysaccharide.** Joseph Granata, Hugo Sanchez, Philip Loeschinger, Anthony Goetz and
Jodi F. Evans, Molloy College, Rockville Centre, Rockville Centre, NY

Physiology, Neuroscience and Clinical

First Place (tie)

Comparative Study of the Response to Light on the Sensory Motor Integration of Gill Lateral Cell Cilia in Bivalve Molluscs
Danellie Semple¹, Johanne Jean-Pierre², Edward J. Catapane¹ and Margaret A. Carroll¹
¹Medgar Evers College and ²Kingsborough Community College, Brooklyn, NY

Thyroid Hormone Induces Vascular Relaxation Through VASP Phosphorylation at Serine239: A Potential Therapeutic Approach to Treat Diabetic Vascular Dysfunction
Sherin Samuel¹ Martin Gerdes² and Maria Alicia Carrillo-Sepulveda²
¹New York Institute of Technology and ²NYIT College of Osteopathic Medicine, Old Westbury, NY

Second Place (tie)

The Neurotoxic Effect of Manganese on Dopamine Post-Synaptic Receptors Are Reversed by Taurine
Loren Dubose, Kurt Loney-Walsh, Margaret A. Carroll and Edward J. Catapane
Medgar Evers College, Brooklyn, NY

Low Level Lead Exposure Differentially Alters Locomotor Activity and Anxiety Dependent Upon Developmental Time Period of Exposure: Implications for Neurocognitive Disorders
Samantha Rubi¹, Nimra Hameed¹, Sukhpreet Kaur², Neena Matthew², Teddy Dacius¹,
Tokunbo Jose¹ and Lorenz S. Neuwirth^{1,2}
¹SUNY Old Westbury, Old Westbury NY and ²SUNY Neuroscience Research Instituted, Old Westbury NY.

PTEN: A Potential Neuroprotective Target in Astrocytes
Dailyn Despradel, Karol Toral and Renée E. Haskew-Layton
Mercy College, Dobbs Ferry, NY

Third Place (tie)

The Structure of the Recurrent Laryngeal Nerve in the Rat
Oluwatobi Olufeko^{1,2}, Inna Rozenberg², Joshua B. Silverman², Krishnamurthi Sundaram²,
Mark Stewart² and Richard Kollmar²
¹CUNY Medgar Evers College and
²SUNY Downstate Medical Center, Brooklyn, NY

**The Stress of Public Speaking Increases Cortisol Levels in Undergraduates:
Is increased Preparation Really the Best Remedy?**
Erin Clinton, Grace Cookson, Stephanie Brown, Daniel Woods and Jodi F. Evans
Molloy College, Rockville Centre, NY

MACUB 2015 Conference

Poster Presentation Award Winners

MASTERS/DOCTORIAL

Developmental Biology and Genetics

First Place (tie)

Does Histone H3 Acetylation Stimulate NuA4 - Nucleosome Interaction?
Tiffany E. Rose and Daniel S. Ginsburg
LIU Post, Brookville, NY

Set3C Stimulates NuA4 Interaction with Nucleosomes and Occupancy Transcribed Genes
Angelo Messina and Daniel S. Ginsburg
LIU Post, Greenvale, NY

Second Place (tie)

An Interconnection Between Rapamycin Resistance Phenotype and Stress Granule Markers in Yeast
Thamer Alanazi, Praveena Dhulipala and Marci J Swede
LIU, Post, Brookville, NY

The Role of DAck in Appetitive Associative Olfactory Learning in Drosophila Larvae
Morgan K. King
Long Island University CW Post, Brookville, NY.

Biochemistry, Biophysics and Biotechnology

Characterizing the Endometrial Microbiome by Analyzing the Ultra-low Bacteria from Embryo Transfer Catheter Tips in IVF Cycles: Next Generation Sequencing (NGS) Analysis of the 16S Ribosomal Gene
Xin Tao^{1,2}, Nathan R Treff² and Tinchun Chu¹
¹Seton Hall University, South Orange, NJ and
²Reproductive Medicine Associates of New Jersey, Basking Ridge, NJ

Microbiology and Immunology

First Place

**Chromatin Condensation Protects DNA from MMS
Sherwin Shaju and Daniel S. Ginsburg
LIU Post, Greenvale, NY**

Second Place (tie)

**Antibacterial Activity and Biofilm Formation Inhibition of tea polyphenols
Gabiella Appice and Tinchun Chu
Seton Hall University, South Orange, NJ**

**Potential Zinc Stress Response Mechanisms in *Synechococcus* sp. IU 625
Robert Newby, Jr. and Tin-Chun Chu
Seton Hall University, South Orange, NJ**

**Comparative Effects of Zinc Chloride on Cyanobacteria *Microcystis aeruginosa*
and *Synechococcus* sp. IU 625
Jose L. Perez and Tinchun Chu
Seton Hall University, South Orange, NJ**

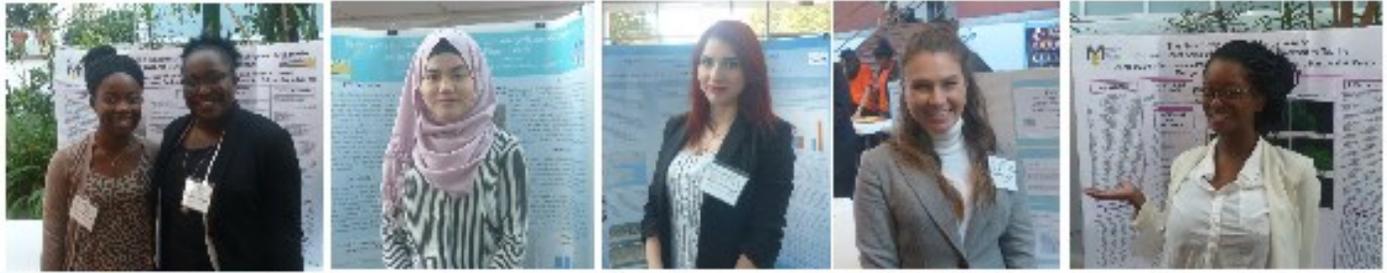
Physiology, Neuroscience and Clinical

First Place

Regulation of Gene Expression by HRE's Under Chemically Induced Hypoxic Mimic Condition. Alberto Herrera, George Coricor and Jane L Ko. Seton Hall University, South Orange, NJ

Second Place

**The Influence of the Rs53576 Oxytocin Receptor Gene Polymorphism on the Modulation of Self-Assessed Stress Levels and Salivary Cortisol in High School Adolescents
X. Wang, V. Dolar, D.S. Ginsburg and A.L. Gucwa
Long Island University, Post Campus, Brookville, NY**

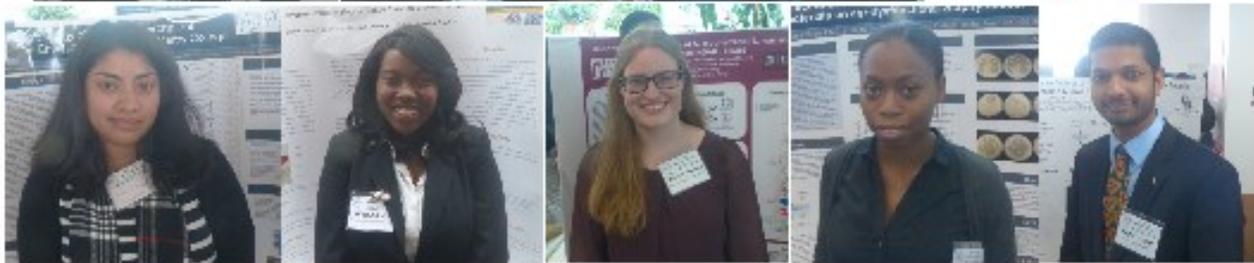


Conference Highlights









MACUB 2016 Conference Poster Abstracts

Examining the Effect of Oxidative Stress on *Tetrahymena thermophila* Sirtuins Exposed to Hydrogen Peroxide. Victor Adedar and Ralph Alcendor, New York City College of Technology, Brooklyn, NY.

Sirtuins are enzymes found in almost all organisms. These function as regulators of cell death, transcription and translation, and mitochondrial biogenesis. Sirtuins have also been shown to be important for longevity. Mice overexpressing sirt6, one of the seven mammalian sirtuins, live 15.8% longer than normal mice. Oxidative stress, OS, is the imbalance between the production of reactive oxygen species, and antioxidants. High levels of OS can disrupt mitochondrial function and can lead to cell death. OS has been connected to many diseases including cancer, diabetes and cardiomyopathies. Although numerous studies have been conducted on OS, the mechanisms involved are still not well understood. This project aimed at examining the effect of OS on sirtuins using *Tetrahymena thermophila* as a model. *T. thermophila* is a fresh water protist found around the world. These organisms are very similar to mammalian cells and, as a result, they have been used extensively for cellular and molecular research. *T. thermophila* are very resistant to conditions that are lethal to mammalian cells. These characteristics make them ideal for studying the effects of high OS. Therefore, we hypothesized that sirtuins are involved in OS regulation in *T. thermophila*. Cells were exposed to varying concentration of H₂O₂, an inducer of OS, for 24 hrs. followed by cell death analysis and gene expression. Cell death as a result of H₂O₂ was concentration dependent with 0.5 mM of H₂O₂ resulting in only 69% of cell surviving, while 1.0 mM resulted in about 50% of cells surviving. The mRNA levels of several *T. thermophila* sirtuins were upregulated in the presence of up to 1.0mM H₂O₂. However, 2.0 mM resulted in significant reduction of sirtuin mRNA levels. These results suggest that sirtuins may be involved in OS in *T. thermophila*.

Tardigrades of Harvard Forest: An Exploratory Report. *Emmanuel G. Agyei¹, William R. Miller² and Meg D. Lowman³, ¹Medgar Evers College, Brooklyn, NY, ²Baker University, Baldwin, KA and ³California Science Academy, San Francisco, CA.

Tardigrades are microscopic limno-terrestrial invertebrates that are found on all seven continents in a variety of substrates, like moss and lichen. Wide-scale distribution of tardigrades is an understudied subject, particularly in the eastern deciduous forests of Massachusetts. Previously, the state only had records of two species. We hypothesize that there are many unidentified species of Tardigrades in forests. To test this we searched for Tardigrades in the Harvard Forest in MA. Tardigrades were collected from the canopies of a wide array of trees through Baker University's Research Experience for

Undergraduate (REU): "Tardigrades in the Canopy" funded by the National Science Foundation. The composition of the species of those trees were used to represent North America's eastern deciduous forest biome. We worked in pairs and utilized the Double Rope Climbing Technique. One hundred and three trees were climbed and samples collected from six different sites in Harvard Forest in MA. Animals were then identified as to species level using a differential interference microscope. Over 5000 tardigrades were collected and thus far fifteen species of tardigrades were found in the canopies. This study expands the known Massachusetts tardigrade species diversity by 650% from 2 to 15. These findings broaden the scientific field's knowledge of tardigrade distribution not only up and along the trunk of a tree, but also on a national scale. This project was funded in part by Funded by NSF:REU #1461005.

Comparison of the Immunomodulation of Macrophages by Lipopolysaccharides (LPS) From Three Different *E. coli* Serotypes. Ayinuer Aierken, A. Lucia Fuentes and Maria Entezari, LaGuardia Community College, Long Island City, NY.

In this study, we investigated the effects of *E. coli* LPS from three different serotypes which are most commonly used as stimulants in immunological studies, 0111:B4, 055:B5 and 026:B6, on cell viability, cell morphology, nitric oxide (NO) production and phagocytosis. Murine-derived RAW-264.7 macrophages were cultured and treated overnight with 100 ng/ml, 500 ng/ml, and 1µg/ml LPS. Cell viability was affected to similar degrees by all serotypes. All three serotypes significantly increased NO production when compared to untreated cells. Furthermore, cells treated with 100 ng and 1µg/ml of LPS serotype 0111:B4, presented significantly higher NO production than both other serotypes, while 026:B6 had the lowest production. Cell morphology was examined using light microscopy following staining. All three serotypes induced an activated phenotype, as evidenced by increased vacuolation, granulation, and presence of lamellipodial extensions. Zymosan particles, derived from the cell wall of *Saccharomyces cerevisiae*, were added following LPS treatment, and phagocytosis was quantified by counting the number of phagocytic cells as well as the number of particles per cell. All three serotypes significantly increased phagocytosis; importantly, this augmentation was significantly higher in cells that were pre-treated with 100 ng/ml and 500 ng/ml of LPS from serotypes 0111:B4 and 026:B6, when compared to the increase in phagocytosis after treatment with LPS from serotype 055:B5. These results stress the importance of considering the particular serotypes used when assessing, or comparing previously reported results on, the effects of LPS in immunomodulation.

An Interconnection Between Rapamycin Resistance Phenotype and Stress Granule Markers in Yeast. Thamer Alanazi, Praveena Dhulipala and Marci J Swede, LIU, Post, Brookville, NY.

Rapamycin is an antifungal that causes cell cycle arrest in yeast and mammalian cells. Rapamycin first binds to the FPR1 (FKBP12) protein and then this drug complex binds with TOR1 signaling complex, causing cell cycle arrest. Mutations in the FPR1, TOR1 and TOR2 lead to rapamycin resistance. Recently, our lab has discovered the XRR1 (exhibits Rapamycin Resistance) gene, which shows a conditional rapamycin resistance phenotype and may interact with the Fpr1 protein. On solid media, the *xrr1?* mutant strain exhibits rapamycin resistance at 30°C while in liquid media the *xrr1?* mutant strain is sensitive to rapamycin at 30°C and is resistant to rapamycin only at 37°C. These results suggest that the XRR1 gene may play a new role in the rapamycin resistance pathway. The Xrr1 protein appears to have a cytoplasmic localization, but shows a punctate pattern after cellular stress. This pattern is similar to proteins that localize to stress granules. The XRR1 gene shows sequence homology to the A1pp macro domain, which interacts with (ADP)ribose, a post-translational modification associated with cellular stress. Macro domain proteins and modified proteins are found in stress granules in mammalian cells. The observation that the Xrr1 protein may locate to stress granules encouraged our lab to study other proteins that localize to stress granules, such as Pbp1 and Pub1. Preliminary data suggests that localization to stress granules is aberrant in *xrr1?* mutants. In addition, data suggest that the *pbp1?* and *pub1?* mutant strains exhibit a rapamycin resistance phenotype on solid media at 30°C. In liquid media, both *pbp1?* and *pub1?* are sensitive at 30°C and 37°C. Continuing experiments will explore the rapamycin resistance phenotypes for these mutant strains at 42°C and 46°C to determine if more extreme stress results in a rapamycin resistant phenotype.

The Influence of Light and Dark Environments on Aggregation Patterns of Turkestan Cockroaches (*Blatta lateralis*) at Different Developmental Stages. Linda Ali and Scott Kight, Montclair State University, Montclair, NJ.

Aggregation is a behavior that results in the formation of a group of conspecifics in close proximity. In arthropods, aggregation may increase survival by reducing water evaporation. This study investigates aggregation behavior in light and dark-mediated environments in Turkestan cockroaches (*Blatta lateralis*), an invasive species in the southwestern United States. We used binary and free choice arena tests to examine patterns of

aggregation of adult females and fourth and fifth instar nymphs. Aggregation behavior occurred more often in illuminated environments compared to dark environments, especially among immatures. The nocturnal nature of cockroaches, as well as negative phototaxis, may be important for inducing group formation. Maternal behavior, however, may increase aggression among females, which in the present study might explain decreased aggregation in some trials. The study also suggests that group size may influence aggregation behavior.

Evidence that Eastern Oysters from Woods Hole, Massachusetts, are a Separate Population from Oysters South of North Carolina. Peter Amoako, Gary Sarinsky and Craig S. Hinkley, Kingsborough Community College, Brooklyn, NY.

Eastern oysters (*Crassostrea virginica*) were found in large numbers in Jamaica Bay, New York, but there are few or none there now. Oysters are important organisms for marine ecosystems as oyster reefs can create habitats for other organisms. Therefore, our lab is interested to repopulate Jamaica Bay with eastern oysters. In order to accomplish this goal we investigated whether populations from Canada to Mexico are genetically different. Previous results in our lab showed there is single polymorphism (T → A) within the cytochrome-c-oxidase I gene (COXI) found in oysters south of North Carolina that was not present in oysters to the north. Last year our lab found one oyster from Tappan Zee, NY, that had the "A" polymorphism so we wanted to test more oysters from the north. In this project, I examined oysters from Woods Hole (WH), Massachusetts. My hypothesis is that the oysters from WH will not have the "A" polymorphism. To test my hypothesis, DNA was extracted from oysters and used to amplify a region of the COXI gene by PCR. An agarose gel was run to verify the correct size of the amplified DNA. Amplified DNA was sequenced by ELIM Biopharmaceuticals and alignment of the DNA sequences showed that none had the "A" polymorphism. These results support my hypothesis. Taken together with our previous results, 1 of 69 northern oysters had the "A" polymorphism while 52 out of 79 southern oysters had the "A" polymorphism. This suggests that the "A" polymorphism may be present only in certain areas in the north or at a very low frequency in the north or both. In the future, we would like to examine oysters from other areas. This work was supported by grants 2R25GM06003 of the Bridge Program of NIGMS and 0537-17-1091 of the CSTEP Program of NYSED.

Antibacterial Activity and Biofilm Formation Inhibition of tea polyphenols. Gabriella Appice and Tinchun Chu. Seton Hall University, South Orange, NJ.

Biofilms are complex microbial communities that adhere to solid surfaces. They are known to be able to survive harsh and stressful conditions and are resistant to antimicrobial therapy. The recent use of natural products as antimicrobial agents has encouraged the investigation of their effects against biofilm formation. Tea, one of the most common beverage, originates from the leaves of the *Camellia sinensis* plant. Most tea can be categorized into one of the two major groups: fermented tea (such as black tea) and non-fermented tea (such as green tea). In this study, the effects of black tea polyphenol, theaflavin (TF), and modified green tea polyphenol, mono-palmitoyl-epigallocatechin-gallate (pEGCG), were used to evaluate the antibacterial activity and the antibiofilm formation on four bacteria. Two gram-positive bacteria, *Staphylococcus epidermidis* (*S. epidermidis*) and *Streptococcus mutans* (*S. mutans*) and two gram-negative bacteria, *Escherichia coli* (*E. coli*) and *Pseudomonas aeruginosa* (*P. aeruginosa*) were selected for this study. The antimicrobial effect of TF and pEGCG was assessed using microplate assay. The qualitative and quantitative biofilm formation assay were investigated by Congo red and resazurin assays, respectively. Antibacterial microplate assay result indicated 0.2% pEGCG and TF inhibit the growth of bacteria, with an LD₅₀ range of 0.05-0.1% for both compounds. Results from biofilm assays showed that theaflavin and pEGCG are highly effective in inhibiting the formation of biofilm at concentrations of 0.05%. The results suggested that tea polyphenols can serve not only as antibacterial alternatives, but also potential antibiofilm agents.

Evaluation of *Mycobacterium smegmatis* as an *In Vitro* Model for Viable But Non-culturable (VBNC) Bacteria. Christina Blonski-Cupo¹, William Matern² and Petros Karakousis², ¹Caldwell University, Caldwell, NJ, and ²Johns Hopkins University, Baltimore, MD.

Tuberculosis is one of the most common infectious diseases worldwide. It is responsible for 1.5 million deaths annually (WHO). The treatment period required to cure Tuberculosis is at least six months for uncomplicated cases. This lengthy treatment period may be due to the presence of viable but non-culturable (VBNC) cells able to tolerate antibiotics. In vitro models of VBNC using *M. tuberculosis* are time consuming and can give inconsistent results. Our study focuses on developing a new, highly consistent in vitro model using *M. smegmatis* to replace VBNC models currently available in *Mtb*. We hope our model will be useful for evaluating novel compounds which reduce the duration of antibiotic therapy in patients with TB.

Direct PCR Detection, Cloning and Characterization of Mold Populations from Soils and Compost. Joy Bochis, Matthew Gardner, Theranda Jashari, Victoria Ellman, Jenifer Vasquez, Stephanie Zapata, Victorya Ramos, Tina Choe, and Luis Jimenez, Bergen Community College, Paramus, NJ.

Molds are somewhat difficult to isolate from environmental samples. Furthermore, identification using phenotypic analysis is inaccurate, slow, and extremely subjective. A procedure was developed to detect and identify mold populations from a New Jersey soil, New York soil, and compost samples. Fungal internal transcribed space sequences (ITS) were amplified by PCR using primers ITS1 and ITS4. Microbial DNA was extracted using the Zymo Microbe DNA MiniPrep protocol. DNA fragments of approximately 640 base pair were detected in all positive samples. Clone libraries were constructed with the amplified DNA fragments by ligating the detected fragments with vector pCR®4-TOPO. Transformations were performed using competent Mix and Go *Escherichia coli* cells. Plasmids were isolated from each clone using the Zippy Plasmid Miniprep and inserts were screened by PCR using M13 DNA primers. All clones with at least an 800 bp insert were DNA sequenced followed by BLAST analysis to determine the identity of the cloned fragments. DNA sequencing of clone libraries showed that compost samples were predominantly comprised of *Thermomyces lanuginosus*. Mold clones from the NJ soil were mostly unidentified species while the most abundant sequences in the NY soil were related to *Cortinarius flexipes*.

Extraterrestrial Hydrology. Sarah Slater Bowker, Robert Link and Noelle Cutter, Molloy College, Rockville Centre, NY.

Terrestrial bodies are found scattered throughout our Solar System, with various compositions and orbits. These factors both affect the likelihood of each body to contain a compound critical to life; water. This project examines various terrestrial bodies throughout the Solar System, explaining and detailing the possibility of each to have water. Examining bodies near to our Earth, and as far out as the Oort Cloud, the goal of the project is to determine the bodies in our Solar System that possess water in any form, or other clues as to how water cycling on Earth may have begun. Bodies examined include planets, dwarf planets, moons, and comets. The bodies that are found to potentially contain water or hydrologic cycles are then further examined, for comparison with known systems here on Earth.

Coordinated Beating of Gill Lateral Cell Cilia of *Mytilus edulis* and *Crassostrea virginica* Involves Neuronal Innervation and Functioning Gap Junctions. Reniece Buchanan, Cheyanne Robertson, Dane Frank, Edward J. Catapane, and Margaret A. Carroll, Medgar Evers College and Behavioral Instruments.

Gill lateral cell (GLC) cilia of bivalves drive pumps providing water currents for feeding, respiration and waste elimination. GLC in *Mytilus edulis* and *Crassostrea virginica* are innervated by serotonergic/dopaminergic nerves. Gills receive branches from the branchial nerve. It is unclear if each GLC is innervated. GLC have gap junctions (GJ). Questions arise if GJ coordinate beating. We hypothesize GJ coordinate GLC cilia beating in *M. edulis* as we found in *C. virginica*. Using immunohistofluorescence we found GJ present. To determine if GJ are involved in coordinating GLC cilia beating we used *M. edulis* preparation, similar to our method with *C. virginica*. Filaments were divided into proximal and distal sections by petroleum jelly that prevented diffusion of drugs from side to side. We stimulated the branchial nerve entering the proximal ends with suction electrodes. Drugs were applied to distal ends. Stroboscopic microscopy was used to measure cilia beating of both ends. Lindane caused dose-dependent decreased beating. Lindane reduces GJ permeability and increases intracellular Ca^{2+} , so we used diltiazem, a Ca^{2+} channel blocker, and found diltiazem did not alter beating, nor the actions of lindane. We also observed in *M. edulis* and *C. virginica* that clusters of connected GLC on the same gill filaments have cilia beating at coordinated rates, but at discoordinated rates with $10^{-5}M$ lindane, suggesting, like cardiac tissue, there are pacemaker cells and follower cells in gill filaments. Results show the physiological functioning of GJ involved in coordinating GLC cilia beating in two bivalve species and neuronal innervation in conjunction with functioning GJ are necessary to coordinate beating rates in both bivalves. Disrupting either one interferes with the normal response. This work was supported by 690340047 of PSC-CUNY, 2R25GM06003 of the Bridge Program of NIGMS and the Carnegie Foundation.

Production of Melanotic Tumors in *Drosophila* as a Result of Juvenile Hormone Mimic Treatment. Raquel Calero and Rebecca Spokony, Lehman College, Bronx, NY.

When *Drosophila melanogaster* undergoes metamorphosis their blood cells transform in shape and become lamellocytes, which encapsulate tissue and produce melanotic tumors. This occurs as a response to infection or overgrowth of a tissue; however, the effect of hormone regulation in this process is relatively unknown. Metamorphosis in *Drosophila* is controlled by two hormones, 20-hydroxyecdysone and juvenile hormone (JH), which regulate changes in gene expression during metamorphosis, and inhibit ecdysone from making these

changes earlier during development, respectively. The formation and development of blood cells occurs in the lymph gland, which differentiates throughout the larval period and its metamorphosis is triggered by a signal produced when ecdysone has reached peak levels. Larval exposure to juvenile hormone mimics (JHM) leads to melanotic tumors. The purpose of this experiment is to characterize variation in lymph gland sensitivity to JHM across a collection of natural caught variants and map the changes in sensitivity to differences in their genotypes. In this experiment approximately 40 *Drosophila* Genetic Reference Panel (DGRP) lines were treated with JHM, methoprene. The production of melanotic tumors among the different genotypes was observed after the treatment. There was significant variation in the incidence and type of tumors produced across the lines examined. We divided the tumors into at least three categories. The first category contains individuals that portray several (more than 1) very small tumors, possibly melanized crystal cells. The second category includes those with a single, big tumor (bigger than a segment). In the third category the shape of the tumors is distinguishably different, the tumors seem to be a chain starting with a big tumor followed by smaller ones trailing down the body of the fly. It is hypothesized that because of this particular shape, these tumors may be a result of melanization of the lymph gland, itself.

Synthesis of Ruthenium Nanoparticle-Single-Walled Carbon Nanotube Composites for the Degradation of Congo Red Dye. Nicholas Carrero, Rawlric Sumner and Tirandai Hemraj-Benny, Queensborough Community College, Bayside, NY.

The Congo red dye, a product dumped by textile factories, is a carcinogen that is toxic to the flora and fauna in our environment. Thus, it has become increasingly imperative to treat these dyes before they enter the environment. Although several physical and chemical methods have been employed to eliminate contaminants from waste water, which includes activated carbon sorption, UV light-degradation, and chemical oxidation-reduction treatments, there is still a need to develop and to improve processes which are rapid and efficient to degrade the dyes. In general, ruthenium is cheaper than other metal catalysts such as Au and Pt, and have demonstrated uniquely high selectivity and catalytic activity. This study aims to: 1) synthesize ruthenium nanoparticles supported on single-walled carbon nanotubes by microwave irradiation in ethanol; and 2) investigate the catalytic properties of the synthesized composites on the degradation of Congo red dye. The synthesized ruthenium nanoparticle-single-walled carbon nanotube composites and their catalytic properties were characterized by High Resolution Transmission Electron Microscopy (HR-TEM), Energy Dispersive X-ray Spectroscopy (EDS), and UV-Vis Spectroscopy. Herein, we report the microwave synthesis of ruthenium nanoparticles of uniform size of ~5 nm which were densely

distributed on the surfaces of single-walled carbon nanotubes bundles. In addition, the Ru nanoparticle-single-walled carbon nanotube composites dramatically improved the degradation of Congo red dye which occurred within a few minutes. Nicholas Carrero and Rawlric Sumner are participants in the NIH Bridges to the Baccalaureate Program at Queensborough Community College.

Traumatic Brain Injury as a Disorder of the Connectome. Merrysha Castillo, Nikoloas Ziogas and Vassilis E. Koliatsos, Wagner College, Staten Island, NY.

Traumatic brain injury (TBI) is associated with a variety of primary and secondary lesions such as contusions and diffuse axonal injury. Although diffuse axonal injury is by definition a network problem because it directly affects axons, contusions are viewed strictly as a focal pathology without the consideration of related connections and circuits. Post-mortem human brains with orbitofrontal contusions or diffuse axonal injury were examined using immunohistochemistry to detect the presence of phosphorylated epitopes of heavy and medium-molecular weight neurofilament proteins (NF-H/NF-M), which marked damaged neurons in interconnected neocortical, limbic, and subcortical areas. Large numbers of phosphorylated NF-H/NF-M-immunoreactive, SMI 310 (+) neuronal cell bodies were found in the ventral group of the lateral nucleus, specifically in the ventral anterior nucleus and ventral lateral nucleus of thalamus for each contusion case. Neuronal cell bodies with p-NF (+) were also identified in the hippocampus, but numbers were smaller. In contrast, frontal sections did not show any p-NF (+) cell bodies. In the diffuse axonal injury case, frontal sections showed robust p-NF (+) cells throughout the cortex. After orbitofrontal contusion, retrograde/transsynaptic degeneration is profound in the thalamus, but less so in the hippocampus, and none in the interconnected frontal cortex. These investigations lay the foundations for elucidation of the neuropathology of traumatic contusions and diffuse axonal injury on the basis of impaired neocortical and limbic connectivity, and promote understanding of behavioral and cognitive changes in patients with TBI.

Development of Peptidyl Ligands Using Solid Phase Peptide Synthesis to Identify Target Receptor Proteins. Suny Chavarria¹, Yerania Aguilar¹ and Sanjai Kumar², ¹Queensborough Community College, Bayside, NY and ²Queens College, Flushing, NY.

Rapid emergence of metabolic disorders necessitates that the underlying signaling events at the cellular level are appropriately understood. Many neuropeptides in the hypothalamus play important roles in regulating food and energy expenditure signaling events. However, the precise mechanism by which they interact with their cognate

receptors remains poorly understood. In this work, a library of peptidyl ligands derived from proSAAS neuropeptide was synthesized using Fmoc-based solid phase peptide synthesis. The synthesized peptide library is currently being purified using reverse-phase HPLC, and characterized by electrospray ionization mass spectrometry. The developed peptide library will then be utilized to identify GPCR proteins that are involved in regulating food and energy expenditure signaling events. Suny Chavarria and Yerania Aguilar are participants in the NIH Bridges to the Baccalaureate Program at Queensborough Community College.

Effects of Alcohol-Based Hand Sanitizer on the Diversity of Skin Microflora. Johnessa S. Clarke, Elizabeth A. Mulligan and Grace Axler-Diperte, Kingsborough Community College, Brooklyn, NY.

Skin microbes play a crucial role in overall health. The skin is the first layer of defense against infectious microorganisms and toxic agents. Other work has suggested that frequently washed hands of healthcare workers are colonized with more pathogenic species than those who wash less frequently. However, many studies have demonstrated that hand washing or the use of alcohol-based hand sanitizer reduces infection rates in various institutional settings. The purpose of this project is to investigate whether the use of alcohol-based hand sanitizer effects the diversity of bacteria on the hands over a single hour of normal activity. Human hands are a conduit for exchanging microorganisms between the environment and the body. My hypothesis is that the diversity of the bacteria will change over time and differ depending on hand cleaning methods. In this study ten volunteers used either hand sanitizer or saline solution, swabbed their hands before and again at times 0, 30 and 60 minutes post washing. Samples were then applied to Tryptic Soy Agar plates and incubated at 37°C overnight. Eight distinctive colonies were chosen from each time point, purified and patched onto selective and differential media and incubated at 37°C. Tryptic Soy Agar, Hektoen Enteric Agar, MacConkey and Sheep's Blood Agar were used to characterize the bacteria. From these samples, selections were made for Gram Staining and microscopic observation. The results showed an increase in hemolytic bacteria for the saline samples, but a decrease in the sanitizer samples. These results support my hypothesis that the diversity of the bacteria on the hands would be different with the hand sanitizer compared to the saline wash. This work is supported by grant 0537171091 of the CSTEP Program of the NYS Department of Education.

The Stress of Public Speaking Increases Cortisol Levels in Undergraduates: Is increased Preparation Really the Best Remedy? Erin Clinton, Grace Cookson, Stephanie Brown, Daniel Woods and Jodi F. Evans, Molloy College, Rockville Centre, NY.

Perceived stress is prevalent among the undergraduate population. When this stress persists, it has the potential to lead to mental health illnesses. Recent research shows 85% of students experience overwhelming anxiety from academic pressures. Physiologically, during stressful events, cortisol levels rise in the body which disrupts homeostasis. The anticipation prior to a class presentation, a form of public speaking, is a common source of perceived stress among undergraduates. The focus of this experiment was to determine if there is a correlation between factors such as increased preparedness, sleep, level of understanding, perceived anxiety and physiological stress parameters. Twenty-eight student volunteers with an impending oral presentation were enrolled from both 100-level and 200-level undergraduate courses. At baseline and on the day of the presentation, salivary cortisol, heart rate, and blood pressure were measured. The participants were also asked to complete the Beck's Anxiety Inventory (BAI). Compared to baseline, cortisol levels on the day of the presentation were significantly increased in both groups. The change in salivary cortisol levels did not correlate with the number of hours spent preparing, the level of understanding nor hours of sleep the night before the presentation. However, the analyses revealed a trend toward an inverse correlation between the self-reported level of understanding and change in cortisol levels. Essentially, students who felt ambiguous toward their level of understanding of their presentation experienced lower changes in cortisol levels when compared to those students who reported a stronger understanding of the material. This study confirms that undergraduates' perceived stress in anticipation of public speaking does manifest in significantly elevated cortisol levels. It does not provide a link between increased preparation and reduction of stress parameters. Future studies could focus on alternative methods such as mindfulness and meditation and their efficacy in reducing undergraduate stress associated with public speaking.

Anterior Cerebral Artery Stroke: A Case Study Created to Understand the Clinical Dysfunctions Related to the Ischemic Brain Regions. D. Colgan, W. Mirza, L. Lorentzen and K. Reilly, Kean University, Union NJ.

A case study was developed by a student team in an undergraduate neuroscience class to link functional brain anatomy to cardiovascular attack in a teaching style format. The student team reviewed the three major cerebral arteries (anterior, middle and posterior) and researched all brain regions that each artery and major bifurcation branches supply. The students then researched

the neurological functions of each brain region. The students selected one artery of interest and a case study was then created that presented the signs and symptoms of embolic anterior cerebral artery stroke. The case study presented a patient, with a history of hypertension and high blood pressure, who suddenly began suffering from a series of ailments such as aphasia, apraxia, and topical anesthesia. Further medical examination performed by the patient's neurologist revealed the diagnosis of Broca's Aphasia, Hemiplegia, and Alien Hand Syndrome. Case study questions were then created to relate the symptomology to the functional anatomy of the brain regions supplied by the artery. In researching and conducting this case study, the students learned to integrate their classroom content in neuroscience and anatomy/physiology with clinical relevance as they designed this case for future use by faculty in an advanced course.

Isolation and Identification of Antibiotic-Resistant Bacteria From New York City Soil Samples. Adolfo Coyotl and Joan Petersen, Queensborough Community College, Bayside, NY.

As an urban environment, New York City puts an enormous amount of pressure on its natural areas. As part of the Soil Joint Seed Project we are interested in the potential effects of antibiotic resistant bacteria on human health and the environment. My research project is focused on determining patterns of antibiotic resistance among bacteria found in New York City soils. Samples were collected from three sites with varying degrees of human influence: Thain Forest (TF-pristine), Central Park (CP-intermediate), and Newtown Creek (NC-heavily polluted). Over 131 pure cultures were isolated on Reasoner's agar (R2A) from dilutions of soil samples. To test for antibiotic resistance, cultures were streaked onto R2A plates containing either penicillin or kanamycin. Gram staining and microscopy were used to determine morphology and Gram reaction of the resistant isolates. Overall there were 96 gram-positive and 35 gram-negative isolates. Endospore-formers were found among all three sites: Newtown Creek had the largest percentage of endospore-formers (65% of isolates). Antibiotic testing showed that there were resistant bacteria in all three sites, with more isolates being resistant to penicillin than to kanamycin. The Newtown Creek site had the most penicillin-resistant isolates (89%). Further studies will involve identification of species by 16S rDNA sequencing along with determination of resistance mechanism using PCR primers specific to resistance genes. The prevalence of the isolates in the natural soil community will be determined by comparing our sequences to metagenomic sequencing results. This research has possible implications for determining sources of antibiotic resistance genes in urban microbiomes.

Immunohistofluorescence Localization of Biogenic Amine Receptors in Ganglia and Tissues of the Bivalve Mollusc, *Mytilus edulis*. Delilah Cummings, Maxine Jacobs, Margaret A. Carroll and Edward J. Catapane, Medgar Evers College and Kingsborough Community College, Brooklyn, NY.

Biogenic amines are neurotransmitters in sensory and motor systems in invertebrates. In bivalves, particularly *Mytilus edulis*, the physiological roles of serotonin and dopamine have been well studied especially with respect to gill and heart. Recently, in the related bivalve *Crassostrea virginica*, other biogenic amines, including GABA and histamine along with their receptors, were demonstrated to be present. These amines are being shown to have physiological roles. We hypothesize *M. edulis* has receptors for the biogenic amines, serotonin, dopamine, histamine and GABA. We tested this using immunohistofluorescence techniques. Primary antibodies to serotonin, dopamine, histamine and GABA receptors, and secondary FITC-labeled antibodies were used to visualize these biogenic amine receptors in cerebral ganglia, visceral ganglia, gill and mantle. Briefly, tissues were dissected, snap frozen, sectioned on a cryostat, fixed with EDAC (N-Ethyl-N'-(3-dimethylaminopropyl) carbodiimide hydrochloride), treated with blockers, and incubated with primary, then secondary antibodies. The results show serotonin 2A-like, dopamine D2-like, histamine H2-like and GABA R1-6-like receptors present in cerebral ganglia, visceral ganglia, gill and in the sensory tentacles of the mantle rim. This comparative study provides new knowledge of the neurobiology of *M. edulis* by confirming the presence of these biogenic amine receptors in the animals' nervous system and innervated organs, in a comparable manner to what has been reported in *C. virginica*. It also provides a foundation to further study the physiological roles for histamine and GABA in *M. edulis*. This work was supported in part by grant 2R25GM06003 of the Bridge Program of NIGMS, grant 690340047 of PSC-CUNY and a Carnegie Foundation award.

Role of Loop 6 in Cyclic-di-GMP Specific Phosphodiesterase in *Shewanella woodyi*. Margaret de los Santos¹, Dominique Ebony Williams² and Elizabeth Boon², ¹Queensborough Community College, Bayside, NY and ²Stony Brook University, Stony Brook, NY.

Biofilms are matrix encapsulated microbial colonies which usually adhere to moist surfaces. Though beneficial to the survival of bacteria, biofilms can carry potentially harmful pathogens which can compromise one's health. Cyclic di-GMP is a bacterial second messenger molecule that has a hand in the formation of biofilms. High concentrations of c-di-GMP cause formation, and in low concentrations the dispersal, of biofilms. Like many other bacteria *Shewanella woodyi*'s cyclic di-GMP processing, bifunctional enzyme has both phosphodiesterase (PDE) and cyclase ability (DGC). Certain bacterial heme proteins have been known to behave as nitric oxide (NO) sensors, such as H-NOX (heme nitric oxide/oxygen binding) proteins. H-NOX proteins have been known to regulate HaCE activity. When nitric oxide is bound to H-NOX, PDE

activity increases which causes biofilm dispersal. This project focuses on uncovering the role of amino acid residues in the loop 6 region, responsible for binding substrate and magnesium ions, of the cyclic di-GMP specific phosphodiesterase in the HaCE. Two different mutants were made, S543A and D535A. Unlike the wild type, enzyme activity of both mutants displayed relatively no activity. H-NOX was also characterized by UV-vis spectrophotometry. Future studies will focus on investigating the activity of the mutants in the presence of H-NOX protein in its reduced unligated and NO bound states. Understanding the roles of the different parts in these complex bacterial signaling pathways may aid in the development of therapeutic intervention methods and antimicrobial treatments to prevent and alleviate biofilms.

Differential D2 Dopamine Receptor Modulation of Selective Cell Populations. Christopher Dessources¹, Samuel R. Cain^{2,3}, Joseph Khafi² and Jeff A. Beeler^{2,3, 1}, LaGuardia Community College, Long Island City, NY, ²Queens College, Flushing, NY and ³Graduate Center City University of New York, NY.

Dopamine D2 receptors (D2Rs) act on many cell populations within the striatum. Some roles for D2Rs include inhibition of striatopallidal medium spiny neurons (MSNs) and regulation of glutamate release from cortical afferents. Global D2 knockdown (D2KD) mice, possessing a stop codon upstream of D2R, show significant attenuation of receptor expression. Generalized knockdown of D2R results in motor deficits and reduced energy expenditure, without other phenotypic aberrations. Presently, a selective D2R rescue allowed explicit examination of the contributions of different striatal cell populations, both in the expenditure of energy and over the course of motor learning. Cre-Lox recombination rescued D2R solely in cells expressing Cre through the removal of the upstream stop codon. Selective, Cre-driven rescue of D2R in striatopallidal MSNs (Adora-Cre) and cortical projections (Rbp4-Cre) was possible due to the limited, population-specific expression of Adora and Rbp4. An open field paradigm was used to measure energy expenditure and the accelerating rotarod tested implicit motor learning both in rescues and knockdowns. Rescued D2R expression in striatopallidal MSNs yielded significant increases in both open field movement and motor learning performance relative to D2KD mice. This likely resulted from increased inhibition of the indirect pathway due to dopaminergic action, causing a net reduction to cortical inhibition. Cortical rescue of D2R, however, did not rescue behavior. Subjects showed no difference in energy expenditure or motor learning relative to knockdowns. We hypothesize that increased presynaptic D2R modulation reduces glutamate release from cortical projections. Consequently, reduced glutamatergic activation of striatal MSNs impairs motor performance. Immunohistochemical staining confirmed that striatal D2R expression was present in both types of rescues. Opposing behavioral results from these rescues highlights the dynamic contributions of D2R in the generation of behaviors and emphasizes the need to examine these receptors as a heterogeneous population.

PTEN: A Potential Neuroprotective Target in Astrocytes. Dailyn Despradel, Karol Toral and Renée E. Haskew-Layton, Mercy College, Dobbs Ferry, NY.

Astrocytes, a specialized glial cell type in the central nervous system, play critical roles in supporting neuronal function and survival. A primary function of astrocytes is to provide antioxidant support to neurons in the form of precursors of the antioxidant molecule glutathione (GSH). Reactive oxygen species (ROS - e.g. superoxide, hydrogen peroxide, and the hydroxyl radical) foster normal brain function by facilitating neuronal communication and regulating cerebral blood flow. However, accumulation of ROS during aging and in neurodegenerative conditions (e.g. Parkinson's disease, Alzheimer's disease, Huntington's disease) leads to neuronal death. Understanding the specific mechanisms by which astrocytes protect neurons from oxidative stress will contribute to the development of novel neuroprotective therapeutics. Our previous data show that the inhibition of protein tyrosine phosphatases (PTP) in astrocytes leads to the robust protection of neurons from oxidative stress. A long-term goal of our laboratory is to identify the specific PTP and PTP-downstream targets that induce astrocytic neuroprotection. We hypothesize that the PTP PTEN (phosphatase and tensin homolog) and its downstream target kinase Akt promote astrocyte-dependent neuroprotection. To study the potential involvement of PTEN and its target Akt, we tested a battery of small molecular PTEN inhibitors in an astrocyte-neuron chicken coculture model. Astrocytes, derived from embryonic day 8 chicken embryos, were plated for two weeks prior to plating neurons derived from embryonic day 5 chicken embryos. To determine an astrocyte-specific mechanism of PTEN, astrocytes were treated with PTEN inhibitors prior to neuronal plating in the presence of the oxidant hydrogen peroxide for 48 hours. Future experiments will determine the role of the downstream PTEN target Akt by specific astrocyte-directed expression of the Akt gene using the GFAP promoter.

Analysis of SIT Protein Expression in Rat Cells and Tissues. Taylor Donnelly, Nicole Rodstrom and Thomas Owen, Ramapo College of New Jersey, Mahwah, NJ.

In studies performed with our collaborators at Temple Medical School, approximately 150 genes potentially involved in the regulation of bone mass in rats were isolated. One of these genes is SIT, a member of a family of transmembrane adapter proteins which recruit other signaling proteins to the cell membrane. Although the published literature about SIT only reports its expression and function in T-cells, recent evidence from our lab demonstrated that SIT is expressed in osteoblasts at the mRNA level and that the bones of SIT knock-out mice show dramatic decreases in the amount of trabecular bone. Our goal is aimed at establishing conditions through which to identify SIT protein in rat cell and tissue samples using Western blot analysis. To begin this work, total cellular protein was prepared from ROS 17/2.8 rat osteosarcoma cells. This protein sample was run on SDS-PAGE and blotted to a PVDF membrane. Both BSA and milk were tried as blocking agents and milk proved superior. Various dilutions of a commercially available goat anti-SIT antibody

were then tried along with various dilutions of a horseradish peroxidase-conjugated donkey anti-goat secondary antibody. The signal was visualized using Clarity substrate and chemiluminescent imaging. Subsequently, total protein samples were prepared from rat stomach, liver, heart, spleen and kidney and analyzed for SIT expression using the optimized conditions. While a strong SIT signal is visible in the ROS cells with a 10 sec exposure, SIT could only be detected in rat kidney tissue and then only after an extended period of exposure (15 minutes). Since SIT is a transmembrane protein, we are planning on preparing membranes from various rat tissues in order to enhance any signal and exploring whether the high abundance of SIT protein in ROS cells is unique to them or is a consequence of their tumor origin.

The Neurotoxic Effect of Manganese on Dopamine Post-Synaptic Receptors Are Reversed by Taurine. Loren Dubose, Kurt Loney-Walsh, Margaret A. Carroll and Edward J. Catapane, Medgar Evers College, Brooklyn, NY.

Manganese, a neurotoxin causing Manganism a Parkinson's-like syndrome, disrupts dopamine neurotransmission in brain. The neurotoxic mechanism is not fully resolved. Reports postulate manganese toxicity is more related to post-synaptic dopamine receptors (D2DR) dysfunction, than degeneration of dopamine neurons. Gill lateral ciliated cells of *Crassostrea virginica* are innervated by cilio-inhibitory dopamine nerves and have D2DR post-synaptic receptors. Our previous physiological work found manganese blocked the cilio-inhibition of dopamine on lateral cells. Our immunohistofluorescence work showed manganese reduced D2DR fluorescent intensity in the cells, suggesting manganese causes a loss of D2DR number or a structural disruption of receptor integrity. There are reports p-aminosalicylic acid (PAS) alleviates some symptoms of Manganism. Our studies with *C. virginica* showed PAS reversed manganese neurotoxicity on the animals' dopaminergic system. The amino acid taurine alleviates symptoms in several neurodegenerative diseases. We hypothesize taurine would reverse manganese neurotoxicity on gill lateral cell. We treated *C. virginica* for 3 or 5 days with manganese (500 μ M) followed by 3 or 5 days with taurine (500 μ M). Gills were excised and treated for immunohistofluorescence as previously done. Fluorescence intensity was quantified using ImageJ from NSF. Manganese treatments caused a 66% decrease in fluorescence compared to controls. Animals treated with 3 days of manganese followed by 3 days of taurine showed only an 18% reduction in fluorescence. Animals treated 5 days with manganese followed by 5 days with or without taurine gave similar results. The study indicates taurine was able to reverse the manganese induced loss of D2DR fluorescence similarly to what occurred with PAS. The neuroprotective mechanism of taurine needs to be further investigated. This study suggests taurine may be a promising therapeutic agent for treatment of Manganism. This work was supported by grants 690340047 of PSC-CUNY, 2R25GM06003 of the Bridge Program of NIGMS and a Carnegie Foundation award.

Common Periwinkles (*Littorina littorea*) from Woods Hole, Massachusetts, Plum Beach in Jamaica Bay, New York, and Fort Wadsworth in the Lower Bay of New York are Not Separate Populations. Mohamed Eid, Gary Sarinsky and Craig S. Hinkley, Kingsborough Community College, Brooklyn, NY.

Common periwinkles (*Littorina littorea*) are marine snails found along the coast of North America from Canada to Maryland. They are indigenous to northern Europe and were introduced to North America sometime during the mid-1800's. Managing these periwinkles is important since the introduction of foreign species often upsets an ecosystem. Since understanding the structure of a population is important for managing the population, our long-term goal is to determine the genetic structure of periwinkle populations. In this study, we compared periwinkles collected from Woods Hole (WH), Massachusetts, to those from Plum Beach (PB) and Fort Wadsworth (FW) in New York to determine if they are part of the same population. We hypothesized that the periwinkles from WH are a separate population from periwinkles of PB and FW. To test this hypothesis, we first PCR-amplified a 700-base-pair region of the cytochrome-c-oxidase I gene using DNA extracted from tissues of twelve snails from WH. The correct size of the PCR-amplified DNA was verified using agarose gel electrophoresis. The DNA was sequenced by ELIM Biopharmaceutical, Inc., and an alignment of DNA sequences was performed using ClustalW. A phylogenetic tree constructed from the sequence alignment by the maximum-likelihood method with distance measurements calculated using the Jukes-Cantor model suggested the periwinkles are from one population. We therefore reject our hypothesis that periwinkles from WH, PB, and FW are separate populations. In the future we would like to compare common periwinkle snails along the East coast of North America. Furthermore, the common periwinkle snail is also found along the west coast of North America so we would also like to compare them to the east coast periwinkles. This work was supported by grant 2R25GM06003 of the Bridges Program of NIGMS and grant 0537171091 of the CSTEP Program of the NYS Department of Education.

Impact of Combined Sewer Overflows on Water Quality and Aquatic Biodiversity at Selected Lower Hudson Raritan Estuary Sites. Carol Ellameh, Merna Bishai, Erick Gonzalez and Meriem Bendaoud, New Jersey City University, Jersey City, NJ.

Regular wastewater is a collection of rainwater runoff, domestic sewage, and industrial waste that collects in a single pipe to a sewage treatment plant, and then discharges to a water body. Excess wastewater after rainfall leads to combined sewer overflows (CSOs) that contain untreated or partially treated waste with high toxicity and increased pathogenic bacterial concentration.

Therefore, CSOs have a high impact on water quality leading to impaired aquatic habitats, compromised drinking water supplies, and endangered human health. Our project focused on studying the bacterial concentrations and diversity at selected lower Hudson Raritan Estuary (HRE) sites in correlation with the location of nearby CSOs and rainfall events. To this end, weekly scientific testing and monitoring of water quality were conducted in the designated lower HRE sites for a period of 10 weeks. Modern bacterial identification techniques were used to determine the concentration of fecal coliform bacteria, a high concentration of which is indicative of poor water quality, as well as the concentration of *Enterococcus* bacteria, an indicator organism for the presence of potential disease-causing organisms. The results of this study will not only inform management practices but will also become an important resource for decision-makers to advance environmental protection.

Taurine Prevents the Neurotoxic Effects of Manganese on the Physiological Response of a Cilio-Inhibitory Dopaminergic System. Krystle Ernest¹, Ernest Fatoke-Osobukola², Margaret A. Carroll² and Edward J. Catapane², ¹Kingsborough Community College and ²Medgar Evers College, Brooklyn, NY.

Manganese, a neurotoxin causing Manganism a Parkinson's-like disease, disrupts dopaminergic neurotransmission. The mechanism is not fully resolved. Unlike Parkinson's, reports postulate the mechanism is more related to downstream neuronal pathways than deficits in nigrostriatal function. Gill lateral cell cilia of *Crassostrea virginica* are controlled by serotonergic-dopaminergic innervations from their ganglia. Dopamine causes cilio-inhibition, serotonin cilio-excitation. Our previous work showed acute and short-term manganese treatments block cilio-inhibition by dopamine and p-aminosalicylic acid (PAS) reversed the neurotoxic action of manganese. Recent reports suggest the amino acid taurine may be worth investigating as a neuroprotective agent because of its efficacy in other neurodegenerative diseases. We hypothesize taurine would reverse the neurotoxic actions of manganese on cilio-inhibitory effects of dopamine in *C. virginica*. We conducted acute experiments testing dopamine dose responses (10^{-6} - 10^{-3} M) on gills treated with manganese (10^{-3} M) in the presence or absence of taurine (10^{-8} - 10^{-4} M). Ciliary activity was measured by stroboscopic. Cilia of control cells responded normally to dopamine. Manganese treatments disrupted the dopamine induced cilio-inhibition and taurine treatments after manganese prevented this neurotoxic effect. Taurine had an ED50 of about 10^{-6} M. We also conducted short-term experiments in which *C. virginica* were treated for 3 or 6 days with manganese (500 μ M) with or without taurine (500 μ M). Taurine prevented the effect of treating whole animals with manganese on disrupting the cilio-inhibitory dose response of dopamine

in gill lateral cells. This study shows taurine effectively prevented manganese neurotoxicity on the cilio-inhibitory dopaminergic system in *C. virginica*. These findings are helpful in furthering the understanding of the mechanism of manganese neurotoxicity and provides evidence suggesting taurine needs to be further investigated as a potential therapeutic agent for Manganism. This work was supported by grants 690340047 of PSC-CUNY, 2R25GM06003 of the Bridge Program of NIGMS and a Carnegie Foundation award.

Development of Kura Clover Intercropped with Field Corn Under Sustainable Tillage Practices. Lina Espinal¹, Michelle Dobbratz² and Scott Wells², ¹New Jersey City University and ²University of Minnesota.

Legume cover crops or green manure are plants that are grown mainly to protect and enrich the soil for a specific period of time. They can occupy a vast variety of niches and are capable of fixing nitrogen in a symbiosis with rhizobia, a type of nitrogen-fixing bacteria that are commonly found in the soil. This symbiotic relationship allows for an increase in the levels of nitrogen in the soil and consequently greater biomass. Researchers at the University of Minnesota have conducted numerous studies with legume cover crops because of their potential to improve soil health by reducing erosion, controlling weeds invasion, reducing soil compaction, building up soil organic matter, and sequestering nutrients, among others. Despite all the environmental benefits provided by cover crops, recent studies in Minnesota have shown that cover crops are grown on only 1.5% of farmland. Farmers are reluctant to opt for this sustainable practice, primarily because there is a fear that cover crops may interfere with cash crop yield and result in a loss of revenue. In an effort to understand and determine the effect of legume cover crops on corn yield, we intercropped perennial legume Kura clover with field corn under four different types of tillage systems; Rotary Zone Tillage (RZT), Shank Tillage (ST), Dual Tillage (DT), Band Kill (BK). We hypothesized that when intercropped with corn, Kura clover can increase nitrogen surplus in the soil allowing for this nitrogen to be used up by the field corn crops. If adopted in corn production, cover cropping legumes can lower the amount of fertilizer needed to grow field corn. Therefore, becoming beneficial for our farmers and for our soils.

Trans-splicing of Interleukin-13 Receptor Alpha Variant 2 into the EGFR Transcript to Block Growth and Reactivate Immunogenic Potential of GBM Cells. Sarah C. Falotico and Martin J. Hicks, Monmouth University, West Long Branch, NJ.

Glioblastoma Multiforme (GBM) is one of the most common malignancies of the central nervous system. It is characterized by increased activation of tyrosine kinase receptors, such as epidermal growth factor receptor (EGFR). Overexpressed EGFR, found in as much as 60% of GBM, leads to tumor growth and proliferation. Changes

in the GBM microenvironment lead to tumor evasion and current therapies for GBM are limited by the blood brain barrier. RNA Trans-splicing therapeutics offers a novel approach to alter overexpressed oncogene transcripts. Pre-mRNA trans-splicing of an alternative polyadenylation signal into the EGFR transcript has the potential to block expression of the transmembrane domain, generating an extracellular peptide rather than a membrane-bound receptor. This shortened and soluble EGFR decoy isoform has the potential to sequester epidermal growth factor. Here, we describe a pre-trans splicing RNA molecule (PTRM) against EGFR that contains the interleukin-13 receptor alpha chain variant 2 (IL13R α 2). This highly immunogenic isoform is selectively expressed in GBM. Additionally, IL13R α 2 elicits a strong cytotoxic T-lymphocyte (CTL) immune response making itself an effective target for immunotherapy. Insertion of the IL13R α 2 immunogen into the PTRM expression vector has the potential to reactivate an immune response directed specifically toward the GBM tumor microenvironment. Delivery of the PTRM-immunogen hybrid therapy using an adeno-associated virus plasmid vector may synergistically inhibit EGFR expression while reactivating the CD-8 T-cells. We have begun preliminary testing of the PTRM expression in tissue culture and cloning of IL13R α 2 into the PTRM vector. Levels of EGFR expression will be evaluated via Western Blot and ELISA in several EGFR expressing GBM cell lines. Acknowledgements: This research was funded by Bristol-Meyers Squibb, Johnson and Johnson, The Independent College Fund of New Jersey, and Monmouth University School of Science.

The Role of *Drosophila* SOCS36E in Linker Histone H1-mediated Heterochromatin Formation and Tumor Suppression. Ki Sum Fan, Andrea Mejia, Meghan Pfau, Nathan Doran, Amber Crockett¹, Arthur I. Skoultchi², and Na Xu¹. ¹LaGuardia Community College, Long Island City, NY, and ²Albert Einstein School of Medicine, Bronx, NY.

The linker histone H1 is a key component of chromosomes and plays a major role in heterochromatin formation. However, how H1 executes these biological roles is largely unknown. Our recent studies showed that H1 interacts with three key factors involved in heterochromatin formation, Su(var)3-9, HP1 and STAT (Lu *et al.*, 2009, Lu *et al.*, 2013, Xu *et al.* 2014). We further discovered that the interaction of H1 and STAT plays an important regulatory role in JAK-STAT-induced blood tumor formation in flies (Xu *et al.*, 2014). To further identify genes that cooperate with H1 in regulation of heterochromatin formation, we completed a mis-expression genetic screen. We ubiquitously mis-expressed 453 distinct genes in control and H1 knockdown flies, by using the EP collection of *P*-element insertions on the second chromosome. We then examined effects of their mis-expression on H1 knockdown-induced lethality. We identified a number of genes whose mis-expression either

decreased or increased lethality induced by H1 knockdown. These genes spanned a wide spectrum of biological activities ranging from cell cycle regulators to chromatin remodelers. One of the suppressors identified in the screen is SOCS36E, a negative regulator of the JAK/STAT signaling. Our studies also showed that SOCS36E not only functions together with histone H1 in mediating fly lethality, but also is required for H1-mediated heterochromatin structure and function. Taken together, our results suggest a role for JAK/STAT signaling and SOCS36E in H1-dependent regulation of essential processes in *Drosophila*.

Neuronal Cell Death and Fluoro-Jade C Staining in Rat Model of Alzheimer Disease. Rashida R. Farokhi¹, Raquel Romaine¹, Sameh Hanna² and Francisco Villegas², ¹Queensborough Community College, Bayside, NY and ²York College, Jamaica, NY.

Alzheimer's Disease (AD) is a progressive degenerative disease of the brain that slowly attacks neurons, impairing a person's abilities to govern emotions, coordinate movement, as well as lose memory. This disease is often characterized by widespread neuronal cell loss, the overabundance of tau protein and amyloid-beta plaques, as well as impaired higher cortical functioning. This study proposes that the use of intracerebroventricular (ICV) injections of the diabetogenic drug, streptozocin (STZ) in rodents in order to produce cellular and behavioral abnormalities that mimic sporadic Alzheimer's disease (sAD). We hypothesized that the highest dosages of STZ injected into the ICV areas in rats would cause more neuronal cell death when observed using Fluoro-Jade C staining techniques. Fluoro-Jade C (FJC) staining is employed to allow for the visualization of neural cell death in the hippocampus and prefrontal cortex. Sprague Dawley rats (n=32) were anesthetized and maintained under isoflurane for the duration of surgery. The control groups (n=7) were subjects injected with a single bilateral ICV infusion of artificial cerebrospinal fluid. The experimental groups received ICV injections of 1 mg/kg STZ (n=10), and 2 mg/kg STZ (n=9). Rats were also fitted with recording electroencephalograph (EEG), screw electrodes, and electromyography (EMG) electrodes for the recording of sleep/wake activity. After approximately 180 days, histology was then conducted to visualize brain sections for cell death using Fluoro-Jade C staining techniques. When the brain tissue was stained, the degenerating neurons were distinguished by FJC-positive neurons. Fluoro-Jade C positive neurons were seen in the prefrontal cortex of the brain in sAD streptozotocin-induced rat model. We conclude that the neurodegenerative process was observed with the high dosages of STZ in the rat model of sporadic Alzheimer's Disease. Data on rats that received varying dosages of STZ ranging from 0, 1 or 2 mg/kg is presented.

Low-level Copper Exposure Causes Cardiac and Neurodevelopmental Defects in the Embryonic Zebrafish. K. Figueroa, J. Pagnotta, I. Mazaharul and A.L. Dell, St. Francis College, Brooklyn, NY.

Neuronal and cardiac birth defects are correlated with pesticide application and water quality. Human exposure to these pollutants may occur through drinking water, or during contact with water in recreational areas, but the molecular pathways that cause these defects are not well defined. GPCR signaling pathways constitute one primary mechanism for cells to respond to their environment. Using the zebrafish retinotectal projection as a model system, we previously showed that GPCR signaling is required for axon guidance during normal development. Retinal ganglion cell (RGC) axons that express a dominant negative G-protein subunit GaS fail to cross the midline and misproject. Our aim was to use this system to dissect the cell signaling events that translate exposure to environmental pollutants into neuronal developmental defects. We measured elevated copper levels at sites along Newtown Creek, one of the most polluted waterways in New York City and designated Superfund Site. We cross checked and augmented our data set with data from the EPA and Newtown Creek Alliance. We then examined the development of zebrafish embryos raised in low concentrations of copper solution. We observed extensive cardiac defects in these embryos as well as neuronal abnormalities. We identified potential gene targets of copper by analyzing GEO datasets, and refined our candidate list by identifying targets expressed in the right time and place to mediate these defects. This work was supported by a St. Francis College Faculty Research Grant. We thank Queens College developmental imaging core for use of the facility and helpful discussions.

Mud Snails (*Ilyanassa obsoleta*) From Woods Hole Massachusetts Do Not Appear to be a Vector for Dermo (*Perkinsus marinus*) Infection of The Eastern Oysters (*Crassostrea virginica*). Christina Florestan, Craig S. Hinkley and Gary Sarinsky, Kingsborough Community College, Brooklyn, NY.

Dermo (*Perkinsus marinus*) is a pathogenic disease that has been found to infect Eastern Oysters (*Crassostrea virginica*) along the Atlantic Coast and is responsible for their high rates of mortality. Dermo affects gill, mantle and digestive epithelium tissue which ultimately interferes with normal functions such as reproduction, respiration and feeding. It is generally thought that Dermo is transmitted from oyster to oyster. However, there is scientific literature that suggests that there may be a possible vector for the transmission of Dermo. Mud Snails (*Ilyanassa obsoleta*) are found in great numbers within the environment of the oysters at Woods Hole, MA. Mud Snails are detritus feeders, eating whatever is found on the top of the mud where they live. It is hypothesized that the mud snails act as a mechanical vector for Dermo in Woods Hole. Tissues were excised from twelve Mud

Snails. DNA was then extracted from the tissue using a DNeasy blood & tissue kit. The polymerase chain reaction (PCR) was used to first amplify the Cytochrome Oxidase 1 (CO1) gene using Folmer's Primer. The amplified samples were then subjected to gel electrophoresis to confirm the correct size (702-bp). Samples were sent for sequencing and subjected to an NCBI blast search. The results verified that the DNA was CO1 from *I. obsoleta*. A second PCR reaction was performed using Dermo specific primers. The amplified materials were then subjected to gel electrophoresis to verify the correct size (304-bp). The results showed that none of the twelve samples or a negative control was amplified but a positive control for Dermo was amplified. We can conclude that the results of this experiment did not support the hypothesis that the mud snails are a vector for Dermo.

Promoter Hypermethylation of SOX9 in Ovarian Cancer Correlates with Platinum-based Chemoresistance. Ryan Frank, Kim Doyle, Maria Samartino and Noelle Cutter, Molloy College, Rockville Centre, NY.

Epithelial ovarian cancer is the most lethal gynecologic malignancy, with approximately 25,000 estimated new cases and over 17,000 deaths in the USA in 2016. Chemoresistance from the commonly prescribed platinum-based drug, carboplatin, is a major contributor to this mortality rate. Increasing evidence has shown that epigenetic alterations, including DNA methylation, play a significant role in cancer, including drug resistance, silencing of tumor suppressors, the activation of oncogenes and the promotion of metastasis. Recent studies have identified genes with CpG islands that undergo epigenetic alterations, particularly methylation, and are associated with the acquisition of chemoresistance in epithelial ovarian cancer. Specifically, SOX9 is a gene with CpG islands that has been shown to be transcriptionally silenced via methylation in resistant epithelial ovarian cancer patients. Therefore, the objective of this study was to determine the potential role methylation has in mediating chemoresistance through cell proliferation and survival through SOX9 suppression (mimicking DNA methylation). Cell survival, senescence, and colony formation assays were performed, along with demethylation treatments and bisulfite conversions. It was determined that methylation induced silencing of SOX9 resulted in altered apoptotic pathways rendering cells more resistant to carboplatin treatment. Furthermore, demethylation of the gene using decitabine reversed this proliferation and survival resistance in cells. Future work is needed to correlate the in-vitro data with in-vivo models to mimic this research and to see if the treatment of epithelial ovarian cancer by DNA demethylating agents could be an alternative adjunct therapeutic strategy for intervention.

Testing the Efficacy of 2 RNA-mediated Interference (RNAi) Strategies in the Nematode *C. elegans*. John Fritsch, Matthew Von Bargen, Austin Marcelo and Samantha Davis, Ramapo College of New Jersey, Mahwah, NJ.

RNA-mediated interference (RNAi) is a process by which double stranded RNA (dsRNA) molecules inhibit gene expression via specific degradation of mRNA transcripts. Since its discovery nearly 20 years ago, researchers have utilized this process to specifically knock-down genes of interest. Several techniques have been developed to induce RNAi in the nematode *C. elegans*. One technique involves injecting dsRNA directly into the *C. elegans*. Another technique requires feeding *C. elegans* with bacteria expressing dsRNA. This study assesses the effectiveness in knocking-down HCP-3, a CENP-A homolog critical to centromeric identity and thus chromosome segregation. Knock-down effectiveness using both techniques will be measured via western blot, immunofluorescent staining, and phenotypic consequences assessed via live imaging and an embryonic lethality assay. Currently, the RNAi feeding protocol has been implemented and the efficacy of the RNAi feeding has been confirmed in all measurable assays. Currently, the dsRNA is synthesized and an effective injection protocol has been established.

A Comparison of the Effectiveness of Alternate Substances to Control the Growth of Bacteria Associated with Wound Biofilm Formation. Monique R.A. Funkhee, Mary T. Ortiz and Loretta Brancaccio-Taras, Kingsborough Community College, Brooklyn, NY.

Biofilms are surface microbial communities enclosed in an extracellular polymeric matrix and are associated with persistent wound infections. Microorganisms associated with biofilms are more resistant to antimicrobial agents because they are protected by the matrix, thus making conventional chemotherapies difficult. With the increasing number of multi-drug resistant bacteria, non-traditional agents with potential anti-biofilm properties that are less expensive, more biocompatible and with less side-effects are being sought as alternatives. The effectiveness of alternative substances was tested using a standard agar diffusion assay to determine if the growth of bacteria associated with wound biofilms could be inhibited. The bacteria tested, *Enterobacter cloacae*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Streptococcus pyogenes* were exposed to three traditional agents: bactine, betadine and hibiclens, as well as six alternative substances: red thyme oil, cinnamon/clove mouthwash, peppermint oil, lavender oil, clove bud oil, and cinnamon. The hypothesis was the alternative substances would be at least as effective as traditional agents to control the growth of bacteria associated with wound biofilms. Zones of inhibition were measured (mm) and averaged (mean \pm SEM). Data were statistically

analyzed using the Mann-Whitney U test ($p=0.05$, two-tailed), comparing the traditional agents with the alternative agents against all five bacteria. The results show the hypothesis was supported for red thyme oil and rejected for cinnamon. Red thyme oil was the most effective and cinnamon was the least effective alternative agent. Future work will include testing traditional and nontraditional agents with the greatest antimicrobial effectiveness and determining the mechanisms by which nontraditional agents work.

Synthesis of Mini-Reporter Construct to Test Gene Transfer of RNA Therapeutics. Kerianne Fuoco, Koushik Muralidharan, Arbaz Khan, Hemangi Patel and Martin J. Hicks, Monmouth University, West Long Branch, NJ.

Glioblastoma multiforme (GBM), a grade IV tumor of the central nervous system, is the most common malignant primary brain tumor in adults. Individuals diagnosed with GBM have a poor life expectancy of approximately 12 months. The poor survival rate is due to a lack of efficacy in current therapies, including chemotherapy and radiation, which are limited by the blood-brain barrier. We are creating novel strategies to bypass these barriers by developing gene transfer vectors to deliver the genetic sequences of RNA therapy molecules to alter the splicing pattern and expression of tyrosine kinase receptors (TKR), creating soluble TKR decoys. In this approach, we expect to modify GBM and CNS cells to deliver the therapeutic anti-cancer molecule into the local milieu. To test this approach, we are creating an *in vivo* tissue culture model. We have designed mini-reporter gene constructs that contain the targeted regulatory elements, including the 5' and 3' splice sites as well as the intronic region of interest of the TKR, vascular endothelial growth factor receptor 2, VEGFR2 (KDR). This mini-reporter construct will test the efficacy of RNA anti-sense therapeutics to block the pre-mRNA splicing event leading to intron retention and alternative polyadenylation signal recognition. Using fluorophores as visual markers, eukaryotic green fluorescent protein (eGFP) will be used to detect the natural exon splicing product, whereas the monomeric red fluorescent protein, mCherry will detect RNA anti-sense mediated intron retention. In this manner, the mini-reporter construct provides a quick and visually measurable test to optimize RNA anti-sense therapies against VEGFR2. In addition to VEGFR2, we will use this mini-reporter design to test the efficacy of RNA therapies directed toward alternative TKRs known to be upregulated in cancer. Acknowledgements: This research was funded by Bristol-Meyers Squibb, Johnson and Johnson.

Examining the Effect of Oxidative Stress on *Tetrahymena thermophila* Sirtuins Exposed to Copper. Edrouine Gabriel and Ralph Alcendor, New York City College of Technology, Brooklyn, NY.

Tetrahymena thermophila, a ciliated protozoan, has been a reliable research model for many years. Studies on *T. thermophila* have made significant contributions to biology such as the discovery of ribozymes and telomere function. *T. thermophila* are ideal research models because of the minimum facility required for growth, maintenance and manipulations. Additionally, the complete *T. thermophila* genome has been sequenced, making it an appropriate model for bioinformatics studies. Overall, the volume of research conducted with these organisms is extensive. However, in the area of stress resistance and aging, *T. thermophila*'s biology has not been exploited. These cells have proficient anti-stress mechanisms. They can survive in sublethal temperature, in extremely low oxygen environment, and starved or poor nutrient conditions. If the condition is conducive, *T. thermophila* can divide indefinitely without showing classic signs of aging such as senescence. The aim of this project is to investigate the role of sirtuins in *T. thermophila* ability to survive high oxidative stress (OS) conditions induced by copper. Cells were exposed to varying amount of copper and incubated for 24 h – 48 h. Cell death analysis showed increase cell death with increasing amount of copper. Only 40% of cells survived after 24 hours in the presence of 1.0 mM of copper. mRNA expression of sirtuins decreased in the presence of 0.1 – 0.5 mM of copper but increased in the presence of 0.8 – 1.0 mM of copper. Similar pattern was seen in the mRNA expression levels of antioxidant genes. These results suggest that sirtuins may be involved in OS induced by high rather than low amount of copper.

New Perspective in the Treatment of Lung Cancer. David Donatucci, Michael Galarraga, Alix Duarte, Christina Faltas, Jan Osea and Natalia Coleman, New Jersey City University, Jersey City, NJ.

It is estimated that over 200,000 new cases of lung cancer will be detected in the US in 2016. Despite intensive research efforts and promising discoveries, approximately 150,000 of lung cancer patients will die in 2016. Therefore, there is an increasing necessity for a new perspective in the treatment of cancer. There is growing evidence of the importance of glutamate signal transduction in cancer. N-methyl-D-aspartic (NMDA) receptors are one of the three glutamate receptors found in the mammalian central nervous system. While it is common knowledge that NMDA receptors are essential for spatial learning and memory, little is known about its function in cancer. We previously showed that human lung cancer cells express NMDA receptors and that NMDA receptors show an anti-proliferative effect. The aim of the current study is to evaluate the NMDA receptor antagonist memantine's ability to reduce Ras-GTP level in human lung cancer cells.

The Role of CHD4 on Histone Recognition. Juan Garcia¹, Yang Gao² and Zhibin Wang², ¹Caldwell University, Caldwell, NJ and ²Johns Hopkins Bloomberg School of Public Health, Baltimore, MD.

The research done deals with CHD4 (Chromodomain Helicase DNA Binding Protein 4), a protein that is part of the NuRD (Nucleosome Remodeling Deacetylase) Complex, a Complex that regulates gene transcription. MEF (Mouse Embryonic Fibroblasts) cells were tested to find how the NuRD Complex interacts and regulates gene transcription. The assay that was used to test this was a ChIP assay (Chromatin Immunoprecipitation assay) and a Western blot, this experiments will show how big the protein CHD4 is and where it attaches in the DNA when it regulates gene transcription. The results from the experiments showed that CHD4 can successfully recognize histone3. Although, further investigation is needed to reveal the mechanism under the current discovery.

Tumor Specific Roles and Interactors of RNA Binding Protein RNPC1a. Kelly Gazca and Emral Devany. Kingsborough Community College, CUNY, Brooklyn, New York.

It is well under investigation that different cancer types have different molecular mechanisms and the treatments should be directed to specific targets in order to ensure efficient and fast diagnosis, treatment and recovery. This project focuses on mRNA binding protein RNPC1a. Interestingly, RNPC1a acts as a tumor suppressor in high invasive breast cancer and as an oncogene in most other cancer types such as low-invasive breast cancer, colon cancer, leukemia, prostate and ovarian cancer. RNPC1a has shown to regulate key proteins such as p53, p63, p73, p21 and HuR. We investigated the mechanism and effects of RNPC1a on mRNA processing in different cancer types (cervical cancer, colon carcinoma, low-invasive and high-invasive breast cancer cell lines were used in the study) under different cellular conditions (normal conditions and UV-induced DNA damage). We either treated the cells with UV irradiation or left them untreated. Total mRNAs were extracted and used for qRT-PCR to detect levels of RNPC1a mRNA and target mRNAs. Total proteins were extracted and used in co-immunoprecipitation assays to detect cancer type specific interactions. Here we show that both RNPC1a mRNA and target mRNA levels are different in different cancer cells. In addition, one of the known interactions of RNPC1a (HuR) only interacts with RNPC1a in colon cancer cell line but not the other cell lines used in this study. These results suggest that RNPC1a itself might be regulated in a tumor-specific manner and/or RNPC1a protein may have different protein interactions in different cancer types. Further studies will contribute to reveal the differences in the mechanism, hence the diagnosis and treatment, of tumor subtypes.

A New Approach to Drug Discovery for the Treatment of Cryptosporidiosis. Mohini Gobin¹, Mary Morada¹, Wesley Van Voorhis², Samuel Arnold² and Nigel Yarlett¹, ¹Pace University, New York, NY and ²University of Washington, Seattle, WA.

Cryptosporidiosis is a potentially lethal diarrheal disease of immunocompromised individuals and young children. In a recent study it was shown to be the third leading cause of death in children from economically low resources countries. Currently only Nitazoxanide is approved by the FDA to treat Cryptosporidiosis, however it is ineffective against the most at risk group of the population. By using new technologies such as hollow fiber technology we are able to imitate the intestine and maintain a long term culture of the parasite *Cryptosporidium parvum* which allows various compounds, such as bumped kinase inhibitors, to be tested over a long period of time. *C. parvum* contains a group of calcium-dependent protein kinases (CDPKs) which are found in plants and ciliates but not in humans or fungi. Here, we describe a series of potent inhibitors that target CDPK1 in *C. parvum* over the mammalian kinases. Furthermore they are able to block *C. parvum* invasion of HCT-8 host cells. The samples are analyzed using qRT-PCR to determine the effectiveness of the applied compound in eliminating the parasite.

Testing Cell Free Extracts of Known and Unknown Bacteria for Inhibition of Biofilm Formation of Pathogenic Bacteria. Erick Gonzalez, Carol Ellameh and Meriem Bendaoud, New Jersey City University, Jersey City, NJ.

Microorganisms can develop biofilms after attaching to surfaces. These biofilm forming cells generate an extracellular polymeric substance matrix. Once the biofilm structure has been established, it provides an environment for microorganisms to exchange genetic material between cells and enable them to become resistant to our immune system and antibiotic treatment. Biofilms play an important role in public health. Most infectious diseases and device related infections, such as catheters at hospitals are caused by biofilm forming pathogenic bacteria. The focus of our research is the Identification and characterization of new anti-biofilm substances. Cell free extracts of known bacteria such as *Bacillus subtilis*, *Lactobacillus casei*, *Escherichia coli* and *Enterococcus faecalis* were tested against pathogenic biofilm forming bacteria. Furthermore, some Unknown bacteria sampled from the Hudson River derived from the Jersey City and Bayonne area were also tested and show strong antibiofilm properties against biofilm forming pathogenic bacteria including *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Erwinia carotovora*, and *Pseudomonas aeruginosa*. Further characterization will be conducted to identify the unknown bacteria and the secreted compounds.

CD105 Deficiency in Mouse Aorta-derived Mesenchymal Stem Cells Promotes An Enhanced Inflammatory Response to Lipopolysaccharide. Joseph Granata, Hugo Sanchez, Philip Loeschinger, Anthony Goetz and Jodi F. Evans, Molloy College, Rockville Centre, Rockville Centre, NY.

Mesenchymal stem cells (MSCs) are being widely studied for their ability to regulate macrophage cell responses. Previous works have demonstrated that mouse aorta-derived MSC (mAo-MSC) support the macrophage inflammatory response. mAo-MSC have been characterized phenotypically for MSC-associated surface antigens and express CD90 and CD105 but do not express CD73. CD105, also known as endoglin, is a co-receptor in the TGF β superfamily of receptors. Mouse adipose-derived MSC lacking CD105 have an increased capacity to regulate T-cells by reducing their proliferation while elevated CD105 expression is consistently associated with inflammatory disease. Therefore, we hypothesized that suppression of CD105 in mAo-MSC will reduce the immunosupportive capacity of the mAo-MSC. We used siRNA to reduce expression of CD105 in mAo-MSC and subsequently examined the effect of this deficiency on their response to lipopolysaccharide (LPS) and their ability to support the macrophage inflammatory response. Contrary to our hypothesis, CD105 deficient mAo-MSC cultured alone and in co-culture with macrophage secreted increased levels of the inflammatory indicators nitric oxide (NO) and interleukin 6 (IL-6) after exposure to LPS. The increase in NO and IL-6 observed in the co-cultures is additive and therefore points to the mAo-MSC as the primary origin. Overall our data suggest that CD105 acts as a regulator of the TLR-4 pathway and may represent an important target for modification of MSC to be used in therapeutics.

Recent Fluctuation of Atlantic Horseshoe Crab (*Limulus polyphemus*) Egg Density on Plumb Beach, Brooklyn, NY. Kelvin Gutierrez and Christina Colon, Kingsborough Community College, Brooklyn NY.

Plumb Beach is a favoured location for horseshoe crabs to spawn during May through July. It was hypothesized that there will be an increase in egg density on both the eastern and western sides in 2016, compared to previous years when there was a downward trend likely exacerbated by Super storm Sandy. Estimating horseshoe crab eggs is done to monitor spawning activity and assess the amount of forage available to migratory birds (Pooler et Al. 2003). Horseshoe crabs have been around for 450 million years. Their blood makes them essential to the biomedical industry and their eggs are important food for migratory shore birds and fish. To monitor egg density, grids were created at the intertidal zone wherein samples were collected randomly, at 5cm and 20cm deep, every two weeks from May to August. Eggs were brought to the lab to be sorted then counted. Data showed a decrease of 12,692 compared to 2015 (41,834), however, 2016 counts (29,142) were higher than 2012 – 2014. Also, the high

survival (17% of hatchlings) was the highest since 2011, thus my hypothesis was in part supported. Furthermore, on the western beach (restored in 2012) there was an increase in density (2,029) and much higher survival than previously observed (33%). The increased density and survival on the restored side supports my hypothesis, and is an indication that the beach restoration was successful. The high survival on the eastern beach is a sign that the habitat is healthy, while reduced egg counts may be due in part to the migration of crabs to the restored western beach; however only time will tell. This work was supported by grant 2R25GM06003 of the Bridges Program of NIGMS and grant 0537171091 of the CSTEP Program of the NYS Department of Education.

Comparison of N-methyl-d-aspartate NR2B Subunit Proportions Between Control, Gonadectomized, and Hormone Replaced Male Rats. Catherine Hernandez¹ and Mary F. Kritzer², ¹Nassau Community College and ²Stony Brook University, NY.

The prefrontal cortex (PFC) mediates cognitive functions that are dependent on dopamine (DA) input coming from the ventral tegmental area (VTA). PFC regulates how much DA it receives. Previous studies show that androgens regulate PFC/DA networks and DA sensitive PFC behaviors by modulating glutamate drive over PFC-to-VTA projections. My study asked whether androgens exert this control by influencing the subunit composition of glutamate receptors on PFC neurons that project to VTA. Specifically I examined the effects of gonadectomy and hormone replacement on NR2B NMDA receptor subunit. In situ hybridization was used to visualize NR2B subunit mRNA. This was combined with retrograde fluorescent labeling of PFC-to-VTA projections neurons. I quantified the proportions of these neurons that contained NR2B mRNA. Results indicated that NR2B subunit is not necessarily hormone regulated in the adult brain. The proportions of the remaining subunits should be quantified to determine the possible influence androgen may have on the subunit composition of NMDA receptor and DA-dependent PFC behaviors.

Regulation of Gene Expression by HRE's Under Chemically Induced Hypoxic Mimic Condition. Alberto Herrera, George Coricor and Jane L Ko. Seton Hall University, South Orange, NJ.

Hypoxia is a condition of inadequate oxygen supply, which can induce cell death. Neuronal cells treated with DFO resulted in hypoxic mimic conditions. Our lab reported previously that the outcome of such a treatment did result in the decrease of cell viability. However, there were still surviving neurons. The surviving cells did not exhibit significant morphological changes under confocal microscopy analysis using annexin-V-FLUOS and propidium iodide staining, indicating that they were not at apoptotic or necrotic stages. These surviving neurons, therefore, developed adaptive responses under hypoxic challenge. Several changes were observed, including an

increase of cellular glutathione levels and the increase of HIF-1 α mRNA levels, a known marker of hypoxia, in surviving cells when compared to control. This is a known mechanism to cope with hypoxia. The increase of kappa opioid receptor expression was also observed in surviving neurons when compared to the control. Clinically, opioids are used to modulate pain sensation, which can result from events such as stroke, trauma and cardiac arrest that lead to hypoxic conditions. There are 4 putative HIF-1 responsive elements found in the 5' upstream of this gene. Two of the four elements showed a significant increase of the reporter activity. To determine if these two elements displayed an interactive effect under hypoxic challenge, these two responsive elements were simultaneously cloned into a reporter plasmid containing the luciferase gene, and resulting plasmids were tested using NMB neuronal cells. Results showed a significant increase of promoter activity under hypoxic challenge. Mutation analysis further confirmed the important role of these elements. Taken together, this study showed the functional roles of two response elements mediating the receptor expression under hypoxic condition.

Antimicrobial Characteristics of Streptomyces. Catherine Immordino, Brenda Torres and Mangala Tawde, Queensborough Community College, Bayside, NY.

Since bacteria co-exist in natural environments along with other microorganisms; interspecies interactions is a major mechanism for many of the unique characteristics that bacteria may possess. Induction of secondary metabolite production as well as other antimicrobial characteristics in Streptomyces bacteria can be studied by growing them in presence of other bacteria. We are interested in studying the mechanisms of antimicrobial characteristics of Streptomyces. Here we show some antimicrobial interactions of Streptomyces against each other as well as other Gram +ve and Gram -ve bacteria. These interactions amongst Streptomyces as well as those with non-Streptomyces indicate secondary metabolites with antimicrobial characteristics. We identified that many of the Streptomyces strains also exhibit resistant to some of the known antibiotics.

Functional Characterization of the Actin-Binding Domain of the Drosophila Protein Dunc115. Nataly Jara¹, Safaa Hassan², Saad Alghamdy³, Roberto Gomez³, Rabina Lakha³, Chris Roblodowski¹ and Qi He³, ¹Queensborough Community College, Bayside, NY, ²New York City College of Technology Brooklyn, NY and ³Brooklyn College, Brooklyn, NY.

Our prior analyses have uncovered the Drosophila gene Dunc-115 as a critical regulator for axonal pathfinding in both the visual system and the ventral nerve cord. Furthermore, our original sequence data reveal that there are three isoforms for Dunc-115: L, M and S for long, medium and short forms, respectively, though the

functional differences among the three isoforms are unclear. The actin-binding results show that only the Dunc-115L is capable of binding to actin and the actin-binding domain in the L form is thus identified. Here we report the results of the mutant analyses from the deletion of the actin-binding domain in Dunc-115L. We have adapted the newly available mutagenesis technique CRISPR-cas9 by using specific oligonucleotide primers to create mutations where only the actin-binding domain of Dunc-115L is eliminated while other isoforms remain intact. The mutants show the phenotype of homozygous lethal, and up to one third of the heterozygous animals have leg defects resembling Distal-less (Dll), implying a possible functional interaction between Dll and Dunc-115. Given the important functions of Dll in the development of limbs and the brain, such a functional interaction between Dll and Dunc-115 suggests a possible functional mode for Dll. Nataly Jara is a participant in the NIH Bridges to the Baccalaureate Program at Queensborough Community College.

The Effect of Carbon Nanoparticles From Supermarket Bread on the Nematode *C. Elegans*. Christine Johnson and Tetyana Delaney, St. Joseph's College, Brooklyn, NY.

Nanoparticles are an area of intense research since it was first discovered millennia ago. Due to nanoparticles being so small, it allows for a faster diffusion time, which could be vital for cancer research and cell therapy. The small size factor, however, can also be a negative quality because it could enter into the cells and could damage DNA as well as alter DNA sequences causing drastic effects such as mutations and genetic disorders. Therefore, it is essential to take into account the toxic effects as well as the beneficial effects. In this experiment, the goal is to see if there is a way to use nanoparticles for biomedical purposes, but to use a nanoparticle that is already known to have no toxic effects. An experiment will be conducted to see how carbon nanoparticles found in food items, particularly supermarket bread, could have an effect on organisms, such as the soil nematode, *Caenorhabditis elegans*. This experiment is based off of a study done to test the presence of carbon nanoparticles in food products and they found that the largest amount of carbon nanoparticles released was found in the crust of supermarket bread as it was heated. Using this study as a backdrop to this experiment, the objective is to observe the effects that carbon nanoparticles from the crust of supermarket bread will have on the soil nematode, *C. elegans*. The factors that would be evaluated include growth, survival, reproduction, movement, and mitochondrial and oxidative stress. The primary goal is to see how the carbon nanoparticles would affect the *C. elegans* and the secondary goal of the experiment is to see that if with decreasing nanoparticle size by heating the bread, if it would increase toxicity levels. Any results from this experiment would further our understanding of naturally derived nanoparticles.

Assessment of Vertebrate and Microbial Diversity in Local Ecosystems Using Environmental DNA (eDNA). Arbaz Khan and Martin J. Hicks, Monmouth University, West Long Branch, NJ.

Oceans and waterways are vast with life. The global preservation of biodiversity in marine environments begins at the local level. The challenge is that life underwater is not easily observable and traditional methods to survey the ecosystems can be disruptive, invasive and time consuming. Advances in DNA sequencing, specifically, next generation sequencing and the tools of genetics and bioinformatics have made it possible to more easily and non-invasively detect the distribution and abundance of the vertebrates and the microbial world that makes up these aquatic systems. We have begun preliminary experiments collecting eDNA water samples from local environments, Lake Takanassee and the waterway that connects to the adjacent ocean environment of Long Branch, NJ. Water samples (1L) were collected in January and June-July of 2016, eDNA was filtered onto nylon membranes, using PowerWater eDNA purification kit, we isolated eDNA from all samples. To detect, identify and differentiate between species, primers with Illumina tag adapters for the 12s ribosomal subunit were added to the eDNA samples via polymerase chain reaction (PCR). After gel electrophoresis verification of amplicon sizes, Illumina tags were added with a second PCR in preparation for Next-Generation sequencing. The sequences were then characterized using the National Center for Biotechnology Information Basic Local Alignment Search Tool (BLAST), which identifies and compares nucleotide sequences to existing sequenced genomes of various species. With a threshold of $\geq 99\%$ similarity, we were able to identify the eDNA of Common Carp, Atlantic Menhaden, American Eel, Harbor Porpoise, Canada Goose, and many other species, including land mammals. This research is supported by the Urban Coast Institute, Monmouth University.

A Comparison of Survival Rates of *Limulus polyphemus* Eggs from Different Regions of Plumb Beach, NY, Both in the Field and in the Lab. Adnan Khoja and Christina P. Colon, Kingsborough Community College, Brooklyn, NY.

Limulus polyphemus, the Atlantic horseshoe crab, is an ancient chelicerate indispensable to humans as well as migratory birds that feed on their eggs. In this experiment, we set out to determine if there was a significant difference in egg viability between a disturbed and an undisturbed section of Plumb Beach, a vulnerable spawning beach in New York. The west side, divided into NYC1, NYC2, and NYC3 is considered more disturbed as it is adjacent to a highway, suffers heavy erosion and most human activity is focused there. The east side, divided into BW, BE, and TC was considered the more natural region. One hundred eggs were retrieved from each site and kept in Petri dishes and their survival rates monitored, while simultaneously doing population surveys on the beach. Because NYC2 had no eggs throughout the experiment, there is no data for this site. Petri dish survival rates were: NYC1: 97%,

NYC3: 93%, BW: 10%, BE: 25%, and TC: 5%. Beach survival, when calculated as the number of live embryos plus live and crushed (from handling) trilobites, was: NYC3 at 84.65%, NYC1 39.87%, BW 17.71%, BE 21.32%, TC 24.80%. When compared to dish survival, this gave a correlation coefficient of 0.817, which approached 0.05 significance (0.878). In all cases, the western beach had greater survival than the eastern side. This was unexpected, because previous research showed that horseshoe crabs prefer the eastern side for spawning. While the western beach appears to be more viable for egg survival, it is important to note that the survival and number of eggs on the eastern beach are still relatively high. Further studies are required to validate these results and determine what factors may cause this differential survival. This work is supported by grant 0537171091 of the CSTEP Program of the NYS Department of Education.

Investigating Dephosphorylation of EGFR by PTP1B. Youngjoo Kim, Leidy Gomez and Aileen Espinal, SUNY at Old Westbury, Old Westbury, NY.

Dephosphorylation is an important covalent modification that plays a key role in the regulatory processes of the cell. The balance between phosphorylation and dephosphorylation is critical in maintaining cellular functions. Protein tyrosine phosphatase 1B (PTP1B) is a member of protein tyrosine phosphatase superfamily and it is able to specifically dephosphorylate phosphorylated tyrosine residues. Past research has linked defective PTP1B to diseases like obesity, diabetes and certain cancers. In addition, studies suggest that PTP1B is able to dephosphorylate epidermal growth factor receptor (EGFR) which plays an integral role in a variety of biochemically mediated cell signals. EGFR belongs to a receptor tyrosine kinase family of enzymes and is an important therapeutic target for various forms of cancers, like non-small lung cancer and breast cancer, specifically. Normal EGFR signaling involves dimerization of EGFR upon EGF binding to the extracellular domain, which causes auto phosphorylation of tyrosine residues in the cytoplasmic domain of EGFR. Phosphorylated tyrosine residues provide docking sites for downstream signaling proteins for initiation of signaling events. Mutation or overexpression of EGFR leads to constitutively active EGFR and affect its downstream signaling, which play central roles in the development of various forms of cancers. The purpose of this research is to further understand the mechanism of action of PTP1B in dephosphorylating EGFR. Our previous study indicated that there is an ordered autophosphorylation in EGFR. In this study we hypothesize that rates of dephosphorylation at each tyrosine residue are different and that they contribute to downstream signaling. We performed a western blot analysis using phospho-specific antibodies. We examined dephosphorylation of EGFR at three different tyrosine residues (992, 1068 and 1173) and observed a different rate of dephosphorylation at each residue. Our study suggests that PTP1B might regulate EGFR signaling pathways by differential inhibition at each tyrosine residue.

The Role of DAck in Appetitive Associative Olfactory Learning in Drosophila Larvae. Morgan K King, Long Island University CW Post, Brookville, NY.

Drosophila Activated Cdc42 kinases (DAck) has been identified as a homolog to human Ack1. It has been found to share the greatest amount of sequence identity in all of its conserved domains. DAck has been found to play an important role in the development of Drosophila melanogaster. In Drosophila, DAck is required for dorsal closure in embryos and spermatogenesis and oogenesis in adults. It has also been reported that Ack1 is highly expressed in the brain in mice, specifically in the hippocampus, and is important for brain development and synaptic function and plasticity. Due to its importance in development and its expression in the brain, we were interested in determining whether DAck plays a role in learning and memory in Drosophila by utilizing a Pavlovian-type learning assay in fruit fly larvae. To test this, we performed an odor-taste learning assay in wild type and DAck86 mutant Drosophila that were homozygous for a loss-of-function allele of DAck to see if they had the ability to form an association between odors and appetitive gustatory reinforcement such as sugar. Wild type and DAck86 third instar larvae were first trained by being placed on an agarose petri dish that contained fructose and were exposed to the first odor of either n-amyl-acetate (AM) or 1-octanol (OCT) and then again in a second petri dish that contained the reciprocal second odor without the sugar reward. Once trained, larvae were tested to determine their preferences and a performance index corresponding to the number of larvae that went to the appropriate odor after training. We found that DAck86 mutant larvae had a significantly lower performance index as compared to wild type larvae ($p < 0.05$), suggesting a learning defect. More work is being done to further investigate the role of DAck in learning and memory.

Prostate Cancer Cells Migration is Regulated by Tyrosine Phosphorylation of Hexim1, Implications in Tumor Metastasis. Karan Kumar and Manya Mascareno, SUNY College at Old Westbury, Old Westbury, NY.

Hexim1, an inhibitor of RNA polymerase II-mediated transcriptional elongation, is a checkpoint for cell proliferation. We studied whether inactivation of Hexim1 via post-translational modification (tyrosine phosphorylation) contribute to de-regulated cell proliferation of prostate cancer cells. It was observed that Hexim1 was a novel target for Jak2 tyrosine kinase. The identification of conserved YXXL motifs on Hexim1 lead to the findings that these motifs were a target for Jak2 kinase activity. To further investigate the significance of the Jak2/Hexim1 axis' role during cell proliferation and cell migration, we incorporated substitution mutations at the tyrosine residues Y168 and Y291 of Hexim1. Substitution mutations myc Y168G Hexim1, and myc-Y168G/Y291E Hexim1 were used to generate stably expressing TRAMP-C2 cell lines. A Wound- Healing assay was conducted using TRAMP-C2 wildtype Hexim1 as well as the TRAMP C2 carrying substitution mutations myc Y168G Hexim1, and myc-Y168G/Y291E Hexim1 clones. A decreased

migration/invasion phenotype was observed in the myc-Y168G Hexim1 and myc-Y168G/Y291E Hexim1 cell lines as compared with the TRAMP-C2 carrying wild type Hexim1. Similarly, the inhibitory role of myc-Y168G Hexim1 and myc Y168G/Y291E during cell migration was significant as compared to TRAMP-C2 wild type Hexim1. Analysis of pY705-Stat3, a downstream target of Jak2 kinase and marker of poor prognosis in prostate cancer, revealed that Hexim1 overexpression inhibited Jak2-mediated Stat3 phosphorylation at pY705- Stat3 during wound healing; however, this inhibitory mechanism was dependent in part on the intact YXXL motifs in Hexim1, as myc-Y168G Hexim1 and myc-Y168G/Y291E were only able to partially facilitate tyrosine phosphorylation of Stat3. A screening of a human array of prostate cancer, using an antibody specific for pY291 Hexim1, revealed phosphorylation of Hexim1 at stages II, III, and IV on the Gleason Scale, suggesting that tyrosine phosphorylation of Hexim1 may inactivate Hexim1 and facilitate cell proliferation as well as cell migration.

Comparison of Debris Accumulation Near the Shore of Kingsborough Community College Before and After Hurricane Sandy. Maria Matevosyan, Mary T. Ortiz and Craig S. Hinkley, Kingsborough Community College, Brooklyn, NY.

In 2006, Kingsborough Community College (KCC) began to participate in the New York City (NYC) Department of Environmental Protection Open Water Project to track the condition of the shores of NYC with respect to debris pollution. On October 29, 2012, NYC experienced hurricane Sandy, a category 1 hurricane. In this study we examined the effects of Sandy on debris accumulation along the shores of KCC. Our hypothesis is that debris accumulation will be less on shores near KCC after Sandy than before. To test our hypothesis, we used data that were collected 2008-2016 at KCC for the Open Water Project and made a comparison of debris accumulation prior to and after Sandy. Data were collected on the eastern side of the KCC campus from observations of three areas—offshore, near shore, and the shoreline. Relative debris accumulation was scored using a number scale from one (very good) to five (poor). We first calculated a mean debris rating (MDR) for each year within each of the three areas. Comparisons of the MDRs using a Mann-Whitney U-test indicated there was a statistically significant difference ($p < 0.05$) between both offshore and shoreline and near shore and shoreline, but no significant difference between offshore and near shore. We then used the individual relative debris accumulation scores to compare debris in each area before (2008-2012) and after (2013-2016) Sandy. A Mann-Whitney U-test with z-scores showed that there was no significant difference ($p < 0.05$) between offshore accumulation ($z = -0.01383$, $p = 0.99202$) or near shore accumulation ($z = 0.78597$, $p = 0.42952$) before and after Sandy. However, there was a significant difference ($p < 0.05$) in shoreline accumulation before and after Sandy ($z = 7.7998$, $p < 0.0001$). Together, these results do not support our hypothesis. This work was supported by grant 0537-17-1091 of the CSTEP Program of NYSED.

Set3C Stimulates NuA4 Interaction with Nucleosomes and Occupancy Transcribed Genes. Angelo Messina and Daniel S. Ginsburg, LIU Post, Greenvale, NY.

Gene expression in eukaryotic cells is regulated in large part by chromatin. For efficient transcription to take place, chromatin must be disassembled. A critical step in chromatin disassembly is histone acetylation, which is regulated by the opposing activities of lysine acetyltransferase (KAT) and histone deacetylase (HDA) complexes. While acetylation is important for transcription, too much acetylation can lead to the production of aberrant transcripts. The NuA4 KAT has been shown to preferentially bind nucleosomes methylated on histone H3 lysines 4 and 36 (H3K4/36me). Nucleosome binding of the Rpd3C(S) and SET3C HDACs is stimulated by H3K36me and H3K4me respectively. Because they have been shown to bind the same histone methylation marks, we wanted to test the hypothesis that one mechanism for the regulation of nucleosome acetylation is through competition between NuA4 and the Rpd3C(S) and SET3C HDACs for nucleosome binding. We analyzed phenotypes, H4 acetylation, NuA4-nucleosome interactions, and NuA4 occupancy at transcribed genes in mutations that affected Rpd3C(S) and SET3C complex integrity (*rco1Δ* and *set3Δ* respectively), SET3C deacetylase activity (*hos2Δ*), and H3K4 methylation (*set1Δ*). We expected that loss of Rpd3C(S) or SET3C chromatin binding, but not deacetylase activity would lead to increased NuA4-nucleosome interactions, more NuA4 occupancy at transcribed genes, and elevated H4 acetylation. Surprisingly, our results suggested that Rpd3C(S) and SET3C stimulate NuA4 interaction with nucleosomes and occupancy at transcribed genes. Loss of *SET1* did not suppress the phenotypes or significantly reduce the elevated H4 acetylation in *set3Δ* cells. NuA4-nucleosome interactions as measured by coimmunoprecipitation were decreased in *set3Δ* cells and to a greater degree in *rco1Δset3Δ* cells. We observed reduced NuA4 occupancy at the *GAL1* ORF in *rco1Δset3Δ* cells with no loss of transcription. Together, these results support a model in which both Rpd3C(S) and SET3C stimulate NuA4 interaction with nucleosomes and recruitment to transcribed genes.

Assessing the Role of Adhesion-GPCRs in Visual System Axon Guidance. Kelly Michalak¹, Joseph Pagnotta¹ and Alison L. Dell^{1,2}, ¹St. Francis College, Brooklyn, NY and ²University of Pennsylvania, Philadelphia, PA.

How do axons form the correct connections during development? Using the zebrafish retinotectal projection as a model system, we previously showed that G-protein coupled receptor (GPCR) signaling is required for axon guidance during normal development. However which GPCRs and ligands contribute to this process is still unknown. We hypothesize that adhesion GPCRs (ADGRs) direct growing axons. To test our hypothesis, we first looked for candidate receptors. We assessed the

expression pattern of ADGR family members by analysis of GEO datasets and the literature, and designed probes for in situ hybridization. We focused on CELSR3 and Latrophilin 1b as our first candidates, due to their expression patterns. CELSR3 is a member of the adhesion-GPCR family with a characteristic a long n-terminal tail decorated with adhesion motifs including EGF-, leucine-rich, and cadherin repeats as well as Ig domains. In the zebrafish retina, CELSR3 is expressed in amacrine cells as well as RGCs. CELSR3 mutants fail to carry out a visually evoked behavior (OKR) but have not been analyzed for guidance defects. We find that CELSR3^{-/-} retinal axons can cross the midline but are defasciculated (4/30 projections p=0.03). This result suggests that CELSR3 may primarily function as an adhesion molecule to maintain axonal bundling during neuronal tract formation. We have extended our analysis to single axon labeling to assess intra-retinal axon guidance as well as topographic mapping in the tectum.

Seals and the City: Pinnipeds are Returning to Urban Waterways. Kelly Michalak^{1,2}, Afia Azaah^{1,2}, Kevin L. Woo^{2,3} and Kristy L. Biolsi^{1,2}, ¹St. Francis College, Brooklyn, NY, ²Center for the Study of Pinniped Ecology & Cognition, Brooklyn, NY and ³SUNY Empire State College, New York, NY.

Urbanization has drastically changed the way in which species interact with their environment. Some generalist species thrive in highly modified habitats, whereas specialists may decline to the point of local extinction. One such group of animals, Phocidae, had once commonly inhabited the waterways of New York City; however, due to a number of potential interacting anthropogenic factors, their presence in this urban habitat was eliminated over a hundred years ago. Recently, the general public began to report sightings of individual seals at various haul out locations along the NYC foreshore. Given these reports, and our own preliminary questionnaire surveys, we conducted naturalistic observations of primarily harbor seals (*Phoca vitulina*) and grey seals (*Halichoerus grypus*) between the wintering field seasons from 2011-2016. Here, we conducted both land-based and boat-based observations at Orchard Beach, and Hoffman and Swinburne Islands. Our results show that seal populations are fairly stable on an annual basis, with a slight increase in the total number of individuals between each field season. The stability in annual populations is encouraging, as it indicates that pinnipeds are indeed returning to previously used locations that are within their natural seasonal range. Moreover, the return of large megafauna to urbanized waterways suggests a likely positive change in ecosystem health – one that is equally favorable for supporting other species. This research was funded by awards to the Center for the Study of Pinniped Ecology & Cognition (C-SPEC), Psychology Department of St. Francis College, SFC Faculty Research Grants, and Faculty Development Awards from SUNY Empire State College.

The Atlantic Oyster Drill (*Urosalpinx cinerea*) Does Not Appear to be a Vector for the Transmission of the Pathogenic Protozoan Dermo (*Perkinsus marinus*) to the Eastern Oyster (*Crassostrea virginica*) in Delaware Bay. Tasmiya Moghul, Craig S. Hinkley and Gary Sarinsky, Kingsborough Community College, Brooklyn, NY.

Eastern Oysters (*Crassostrea virginica*) remove organic and inorganic particles from the water and provide a habitat that many organisms depend on to survive. Dermo (*Perkinsus marinus*), a pathogenic protozoan is a major factor causes the disruption of epithelium and connective tissue in oysters which leads to its death. Dermo cells are released into the environment in feces or from decaying tissues. The Dermo is transferred from oyster to oyster Literature suggests that a vector may be involved. The Atlantic Oyster Drills (*Urosalpinx cinerea*) are a major enemy of the oysters. Oyster drills attack by making a small hole in the shell, and then digesting the soft meat of the prey. We hypothesize that the oyster drills may be a vector for Dermo. Dermo has been observed in Delaware Bay since the 1950's. This research is designed to test the presence of Dermo from oyster drills collected in the bay. Tissue was excised from twelve oyster drills. DNA was extracted from the tissue. PCR reactions were used to amplify the mitochondrial cytochrome oxidase1 (CO1) gene. Gel electrophoresis was run on the PCR product to demonstrate that DNA was extracted. Eleven of the twelve tissues were amplified. The amplified products were sequenced and subjected to a NCBI Blast search which confirmed that the DNA was the CO1 gene from *U. cinerea*. Extracted DNA plus a Dermo positive control and a negative control were amplified using a Dermo-specific primer. We ran gel electrophoresis to see if Dermo was present in the DNA. Dermo was not found in any of oyster drills tested or in the negative control. However, the positive control was amplified. The results of this experiment did not support our hypothesis that the oyster drills are a vector for the transmission of Dermo to oysters.

Quantitative Measure of Vascular Smooth Muscle Relaxation in Primary Rat Descending Aorta Cells. Candace Morales-Wilde, Brianna Dzedzic and Thomas Owen, Ramapo College of New Jersey, Mahwah, NJ.

Prostaglandins are small lipids derived from arachadonic acid in the cell membrane through a series of enzymatic steps. Prostaglandin E1 and E2 stimulate the differentiation of mesenchymal stem cells into osteoblasts and thus could be potential treatments for osteoporosis. Unfortunately, the interaction of either of these molecules with the cell surface receptor EP4 also results in a decrease in blood pressure through vasodilation. It is possible that medicinal chemistry efforts could solve this problem, but an *in vitro* method for measuring vasodilation would be needed. Our work is focused on preparing primary cultures of rat descending aortic smooth muscle cells via a published procedure, performing molecular characterization to ensure that the cells are genuine

vascular smooth muscle cells, and then to develop and implement a method to stain the nuclei of the cells in order to measure any changes in their relative positions following PGE1 or PGE2 treatment. Characterization of the cells will be initially done by RT-PCR for the $\alpha 1B$ and $\alpha 1D$ adrenergic receptor subtypes which have been shown to be expressed by vascular smooth muscle cells. RNA from rat renal arteries will be used as a positive control. An antibody to α -SM-actin will be utilized to perform immunofluorescent staining of VSMCs in cell culture. VSMC nuclei will be stained using Hoechst dye, which makes it possible to view distances between individual VSMC nuclei as well as changes in the distance when VSMCs are exposed to PGE1 or PGE2. An increase in nuclei distance would correspond to vasodilation and therefore a decrease in blood pressure. Sodium nitroprusside will be used as a positive control in experiments testing PGE2 and PGE1 as it is a vasodilator that releases NO which activates guanylyl cyclase and forms cGMP, a molecule that inhibits calcium entry into the cell thereby inducing vascular smooth muscle.

Short-Term Effects of Alcohol-Based Hand Sanitizer on Bacterial Numbers. Arwa Najeeb, Elizabeth A. Mulligan and Grace L. Axler-DiPerte, City University of New York: Kingsborough Community College, Brooklyn, NY.

Ten volunteers participated to determine what the effects of alcohol based hand sanitizers are on the number of bacteria on the hands over a brief period of normal activity. Five of the volunteers were given hand sanitizer (HS) and the remaining Isotonic Saline (IS). They were asked to swab their hands before using the solution ($t=B$), right after ($t=0$), 30 ($t=30$) minutes after, and 60 ($t=60$) minutes after cleaning the hands. These swabs were then plated onto Tryptic Soy Agar (TSA) and the number of colony-forming units (cfu) for each time point was determined. My hypothesis was that the number of colonies would decrease from $t=B$ to $t=0$ drastically and increase from $t=0$ to $t=60$ for the volunteers with HS while the volunteers with IS would remain the same. The group using HS showed a statistically significant (Mann-Whitney U-test, significance= 0.05, two-tailed) reduction in the number of cfu from time $t=B$ to $t=0$, as expected ($U_{(B-0)}=1$), however at $t=30$ minutes post-use the number of cfu was not significantly different from $t=B$ ($U_{(0-30)}=3$). Furthermore, at $t=60$ min, any reduction seen at $t=0$ was gone ($U_{(0-60)}=2$; $U_{(B-60)}=9$). No significant difference in cfu was observed between any time points for IS. Using the Kruskal-Wallis method the IS resulted in no significant difference in cfu between the five volunteers ($p=0.87$ and $H=11.35$). There was also no significant difference in cfu between the five HS volunteers ($p=0.048$ and $H=11.35$). Therefore, the use of hand sanitizer appears to confer a short-lived benefit in reducing the number of bacteria on the hands, however by 30 minutes post-use the benefit is absent and not different from the use of isotonic saline. This data supports my hypothesis.

Unraveling the CHD5 Interactome to Identify Anticancer Drug Targets. Ramish Nasir and Shilpi Paul, SUNY Old Westbury, Old Westbury, NY.

Chromodomain Helicase DNA-binding domain 5 (CHD5) was identified as a novel tumor suppressor mapping to 1p36 found to be deleted frequently in a variety of human cancers. CHD5 status has been identified as a prognostic indicator of survival following anti-cancer therapy for several cancers, opening the possibility that therapies that induce wild-type CHD5 expression can enforce tumor suppression. However, CHD5 levels must be carefully controlled to avoid the deleterious effect of cellular senescence. As such, identifying CHD5 interactome may prove useful targets for anti-cancer therapies. CHD5 was identified to interact with chromatin and this chromatin interaction was identified critical for CHD5 to mediate tumor suppression (Paul *et al*, 2013). Chromatin regulatory proteins are present in large multisubunit complex(s) and CHD5 has been reported to exist in large NURD-like protein complexes. Here in this study, we took an initial literature based approach to identify potential effectors of the CHD5-interactome by screening two high-throughput data sets (MudPIT and yeast two-hybrid; Paul, unpublished data). We identified potential CHD5-partners implicated in regulatory processes like transcription, protein post-translational modification, chromatin modification, RNA processing, etc. Using molecular and cell biology tools, we are currently validating the candidate CHD5-interactors. Our evidences support that CHD5 interactome consist of factors that function as histone dephosphatase, deacetylase and methyltransferase). One of the potential CHD5 interactor is SUV39H, a histone lysine N-methyltransferase also identified as an anticancer drug target for prostate cancer (Watson *et al*, Oncogenesis 2014). Our data provides evidence that CHD5 is recruited to regions enriched in H3K9me3, further supporting that SUV39H as a potential player in CHD5 interactome. We are using multiple strategies to determine the role of SUV39H in CHD5's tumor suppressive mechanism. This study will help us to identify SUV39H as a novel drug target for CHD5 related cancers.

Induction of Pre-mRNA Trans-Splicing and Intronic Poly Adenylation in the EGFR Transcript to Form a Shortened Soluble Decoy in Human Glioblastoma Cells. Peter Nekrasov and Martin J Hicks, Monmouth University, West Long Branch, NJ.

Therapies for glioblastoma multiforme (GBM), the most common central nervous system malignancy, are limited by the blood-brain barrier and therapeutic persistence. Tyrosine kinase receptors (TKRs), such as epidermal growth factor receptor (EGFR), are overexpressed in GBM and drive cell proliferation. Previous research examining aberrant transcripts in cancers has identified alternative intronic polyadenylation in the EGFR transcript, resulting in truncated EGFR

isoforms that function as soluble extracellular decoys and sequester EGF. To take advantage of this natural phenomenon, we engineered a pre-trans-splicing molecule (PTM) with simian virus 40 polyadenylation signal, U7 snRNA, and splicing elements to splice into and halt the EGFR transcript upstream of the transmembrane domain. The binding domain, a complementary target sequence playing a large role in trans-splicing efficiency, was designed according to splicing motifs in the EGFR pre-mRNA transcript. Three PTM versions were synthesized: double binding domain, single binding domain, and a bicistronic vector with two distinct antisense targets. These vectors will be tested to examine transmembrane domain skipping, enhancement of trans-splicing efficiency over cis-splicing, and activation of pseudoexons 15A and 15B with alternative intronic polyadenylation signals. Transfection of cancer cell lines A172 and U87 using the three versions of PTM in an adeno-associated virus plasmid shows expression and EGFR transcript interference. Funding Sources: Johnson & Johnson, Bristol-Myers Squibb, Independent College Fund of New Jersey, and Monmouth University School of Science Summer Research Program.

Potential Zinc Stress Response Mechanisms in *Synechococcus* sp. IU 625. Robert Newby, Jr. and Tin-Chun Chu, Seton Hall University, South Orange, NJ.

Cyanobacterial harmful algal blooms are becoming increasing occurrences globally due to rapid eutrophication resulted from climate change, agricultural run-offs, and other industrial contamination sources. Heavy metals such as zinc are among many of the elements that are contributing to the eutrophication events. In this study, we've investigated the potential zinc stress response mechanism using a model unicellular cyanobacterium *Synechococcus* sp. IU 625 (*S. IU 625*). *S. IU 625* in medium containing ZnCl₂ at levels of 0, 10, 25, and 50 mg/L were collected and monitored over 29 days. Viability analysis with SYTOX[®] shows that *S. IU 625* is capable of surviving in all concentrations up to 8 days, however after this point 50 mg/L demonstrated a lethal effect. *S. IU 625* cultured in 25 mg/L shows several divisionary and membrane defects, as detected with scanning electron microscopy; and pigment deficiencies as measured with flow cytometry. Differences in size, chlorophyll a, and allophycocyanin intensity were observed in the cells exposed to 25 mg/L ZnCl₂. Increasing fluorescent signal from zinquin also indicated an increasing intracellular zinc concentration in this population. The results suggested that internal binding proteins, efflux mediated channels, and membrane composition changes could be part of the zinc stress response mechanism in freshwater cyanobacteria.

The Structure of the Recurrent Laryngeal Nerve in the Rat. Oluwatobi Olufeko^{1,2}, Inna Rozenberg², Joshua B. Silverman², Krishnamurthi Sundaram², Mark Stewart² and Richard Kollmar². ¹CUNY Medgar Evers College and ²SUNY Downstate Medical Center, Brooklyn, NY.

Injuring a recurrent laryngeal nerve (RLN) alongside the trachea causes unilateral vocal-fold paralysis, hoarseness, shortness of breath, and possibly bacterial pneumonia. Potential drug treatments that promote RLN regeneration could be tested in rats as animal models. Our previous attempts to damage the RLN in rats, however, caused highly-variable degrees of injury. To improve our surgical approach, we investigated the anatomy of rat RLNs relative to the trachea. Adult Sprague-Dawley rats were euthanized by deep anesthesia and transcardial perfusion with saline and formaldehyde fixative. The trachea with both RLNs was sectioned transversely at about 50 μm thickness. Neurons were labeled with anti-neurofilament antibodies, and myelin with a lipophilic fluorophore. Brightfield and fluorescence microscopy were used to image a range of magnifications, from whole sections to individual nerve bundles. Morphological parameters, such as distances and angles between the RLNs, trachea, and esophagus, were measured with image-analysis software. Our results indicate that the RLNs change their position with respect to the trachea from dorsomedial rostrally to purely medial caudally. The right RLN becomes more accessible on the tracheal surface caudally, whereas the left RLN remains protected in a groove between trachea and esophagus. Along the trachea, both RLNs are associated at different locations with a neurovascular bundle or an overlying fat pad and sprout multiple branches. The RLN bundles contain two classes of neurons that are distinguished by the thickness of their axons and myelin sheaths and are grouped together within the nerve bundle. We conclude that the structure of the RLN in the rat is more elaborate than previously described. Our result suggests that a reliable procedure for RLN injury requires crushing or bisecting both the neurovascular bundle and the overlying fat pad to ensure that all branches are reliably damaged.

Examining the Effect of Oxidative Stress on *Tetrahymena thermophila* Sirtuins Exposed to Starvation. Kabiru Omalaja and Ralph Alcendor, New York City College of Technology, Brooklyn, NY.

Tetrahymena thermophila are free-living ciliate protozoa that can be found in fresh water around the world. These organisms are model organisms for biomedical and toxicology studies. Studying these organisms has led to important findings such as mechanisms controlling cell cycle and discovery of the structure of telomeres and telomerase. *T. thermophila* are known to be very resistant to temperature and other environmental conditions. Therefore they are ideal for examining the effect of oxidative stress, OS. Oxidative stress is the imbalance between levels of free radicals and the ability of the body to neutralize these free radicals. High levels of OS can prevent proteins, DNA and other

important molecules from functioning. Sirtuins are a family of proteins found in almost all living organisms. These proteins have been implicated in a wide variety of cellular functions such as mitochondrial biogenesis, cell death and longevity. Several studies have shown sirtuins function in regulation of OS. However, in *T. thermophila*, the role of sirtuins in regulation of OS has not really been examined. This project aims to examine the effect of OS on *T. thermophila* sirtuins. Cells were exposed to normal and starved conditions for 24-48 h. followed by cell death and mRNA analysis. Starvation led to a significant reduction in cell number and metabolic activity. Compared to control, after 24 h there was more than 75% reduction in metabolic activities. Furthermore, the expression levels of most sirtuins were significantly reduced after 24 to 48 h in starved conditions. Interestingly, compared to 24hrs in normal conditions, 48 h in normal conditions resulted in higher sirtuins expression levels. These results suggest that *T. thermophila* sirtuins may play a very different role in normal and starved conditions.

Preferential Killing of v-Ras-transformed NIH 3T3 Cells by Inhibiting Activated Cdc42-Dependent Kinase (ACK) Ruth Opoku, Nadia Burgos, Ijaz Ahmed, Raj Rajnarayanan and Alam Nur-E-Kamal, Medgar Evers College, Brooklyn, NY.

Hypothesis: The Ras GTPase family has been found to regulate various functions of mammalian cells. Oncogenic mutants of Ras GTPases have been demonstrated to be associated with the development of various types of cancer in human. We propose a hypothesis that cancer cells are more dependent on Ras-Cdc42-ACK signal transduction pathway for their survival. We have previously demonstrated that overexpression of Cdc42 kinase domain of ACK induce death of v-Ras transformed NIH 3T3 cells, but not the parental NIH 3T3 cells. In this report, we studied the effect of tyrosine kinase inhibitors on growth of v-Ras-induced and parental NIH 3T3 cells. Method: We have seeded v-Ras transformed NIH 3T3 cells into culture dishes and incubate overnight at 37°C in standard culture condition. Then tyrosine kinase inhibitors of MG series were added individually or in different combination at various concentrations. Cell growth was monitored by MTT assay in every 24 hours for three days. We determined IC50 for each inhibitor. We have assayed markers of apoptosis in ACK inhibitor treated cells. Results: It was found that some inhibitors of activated Cdc42-associated Kinase (ACK) induced Ras-transformed cell death, while some inhibitors did not exhibit v-Ras cell death activity. The cell death activity of these inhibitors exhibited very weak activity against parental NIH 3T3 cells. We also demonstrated that cell death was associated with induction of apoptosis. We will present the results obtained from this study. Conclusion: Our results indicated that ACK inhibitors induce preferential death of v-Ras transformed cell. Development of ACK kinase inhibitors might lead us to design new experiments to study the potential of such inhibitors for the treatment of Ras-induced cancer.

Project Feederwatch: A Tale of Two Seasons. Vasilios Orogas, Abdul-Mumin Sanni-Adam, Catalina Melendez, Liborio Adorno, Lauren Chukrallah, Dolly Basaldua, Adewale Busayo, Frances Raleigh and Katherine Wydner, Saint Peter's University, Jersey City, NJ.

The second season of Project FeederWatch at Saint Peter's University was recently concluded. Following a protocol developed by Cornell University, FeederWatch is a winter survey of birds that provides information about changes in bird distribution and abundance across North America. A FeederWatch area was monitored between Gannon Hall, Pope Hall, and the Yanitelli Recreation Center on two consecutive days, Thursday and Friday, for 20 weeks between November 2015 and April 2016. A tube feeder was filled with birdseed and one to two suet blocks were provided as continuously as possible. Data was recorded on species of birds, highest number of each species, and physical factors such as weather conditions and snow cover. Although our observations are most informative in their contribution to the North American database, we have analyzed our local data and made comparisons between the seasons 2014-2015 and 2015-2016. In both seasons, the most abundant species reported was the house sparrow (*Passer domesticus*). A total of 6 species were reported this season as compared to 11 species during 2014-2015. A leucistic (white-backed) female house sparrow identified during 2014-2015 regularly continues to visit the FeederWatch area. Comparisons are made between our urban data and the cumulative data reported for the state of New Jersey.

Recovery of a Juvenile Horseshoe Crab (*Limulus polyphemus*) Population on an Eastern Tidal Flat of Plumb Beach. Malokhat Ortikova and Christina Colon, Kingsborough Community College, Brooklyn, NY.

The goal of this research was to monitor the population of juvenile horseshoe crabs (*Limulus polyphemus*) on Plumb Beach. It was hypothesized that juveniles will increase in the tidal creek and reappear on the east tidal flat. This population was healthy in 2011 but declined to zero perhaps due to Superstorm Sandy and a beach re-nourishment project. An adjacent tidal creek supports a population which may be a refuge from these disturbances. Horseshoe crabs have been around for 450 million years, and are essential to the biomedical industry. Their eggs are also an important food source for shorebirds and shore zone fishes (Botton and Loveland 2006). To provide accurate counts, 15 minute timed visual surveys for juveniles were conducted at low tide every two weeks from May 9th to August 17th on east and west tidal flats and the tidal creek. Juveniles were captured, measured with calipers (prosoma width) then released. Data showed 206 juveniles in 2016 in the tidal creek (47 per hour search effort) which peaked on August 1st. This is about half of the 400 found in 2015 (78 per hour search

effort), which also peaked on July 29th, so my first hypothesis was not supported. However, on August 17th, 71 young of the year were found on the eastern beach, which supports my other hypothesis. No hatchlings were found on the western tidal flat despite counting 260 hatchlings (520/search hour) in 2014. Several factors that may reduce survival are Nitrogenous waste, which causes algal blooms and hypoxia, which may lead to disease outbreaks (Braverman *et al.* 2012), thus next year water chemistry such as DO and N levels will be monitored. This work is supported by grant 0537171091 of the CSTEP Program of the NYS Department of Education.

Post-transcriptional Processing of the Mitochondrial ATP8 Gene: A Potential Cause for S-type Cytoplasmic Male Sterility in Maize. Avisek Parajuli and Terry L. Kamps, New Jersey City University, Jersey City.

CMS is a maternally inherited trait that prevents the production of viable pollen. This phenotype is usually associated with the expression of a chimeric region within the mitochondrial genome. These chimeras arise from genomic rearrangements that are due to recombination events among the multiple repeat regions typical of plant mtDNA. Mitochondrial biogenesis and gene expression is determined by nuclear-mitochondrial interactions and the CMS phenotype can be suppressed by nuclear restorer-of-fertility (Rf) genes. Most Rf genes have been found to be within the large family of RNA editing pentatricopeptide (PPR) genes. Their action is consistent with CMS phenotypes being associated with post-transcriptional changes that affect protein accumulation. A reduction in accumulation the ATP8 protein, a subunit of mitochondrial ATP synthase, has been observed during pollen development of maize CMS-S cytotypic as compared to pollen development in plants with a normal fertile cytoplasm. We examined the published whole genome sequencing of the maize mitochondrial genomes. The results showed the *atp8* locus is downstream of an open reading frame locus, *orf-111* and a short intergenic sequence. This organization has potential for the *atp8* locus to be expressed as a chimeric locus that would be consistent with most CMS systems. We are conducting cDNA analysis to examine the expression and processing of this region of the maize mitochondrial genomes from normal and CMS-S cytotypes. We will be reporting on RNA editing and processing results revealed by PCR products and sequencing analysis and their potential effects on ATP8 accumulation during pollen development.

Design of a Gene Transfer Vector to Deliver a Stabilized Anti-EGFR RNA Aptamer to the Glioblastoma Microenvironment. Sachin Parikh and Martin J Hicks, Monmouth University, West Long Branch, NJ.

Glioblastoma multiforme (GBM) is an incurable and aggressive type of brain tumor. It is the most common central nervous system (CNS) malignancy with a median survival of only 14 months. The epidermal growth factor receptor (EGFR) is a type of tyrosine kinase receptor (TKR) dysregulated in about 60% of GBM tumors. EGFR amplification and over-expression leads to uncontrolled growth and proliferation of GBM. Although a great deal is known about the biology exhibited by EGFR-activated GBM, the application of therapies against the biologic processes is limited by the blood-brain barrier, which restricts systemically administered therapies from reaching the brain. We are creating an in vivo tissue culture model to develop a novel strategy to bypass these barriers by developing a gene transfer vector to deliver the genetic sequences of a known anti-EGFR RNA therapy aptamer that binds with high affinity against EGFR. In this approach, we will use a gene transfer system to modify GBM and CNS cells to express the therapeutic anti-cancer RNA aptamer molecule, and using an extracellular RNA "exRNA" localization element, the RNA aptamer will be transported and spread throughout the tumor microenvironment where EGFR is abundant. In addition, we have added an RNA structural element (an inactivated hammerhead ribozyme) important for the stabilization of the RNA therapeutic molecule. Acknowledgements: This research was funded by the Monmouth University School of Science.

The Effects of Bisphenol A and Bisphenol F on the Reproductive System Using *Caenorhabditis Elegans*. Suyapa Penalva-Lopez, Merna Sawaged, Katherine Leonor, Saint Peter's University Jersey City, NJ.

Bisphenol A is a chemical compound that has been used for over 40 years to harden plastics. This compound is present in plastics such as water and baby bottles. Recent research has determined that such chemicals can have a negative effect on the endocrine system, as well as enhancing PCOS. Concerns of the toxicity of these chemical compound led companies to replace bisphenol A to other compounds. These replacing agents are also part of the bisphenol family. One replacements agent that is currently being used to replace BPA is Bisphenol F which is known as BPF. Limited literature research gives no certainty that BPF is less harmful than BPA. The main focus of this study is to determine the effects of acute and chronic exposure of bisphenol A and bisphenol F on the reproductive function over several generations using *C. elegans* as a model system. The following techniques (egg-laying, hatching rate, reproductive rate and the fertilization assay) were used to determine the toxicity effects of the bisphenol family. The data suggested that

both BPA and BPF disrupt the functions of the reproductive function and both bisphenol analogs may have similar toxicity effects. It is yet to be elucidated whether both bisphenol analogs target the same mechanism of action and whether this is similar to human

Comparative Effects of Zinc Chloride on Cyanobacteria *Microcystis aeruginosa* and *Synechococcus* sp. IU 625. Jose L. Perez and Tinchun Chu, Seton Hall University, South Orange, NJ.

The increasing occurrence of harmful algal blooms has been found to have direct negative impacts on water usage, human health, and a healthy environment. Previous studies have shown that climate change, eutrophic states, and the presence of runoff compounds have presented advantageous conditions for large cyanobacteria communities over many aquatic organisms – leading to greater toxin presence in the aquatic environment. In this study, we demonstrated the effects of zinc chloride on the growth, morphology, and production of zinc responsive genes in freshwater cyanobacteria, *Microcystis aeruginosa* and *Synechococcus* IU 625. Various concentrations of zinc chloride ($ZnCl_2$): 0, 10, 25 and 50 mg/L were used to evaluate growth for both *Microcystis aeruginosa* (*M. aeruginosa*) and *Synechococcus* sp. IU 625 (*S. IU 625*) cultures. The results indicated that both cultures showed more growth in 10 mg/L $ZnCl_2$ compared to controls (0 mg/L). While 25 mg/L and 50 mg/L $ZnCl_2$ are shown to be the Inhibitory Concentration (ID_{50}) and Minimum inhibitory concentration (MIC) for *S. IU 625*, *M. aeruginosa* seemed to be more sensitive to the zinc toxicity. The growth of *M. aeruginosa* was completely inhibited by 25 mg/L $ZnCl_2$. Morphological defect were observed in increased concentration of $ZnCl_2$, which may elucidate the zinc stress response mechanism in cyanobacteria.

Evaluation of Cytokines in Autoimmune Retinopathy. Pamela D. Marte Perez^{1,4}, Kelly Baloga², Doreen N. Nguyen², Christopher D. Heaney¹, Sapna Gangaputra³, Nida Sen³, John Hooks³ and Barbara Detrick¹, ¹The Johns Hopkins University, ²The Johns Hopkins Hospital, ³National Eye Institute, NIH and ⁴Caldwell University, Caldwell, NJ.

Autoimmune Retinopathy (AIR) is a retinal degenerative disease associated with an autoimmune component. Ocular findings are characterized by vision loss and retinal defects as well as the presence of anti-retinal antibodies. Cytokines are cell signaling proteins which are important regulatory molecules in immune responses. Cytokines have a variety of functions. IL-6, a potent cytokine is recognized as a biomarker in several diseases because of its potential to regulate inflammation. The purpose of this study was to identify the presence of cytokines in AIR patient serum that may contribute to inflammation. The initial screen identified the upregulation

of IL-6 in serum of AIR patients. A total of thirty one serum samples obtained from twenty-five patients with a diagnosis of AIR and 20 serum samples from healthy individuals (no prior history of ocular disease) were evaluated. Serum levels of IL-6 were determined using standard commercially available EIA kits (R&D Systems, Minneapolis, Minnesota). In addition to the standard EIA method, serum specimens were screened using the 6-plex bead array assay kits (Bio-Rad Hercules, California). In this study the following cytokines were analyzed: IFN-gamma, IL-6, GM-CSF, CXCL9, IL-1 beta and IL-10. Initial screening of AIR patient sera revealed elevated levels of IL-6, IL-10, and CXCL9. In order to quantify IL-6 more accurately, it was measured using EIA. IL-6 was detected in 45 % of sera from AIR patient samples. In contrast, IL-6 was not detected in 20 sera from normal individuals. Comparison of serum IL-6 with disease activity revealed that it was more frequently detected at high levels in severe disease. IL-6 can be frequently identified in sera from AIR patients. Moreover, the presence of IL-6 is associated with increased disease activity. This data indicates that the cytokine IL-6 may contribute to the inflammation process in this disease.

The Effect of Lowering VHL on PC -12 Cells. David Perrotto and Alan Schoenfeld, Adelphi University, Garden City, NY.

The von Hippel-Lindau (VHL) tumor suppressor gene is found on chromosome 3p25.5 and is associated with the inherited disease VHL disease. Germ line mutations in VHL predispose the affected individual to develop both malignant and benign tumors in the kidneys, retina, pancreas, central nervous system, testes, inner ears, and adrenal glands. Although loss of VHL causes adrenal gland tumors (pheochromocytoma), there does not exist a good cell model to study this aspect of VHL disease. The CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats)-Cas 9 system is responsible for guiding the Cas 9 protein for sequence specific recognition and cleavage of target DNA complementary to the guide sequence. Here, the CRISPR-Cas system will be used to cause mutational inactivation of the VHL gene in cells of adrenal origin. Thus, this research will attempt to see the effects of using the crisper cas 9 system to lower the expression of the VHL gene in PC-12 adrenal neural cells

The Effect of Benzethonium Chloride and Natural Antibacterials on AGR-dysfunctional *Staphylococci*. Bien N. Pocorni, Geetha Surendran and Davida Smyth, Mercy College, Dobbs Ferry, NY.

The *Staphylococcal* accessory gene regulator (*agr*) is a quorum sensing system that regulates many colonization and virulence factors. Depending on the conditions, *agr*-activity may influence antibiotic resistant properties. *Agr*+ bacteria show a decreased expression of cell surface proteins and an increase in secreted virulence factors.

However, recent studies indicate that *agr* dysfunctional mutants are less susceptible to vancomycin treatment and seem to be the cause of most complications in healthcare environments. These facts brought us to question whether *agr* mutants are also less susceptible to common anti-bacterial agents such as benzethonium chloride; a quaternary ammonium compound that can be found in a range of cleaning, disinfecting and personal care products since the drawback on triclosan. In addition, we also examined the effectiveness of specific natural products said to have anti-bacterial properties such as garlic, onions, apple cider vinegar, ginger, turmeric, cinnamon and Manuka honey. The experiment was done on strains 6734 (*agr*+), 7206 (*agr*-) and 25923 (wildtype). Our current results indicate that benzethonium chloride successfully inhibits the growth of all three strains even at a MIC of .00001 g/ml. We observed great effectiveness of garlic on strains 6734 and 7206, which did not affect the wildtype at all surprisingly. The results of this study show that both *agr*+ and *agr*- *staphylococci* show that both *agr*+ and *agr*- *staphylococci* are equally susceptible to benzethonium chloride and garlic. Only when grown in the presence of apple cider vinegar did the *agr*- type prove more susceptible than the *agr*+ type. As we continue this comparative research, we will examine the susceptibility of multiple *agr*- and *agr*+ *staphylococcal* strains at a wide range of benzethonium concentrations. In addition, we will also look into the active ingredients of garlic, apple cider vinegar and other spices to find out if we can amplify their antibacterial functions.

HDAC9 Linked to Lysine Acetylation Levels: a Potential Mechanism in Diabetic Nephropathy, Simran Alexandria Polce and Maria Alica Carrillo Sepulveda, NYIT-COM Old Westbury NY.

Obesity has reached epidemic levels in the United States, becoming a leading public health concern and major risk factor to the development of type II Diabetes (DM II). DM II is a metabolic disease primarily characterized by high levels of glucose. Diabetic nephropathy is one of the major complications faced by diabetic patients. It is characterized by fibrosis, mesangial matrix expansion and loss of proper glomerular filtration, and vascular renal dysfunction. The mechanisms involved in these pathological conditions are still not well known. Recent evidence has indicated that some post-translational modification, such as acetylation, is associated with diabetes. Based on recent published findings correlating increased lysine acetylation levels with kidney disorders and our preliminary data showing that levels of lysine acetylation are augmented in diabetic kidneys, we hypothesize that HDAC9, one of the histone deacetylase enzymes regulates lysine acetylation levels in diabetic kidneys. To address our hypothesis, adult db/db mice, a model of obesity-type II diabetic, were utilized in this current study. Db/db mice showed robust increased body weight (39.7642g vs. 19.5000g, $p < 0.05$), kidney

weight (0.2388g vs. 0.1639g, $p < 0.05$) and blood glucose levels (265.1690mg/dl vs. 114.5556mg/dl, $p < 0.05$) compared with the control group. These results validate the diabetic model used. Proteins were isolated from diabetic kidneys to further determine HDAC9 levels through western blot. Levels of HDAC9 in kidney tissue from db/db mice were significantly lower compared with the control group, indicating that diabetic milieu modulates the expression of epigenetic enzymes. Together these results suggest that HDAC9 may be playing a role in controlling the levels of lysine acetylation in diabetic kidneys. Thus, targeting HDAC9 may offer novel therapeutic strategies for treating diabetic nephropathy. As a future direction, this study will be testing whether overexpression of HDAC9 will ameliorate kidney function in diabetic conditions.

Urban Tree Canopy Assessment. Elizabeth Ramirez and Coleen Di Lauro, Bergen Community College, Paramus, NJ.

Urban landscape can affect the air quality, flow and excrete aeroallergens Invasive species. Air pollution has gained significant attention through the American Lung Association; Hudson County quality of air ranks an F. Studies has shown trees improve the quality of air. Trees absorb the greenhouse gases such carbon dioxide (CO_2), nitrogen dioxide (NO_2), and harmful microscopic particles from ozone layer. Do trees worsen the quality of air by trapping pollutants and confining them to street level? Our findings revealed from the Hudson County Parks Department. This is just the initial phase of the project with as of last year 14% of the total area mapped. Even so we gathered a lot of excellent data. We notice a fungus affecting almost all the London Plane trees. This is significant because London Planes have a high carbon abatement rate (Kovacs) and were heavily planted. But we should not over plant them or we may loss a majority of our trees to a blight! (Davis) Other preventable problems like invasive species can be solve by educating the community about them.

Ebola Virus: As an Example of the Biology and the Epidemiology of a Virus to be Used as a Teaching Guide. L. Reilly, D. Colgan, W. Mirza and L. Lorentzen, Kean University, Union NJ.

The goal of this project was to raise awareness about the acute dangers a virus such as Ebola poses not only to local communities but ultimately to the world at large. The outcome of the project was a compilation of information resulting in an easy resource document to share (at the MACUB conference) with academics who teach college freshman biology or high school AP biology to incorporate, if they wish, into the unit that discusses viruses. Ebola is a potentially deadly virus with no current cure and has shown recurring outbreaks since 1976.

Ebola works similar to many viruses that incorporate DNA into a cell with the purpose to weaken, manipulate and ultimately destroy the host. The virus targets macrophages and dendritic cells to spread the virus to most organs of the body causing cell death, organ failure and hemorrhagic bleeding. The simple unit plan includes the biology of the virus, its transmission, signs and symptoms, current and potential treatments, the history and the spread of the epidemic, the Center for Disease Control's strategies to contain epidemics and a suggested list of questions for class discussion.

Effect of Single Walled Carbon Nanotubes on Breast Cancer Cell Migration. Belkis Rodriguez, Jonathan Edouard, Mathiu Perez, Sunil Dehipawala, Tirandai Hemraj-Benny and Regina Sullivan, Queensborough Community College, Bayside, NY.

Biomedical applications of single walled carbon nanotubes (SWCNT) have the potential to expand treatment options for cancer patients. Carbon nanotubes have a high surface area to volume ratio which allows for surface functionalization. The size of these nanotubes facilitates use as a drug delivery system as well. Recent studies have shown that unfunctionalized nanotubes enter cells via endocytosis. In addition the nanotubes may enter cells through cellular gap junctions and ion channels. In previous studies we have shown that nanotubes are not cytotoxic in low concentrations. Currently we are testing the hypothesis that unfunctionalized single walled carbon nanotubes incorporate into the actin cytoskeleton and decrease migration of triple negative breast cancer cells. However our studies have been limited by aggregation of the nanotubes in aqueous solutions which decreases cellular uptake and increases cytotoxicity in in vitro studies. Coating single walled carbon nanotubes with collagen has been shown to facilitate cellular uptake thus allowing for intracellular associations to be investigated. This method has limitations due to the acidic pH of the collagen solution. In this study, we compared the effect of collagen coated single walled carbon nanotubes with debundled single walled carbon nanotubes on breast cancer cell migration. Migration assays were performed and revealed that breast cancer cells treated with collagen coated SWCNT as well as the debundled SWCNT has a reduced rate of migration. These results suggest that the SWCNT may be incorporating into the actin cytoskeletal disrupting rearrangements that are required for the metastatic process. In future studies we plan to measure Young's modulus which is an indicator of the degree of flexibility which in turn can be correlated with changes in the actin cytoskeleton. The study will be expended to include other types of cancer cells as well noncancerous cells and may reveal potentially novel cancer treatments.

Memory Impairments During Cognitive Tasks in a Streptozotocin Induced Rat Model of Sporadic Alzheimer's disease. Raquel Romaine¹, Rashida R. Farokhi¹, Farrah Hussain¹ and Francisco Villegas², ¹Queensborough Community College, Bayside, NY and ²York College, Jamaica, NY.

Alzheimer's Disease (AD) is the most common neurodegenerative disorder, leading to cognitive deficits, behavioral impairment and dementia. Data has shown that there are strong correlations between sporadic Alzheimer's Disease (sAD) and Diabetes II. The neurotoxin, Streptozotocin (STZ) was employed via intracerebroventricular (ICV), to mimic sAD by causing cellular and behavioral abnormalities. Rat models of sAD were created using ICV-STZ, and were used in the present study to observe the disruptions caused in working memory, and reference memory using the object discrimination test and Morris water maze. Rats were anesthetized and injected with STZ dissolved in artificial cerebral spinal fluid (aCSF) into their lateral ventricles. The control groups were rats that received ICV injections of aCSF (n=7), and the experimental groups were rats that received ICV injections of 1 mg/kg STZ (n=10), and 2 mg/kg STZ (n=9). No significant differences were found between the control group and groups injected with STZ in the Morris water maze 'working memory procedure'. However, the group of rats that received 2 mg/kg of STZ visited fewer quadrants than the control group and the group that received 1 mg/kg of STZ. The groups of rats injected with higher dosages of STZ were hypothesized to have memory deficits, but the results show little difference in the number of quadrants visited when compared to the control groups. The object discrimination data showed that the 2 mg/kg of STZ injected group visited the novel objects slightly more than the subjects in the control group and the 1 mg/kg of STZ subjects, but not enough to show a significant difference in memory impairment. There appeared to be a trend in memory deficits with higher dosages of STZ, thus 3 mg/kg of STZ will be used in future experiments to examine cognitive decline commonly associated with AD and Diabetes II.

Live Imaging and Analysis of Wild-Type Versus CENP-A Depleted Embryos in the Nematode, *Caenorhabditis elegans*. Brianna Romer and Joost Monen, Ramapo College of New Jersey, Mahwah, New Jersey, NJ.

The centromeric protein CENP-A is critical for kinetochore deposition and chromosome segregation in mitosis. In the nematode *Caenorhabditis elegans* two CENP-A homologs exist, HCP-3 and CPAR-1. HCP-3 plays a conserved centromeric role critical for mitotic chromosome segregation, whereas CPAR-1's role has yet to be determined. To demonstrate the function of HCP-3, an RNAi feeding protocol was used to deplete HCP-3 from the embryos of a wild-type GFP::Histone-H2b/GFP::gamma-tubulin expressing strain. Both wild-type [n=11] and HCP-3 [n=6] depleted embryos were imaged

on a Zeiss Axiovert 200M epi-fluorescent microscope using time-lapse microscopy at 30 second intervals. HCP-3 depleted embryos display a clear chromosome segregation defect and fail to properly divide. Close examination of spindle pole dynamics indicate that in HCP-3 depleted embryo, spindle poles separate prematurely likely due to the failure of the spindle to attach to chromosomes. These experiments are consistent with previous studies and provide a robust and reproducible assay to test RNAi depleted perturbations and analyze chromosome and spindle pole dynamics in live embryos. Future experiments will include a determination to the extent at which HCP-3 is depleted under RNAi condition as well as a similar characterization of CPAR-1 depleted embryos to test the functional role of CPAR-1 in early embryonic development.

Does Histone H3 Acetylation Stimulate NuA4 - Nucleosome Interaction? Tiffany E. Rose and Daniel S. Ginsburg, LIU Post, Brookville, NY.

Nucleosome acetylation is a key step in chromatin disassembly during transcription. The process is regulated by the opposing activities of lysine acetyltransferase (KAT) and histone deacetylase complexes. Transcription cannot occur without enough acetylation, while too much acetylation can lead to production of aberrant transcripts. The two major KAT complexes in yeast are NuA4 and SAGA. NuA4 acetylates histones H4 and H2A while SAGA acetylates histones H3 and H2B. We have previously shown that they work together to stimulate both transcription initiation and elongation. NuA4 binding to nucleosomes is stimulated by histone methylation, while the SAGA bromodomains mean that its interaction with nucleosomes is stimulated by NuA4-mediated histone acetylation. The Yaf9 protein in NuA4 has a YEATS domain that has been shown to bind acetylated histones and global H4 acetylation decreases when the KAT subunit of SAGA is deleted. The goal of this project was to test the hypothesis that NuA4 interaction with the nucleosome is in part stimulated by SAGA-mediated H3 acetylation. We examined H4 acetylation and NuA4 occupancy at the GAL1 gene in strains lacking either ADA1, important for SAGA integrity, or GCN5, the SAGA KAT subunit. Additionally, we looked at H4 acetylation in strains in which the H3 lysines acetylated by SAGA were mutated. Loss of H3 acetylation led to an increase in the amount of global H4 acetylation in bulk histones. Additionally, gcn5Δ cells demonstrated increased NuA4-nucleosome interactions. In contrast to this we found that NuA4 occupancy decreased at the GAL1 UAS and 3'ORF in both ada1Δ and gcn5Δ cells. This suggests that NuA4 recruitment to transcribed genes may be stimulated by H3 acetylation. These results provide preliminary evidence that the relationship between NuA4 and SAGA may be more complex than previously thought with positive feedback leading to nucleosomal acetylation rapidly spreading across a transcribed gene.

Low Level Lead Exposure Differentially Alters Locomotor Activity and Anxiety Dependent Upon Developmental Time Period of Exposure: Implications for Neurocognitive Disorders. Samantha Rubi¹, Nimra Hameed¹, Sukhpreet Kaur², Neena Matthew², Teddy Dacius¹, Tokunbo Jose¹ and Lorenz S. Neuwirth^{1,2}, ¹SUNY Old Westbury, Old Westbury NY and ²SUNY Neuroscience Research Instituted, Old Westbury NY.

Lead (Pb) is a developmental neurotoxin that causes lifelong cognitive dysfunction. However, certain motoric behaviors and underlying emotional states may obscure behavioral measures as they relate to higher order cognitive processing. Here we examined the effects of an environmentally relevant Pb exposure (25 ppm Pb-acetate in drinking water) in Long Evans Hooded rat's locomotor and anxiety behaviors through the Open Field (OF) and Elevated Plus Maze (EPM) as a function of developmental time period of exposure. Control rats evidenced typical levels of locomotor activity and anxiety, whereas rats exposed to Pb in Early Postnatal development (EPN) exhibited hyperactivity and Perinatal (Peri) exposed rats showed hypoactivity in the OF. Interestingly, in the EPM no differences in male rat's anxiety levels irrespective of developmental time period of Pb exposure. However, the female rat's showed a development time period effect, whereby EPN and Peri-PND0 rats had elevated anxiety in contrast to Control and Peri-PND22 rats. Results show Pb toxicity, dependent upon the developmental time period of exposure and sex has different susceptibilities and specific deficits that may impede their ability to perform other learning and memory tasks assessing their cognitive functions. As such, this preliminary data cautions the field to carefully evaluate the sex specific roles and the time of development as important differential and predictive factors of later life cognitive outcomes in response to Pb exposure. The goal of this work is to offer more accurate neurotoxicity assessments for appropriate generalizability across and between animal and human models in re-characterizing sub-clinical early warning signs of low level neurotoxic neuro-behavioral markers of intellectual disability.

***Toxoplasma gondii* is Not Found in The Eastern Oyster (*Crassostrea virginica*) in Jamaica Bay, New York. Izzah Sabir, Craig Hinkley and Gary Sarinsky, Kingsborough Community College, Brooklyn, NY.**

Toxoplasma gondii is a parasitic protozoan which is known to cause the disease Toxoplasmosis. *T. gondii* can be passed from animals to humans. It may cause brain damage in the human fetus and young children, muscle pain in adults, headaches and a fever which could last for weeks. Animals such as cats are hosts for *T. gondii*. Though they are not affected by this protozoan, their feces contain the protozoan which is known to end up in greater bodies of water or soil and which then transfers into organisms. Since oysters are filter feeders, they breathe in the parasite infected water. If they become infected, the parasite will transfer to humans who consume them. Jamaica Bay is a 173 sq. mile body of water which is

surrounded by Queens, Brooklyn, and Rockaway. The habitats in the bay are home to 91 fish species, over 300 bird species, and many other small mammals. We hypothesize *T. gondii* is present in Jamaica Bay and therefore will be present in oysters. Ten Eastern Oysters (*Crassostrea virginica*) naturally found in Jamaica Bay were collected and their tissues were extracted. PCR reactions with Folmers primer was used to amplify the mitochondrial cytochrome oxidase 1 (CO1) gene. Gel electrophoresis of the amplified product was performed to show that DNA was isolated. A *T. gondii* specific primer was then used to amplify extracted DNA from the ten oysters plus a positive and negative control. Only the positive control was amplified. The negative control and the ten samples were all negative. The oysters tested did not have *T. gondii*. The results of sequencing the DNA showed that all of the samples amplified the mitochondrial CO1 gene from *C. virginica*. Since *T. gondii* was not found to be present in the oysters tested the hypothesis was not supported.

Use of a Hollow Fiber Bioreactor for Analysis of Antiviral Compounds on the Cryptosporidium virus. Gabrel Samantha, Mary Morada and Nigel Yarlett, Pace University, New York, NY.

Cryptosporidium parvum is an intestinal parasite causing significant morbidity and mortality in economically poor countries. It is listed in the top three pathogens causing severe pediatric intestinal disease for which no reliable effective treatment exists. The parasite harbors an intracellular virus (*Cpv*) that may play a role in the pathology of the disease. Until recently it was not possible to grow the parasite in vitro to understand the pathology of the disease. Studies performed at the Haskins Laboratories of Pace University resulted in the development of a hollow fiber bioreactor that enabled long-term growth of the parasite under conditions that mimic the intestinal tract. Using this system we are able to test various antiviral compounds with the following aims: i) can we create a *C. parvum* *Cpv*- strain; ii) does removal of the *Cpv* affect growth of the parasite; iii) is the *C. parvum* *Cpv*-strain able to cause disease in the host. Two antiviral agents were tested, gliotoxin and phosphonoacetic acid. Gliotoxin dramatically reduced the *Cpv* (<10% of controls) but did not prevent growth of *C. parvum*. However, after 3 days of treatment with gliotoxin bizarre morphological forms of *C. parvum* were observed that may indicate the antiviral has an effect on specific life cycle stages of the parasite; or it may be due to loss of the virus. Moreover, phosphonoacetic acid also reduced the *Cpv* (<1% of controls), however by day 3 it also significantly reduced *C. parvum* (*C. parvum* oocysts undetectable by microscopy). Removing gliotoxin or phosphonoacetic acid resulted in *C. parvum* recovery to 90% of controls in 2 and 5 days, respectively. Studies are continuing using longer exposure to determine if it is possible to produce a *Cpv*- strain that can be used to determine infectivity using a T-cell deficient mouse model.

Thyroid Hormone Induces Vascular Relaxation Through VASP Phosphorylation at Serine239: A Potential Therapeutic Approach to Treat Diabetic Vascular Dysfunction. Sherin Samuel¹, A. Martin Gerdes² and Maria Alicia Carrillo-Sepulveda². ¹New York Institute of Technology and ²NYIT College of Osteopathic Medicine, Old Westbury, NY.

Vascular complications are a major problem faced by diabetic patients, especially the impairment of vasorelaxation. Recent studies have indicated that thyroid hormone, triiodothyronine (T3), has anti-diabetic effects. T3 is known to rapidly relax vascular smooth muscle cells (VSMCs) via nitric oxide (NO) mechanisms. However, the endothelial-smooth muscle cells interaction mechanisms have not been well explored. An association between VASP phosphorylation at serine 239, a substrate for cGMP-dependent protein kinase (PKG), and VSMC relaxation has been demonstrated. Thus, we hypothesized that NO/PKG/VASP signaling pathway may be involved in T3-induced vascular relaxation. In vitro studies showed that human endothelial cells (EC) treated with 0.1 μ M T3 for short-time (2 to 60 minutes) display significant phosphorylation of Akt (1.7 \pm 0.1 fold of increase vs. unstimulated cells) and eNOS (1.9 \pm 0.2 fold of increase vs. unstimulated cells). Moreover, T3 robustly increased levels of NO (2.3 \pm 0.2 fold of increase vs. unstimulated cells; $p < 0.001$) in EC. Additionally, T3 treatment increased phosphorylation of VASP at serine 239 in human VSMCs (2.0 \pm 0.2 fold of increase vs. control; $p < 0.05$), which was diminished in the presence of 1 μ M KT5823, a selective PKG inhibitor. Rat aortas incubated with 0.1 μ M T3 for 24 hours showed a significant increase in PKG expression (1.8 \pm 0.1 fold of increase vs. unstimulated aortas; $p < 0.05$). Endothelium-dependent and -independent relaxation was assessed in rat aortas treated with 0.1 μ M T3 for 20 minutes. Aortas treated with T3 exhibited greater sensitivity (EC50) to acetylcholine (EC50 value: 7.80 \pm 0.07 vs. 7.10 \pm 0.05 control, $p < 0.0001$) and sodium nitroprusside (EC50 value: 8.12 \pm 0.03 vs. 7.6 \pm 0.02, $p < 0.0001$). T3-induced vasorelaxation independent of endothelium was reduced in the presence of 1 μ M KT5823 (EC50 value: 7.8 \pm 0.02, $p < 0.05$). Our results suggest a novel NO/PKG/VASP molecular mechanism underlying T3-induced vascular relaxation. Strategies utilizing T3 in safe dose pose a promising approach as an adjunct therapy to treat vascular dysfunction in diabetes.

Juvenile Hormone Sensitivity of an Enhancer Region on the Foraging Gene. Marium Sarder and Rebecca Spokony, Baruch College, New York, NY.

Juvenile hormone (JH) and ecdysone work in conjunction to regulate insect physiological development. The ecdysone pathway has been well researched; however the molecular pathways of JH are largely unknown. A goal for this experiment is to determine if foraging, a JH target gene in other insects, is a JH target gene. The main objectives of this experiment were to determine if (1) JH affects transcription (2) if JH regulated

enhancers are also regulated by ecdysone. This was examined by generating transgenic flies with a reporter gene, LacZ, attached to a putative enhancer from the foraging locus. Control transgenic flies had an enhancer region with the full Ecdysone Receptor (EcR) binding motif, whereas mutants had an altered, nonfunctional EcR binding site. Third instar larvae with the reporter gene construct were treated either with ethanol, or JH mimic: methoprene; they were then dissected and stained. Results indicate that the enhancer regulates transcription in a temporally, spatially, sex specific, environmentally and ecdysone receptor dependent manner. LacZ expression was found in ecdysone-regulated locations, such as the eye disc morphogenetic furrow, midgut, and imaginal discs. Males displayed higher LacZ expression, and greater sensitivity to methoprene than females. After mutating the EcR binding site, LacZ expression was lost in some tissues where it was expressed in the full enhancer flies. Methoprene treatment restored LacZ expression in some tissues where it was lost after mutating the EcR motif. These results strongly suggest the enhancer is regulated by both JH and ecdysone, and that foraging is a JH target gene. Flies without EcR will be examined to further test this hypothesis. We predict that the absence of EcR will lead to a large decrease in JH sensitivity. Foraging expression will be examined in tissues that display the most enhancer activity to confirm the target gene identity.

Optimizing Cross-linking Efficiency of Triblock Protein Polymers for Hydrogel Fabrication. Nicole L. Schnabel¹, Andrew Olsen¹, Yao Wang¹ and Jin K. Montclare^{1,2,3}, ¹NYU Tandon School of Engineering, Brooklyn, NY, and New York University^{1,2,3}, New York, NY and ³SUNY Downstate Medical Center, Brooklyn, NY.

Many naturally occurring adaptive and responsive biological systems, such as the gecko's adhesive capabilities and the self-cleaning mechanism of the lotus leaf, can be engineered as biologically-inspired synthetic materials. Using protein hydrogels as a building block, we aim to achieve patterning and mimic the properties delivered by these systems. Our lab has generated a chemo-mechanically responsive triblock polymer that comprises two domains, one derived from the non-collagenous glycoprotein cartilage oligomeric matrix protein (C), capable of binding small molecules, and the other from the elastin-like polypeptide motif (E), which exhibits an inverse transition temperature. This triblock polymer is rationally designed, in which the two domains are linked into a C-E-C sandwich type of polymer, combining the respective functions of each domain. Free standing protein hydrogels have been generated by conjugating the CEC triblock polymer protein with a photo-crosslinkable diazirine group (CEC-DAz) followed by UV-mediated crosslinking. This CEC-DAz hydrogel has the potential to be responsive in dynamic environments of temperature as well as humidity content and capable of

small molecule binding for drug delivery purposes. Furthermore, we have optimized the most effective expression of the CEC protein through a side-by-side comparison of isopropyl β -D-1-thiogalactopyranoside (IPTG) and auto-induction expression, determining the higher effectiveness of auto-induction by the increased pure protein yield. We have assessed the efficiency of the crosslinking modification with respect to protein concentration yield and length of UV exposure. Further studies to fabricate the CEC polymer into various patterns on substrates are underway.

Analysis of the Relationship Between PKC Iota (PKC ι) and VHL-mediated Cellular Phenotypes in 786-O Cells. Alissa Schurr and Alan Schoenfeld, Adelphi University, Garden City, NY.

Von-Hippel Lindau (VHL) disease is a familial cancer syndrome that predisposes individuals to renal cell carcinoma as well as other tumor types. PKC iota (PKC ι), an isoform of protein kinase C, has been shown to bind pVHL; however, the significance of this interaction remains unknown. By using the CRISPR genome engineering system, retroviral LentiCRISPR plasmids were transfected into both VHL-null parental cells and 786-O cells with VHL introduced in order to produce knockouts of PKC ι . The colonies with successful production of these knockouts have been identified via western blotting, and various assays have been performed for cellular phenotypes that have been previously shown to be affected by both VHL and atypical PKC (aPKC). It is hoped that this experimentation will provide a clear determination of the relationship between VHL and PKC ι .

Comparative Study of the Response to Light on the Sensory Motor Integration of Gill Lateral Cell Cilia in Bivalve Molluscs. Danellie Semple¹, Johanne Jean-Pierre², Edward J. Catapane¹ and Margaret A. Carroll¹, ¹Medgar Evers College and ²Kingsborough Community College, Brooklyn, NY.

Gill lateral cells (GLC) of bivalves *Crassostrea virginica* and *Mytilus edulis* are innervated by serotonin/dopamine nerves. Motor aspects of GLC innervation are well studied, but not the sensory. We found light, food, crab extract and histamine initiated a sensory-motor integration response between mantle rim sensory tentacles and GLC cilia. Histamine is a neurotransmitter in photoreceptors. Near the cerebral ganglia, *C. virginica* and *M. edulis* have cerebral ocelli (eyespot) that have rarely been studied other than histological/anatomical reports. We hypothesize cerebral ocelli in both bivalves are light sensitive and involved in histamine mediated sensory-motor integration effecting GLC. We used whole animal with intact gill innervation to access actions of light and a histamine H2 receptor antagonist on cerebral ocelli. Cilia beating was measured by stroboscopic microscopy. An eleven lumen, ¼ inch diameter spotlight was used to stimulate cerebral ocelli. Stimulating cerebral ocelli with

light decreased cilia beating in *C. virginica*, but increased beating in *M. edulis*. In both animals, excising cerebral ocelli prevented the response to light. Famotidine, a histamine H2 receptor antagonist, blocked the response to light. We also tested light on mantle rim sensory tentacles in *M. edulis* and found light caused cilio-excitation, opposite to the cilio-inhibitory in *C. virginica*. Since *C. virginica* is nocturnal, while *M. edulis* is not, opposite responses are not surprising. The study demonstrates sensory-motor integration between mantle rim sensory tentacles and the motor response of GLC cilia, and adds new knowledge of the physiological role of cerebral ocelli in a class of animals where it has not been well studied. It supports the hypothesis histamine is the sensory neurotransmitter for photoreceptors in cerebral ocelli and mantle rim in these bivalves. This work was supported by 690340047 of PSC-CUNY, 2R25GM06003 of the Bridge Program of NIGMS and the Carnegie Foundation.

HMGB1 Inhibits Phagocytosis and Migration in LPS-stimulated Microglial Cell. Jordy Sepulveda, Maria Entezari and Lucia Fuentes, LaGuardia Community College, Long Island City, NY.

Neuro-inflammation and accumulation of A β -containing amyloid plaques are critical components of the pathogenesis of Alzheimer's disease (AD). It was shown that high-mobility group box1 (HMGB1) is extracellularly associated with A β plaques in AD brain. Activated microglia are able to migrate to the sites of A β deposition and eliminate A β by phagocytosis. The impairment of microglia migration and A β phagocytosis appear to be closely involved in the progression of AD pathology. However, the underlying molecular mechanisms responsible for disease progression are still unclear. We previously demonstrated the importance of HMGB1 in the impairment of phagocytosis ability in the peritoneal macrophages. Therefore, we hypothesized that HMGB1 contributes to microglial dysfunction under neuro-inflammation condition. In this study, we examined the effect of HMGB1 on migration and the phagocytotic ability of LPS-stimulated BV2 microglial cells. The migration and phagocytosis function evaluated by in vitro wound healing assay and phagocytosis assay respectively. Our data show that treatment with 10 and 100ng/ml of recombinant HMGB1 significantly decreased the migration and phagocytosis ability in BV2 cells. Interestingly, HMGB1 at both concentrations inhibited the effect of LPS-induced up-regulation of phagocytosis in BV2 cells. Moreover, treatment with recombinant HMGB1 in BV2 cells accompanied by the expression of Scavenger receptor II, CD36, and Toll-like receptor 4 (TLR4) on these cells. These results suggest that activation of CD36/TLR4 signaling pathways contributes to the HMGB1-induced microglia dysfunction. Thus, inhibiting of HMGB1 may provide a therapeutic target for enhancing of microglia's ability to migrate and phagocytose in AD.

Chromatin Condensation Protects DNA from MMS. Sherwin Shaju and Daniel S. Ginsburg, LIU Post, Greenvale, NY.

Chromatin is the complex of proteins and DNA that makes up eukaryotic chromosomes. The basic unit of chromatin is the nucleosome, which consists of ~150 bp of DNA wrapped around the core histone proteins. From nucleosomes separated by ~100 bp of naked DNA (beads on a string), chromatin forms higher order structures with the addition of other proteins, resulting in the condensed metaphase chromosome, which is the most compact form of chromatin. Chromatin helps the DNA fit inside the nucleus as well as regulating what can access the DNA. Replication, repair, and transcription are all impeded by chromatin. These processes only occur efficiently when chromatin is disassembled. Just as chromatin limits how proteins can access the DNA, we hypothesized that it would also protect DNA. We used the yeast *Saccharomyces cerevisiae* to study how chromatin affects cellular sensitivity to the DNA alkylating agent methyl methanesulfonate (MMS). MMS alkylation can lead to DNA mismatches, nicks, and double strand breaks, resulting in mutation. Using growth assays, we found that active transcription significantly increases the sensitivity of a gene to MMS. Consistent with this, we found that increased chromatin compaction due to nocodazole-mediated metaphase arrest reduced the effects of MMS. Because transcription led to an increase in MMS-mediated DNA damage, we proposed that transcribed genes would be subject to increased damage under normal growth conditions, requiring recruitment of DNA repair factors. We found by chromatin immunoprecipitation that the Rad51 DNA repair protein is recruited to the GAL1 gene during transcription, although its occupancy was not stimulated by MMS treatment. This suggests that there is enough damage occurring during normal transcription to require DNA repair factors. Together, these results support our model that chromatin helps protect DNA from damage in addition to regulating other DNA-based processes

Using CO1 vs. 16S rRNA DNA Barcoding for Taxon Clustering and Phylogenetic Resolution in Invertebrates. Harpreet Singh, Thomas Smith, Julian Stark and Nidhi Gadura, Queensborough Community College, Bayside, NY.

The mitochondrial CO1 gene has long been the standard locus for DNA barcoding of animals. Its mutation rate is while its sequence remains highly conserved between closely-related species. On the other hand, 16S rRNA is a commonly used region in DNA barcoding to identify a particular bacterial species. Some studies have used mitochondrial 16s rRNA gene for invertebrates because it is a powerful tool for genetics and phylogenetic studies. We hypothesize that in a parallel study, we will be able to resolve major taxonomic groups and closely related taxa using both CO1 and 16S rRNA barcoding regions. In this study, CO1 and 16S rRNA barcodes were

amplified from a diverse array of 37 invertebrate species. Samples were acquired and identified from several entomology research laboratories and from QCC campus. DNA was extracted from samples, and the barcoding region was amplified by PCR using appropriate primers. The forward and reverse strands of the PCR products were then sequenced and aligned in order to build consensus sequences, and the correct identifications of the samples were confirmed using BLAST. The consensus sequences were then exported to MEGA where they were aligned using MUSCLE. Using these CO1 and 16S rRNA barcoding sequences, phylogenetic trees were constructed with software available from Cold Spring Harbor and MEGA. The efficacy of the CO1 and 16S rRNA gene as a universal invertebrate barcoding region was then examined by comparing the resulting phylogenetic tree to established phylogenetic relationships between the species involved in this study. Both CO1 and 16S rRNA data show appropriate taxon clustering and phylogenetic resolution from the samples tested. A combined analysis using a subset of taxa which amplified for both the CO1 and 16S rRNA regions gave more robust results under both maximum parsimony and maximum likelihood analyses.

Efficacy of Antisense Guided Pre-mRNA Splicing on Epidermal Growth Factor Receptor Expression in Human Glioblastoma Cells. Nicole Sivetz. and Martin J. Hicks, Monmouth University, West Long Branch, NJ.

Glioblastoma multiforme (GBM), the most common central nervous system (CNS) malignancy, is characterized by overexpression of the membrane bound epidermal growth factor receptor (EGFR). Activated EGFR promotes GBM tumor proliferation and growth. Current prognosis for patients receiving standard care is approximately fourteen months. GBM tumors, protected by the blood brain barrier grow aggressively extending into healthy tissue. Our novel approach to deliver DNA encoding anti-sense RNA molecules to alter pre-mRNA splicing of the EGFR mRNA transcript in GBM cells has the potential to bypass this barrier. In the strategy presented, we have designed a pre-trans-splicing RNA molecule (PTRM) to deliver a polyadenylation signal (PAS) into the EGFR pre-mRNA transcript upstream of the exon corresponding to the transmembrane domain, altering the mature EGFR transcript. In our design, optimization of the EGFR antisense binding domain and a U7 snRNA-SmOpt localization signal will enable the PTRM to compete against the downstream 3' splice sites of the EGFR transcript, generating a shortened mRNA transcript. This shortened transcript would translate into a non-membrane bound soluble peptide decoy sequestering extracellular epidermal growth factor and inhibit activation of EGFR. The PTRM therapy construct was cloned into an adeno-associated viral plasmid vector and delivered to GBM cell lines. Total RNA was isolated from cells and reverse

transcribed using a random primer mix and target-specific primers to generate cDNA. PCR with specifically pre-designed primer sets will be used to detect therapy expression and alternative splicing of EGFR transcripts. Our novel approach to harness the cellular pre-mRNA splicing machinery and gene therapy to generate a targeted therapeutics may be an effective strategy in the treatment of GBM. This research was funded in part by Bristol-Myers Squibb, Johnson & Johnson, and the Independent College Fund of New Jersey.

Protein Concentration Determination in *Petunia hybrida* Flowers. Angel Jizzelle Smith and Farshad Tamari, Kingsborough Community College, Brooklyn, NY.

The developmental aspects of *Petunia hybrida* has not been well studied. Our goal is to determine the protein concentration ([protein]) in each tissue at different developmental stages (i.e. bud stages at four (-4), three (-3), two (-2) and one (-1) days before anthesis) for both reproductive and non-reproductive tissue, as well as in the mature flowers. We hypothesize that 1. The changes in protein quantities throughout development differ for reproductive tissues compared to non-reproductive tissues. 2. Proteins quantities in the reproductive tissues are temporally controlled and increase steadily throughout development; but reach their peak when the organ is reproductively active. To achieve our goal, flowers were dissected and 10 mg of each reproductive and non-reproductive organ was extracted in PBS. Samples taken from each flower included the sepals, petals, styles/stigmas, ovaries, and anthers. A conventional Bradford protein assay was used to determine the total amount of protein in each sample. Basic data manipulation was performed using Microsoft Excel 2013. All statistical analyses were performed using SigmaPlot 12. The highest [protein] was obtained in reproductive tissues and varied considerably in each. The highest [protein] was observed in anthers ($14.51 \pm 0.55 \mu\text{g}/\mu\text{L}$). As expected, for anthers the highest [protein] was obtained at dehiscence, while a buildup of proteins is observed from -3 to -1 days before anthesis (7.58 ± 1.48 and $10.887 \pm 0.77 \mu\text{g}/\mu\text{L}$, respectively). Also, as expected for ovaries, there is a build-up of proteins throughout development, ranging from $7.64 \pm 0.33 \mu\text{g}/\mu\text{L}$ to $10.69 \pm 2.02 \mu\text{g}/\mu\text{L}$. In pistils, [protein] builds up from -4 to -2 days before anthesis (from $6.30 \pm 0.87 \mu\text{g}/\mu\text{L}$ to $9.14 \pm 1.66 \mu\text{g}/\mu\text{L}$), but then drops back to $6.99 \pm 0.38 \mu\text{g}/\mu\text{L}$. Data provided in this research will shed light into the overall protein expression throughout development for this commercially important *P. hybrida*.

Migration of Pc3 Cells is Regulated by Hexim 1. Awa Sow and Manya Mascareno, SUNY Old Westbury, Old Westbury, NY.

An important checkpoint protein during cell proliferation is Hexim 1, which acts as an inhibitor for RNA Polymerase II-mediated transcriptional elongation. The post translation modification, tyrosine phosphorylation, of Hexim 1 was studied as it related to the Jak 2 tyrosine kinase and its inactivation effects. Hexim 1 was noted to be an ideal target for the Jak 2 kinase based on the identification of identical conserved YXXL motifs between Hexim 1 and Jak 2. The conserved motifs were target sites for Jak 2 kinase activity of tyrosine phosphorylation, and so it was hypothesized that the Jak 2 kinase phosphorylates Hexim 1. In order to test the significance of these conserved motifs of Hexim 1 substitution mutations of myc- Y291E and myc Y167G/Y291E were introduced in PC3 cells. Stable PC3 clone cell lines were created and maintained to overexpress wild-type Hexim 1, and to incorporate substitution mutations of myc- Y291E and myc Y167G/Y291E. To test the role, if any of the Jak 2/Hexim 1 axis a migration and invasion study was conducted using the PC3 cell lines. A wound healing assay was conducted with each of the three clones and showed that cell migration was inhibited in the single (myc- Y291E) substitution and further inhibition of migration was seen in the double (myc Y167G/Y291E) substitution mutation when compared to the overexpressed wild-type Hexim 1 cell line. Noting that the inability for Hexim 1 to be phosphorylated at these conserved YXXL motifs decreases the ability of the cells to migrate. Additionally, analysis of Vimentin, a protein marker for epithelial to mesenchymal transitions, was found to be enhanced in the overexpressed wild type Hexim 1 PC3 cell line. A transwell invasion assay was conducted using the same PC3 clones discussed earlier and found that the overexpression of wild - type Hexim 1 inhibited cell invasion and further inhibition was noted for the single substitution mutation and double substitution mutation. To conclude it was shown that mutations within the conserved YXXL motifs of Hexim 1 inhibited cell migration as well as cell proliferation. Since these motifs are identical to those found on the Jak 2 kinase it is thought that Jak 2 may be phosphorylating Hexim 1.

Pesticides and Alzheimer's Disease; High-mobility Group Box1 Protein (HMGB1) Pathway and Microglis Dysfunction. Erick Subillaga¹, Maria Entezari² and Mohammad Javdan¹. ¹Queensborough Community College, Bayside, NY and ²LaGuardia Community College, Long Island City, NY.

The etiology of most neurodegenerative disorders, such as as Alzheimer (AD) diseases attributes to genetic predispositions and exposure to harmful environmental factors. Limited studies have been documenting the low dose exposure to pesticides as one of the risk factors in developing of AD. The molecular mechanisms are not fully understood. The common features of AD is inflammation and protein aggregation. Microglia within the central nervous system playing a central role in response to chronic injury or inflammation. Microglia should constantly clear protein aggregations which are highly toxic to nerve cells. Over-activation of microglia have been well studied in developing AD. Therefore, we hypothesized that dysfunction of microglial cells could link the development of AD to the pesticide exposure. To test our hypothesis, we treated BV2 microglial cells with 1 and 5µg/ml of Permethrin for 24 hours. Permethrin as a pesticide which is used widely in and around households, including on pets, in mosquito control, and in agriculture. We assessed the effect of Permethrin on the cell viability, morphology and phagocytotic function of BV2 cells by performing the MTT assay, and the phagocytosis assay respectively. Our results showed Permethrin significantly reduced cell viability of microglia and drastic changes in their morphology compared to the control group. Phagocytosis was determined following Permethrin treatment by counting the number of phagocytic cells as well as the number of particles per cell after incubation with zymosan. Permethrin, significantly reduced the number of particles that were ingested by microglia compared to the control group. These data help us to clarify the role of pesticides as environmental risk factors in the genesis of neurodegenerative diseases and understand the particular molecular mechanisms that are involved in neurodegeneration and pesticide exposure. and with western blot assay we got proof of HMGB1 pathway in effects of Pesticides on microglia.

Novel Microwave Prepared Single-Walled Carbon Nanotubes-Ether Ionic Liquids Mixtures as Electrolytes for Dye-Sensitized Solar Cell Applications. Rawlric Sumner, Sharon Lall-Ramnarine and Tirandai Hemraj-Benny, Queensborough Community College, Bayside, N.Y.

There is a great need for the development of renewable green energy sources. In recent years, the use of dye-sensitized solar cells to replace silicon based solar cells have attracted much attention. However, more efficient electrolytes need to be developed to facilitate the increased commercialization of dye-sensitized solar cells. In this comparative study, the properties of single-walled carbon nanotubes (SWNTs) in methylimidazolium ionic

liquids (ILs) bearing ether and alkyl side chains were investigated as potential electrolytes for dye sensitized solar cell applications. The ionic liquids were prepared by reaction of 1-methylimidazole with the alkylhalide or alkoxyhalide to yield the halide salt. The halide salt was then converted to the bis(trifluoromethylsulfonyl)amide (NTf₂) IL. The structures of the ILs were confirmed using H-1 and C-13 Nuclear Magnetic Resonance (NMR) spectroscopy. SWNT- IL composites were prepared by cost and time efficient microwave irradiation. The conductivity of ILs and SWNT-IL mixtures were measured using a conductivity meter in a low moisture environment. Preliminary, conductivity values greater than 3.0 mS/cm were obtained for SWNT-IL mixtures making them promising electrolytes for electrochemical devices such as dye-sensitized solar cells. Rawlric is a participant in the NIH Bridges to the Baccalaureate Program at Queensborough Community College.

Identification and Characterization of Cyanophage, Cyanobacteria and potential Cyanotoxins in New Jersey Freshwater Bodies. Sally Tarabey, Christian Rios-Ruiz and Tinchun Chu, Seton Hall University, South Orange, NJ.

Lake eutrophication along with the rise of Earth's temperatures has led to growing environmental and public health concerns regarding the development of cyanobacterial harmful algal blooms (CHABs), especially in recreational and drinking water sources. Cyanobacteria, a photosynthetic prokaryotic microorganism, is what makes up these blooms and is found to be able to produce toxins that impact both humans and animals. This study investigated different approaches in detecting, identifying, and evaluating water bodies potentially containing such HABs. Water samples were collected from various water bodies throughout New Jersey. DNA Chelex extraction was performed on the water samples. Polymerase-chain reaction based assays were carried out using phyto-specific primer sets Psf/Ur and microcystin targeting Msf/Msr, followed by gel electrophoresis. Results indicated the presence of photosynthetic organisms in four water sites and microcystin-producing species in the Greenwood Lake water site. Flow cytometric assays were used to identify the genus of cyanobacteria based on the forward-scatter (FSC)/phycoerythrin (PE) profile. The results showed presence of *Synechococcus*, *Microcystis*, and *Cylindrospermum*-like species in Deverson and Rainbow Lake, *Microcystis* and *Synechococcus*-like species in Greenwood and Mirror Lake. Viral Plaque assays were done ineto detect cyanophage present in the water samples. Viral plaque assays were also carried out and the results indicated that the presence of cyanophage, host-specific predators of cyanobacteria, in all sites except Greenwood Lake.

The Role of Social Learning on Ethanol Preference and Courtship Behavior in *Drosophila melanogaster*. Erica Taveras, Matthew Romero, Arifa Altaf and Preethi Radhakrishnan, LaGuardia Community College, Long Island City, NY.

Our research has very important, broad implications of how social learning and behavior can modulate molecular changes within organisms thereby changing fitness and affecting reproductive potential. To attract a female *D. melanogaster* males perform an array of courtship behaviors to attract potential mates. Although these courtship behaviors are innate in males, particular social experiences can modify these behaviors and performance. We used three distinct sexual experiences to rear and generate three treatments of male flies. The first, 'successfully mated' treatment was subjected to free mating for 6-hour long sessions over 4 days. The second, 'rejected-isolated' treatment was subjected to 1-hour sessions with an hour of isolation intermissions 3 times a day over 4 days. The third, control treatments of 'non-mated' males were not given access to any females. After four days of courtship conditioning, males were assayed for 1) ethanol preference, 2) courtship behavior and 3) fertility. Our experiments reveal that social learning over a four-day period has an interesting correlation with courtship behavior. We find a surprising link between social learning, stress and varied NPF (Neuropeptide F) levels within the male flies, which thereby influence mating behavior and subsequent fertility.

EEG: Recognizing Brain Wave Patterns To Measure Degrees of Multiple Intelligences Using Howard Gardner's Theory and BIOPAC EEG Signals Acquisition Software Via Bioelectric Recording Electrodes. Alexander Thomas, Sheron Mehak, Daniel Rezae, Jonny Lazarte, Sara Lamcaj, Sana Kahn and Jenny Rose, Bergen Community College, Paramus, NJ.

An electroencephalogram (EEG) is a test used to detect electrical activity of the brain. This procedure tracks and records brain wave patterns. Small metal discs with thin wires (electrodes) are placed on the scalp, and then signals are sent to a computer to record the results. This Neurophysiology Project will be recording the electrical activity of the brain through EEG tests under the Biopac EEG Acquisition software using various stimuli. Identifor.com was the website used, based on Howard Gardener's Theory of Multiple Intelligences for a series of computer games to identify an individual's skills, abilities and interests. Brain waves are displayed on the screen as the subject played eight separate games. Each game represents one of Howard Gardner's intellect categories. The recorded brain waves are then analyzed to determine a correlation between success rates and specific wave forms. This study will reveal which brain waves are necessary for optimal learning and will give insight into how the brain acquires and retains information.

Characterizing the Endometrial Microbiome by Analyzing the Ultra-low Bacteria from Embryo Transfer Catheter Tips in IVF Cycles: Next Generation Sequencing (NGS) Analysis of the 16S Ribosomal Gene. Xin Tao^{1,2}, Nathan R Treff² and Tinchun Chu¹, ¹Seton Hall University, South Orange, NJ and ²Reproductive Medicine Associates of New Jersey, Basking Ridge, NJ.

There is growing interest in reproductive tract microbiome. The vaginal and placental microbiome have been partially characterized and shown to be related to obstetric outcomes. In this study, we developed a sensitive method to analyze 16S ribosomal RNA profiling from ultra-low bacterial counts, then studied the endometrial microbial environment by analyzing catheter tips after embryo transfers during in vitro fertilization (IVF). An extensive validation was performed on extracted DNA and culture lysates without DNA isolation from the single- or poly-microbial samples of *Escherichia coli*, *Proteus vulgaris*, *Staphylococcus epidermidis*, and *Bacillus cereus* by Illumina 16S V4 metagenomics workflows. The direct lysis method can reliably detect the genus or species taxonomic level for both single- and poly-microbial samples up to as little as 60 bacterial cells in the culture lysate. Over 99% total operational taxonomic units (OTUs) were assigned to correct genus or species. The endometrial microbiome at the time of embryo transfer was characterized by analyzing catheter tips with Illumina V4 metagenomics for 70 patients who underwent IVF cycles. *Lactobacillus* spp. were detected in all 70 samples. Other vaginal bacteria (*Corynebacterium*, *Bifidobacterium*, *Staphylococcus*, and *Streptococcus*) were also detected. The Illumina V4 metagenomics workflow with preamplification provided a rapid and sensitive method for the identification of bacterial genus or species in single- or poly-microbial samples and clinical embryo transfer specimen, which may shed the light to the relationship between endometrial microbiome and reproductive success.

Characterizing the Role of O-GlcNAcylation in Cardiomyopathy: Constructing a Transgenic Mouse. Anthony Tucker-Bartley, Rotimi Mesubi, Elizabeth Luczak and Mark E. Anderson, Wagner College, Staten Island, NY.

Cardiac disease is currently one of the leading causes of mortality in the United States. The emergence and proliferation of cardiomyopathy can be influenced by the co-morbidity of risk factors such as Diabetes. Prior research suggests that O-GlcNAcylation is increased in individuals who suffer from diabetes. The two enzymes responsible for this process, OGT and OGA are not fully understand. In order to study their role in the development of cardiomyopathy a transgenic mouse model of each gene was developed. The human variant of OGT & OGA were cloned into mouse vectors to simulate the effects of enzymatic overexpression of these genes. This paper address the process of cloning these genes and the future applications of this novel transgenic model of cardiomyopathy.

Examining the Effect of Oxidative Stress on *Tetrahymena thermophila* Sirtuins Exposed to High Glucose. Masood Usman and Ralph Alcendor, New York City College of Technology, Brooklyn, NY.

Sirtuins are a family of proteins that exist in almost all life forms. They are involved in important processes and functions such as lifespan expansion, protection from cell death and regulation of important cellular processes. Although sirtuins have been studied extensively, little is known on their role in oxidative stress in *T. thermophila*. *T. thermophila*, a ciliate protozoan, has been used as a model for studying the effects of a wide range of factors and stresses. These cells are very easy to work with and they are great models for eukaryotic cells, especially human cells. One of the most popular uses of *Tetrahymena* is to examine the toxicity of drugs and heavy metals. Oxidative stress is the imbalance between toxic form of oxygen or reactive oxygen species, and antioxidants. High levels of oxidative stress results in cell death, and damage to proteins and nucleic acids. Oxidative stress has also been shown to be involved in diseases such as cancer and Alzheimer's disease. Although oxidative stress has been studied extensively, very little is known on *T. thermophila*'s response to oxidative stress. Therefore, the goal of this project was to examine the effect of oxidative stress on sirtuins in *T. thermophila*. We hypothesis that sirtuins are involved in oxidative stress activities. Cells were incubated in glucose free and high glucose media for 24 – 48 h. high glucose has been shown to increase the levels of OS in cells. Following incubation, cell death and mRNA levels for selected sirtuins were assessed. Cell activity and cell number were both higher in media with glucose, 0.25, 0.5, 0.75 and 1.0%, compared to glucose free media. Sirtuin mRNA expression were consistently higher in glucose media. mRNA of antioxidant genes, were also higher in media with glucose compared to glucose free media. These results suggest that

A Comparison of Chlorophyll in Native and Invasive Species. Christiana Vazquez, Noran Abo--Donia, Annarose Levenberg, Noemie Rivera and Kathleen Nolan, St. Francis College, Brooklyn, NY.

The chlorophyll a content was calculated in three native species (red oak, ash, and shagbark hickory) and three invasive species (Japanese knotweed, porcelain vine berry, and pokeweed). Values were compared at two different times during the fall to see if photoperiod had an effect on chlorophyll content. The absorption and fluorescence spectra were also presented for each sample. Spec-20's were initially utilized for the first few samples, and then we switched to the Vernier Spectro-vis Plus for ease and time-saving in data collection. Future studies will include using chlorophyll a as a tool in water quality analysis.

The Changing Microbiome of Mercy College. Natalie Vegas and Davida Smyth, Mercy College, Dobbs Ferry, NY.

Recently at Mercy, a new dorm building was built called Hudson Hall, across from the old building Main hall. We aimed to determine if Hudson hall, before all the students moved in, would contain different microorganisms that are not human associated. We also wanted to see if any the students moved in, if this changed and if there was any transmission between Hudson hall and Main hall on the hands of the human residents. To examine what type of microorganisms were found before and after Hudson hall was occupied, we swabbed sites such as elevator buttons and door handles before the opening of the building. We swabbed 10 sites in Hudson hall that we believed would be high contact areas: three doors (two in ground floor and one in second floor) and four elevator buttons in the main elevator. A week after the students moved in, we sampled the same sites again to compare. After we swabbed them, we extracted the DNA from the swabs using a DNA extraction kit from MO BIO. The DNA we gathered was amplified using Polymerase Chain Reaction and deep-sequenced by Mr DNA. The 16S rRNA sequencing revealed a wide variety of microorganisms and indicated that there was a change in the bacterial residents of Hudson hall before and after the student residents moved in. A grand total of 1,066 different types of bacteria were present before and after. The data implies that there was transmission of bacteria from the hands of humans to the building. We have since begun to culture the bacteria in the various sites and identified that the viable bacteria found do correlate with the 16S rRNA data observed in the sequencing.

One Plus One is Greater than Two: Optimizing Synergistic Antibacterial Effect Between Tea Polyphenols and Two Antibiotics. Jonathan Valsechi-Diaz and Tinchun Chu, Seton Hall University, South Orange, NJ.

In September of 2016, the United Nations called antibiotic resistance the greatest and most urgent global risk. Even if antibiotics were used properly, bacteria would still slowly become untreatable. Past studies have suggested that, in some cases, natural compounds used in tandem with the regular dosage of antibiotics create a synergistic effect able to significantly inhibit bacterial growth. For this study, Erythromycin, an antibiotic shown to gain up to a 183.33% increase in zone of inhibition with black tea polyphenols, and Streptomycin, an antibiotic which multiple bacterial species are susceptible to, were combined with one of two natural compounds, black tea polyphenol Theaflavins (TFs) and lipid-soluble green tea polyphenols (pEGCG), to be tested on an array of model bacteria. These included the gram positive species *B. megaterium*, *S. epidermidis*, and *S. mutans* as well as the gram negative species *P. vulgaris* and *P. aeruginosa*. In the cases where 15 µg of Erythromycin or 10 µg of Streptomycin with a natural compound was able to greatly inhibit bacterial growth, the same combination was tested with half that dosage of antibiotic. In some cases, half of the standard dosage of antibiotic gave an LD₅₀ when combined with a low concentration of natural compound.

Effects of Tip60 and Paclitaxel on Breast and Lung Cancer. Himali Vashishtha and Daniel S. Ginsburg, LIU Post, Brookville, NY.

Tip60 is lysine acetyltransferase (KAT), coded by the KAT5 gene, which is involved in transcription, DNA repair, and apoptosis. It acetylates histones, the ATM protein kinase, and the p53 tumor suppressor, among other targets, making it a key regulator of cell homeostasis. Tip60 has been shown to be a tumor suppressor in most cancers but an oncogene in prostate cancer. Some of Tip60's functions take place in the nucleus while others occur in the cytoplasm. We were interested in investigating whether the nuclear localization of Tip60 is important for its tumor suppressor function. We hypothesized that decreasing Tip60 nuclear localization would reduce its antiproliferative effect in breast and lung cancer cells. To test this hypothesis, we mutated a putative Tip60 nuclear localization signal (NLS) and measured nuclear localization by immunofluorescence, as well as proliferation of two different transiently transfected breast and lung cancer cell lines. Tip60 lacking the putative NLS (Tip60 Δ NLS) had a significantly different localization pattern compared to the wild type Tip60 in all of the cell lines we tested, suggesting that we mutated the correct NLS. Although Tip60 Δ NLS showed significantly less nuclear localization than WT Tip60, neither construct reduced proliferation of breast and lung cancer cell lines in combination with paclitaxel treatment. These results suggest that we have identified the Tip60 NLS, but that the effects of changing Tip60 localization are more subtle than large changes in cell growth.

Seizure--associated Central Apnea in an Rat Model: Evidence for Resetting the Respirator Rhythm and Activation of the Diving Reflex. S.M. Villiere¹, K. Nakase², R. Kollmar², J. Silverman², K. Sundaram² and M. Stewart², ¹City University of New York Medgar Evers College and ²State University of New York Downstate Medical Center, Brooklyn, NY.

Respiratory derangements, including central and obstructive apnea, are consequences of seizure activity in epilepsy patients and animal models. Irregular, tachypnic breathing during seizure activity indicates an impact of seizure spread on respiratory rhythm generation. We studied episodes of central apnea, defined as periods > 1 second with no evidence airflow during plethysmography, in rats anesthetized with urethane and given parenteral kainic acid to induce recurring seizures. Our aim was to determine when in the respiratory cycle apneic periods start and if apneic periods end with 1) a reset of the breathing cycle to a common phase at apnea offset, 2) a continuation from the point at which the respiratory cycle was aborted at apnea onset, 3) a continuation from a point predicted by a continuation of the breathing pattern preceding apnea, or 4) a random re-entry into the respiratory cycle. One hundred ninety central apnea episodes (1.04 to 36.18 seconds, mean: 3.2 \pm 3.7

seconds) were recorded during seizure activity from 12 rats. The majority of apneic period onsets occurred during expiration (125/161 apneic episodes, 78%), particularly late expiration (82/125, 66%). Apneic period offsets were more uniformly distributed between early and late expiration (27%, 34%) and inspiration (16%, 23%). Central apneic periods were associated with a burst of high frequency activity in the EEG. These bursts could be used to identify periods where airflow was maintained, indicating that periods of no airflow were likely due to activation of the diving reflex. Events that were not masked by the diving reflex were completely stereotyped within individual animals, indicating a reset of the respiratory rhythm which was highly variable after about 1 second. We conclude that seizure-associated central apnea is associated with 1) a reset of the respiratory rhythm, and 2) activation of the diving reflex to suppress respiratory behavior.

Calcineurin B Subunit CA²⁺-Binding Domains Differentially Contribute to Phosphatase Function in Yeast. Anthony Wallitsch¹, Sean Connolly¹, Eric Muller¹ and Tami Kingsbury². ¹Iona College, New Rochelle, NY and ²University of Maryland Baltimore County, Baltimore, MD.

Cytoplasmic increases in intracellular Ca²⁺ result in the activation of the protein phosphatase calcineurin. Calcineurin is a heterodimer composed of a catalytic subunit (CNA) and a smaller regulatory subunit (CNB), which binds to Ca²⁺ and subsequently initiates phosphatase activity. CNB possesses four Ca²⁺-binding EF hand domains. To understand the contribution of individual EF hand domains to calcineurin function *in vivo*, a set of CNB1 alleles with mutations in EF1, EF2, EF3 or EF4 were generated. The mutant alleles were tested for their ability to support normal calcineurin function. Quantification of calcineurin-dependent reporter gene expression demonstrated that mutation of EF1 or EF2 substantially reduced calcineurin's ability to stimulate the reporter. Mutations in EF3 partially reduced the calcineurin dependent gene transcription but EF4 was not statistically different from wildtype. These results were consistent in assays that expressed calcineurin from a low copy or high copy plasmid. In addition, the use of a large extracellular concentration of Ca²⁺ to induce signaling along with more physiological treatments that are known to result in Ca²⁺ influx yielded similar results. Thus, EF1, EF2 and to a lesser extent EF3 play a role in phosphatase activation by intracellular Ca²⁺ and downstream events that promote calcium sensitive gene transcription.

The Influence of the Rs53576 Oxytocin Receptor Gene Polymorphism on the Modulation of Self-Assessed Stress Levels and Salivary Cortisol in High School Adolescents. X. Wang, V. Dolar, D.S. Ginsburg and A.L. Gucwa, Long Island University, Post Campus, Brookville, NY.

The hypothalamic-pituitary-adrenal (HPA) gland axis regulates endocrine stress responders such as cortisol, the major glucocorticoid hormone. Cortisol is involved in important physiological functions such as the regulation of glucose, calcium absorption and blood pressure. Cortisol is also released in response to stress-related events. Although it typically exhibits a diurnal pattern of release, it has been shown that clinical depression and psychological stress could alter cortisol levels. Another hormone involved in the stress response is oxytocin, stored and released by the posterior pituitary gland. It has been shown that oxytocin can decrease anxiety and protect against stress, particularly during times of social support. A single nucleotide polymorphism (SNP) Rs53576 was identified in the oxytocin receptor. Individuals with the G allele have been reported to be more optimistic and empathetic and handle stress well; however, those with the A allele were found to be increasingly reactive to stress. We wanted to investigate the relationship between cortisol, genetic variation in Rs53576 and self-perceived stress in high school adolescents as determined by survey analysis. Salivary cortisol levels were measured by an enzyme immunoassay kit and DNA was extracted from saliva, subjected to PCR, then used for restriction fragment length polymorphism analysis. Ordinary least squares and logistical regression models were used to determine the relationship between measured salivary cortisol levels, Rs53576 alleles and self-perceived stress. Although we would typically expect decreased cortisol levels in those with the G allele, we found no relation between salivary cortisol levels, Rs53576 and stress. One possibility is that our findings were influenced by the psychosocial and physiological changes that occur during adolescence and that stress responsiveness is still being formed. A more comprehensive study involving a larger cohort may better indicate whether these measures may be used as functional markers for stress tolerance in high school adolescents.

The Abundance, Host Preferences, and Economic Importance of Horse Flies (Insecta: Diptera: Tabanidae) in Sub Saharan Africa: A Case Study in Ghana, West Africa. Kerry-Ann Wilson, James A. Timbilla and Scott C. Sherman, Queensborough Community College, CUNY Bayside, NY.

Horse Flies (Insecta: Diptera: Tabanidae) are distributed in many biogeographical regions of the world including Sub Saharan Africa. The family Tabanidae is adapted to many habitats with considerable biological and economic importance. The adult hematophagous females of most known species prey on humans, domestic animals, and wildlife and also act as vectors of diseases such as anaplasmosis, anthrax, equine infectious anemia, loiasis,

surra, and tularemia. A case study in Ghana, West Africa began in 2014 on the perceived importance of three genera (*Atylotus* Osten Sacken, 1876; *Haematopota* Meigen, 1803; and *Tabanus* Linnaeus, 1758) of Tabanidae to humans, and to livestock care using a reconnaissance survey questionnaire developed by the authors. The questionnaire included Colored photographs to help agricultural, veterinary extension officers and farmers identify the fore-mentioned genera. Data collection included occurrence, seasonal abundance, livestock host preferences, and their economic importance. The results from 2014-2016, covering the Sahel and Semi-Deciduous ecological zones of Ghana indicate that the flies are most abundant during the wet season, which correlate with the rainfall distribution. In the Sahel ecological zone the Tabanidae are known to suck blood from and cause harm to cattle, sheep, horses, goats, donkeys, and humans while only cattle, donkeys and humans are affected in the Semi Deciduous ecological zone. The economic impact of horse flies on humans and livestock, and prospects for their control in sub-Saharan Africa are discussed.

The Population Dynamics of the Eastern Painted Turtle, *Chrysemys picta picta*, From a Connecticut Farm Pond. Amber Xaypanya, Christopher Meredith, Jennifer Tatnall and Antonios Pappantoniou, Housatonic Community College, Bridgeport, CT.

Aspects of the population dynamics of the Eastern Painted Turtle *Chrysemys picta picta* from a Connecticut farm pond were studied. Turtles were collected using hoop traps. Individual turtles were measured, weighed, had their gender determined, marked with a unique alphabetic identifier and returned back to their place of capture. Results of this study showed that female Painted Turtles averaged 127.9mm carapace length and weighed an average of 321.8 grams while males weighed an average of 268.5 grams and had an average carapace length of 124.2 mm. The female:male ratio was as 0.77:1. There was a decline in the catch per unit effort (CPUE) during the 2015 and 2016 collecting seasons. The decline in the CPUE may reflect the severe winters between 2013-2015.

Nesprin-1 Regulation of Organelle Positioning During Endothelial Cell Migration. Nicole Williams, Zayna King and Monn Monn Myat, Medgar Evers College- CUNY, Brooklyn, NY.

Cell migration is important for the development of multicellular organisms, immune response, wound healing, cancer metastases and angiogenesis. Here, we are studying the role of Nesprin-1, a family of proteins known to link the nucleus to the cytoskeleton (Rajior and Shanahan, 2013) in endothelial cell migration during angiogenesis. Our hypothesis is that Nesprin-1 affects organelle repositioning and the microtubule (MT) cytoskeleton during endothelial cell migration. We detected endogenous Nesprin-1 in HUVEC (human umbilical vein endothelial) cells using immunostaining which showed the localization of nesprin-1 in the nucleus and the cytoplasm. Through gm130 staining of the Golgi, this led to our discovery that

the Golgi is unchanged following siRNA mediated knockdown of nesprin-1. We also provide evidence that Nesprin-1 knockdown affected centrosome repositioning — centrosomes were localized further away from the nucleus. In current and future studies we will 1) determine the organization of the MT and actin cytoskeleton during angiogenesis and 2) test if Nesprin-1 depletion disrupts MT organization in the angiogenic sprouts. HUVECs will be untreated and treated with nocodazole and/or Taxol to depolymerize and stabilize MT, respectively. HUVECs will also be treated with latrunculin B and Jasplakinolide to depolymerize and stabilize actin, respectively. Our findings will further our understanding of cell migration in angiogenesis which can benefit anti-angiogenic therapy for cancer.

Identification of Plankton Species in the Hempstead Harbor Region Using DNA Barcoding. Oscar J. Zagalo and Nidhi Gadura, Queensborough Community College, Bayside, NY.

The Coalition to Save Hempstead Harbor (CSHH) has been working on the development of a report card for Hempstead Harbor, New York, with the purpose of clearly communicating the health of the harbor to the general public. Last summer there were reports of sludge and large pollen slicks mixed possibly with an algal bloom in the harbor. Water clarity suddenly came forth as a primary indicator of the health of the harbor, overshadowing the work that has been done with dissolved oxygen and bacteria. Samples were collected and tested for bacteria, but other tests needed to be done in order to definitively eliminate other forms of pollution. The body of water has been monitored and measured for water temperature, salinity, dissolved oxygen (DO), pH, secchi depth, turbidity (in NTUs), nitrates, and ammonia. All parameters were consistent throughout the season except for turbidity. We hypothesized that different plankton species might be responsible for the increase in turbidity. This research project was done to better understand this phenomenon by looking at water samples collected from several stations over the summer in order to identify the plankton that may be causing a bloom. DNA was extracted from the water samples, and appropriate primers were used for PCR to amplify the *rbcl* gene region that has been established for DNA Barcoding. Sequence analysis was done using DNA Subway program developed by Cold Spring Harbor Laboratory. Our results revealed *Bathycoccus prasinos* (Picoplankton) that was most frequently found in most sites of the water samples collected. However, to our surprise species like *Cucumis sativus* and *Cucumis melo* were found in water samples as well.

Direct PCR Detection, Cloning, and Characterization of Bacterial RubisCO Genes from New Jersey Soils. Stephanie Zapata, Anna Gonzalez, Margarita Kulko, Ryan Kim, Theranda Jashari, Aidan Holwerda, Tina Choe and Luis Jimenez, Bergen Community College, Paramus, NJ.

Ribulose-1,5-bisphosphate carboxylase/oxygenase, commonly known by the abbreviation RubisCO, is an enzyme involved in the first major step of carbon fixation, a process by which atmospheric carbon dioxide is converted by bacteria to energy-rich molecules such as glucose. Microbial DNA was extracted from temperate soils using the Zymo Microbe DNA MiniPrep protocol. RubisCO gene sequences were amplified by PCR using degenerate primers cbbLG1F and cbbLG1R. DNA fragments of approximately 800 base pair were detected in all positive soil samples. Clone libraries were constructed with the amplified DNA fragments by ligating the detected fragments with vector pCR®4-TOPO. Transformations were performed using competent Mix and Go Escherichia coli cells. Plasmids were isolated from each clone using the Zippy Plasmid MiniPrep and inserts were screened by PCR using M13 DNA primers. More than 100 clones were screened for the presence of RubisCO genes with 52 clones showing a positive reaction. DNA sequencing and BLAST analysis determined the identity of the cloned fragments. DNA sequencing of clone libraries showed that 88% of the sequences were related to Proteobacteria and 12% to Actinobacteria. The number one bacterial species detected were *Variovorax paradoxus* with 17% of clones. Other bacterial species detected were, *Bradyrhizobium elkanii*, *Pseudonocardia dioxanivorans*, *Rhodopseudomonas palustris* and *Starkeya novella*.

MACUB 2016 Conference Member Presentations

The Multicultural Lab: An Interactive Workshop on Ancestral origins, Race and Mitochondrial DNA. Sara Danzi-Engoron and Larisa Honey, Queensborough Community College, Bayside, NY.

This workshop was designed to address misperceptions about race and genetics while drawing on the strengths and opportunities of Queensborough's multicultural experience. The integration of knowledge and methodologies from biology and cultural anthropology are key elements of the workshop. In Dr. Honey's Anthropology class, students learn about evolution, natural selection, the construction of the concept of 'Race,' and the social consequences of racist policies that persist despite the fact that there is no genetic basis for race. Over each of the last 4 semesters, students from one of Dr. Honey's Anth101 classes came to the laboratory with Dr. Danzi Engoron to extract their own mitochondrial DNA (mtDNA) from cheek cells in saliva for sequencing and analysis to determine each individual's ancestral origins. Students learn that we are all almost identical in our DNA sequences and while we can determine certain ancestral origins from these sequences, it can be difficult to identify which individuals we will be most closely related to (genetically) based on physical characteristics. The workshop culminates in a classroom activity, which includes analysis of the relationships between all students in the classroom and comparisons to DNA sequences and haplogroups from all over the world. Students have embraced this activity and their new understanding of the superficiality of physical characteristics. In the past, we were limited to including only one class of 20-25 students each semester. We have now modified our protocol, acquired the necessary supplies and set up a mobile laboratory that allows us to bring the lab to the classroom. We have included almost 70 students this semester alone. This expanded workshop has allowed us to develop a pedagogical study to explore the impact of the students' personal involvement in the learning process.

Draw-Pair-Share - Activating Learning in First-year Biology and Anatomy and Physiology. A. L. Dell¹ and I. V. Ellison², ¹St. Francis College, Brooklyn, NY and ²Mercy College, Dobbs Ferry, NY.

College biology classes have a high drop-fail-withdraw (DFW) rate and a high exam failure rate while continuing to rely heavily on traditional lecture format, which does not engage students with diverse educational backgrounds and learning styles (Freeman et al. 2011). We previously showed that incorporating short draw-pair-share exercises into lecture helped students learn challenging concepts, such as the generation of the action potential. Students who engaged in these exercises and then explained their work to student partners outperformed their "listener" peers on both long- and short-term assessments. A bonus for instructors is that the drawings are a means to quickly assess student misconceptions. However, in allowing students to self assign as drawers or listeners, we may have inadvertently skewed our results, with more engaged and confident students taking the lead. Here we report the outcomes of an improved study design that aimed to engage passive students, as well as assess concept retention at additional time-points. We expanded this approach to a first year Anatomy and Physiology course for Nursing and Pre-Nursing students. Our results suggest that "draw-pair-share" is a valuable approach to engage struggling students, and contribute to deep learning across biological curricula.

Undergraduate Research: A Sustainable Anchor for Queensborough STEM Students. Nidhi Gadura, Queensborough Community College, Bayside NY.

This session will discuss how with the help of U.S. Department of Education MSEIP grant, undergraduate research is used successfully to engage underprepared and underrepresented students in STEM research. Attendees will walk away with research models they can use on their campus to increase STEM involvement of students, especially at predominantly teaching institutions. Best practices section will include: a) Using Cold Spring Harbor - Urban Barcoding Project, as a model system to engage freshman in research; b) Using updated curriculum to fund equipment in the labs; and c) Administration buy-ins include doubling the teaching space as research space. The main intended outcomes of this session are three fold; 1) To present the audience with results from our three tiered model used to engage freshman and sophomores in research at a community college, 2) To share best practices and undergraduate research models that can be duplicated on other campuses with limited resources, and 3) Share strategies on overcoming challenges when starting novel projects on campus which includes faculty development workshops.

The Effects of Antioxidants on Macrophage Function. Kathy Flores, Jaswinder Kaur and Tonya Hendrix, LaGuardia Community College, Long Island City.

The innate arm of the immune system is the first line of defense against disease-causing agents or pathogens. Macrophages, phagocytes of the innate immune system, recognize, engulf, and then destroy broad classes of pathogens. These processes require that the macrophages produce oxidants. However, oxidative stress caused by exposure of cells and tissues to oxidants results in signs of aging which include reduced functioning of the immune and cardiovascular systems as well as diseases like cancer. This goal of this study is to describe the function of macrophages in the presence of antioxidants. In particular, we are measuring the ability of a cell line of macrophages, RAW cells, to phagocytose particles of zymosan (derived from the cell wall of yeast) at varying concentrations of the antioxidant, N-acetyl cysteine (NAC). RAW cells phagocytose better at 10mM NAC; above and below this value phagocytosis decreases. At 50mM NAC, phagocytosis is minimal. The data supports our hypothesis that the beneficial effects of NAC are limited. This study suggests, that while low levels of antioxidants are beneficial for immune cell function, higher levels are not.

Evolutionary Perspectives in Health and Disease. Nasreen S Haque, New York Medical College, Valhalla, NY.

Evolutionary medicine offers a unique perspective in understanding health and disease but is ignored by the medical community and often omitted in scientific programs. The result is that an uninformed generation of physicians/ scientists continue to emerge who ignore evolutionary perspectives; missing the opportunity of including the fundamental principle of biology in healthcare and research. Thus, there is a need for training programs where evolutionary perspectives are included in the school curriculum. Essentially, such programs would offer the necessary background on evolutionary mechanisms and provide tools for development and testing of ideas. However, these programs must also be designed keeping the student perspective in mind. The students comprise of individuals with different background, experiences and learning skills with diverse career objectives. The key to retention and successful outcomes depends upon acknowledging individual viewpoints within groups, encouraging strategic partnerships and providing long term commitment to continued education. Encouraging collaboration within and outside the groups allows for sharing of ideas and combining individual skills with more predictable outcomes. Recognizing the potential of each individual within a group early in this process will allow better use of resources in long term partnerships. This can be done by forming a global network(s) of scholarship and communication such as the EvolMed Basics global network that we have developed at New York Medical College. Once these practices are in place, increased participation within the medical and scientific community is expected.

The Use of DNA Barcoding to Teach Students the Importance of Classifying Biodiversity. Antonia M. Florio, St. Francis College, Brooklyn NY.

Students can develop an awareness and concern for biodiversity loss by learning to identify the organisms around their neighborhoods or campuses. In response to this, we have implemented DNA barcoding curriculum at St. Francis College (SFC) to identify species in New York City (NYC). Classifying species based on morphology can prove challenging for students because they often become frustrated by species descriptions and taxonomic keys. The use of DNA barcoding has been championed as a way to overcome this, while also providing an inquiry-based approach for student-driven research. DNA barcodes are short DNA sequences that contain enough polymorphisms to identify organisms to the species level. Students propose projects, collect samples, extract whole genomic DNA, and use PCR to amplify the appropriate gene for their taxonomic group (plants: chloroplast genes *rbcL* and *matK*; animals: mitochondrial *COI*; fungi: nuclear ITS; and bacteria: 16S rRNA). Successful PCRs (confirmed by gel electrophoresis) can then either be sequenced in-house or sent away to a company for Sanger sequencing. Students clean their data using any sequence editing program, and perform BLAST searches through Genbank to identify their samples. Students can also learn how to resolve evolutionary relationships by generating multiple sequence alignments and phylogenetic trees. This presentation will summarize how we have used DNA barcoding at SFC to introduce high school and undergraduate students to classifying NYC biodiversity. The pros and cons of the method are presented, and a framework for implementing the project over a semester is proposed.

The Highly Unusual Nasal Anatomy of Two Brontothere Genera, Extinct Relatives of the Horse and Rhino (Mammalia, Perissodactyla, Brontotheridae), Bryn J. Mader, Queensborough Community College, Bayside, NY.

The nasal anatomy of two brontothere genera (extinct relatives of the horse and rhino) is described, and appears to be unique among all Mammalia. The air stream through the nasal cavity, rather than being directed laterally to either side of the nasal septum, appears to be channeled through the center of a highly modified vomer (one of the bones that forms the septum). The internal nostrils (nares) are essentially closed and the functional air passageway has been re-directed backwards into the area of the nasopharynx, apparently exiting through an opening known as the vomerine notch. This unusual morphology occurs in the brontothere genus *Metarhinus* and it is developed to an extreme in its contemporary relative *Sphenocoelus*. The functional significance of so unusual a feature is uncertain, but may have to do with the supposed amphibious habits of these genera, or perhaps, was an adaptation enhancing olfaction. Brontotheres (also known as Titanotheres) lived during the Eocene Epoch, about 56 - 34 million years ago, in both North America and Central Asia. They belong to the family Brontotheriidae of the Order Perissodactyla (horse/rhino group). Both *Metarhinus* and *Sphenocoelus* lived during the Uintan North American Land Mammal Age (approximately 42-46 million years ago) and are members of the brontothere subfamily Dolichorhininae.

Integrating Epigenetics and Epigenomics in Teaching Discovery-based Laboratory. Shilpi Paul, SUNY College at Old Westbury, Old Westbury, NY.

To initiate critical thinking among students and engage them through the fun of discovery in learning science, there is an immense importance of designing discovery-based or guided-inquiry based laboratories and integrating them in semester long curriculum. This workshop will provide an overview of epigenetic mechanisms and using epigenomics tools to explore their importance in a wide range of biological processes, from cancer to behavior science and environmental biology. Epigenetics is the study of the heritable changes in gene expression without any alteration in the DNA sequences. This workshop will emphasize on the role of the epigenetic factors involved in determining various biological phenomenon. Educators will be able to explore the vast data sets and tools available to study Epigenomics and integrate the tools in classroom teaching to answer some important fundamental biological questions. Several biological questions will be discussed to identify potential discovery-based lab projects by utilizing molecular, biochemical and epigenomic tools.

Ontogeny and Histochemistry of Translator Apparatus in *Calotropis procera* - a Milkweed fromTropics. Kumkum Prabhakar, Nassau Community College, Garden City, NY.

Survival of plants in any ecosystem is based on how well they can protect themselves from predators and on the mode of pollination that ensures reproductive success. These adaptations may be morphological, embryological, anatomical or physiological. The milkweed family has many members that have evolved to survive extreme conditions in the tropics by modifying the way plants get pollinated. Orchidaceae also has some members with pollen aggregates known as pollinia. Orchidaceae and Asclepiadaceae were, at one time, the only families known to have complex aggregates of pollinia. With re-categorization of some plants from Asclepiadaceae to Apocynaceae, pollinia are reported in Apocynaceae too. Monarch butterfly and milkweed's symbiotic association is well known in North America. An Indian Milkweed, *Calotropis procera*, found in arid conditions in the tropics, is a short shrub with phytotoxic leaves, lavender flowers, and milky thick white latex. Rare visit by pollinators in addition to other abiotic factors might have caused *C. procera* and some members of Asclepiadaceae (milkweed family) to have pollen packaged in the waxy sacs of pollinia. These pollen sacs are attached to a complex carrier known as translator apparatus. Detailed insight on the ontogeny and histochemistry of the translator apparatus and importance of organismal botany for DNA barcoding will be presented. In addition to the anatomical details of translator apparatus formation, this PowerPoint presentation will include discussion on re-categorization of members from Asclepiadaceae into Apocynaceae and evolutionary relationship with Orchidaceae. This discussion will be based on the phylogenetic trees constructed using SmartBlast and DNA subway of some representative members of these families to explore their relationship.

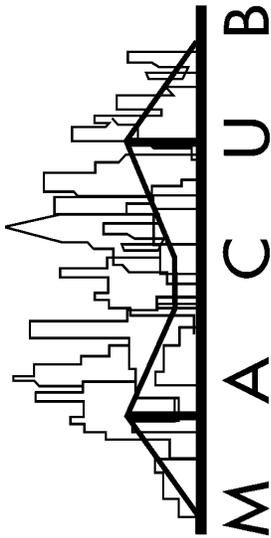
The Shoemaker Holly Forest, an Old Growth *Ilex opaca* Forest, Seaville, New Jersey. Richard Stalter, Shadin Aktak, Elizabeth Agolli, Kelly Crevani, Michael Labarbera, TuLoan Ly, Jaisima Minhas and Alice Cipriano Sivek, St. John's University, Queens, NY.

The objective of this study was to determine the arborescent composition and dominance of tree species at the Shoemaker Holly Rest Area, 39° 13' 44" N, 74° 22' 55" W., Seaville, New Jersey, mile marker 22.8 on the Garden State Parkway. This is the site of New Jersey's Shoemaker Holly, the state's oldest and largest (diameter) *Ilex opca*. Trees 1.37 meters above the ground with a trunk diameter 7.6 centimeters or greater were sampled using the point centered quarter method. Points were selected at the open woodland south of the Shoemaker Holly and again at the woodland north of the Shoemaker Holly. Number, relative number, frequency, relative frequency, basal area, relative dominance, and importance value were calculated for the trees encountered in our sample. *Ilex opaca*, American Holly, was the most abundant tree at site, composing 79% of the sample, and attained the highest relative dominance (percent basal area) and importance value with values of 55.1 and 189.9 respectively. *Ilex opaca* will most likely remain dominant here because of its longevity and lack of competition with arborescent species.

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