6th Annual S.T.E.M. Research Symposium

This research symposium is aimed at engaging the Barry community in learning about and share in the excitement of ongoing discoveries and research within the S.T.E.M. disciplines (Science, Technology, Engineering, and Math). Undergraduate students will present posters related to their past and current research in biology, chemistry, computer science, health science, information technology, mathematics, psychology, and physics.

DAY

Wednesday, April 16, 2014

TIME

9:00 AM - 1:00 PM

PLACE

Andreas 111

Barry University, Miami Shores, FL

Organized by Members of Barry University's STEM Committee:

Sumera Ackbarali MS, Khaled Deeb PhD, Sabrina Des Rosiers PhD, Maurizio Giannotti PhD, Christoph Hengartner PhD, Ricardo Jimenez PhD, Peter Lin PhD, Adina Oprisan PhD, Zuzana Zajickova PhD, & Anita Zavodska PhD

We gratefully acknowledge:

Sponsors from Barry University: Department of Biology; Department of Math and Computer Sciences; Department of Physical Sciences; School of Professional and Career Education.

Dedication of research mentors, support staff and undergraduate researchers.

Special thanks for assisting with the Symposium to:

Institutional Advancement, Department of Marketing and Communications, Dr. Flona Redway, Director of RISE/MARC programs, Ms. Michelle Aznarez, Technical coordinator of MARC program, Ms. Beth Culverson, Administrative Assistant III at Department of Physical Sciences, and undergraduate student volunteers.

BARRY UNIVERSITY - COLLEGE OF ARTS & SCIENCES

Department of Biology

1. Identification and characterization of genetic interactions between *cdc13-1*and y*KU80* in *Saccharomyces cerevisiae.*

Wesam Azaizeh, Jovans Lorquet, Alice Nakasone, KeiAuyndria Edwards, Sue -Ann Flores, Lauren Sanchez, Maxime Jean, Christoph Hengartner, and Leticia Vega (Department of Biology, College of Arts & Sciences, Barry University, Miami Shores, FL)

Telomeres are the physical ends of eukaryotic chromosomes that protect DNA ends from degradation and from end-to-end fusion. Telomeres consist of stretches of repeated C/G-rich DNA ending with 3' single stranded G-rich overhangs. The enzyme telomerase and accessory proteins such as Ku and Cdc13p maintain and facilitate telomere functions. In *S. cerevisiae*, *cdc13-1* is a temperature sensitive allele of Cdc13p, an essential telosome protein that binds to single-stranded G-tails to prevent telomere degradation. The Ku heterodimer, composed of Ku70 and Ku80, functions in DNA non-homologous end joining, recombination and telomere end protection. Yeast cells lacking Cdc13p or the Ku complex have uncapped telomeres and long single-stranded G-tails. This study examines the effects of mutations in yKU80 on *cdc13-1* strains. We introduced a library of 125 mutant yku80 alleles into the *cdc13-1* background by plasmid shuffle and determined the effects on viability and telomere end protection of the various yku80 mutant alleles. We found that 30 out of 125 yku80 alleles tested increased the temperature sensitive phenotype of *cdc13-1* strains, suggesting a telomeric end protection role for these mutant yku80 alleles. We are currently characterizing the telomere phenotypes of double mutant strains by Southern blot analysis. Initial characterization of *cdc13-1*, *yku80* double mutant strains show that temperature sensitivity may be uncoupled from telomere shortening in *cdc13-1*, yku80 double mutant strains.

Support by the NIH-NIGMS RISE Grant, R25 GM059244-14, Department of Biology, Barry University.

2. The importance of voltage-operated calcium channels (VOCC's) in astrogliosis.

Precious de Verteuil[1], Diara Santiago[2], Vilma Spreuer[2], Courtney Benson[2], Veronica Cheli[2], and Pablo Paez[2] ([1]Depatrment of Biology, Barry University, Miami Shores FL; [2]University at Buffalo, Buffalo, NY)

Astrocytes are glial cells within the central nervous system (CNS) that provide trophic support of the CNS immune response. Under pathological circumstances, astrocytes respond via "astrogliosis". During astrogliosis, astrocytes exhibit changes in cellular morphology and protein expression profiles. Initially, astrogliosis is necessary to stop damage from spreading to multiple areas within the CNS; however, increased astrogliosis promotes the formation of scar tissue--preventing undamaged neurons from reestablishing connections post injury. In this study we hypothesized that L-type voltage-operated calcium channels (VOCC) expressed on astrocytes are essential for astrogliosis. We tested our hypothesis using two astrocyte cell lines that varied in expression level of VOCC. Initially, we mimic a bacterial infection by treating cells with the endotoxin lipopolysaccharide (LPS). Our results show that the astrocyte cell line with high expression of VOCC showed significantly greater expression of classical astrocyte reactivity markers (i.e. GFAP and S100β) than control cells after LPS treatment. Importantly, LPS treatment was unable to induce astrogliosis in the cell line with low levels of VOCC expression, suggesting that the presence of these calcium channels is essential for astrocyte activation. Furthermore, we treated cultures with LPS in the presence of verapamil, a specific VOCC inhibitor. As expected, the presence of verapamil in culture medium drastically reduces the number of GFAP and S100β positive cells. In summary, our results suggest that VOCC may play a fundamental role in induction of reactive astrocytes, and indicate that inhibition of these calcium channels may be an effective way to prevent astrocyte activation.

Supported by Zannoni Summer Undergraduate Research Fellow (SURF) Award, University at Buffalo; ASPET, The American Society for Pharmacology and Experimental Therapeutics, University at Buffalo.

3. Genetic interactions of telomere binding proteins in yeast.

KeiAuyndria Edwards, Alice Nakasone, Wesam Azaizeh, Lauren Sanchez, Sue-Ann Flores, Jovans Lorquet, Maxime Jean, Christoph Hengartner, and Leticia R. Vega (Department of Biology, Barry University, Miami Shores, FL)

Telomeres are the physical ends of linear eukaryotic chromosomes that protect DNA ends from degradation and from anomalous end-to-end fusion with other chromosomes. Telomeres are composed of repeated TG rich DNA sequences that end with 3' single stranded G-rich overhangs. The enzyme telomerase and accessory proteins such as Ku and Cdc13p maintain telomeres and facilitate telomerase function. In the budding yeast, S. cerevisiae, Cdc13p is an essential protein that binds to the single-stranded G-tails to prevent their degradation. Cdc13p also functions to recruit the telomerase enzyme to telomeric ends. The cdc13-1 allele is a temperature sensitive allele of CDC13 that is defective in telomere capping. cdc13-1 mutants strains are inviable at temperatures above 30°C. The Ku70/Ku80 heterodimer also plays an important, but non-essential role in the protection of telomeres from degradation and participates in telomerase recruitment. Ku also plays additional roles in DNA metabolism including: non-homologous end joining and recombination. Yeast cells lacking Cdc13p or the Ku complex have uncapped telomeres and exhibit long, single-stranded G-tails. This study examines the effects of mutations in yKU80 on cdc13-1 strains. Using a genetic library of yku80 mutations, we introduced 125 mutant yku80 alleles into the cdc13-1 background by plasmid shuffle and determined the effects on viability and telomere end protection of the various yku80 mutant alleles in cdc13-1 strains. 30 out of 125 yku80 alleles tested increased the temperature sensitive phenotype of cdc13-1 strains, suggesting a telomeric end protection role for these mutant yku80 alleles. We are currently characterizing the telomere phenotypes of double mutant strains.

Supported by NIH-NIGMS RISE Grant, R25 GM059244-14, Barry University.

4. Do male house crickets (*Acheta domesticus*) communicate honestly to females?

Fabio Frech, Thalia Altamirano, Vania Arboleda, Diana Cordero, Edgar Garcia, Kamren Livingston, Shaynell Monreal, Andrea Roberti, Raynelle Salters, Domingo Sosa, Riann Zabaleta, and Michael P. Robinson (Department of Biology, Barry University, Miami Shores, FL)

Sexual selection theory predicts that females should prefer mates that have better genes because their offspring will inherit those genes and be more successful. In many species males signal to potential mates and females use that signal to their mates. This creates a potential conflict. Females are under pressure to respond to honest signals (i.e., those that carry reliable information) and ignore dishonest signals. Males, however, should produce signals that convince females to mate regardless of the male's actual quality (i.e., all males produce similar signals with some being dishonest). Signaling systems persist despite this conflict, because many signals are apparently too costly to be cheated by low-quality males and thus the signals remain honest. We are examining the relationship between the quality (i.e., degree of symmetry) of male house crickets (*Acheta domesticus*) and their courtship vocalizations to determine if this is an honest signal. In theory, symmetry indicates developmental stability (i.e., better genes) and symmetrical mates should be preferred. Previous results have indicated that symmetry in male house crickets is positively correlated with increased immune function. We recorded and analyzed the vocalizations of males. In addition, we took various morphometric measurements of the males including weight and measurements of the wing and legs. We then used these measurements to calculate fluctuating

asymmetry. We will discuss the signal's relationship with symmetry and its potential for use by females in mate choice.

Supported by the NIH-NIGMS MBRS RISE: R25 GM059244-13 award to Barry University.

5. Determining the role of hyaluronic acid synthase 2 (Has2) on metastasis of lung adenocarcinoma.

Talia Guardia[1], Vasilena Gocheva[2], and Tyler Jacks[2] ([1]Department of Biology, Barry University, Miami Shores, FL; [2]The David H. Koch Institute for Integrative Cancer Research at MIT, Cambridge, MA)

Lung cancer is the leading cause of cancer deaths worldwide, and metastasis accounts for over 90% of these mortalities. To better understand the molecular mechanisms of lung adenocarcinoma, we have turned to a genetically engineered mouse model with oncogenic K-ras activation and deletion of tumor suppressor p53, which closely recapitulates the histopathological features of the human disease. Previously, we had isolated and characterized tumor cell lines from primary non-metastatic and metastatic tumors and associated metastases. The top gene expression change between these lines was the expression of hyaluronic acid synthase 2 (Has2). Has2 is one of the synthases of hyaluronan (HA), a cell-surface-associated polysaccharide, which is a major component of the extracellular matrix. Several studies have reported an association between HA levels and tumor cell invasiveness; however, whether HA plays a role in metastasis of lung adenocarcinoma has not been examined. In this study, we investigated if overexpression of Has2 in non-metastatic cells increases their metastatic potential and if knockdown of Has2 in metastatic cells decreases their metastatic potential. However, for overexpression we did not see any change in Has2 expression at the mRNA or protein level. For knockdown, we had a 47% reduction in expression compared to the control. Based on proliferation and scratch assays performed using these knockdown cells, we saw a decrease in the proliferation rate under stress, and surprisingly, a slight increase in migration ability. Using these methods to alter the levels of Has2 expression will provide us with a better understanding of the molecular mechanisms underlying metastasis.

Supported by Massachusetts Institute of Technology Summer Research Program HHMI grant # 52006930.

6. Investigation of the wound healing process of adult *Danio rerio* wildtype zebrafish.

Victoria Hoelscher, Kevin Williams, Precious de Verteuil, and Brenda Schoffstall (Department of Biology, College of Arts and Sciences, Barry University, Miami Shores, FL)

Danio rerio (zebrafish) share many physiological and genetic characteristics with humans, making them an attractive model system for scientific research. Zebrafish have been shown to completely regenerate significant portions of heart, fin, and tail tissues without loss of function or formation of permanent scar tissue. However, zebrafish have not yet been established as a model to study regeneration in skeletal muscle and surrounding tissues. Specifically, the healing response following deep tissue burn puncture wounds has not yet been described. We hypothesize that zebrafish should completely regenerate skeletal muscle and surrounding tissues in response to this type of injury—making them an interesting wound-healing model. Our investigations have focused on determining the length of time necessary for zebrafish to fully recover from deep tissue burn puncture wounds. We first standardized methods for introducing a hot puncture wound completely through the myotomal muscle of the zebrafish, one millimeter below the dorsal fin. Healing was tracked by photographing wounded fish daily until healing appeared complete upon gross examination. Our results indicate that deep tissue burn puncture wounds require thirty days to heal, with minimal to no external scarring visible. Our preliminary results support the use of zebrafish as a model to investigate molecular regeneration processes following deep tissue burn puncture wounds.

Future studies will include time point sampling for analysis of the wound healing process at tissue and molecular levels. Findings could translate into applications for treatment of burn puncture wounds to skeletal muscle in humans, such as those inflicted during military combat.

*Supported by NIH-NIGMS MARCU*STAR Grant, T34 GM008021-29, Barry University (PD); DARPA Grant BAA 10-55, G. Packert and Barry University (VH); ASCB MAC Linkage Fellowship, B. Schoffstall.*

7. An analysis of the methylation response following ethanol exposure.

Shanika Kingston, Rafael Brango, Alec Davila, Anna-Lecia Lyn-Cook, Laura Mudd, and Stephanie Bingham (Department of Biology, Barry University, Miami Shores, FL)

Epigenetic modification involves intricate sets of chemical reactions that are responsible for the development and maintenance of every organism. It plays key roles in packaging and interpreting the genome, mainly during embryogenesis. Epigenetic mechanisms identified so far include sumoylation, methylation, acetylation, phosphorylation, and ubiquitylation. Each mechanism may either activate or inactivate gene function resulting in the regulation of patterns of gene expression without modifying the actual gene sequence. An intensification or reduction of these processes leads to aberrant epigenetic regulation, which may cause apoptosis, mitogenesis, or other malfunctions in tissue and cell development. Exogenous influences, including environmental and chemical exposures, have been linked to the modifications in customary epigenetic functions. We hypothesized that prolonged ethanol exposure may inhibit the function of genes deemed critical in nervous system development. An analysis of genetic methylation in response to ethanol exposure in genes responsible for nervous system development was performed using Real-Time PCR to test the hypothesis.

Supported by NIH-NIGMS RISE Grant R25 GM059244-14, Barry University; Department of Energy Grant DE-FG02-06CH11438; Department of Biology, Barry University.

8. Characterization of novel genes required for meiotic silencing by unpaired DNA.

Shanika L. Kingston[1], Logan M. Decker[2], Erin C. Boone[2], Hua Xiao[2], and Patrick K. T. Shiu[2] ([1]Barry University, Miami Shores, FL; [2]University of Missouri, Columbia, MO)

In many organisms, an abnormal gene copy number is a red flag for mischief, perhaps indicating a selfish element on the move. In the fungus Neurospora crassa, a gene not paired with a partner during meiosis is silenced by a genome surveillance mechanism known as meiotic silencing by unpaired DNA (MSUD). MSUD utilizes common RNAi proteins (such as Dicer and Argonaute) to silence its target sequences. Recently, we conducted a reverse genetic screen to identify new mutants defective in this process. The aim of this study is to characterize two of these newly discovered candidates, sad-f' and sad-g' (suppressor of ascus dominance-f' and -g'). Our silencing assays indicate that the two MSUD mutants dominantly suppress the silencing of different unpaired genes in varying degrees. Our results also show that while neither gene is required for vegetative growth, sad-f' is essential for sexual development. SAD-G' is localized in the nucleus.

Supported by NSF-REU, University of Missouri.

9. Investigation of IGF2 and YAP1 gene expression in proliferating zebrafish hearts.

Nicole H. Lopez and Brenda Schoffstall (Department of Biology, College of Arts and Sciences, Barry University)

Danio rerio (zebrafish) hearts respond to excessive cardiac overload stress with cardiomegaly via cardiomyocyte proliferation. Human hearts, on the other hand, have very little capacity for efficient cardiac cell division. Using our zebrafish model, we hope to identify specific gene pathways that act as the "switch" that "turns on" efficient cardiomyocyte proliferation. It is possible that these same signaling pathways exist in humans, but are "turned off" or blocked after heart development. We have determined that after 2 weeks of forced,excessive overload stress, zebrafish cardiomyocytes begin to actively proliferate. In this experiment, we have collected preliminary data concerning expression of IGF2 and YAP1—two genes previously identified as possible candidates for the cardiomyocyte proliferation switch. With the use of specifically designed RT^2 qPCR Primer Assays (Qiagen) in the conventional polymerase chain reaction (PCR), we have demonstrated expression of IGF2 and YAP1 in time point samples taken from zebrafish during a 4-week trial of forced exercise. Comparisons were made between exercised and non-exercised (control) fish. β-Actin and cardiac Troponin sequences served as positive controls to verify expression in cardiac tissue. Results from these PCR evaluations will be used to design quantitative PCR experiments to determine if these genes are upregulated during the proliferation phase. Currently, we are troubleshooting the PCR design using these preliminary data sets. Upregulation of either IGF2 or YAP1 in proliferating hearts may indicate that one of these genes has potential to be used as a "therapeutic switch" to turn on cardiomyocyte proliferation in human hearts.

*Supported by NIH-NIGMS MARC U*STAR Grant, T34 GM008021-29, Barry University and Barry University Faculty Incentive Grant, B. Schoffstall.*

10. Whitefly vector dietary conditions associated with the inoculation of foregut borne criniviruses.

Nicole H. Lopez[1], Angel Y. S. Chen[2], and James C. K. Ng[2] ([1]Department of Biology, Barry University, Miami Shores, FL; [2]Department of Plant Pathology & Microbiology, University of California, Riverside, CA)

As is typical of insect transmitted viruses, successful transmission of criniviruses entails three critical but poorly understood processes: virus acquisition, retention, and inoculation. Criniviruses, grouped within the family Closteroviridae, are plant viruses transmitted by whitefly vectors of the Bemisia tabaci species complex in a manner that involves the retention of virions in the vector's foregut, but does not require virus circulation through the vector. This study focuses on the inoculation stage of transmission for two criniviruses: Lettuce infectious yellows virus (LIYV), which is transmitted specifically by B. tabaci biotype A; and Lettuce chlorosis virus (LCV), which is transmitted by both B. tabaci biotypes A and B. This investigation seeks to identify conditions (pH and plant host components) that may play a role in mediating the inoculation of these criniviruses. Viruliferous whiteflies were fed liquid diet at pHs of 4, 7.4, and 9 (to be supplemented with or without sap purified from uninfected plants) to determine if and how effective viruses were inoculated under these conditions. Our experiments are aimed at testing the hypothesis that the inoculation/release of criniviruses that are retained in their vectors' foreguts is influenced by the pH and/or contents of the plant hosts on which the vectors feed. While the detection method for LCV is still under optimization, our results showed that purified LIYV virions were acquired by B. tabaci biotype A through the membrane feeding. In addition, detection of LIYV by RT and nested PCR in the inoculation buffers at pH 4 and 9 but not pH 7.4 suggested that pH is a potential factor to trigger the inoculation of virus by viruliferous whiteflies.These findings have potential consequences on the improvement of plant health and productivity upon which humans and animals depend for their survival.

*Supported by the NIH-NIGMS MARCU*STAR Grant, T34 GM008021-29, Barry University.*

11. *In vivo* characterization of Protein Interactions in S. *pombe* through confocal microscopy.

Jovans Lorquet[1], Julien Berro[2], and Thomas Pollard[2] ([1]Department of Biology, Barry University, Miami Shores, FL; [2]Department of Molecular Biophysics and Biochemistry, Yale University)

Protein interactions are essential to the understanding of cellular functions. Most techniques to characterize and quantify protein-protein interactions are done *in vitro* and require protein purification (pull down assays, NMR, X-ray etc…). This study describes a new *in vivo* approach to characterize protein interactions using *Schizosaccharomyces pombe* as a model organism. Our system relies on fusing a protein of interest to a spindle pole body (SPB) protein to enable us to anchor the protein of interest at the SPB. A putative target protein is tagged with GFP. Interaction between the protein of interest and the target protein is monitored by screening for fluorescence at the spindle pole body using microscopy. To validate our approach, we tested the interaction of the Actin-capping proteins, Acp1 and Acp2 in our assay. Using a PCR based approach, we fused our protein of interest (Acp2p) to specific anchor proteins of the spindle pole body. Acp1-GFP was used as a target. We examined the ability of two SPB proteins to function as anchors. Thus far, we have constructed 8 strains of which 4 were found to exhibit localized protein interactions. We are currently planning on coupling our new approach to quantitative fluorescence microscopy methods to quantify *in vivo* the stoichiometry of these protein complexes. This *in vivo* approach may allow us to analyze interactions in a physiological setting.

Supported by the Raymond and Beverly Sackler Institute for Biological, Physical and Engineering Sciences, Yale University. NSF, NSFDBI-1156585.

12. Identifying UV-reflecting patterns in fish by using ultraviolet photography.

Kevin S. McCarty and Michael P. Robinson (Department of Biology, Barry University, Miami Shores, FL)

Ultraviolet (UV) radiation (200-400 nm) is located towards the short wavelength, high frequency end of the electromagnetic spectrum. Although ultraviolet radiation is not visible to the human eye some animals, including many coral reef fishes, use it as a potential source of communication. Two species of Pacific coral reef damselfishes (Pomacentridae) possess complex ultraviolet facial patterns. These ultraviolet facial color patterns can be used for territorial aggression, identification, or other forms of communication. Damselfishes have evolved a range of social systems and color patterns in visible light. UV-reflectance has the potential also to vary with social system. To gain a better understanding of the variation in UV-reflectance, we followed a methodology described here for photographing these fishes. This is a modification of an earlier methodology developed by Siebeck (2004). The fishes were placed into a small single sided UV-transparent Lexan aquarium, which enabled us to photograph the fishes without harming them. A Nikon D3000 digital SLR camera was modified by having its internal filter removed and replaced by a UV-transmitting filter. This filter allowed only ultraviolet radiation between 250 to 400 nm (with a peak near 360 nm) to pass through to the camera sensor. We photographed both sides of the fish allowing a comparison of asymmetry.

Supported by a Barry University Faculty Senate Mini-Grant.

13. Quantitative and qualitative analysis of biofilm production in *S. aureus.*

Alice Nakasone[1], Jarred Fiedler[1], Federico Lin[1], Jody-Ann Chinloy[1], Leticia Vega[1], Christoph Hengartner[1], and Gerhild Packert[2] ([1]Department of Biology, College of Arts & Sciences; [2]College of Health Sciences, Barry University, Miami Shores, FL)

Biofilm formation is a survival mechanism some bacteria have evolved. Biofilms allow bacteria to adapt and survive host defense systems and harsh environmental conditions. As such, biofilms contribute to

bacterial pathogenicity. A crucial component of biofilms is the polysaccharide matrix, which allows adherence of bacteria to surfaces. The focus of this study was to explore different qualitative and quantitative methods to analyze the biofilms formed in static cultures of *Staphylococcus aureus*, a clinically important Gram-positive facultative aerobe. Biofilm formation in DIFCO nutrient and thioglycollate broths were examined and compared via microtiter plate assays and microscopy. A crystal violet staining method determined that only 24 hours was required for biofilm formation and that the thioglycollate broth promoted biofilm production. Additionally, to determine the presence of a polysaccharide matrix, *S. aureus* biofilm cultures in a microtiter plate assay were treated with diastase. Samples treated with diastase had less measureable biofilm when measured by absorbance following crystal violet staining and by microscopy than untreated controls. These data suggest that under our laboratory conditions S. aureus produces a polysaccharide rich biofilm matrix. These studies will help us with our long-term goal to identify chemicals and treatments that may inhibit biofilm formation.

Supported by the DoD-DARPA Grant, HR0011-11-1-006 , College of Health Sciences, Barry University, and NIH-NIGMS RISE Grant, R25 GM059244-14, Department of Biology, Barry University.

14. Retinoic acid down-regulates CDKs and induces differentiation of TF-1a cells.

David Novo, Talia Guardia, and Tang Hu (Department of Biology, Barry University, Miami Shores, FL).

All growing cells undergo a cell cycle. In mammalian cells the eukaryotic cell cycle is divided into four distinct phases: G1, S, G2 and M phases. Viable cells may also remain for prolonged periods in a resting state called G0 phase. In G0 phase the cells either have irreversibly exited the cell cycle for a particular process or function, such as differentiation, or return to G1 with appropriate stimulation. The cycle of a normal cell is controlled by a group of proteins termed cyclin-dependent kinases (CDK). Cdk only becomes active when bound to a regulatory subunit called cyclin. All-trans retinoic acid (ATRA), the metabolite of vitamin A, has been reported to induce several Acute Myeloid Leukemia (AML) cell lines to differentiate to granulocyte-like cells, which is mediated by the nuclear retinoid acid receptors (RARs) and retinoid X receptors (RXRs). These heterodimeric complexes regulate DNA transcription by binding to the promoter regions of various target genes in the presence of co-activators and induce transcription of target genes. TF-1a is a leukemia cell line established in 1998 (1). In this study, we investigated whether ATRA could induce differentiation of TF1-a cells and a possible link between the differentiation and expression of CDKs. Our preliminary data shows that vitamin A inhibits cdk4 and cdk6. At a high concentration (10-4M) ATRA induced significantly induced granulocytic differentiation of TF-1a cells, evidenced by reduced size and increased ratio of nucleus to cytoplasm, and macrophage-like changes. The cells treated with DMSO (solvent control) failed to induce differentiation of TF-1a cells. The correlation of inhibition of the CDKs and TF-1a cell differentiation is currently under investigation.

Supported by Faculty Incentive Grant and NIH-NIGMS MBRS RISE: R25 GM059244-13, Barry University.

15. The effects of embryonic ethanol exposure on cardiac function.

Peter Nwokoye, Mandy Carper, and Stephanie Bingham (Department of Biology, Barry University, Miami Shores, FL)

It is well established that embryonic ethanol exposure can have detrimental effects on tissue morphogenesis leading to developmental delays and cognitive deficits termed Fetal Alcohol Spectrum Disorders (FASD), of which Fetal Alcohol Syndrome (FAS) is the most severe form. Using zebrafish as a model system for FASD, we performed an investigation of the morphological effects of ethanol exposure

during embryogenesis. Analysis of ethanol-treated zebrafish embryos revealed striking effects on cardiac morphogenesis. As a follow-up to this observation, we examined cardiac function and discovered deficiencies in cardiac output. To further study the mechanisms underlying these abnormalities, several supplements that could potentially mitigate the observed cardiac defects will be tested. In addition, the expression of cardiac markers, such as *gata5* and *nkx2.5*, will be monitored following ethanol exposure.

Supported by: Department of Biology, Barry University; Faculty Incentive Grant, Barry University; Department of Energy Grant DE-FG02-06CH11438, Barry University.

16. Culture, isolation, and comparison of the microbiome of dissected guts from Opiliones, *Leiobunum* sp. and *Gasteracantha cancriformes.*

Jessica Ricketts, Nicholas Morales, and Brenda Schoffstall (Department of Biology, College of Arts and Sciences, Barry University)

Harvestmen (Order Opiliones, Class Arachnida) arachnids are closely related to "true spiders" (Order Araneae). While harvestmen are scavengers who masticate and digest solid food, true spiders use venom to externally pre-digest prey. To date, classification of the normal digestive flora of Opiliones has not been published. Our overall hypothesis is, given their internal digestion process, harvestmen will exhibit greater diversity of digestive microbiota in contrast to true spiders. We performed a screen to investigate this hypothesis by conducting aseptic dissection and isolation of the guts from 10 organisms of each species. Aerobic non-fastidious culture isolates from Opiliones *Leiobunum* sp. guts exhibited much greater diversity than those from our representative "true spider", *Gasteracantha cancriformes*. From *Leiobunum*, we identified eight species including *Pseudomonas luteola, Staphylococcus aureus,* and *Enterococcus faecium*. Four others were presumptively identified based on colony morphology and gram stains. In stark contrast to this diversity, we only isolated six different aerobic, non-fastidious colony types from dissected guts of *Gasteracantha cancriformes*. Of these, we identified S*taphylococcus hominis*, *Staphylococcus epidermis* and *Micrococcus spp*. Thus, our culture results have indicated a much more diverse microbiota in the gut of harvestmen when compared to that of true spiders. Our results contribute to current understanding in Opiliones ecology, and are important for expanding the relatively small body of knowledge published about these fascinating arachnids. We hope that results from this project might introduce these organisms as a new invertebrate model for translational studies in gut microbiome research.

Supported by ASCB MAC Linkage Fellowship, B. Schoffstall.

17. Exploration of neuronally enriched mRNAs in *C. elegans.*

Peter Rodriguez[1], Xico Gracida[2], and John A. Calarco[2] ([1]Department of Biology, College of Arts and Sciences, Barry University, Miami Shores, FL; [2]FAS Center for Systems Biology, Harvard University, Cambridge, MA)

Disruptions in neuronal gene expression can affect the onset and progression of neuronal disorders. We have utilized *Caenorhabditis elegans* as a model organism for studying neuronal traits. Using the translating ribosomal affinity purification (TRAP) method, we have previously identified neuron-specific, translated mRNAs with enriched expression in the nervous system of *C. elegans*. From these genes we selected eight candidates (c34d4.1, elc-2, cah-2, f13e9.15, del-2, klp-13, y53g8al.1, and drn-1) for further characterization. Our working hypothesis is that if any of these particular genes are deleted, we will be able to detect loss of specific function in *C. elegans* neuronal phenotypes. To investigate this hypothesis, we have begun to classify initial expression of the candidate genes in neuronal tissue, and evaluate our

initial attempts to knock out gene expression. Following transfection of *C. elegans* with GFP tagged candidates, we confirmed expression of tagged transcripts for elc-2, myo-3, drn-1, and f13e9.151 in neuronal cells of the F1 generation using RT-PCR. Localized expression of GFP-elc2 protein was identified via fluorescence microscopy. In addition, we used the CRISPR (clustered regularly interspaced short palindromic repeats) technique to attempt to disrupt expression of our candidate genes in *C. elegans*. Following Sanger sequencing of appropriate regions, gene sequences were analyzed for disruptions at the targeted loci. For candidate genes c34d4.1, elc2, cah-2, del-2, and f13e9.15, analysis revealed that we have been thus far unsuccessful in knocking down gene expression using the CRISPR method. Work on this project is ongoing; future work will focus on verification of all eight candidates in neuronal tissue, and continued attempts to knock down expression of candidate genes using the CRISPR method.

18. Karyotyping to investigate nondisjunction in the zebrafish model system.

Mariana Ruiz, Kellina Langaigne, Nicole Schtupak, Chris-Ann Xavier, Christina Dampman, and Stephanie Bingham (Department of Biology, Barry University, Miami Shores, FL)

Nondisjunction is the misalignment and subsequent unequal distribution of chromosomes in daughter cells prior to, or during embryogenesis. It is linked to a significant proportion of birth defects and miscarriages each year. We are using karyotyping as a means of detecting chromosomal abnormalities associated with nondisjunction. Karyotyping is a procedure that is used to visualize and classify the chromosome counterparts (homologs) different characteristics such as size and shape. Though factors such as maternal age have been identified as contributing factors to the incidence of nondisjunction, the underlying causes are largely unknown. We are therefore using zebrafish as a model system for testing various environmental stresses that may lead to chromosomal abnormalities. We hope use of this model will provide clues to the initiation of nondisjunction and the associated chromosomal abnormalities in humans.

Supported by NIH-NIGMS RISE Grant, R25 GM059244-14, Barry University; Department of Biology, Barry University; Department of Energy Grant DE-FG02-06CH11438.

19. Expression of IGF2 and YAP1 proteins in zebrafish proliferating cardiomyocytes.

*Johan Sanchez, Peter Rodriguez, and Brenda Schoffstall (*Department of Biology, College of Arts and Sciences, Barry University)

Every year, approximately 1 million people die from heart related disorders; by 2020 it will be the leading cause of death around the world. The inability for an adult heart to regenerate after an infarction makes saving lives affected by heart disease even more difficult. Previous research has showed that human cardiac cells are only able to proliferate until about 20 years of age, with the highest percentage of cell division between birth and the first year of life. *Danio rerio* (zebrafish) hearts, on the other hand, respond to excessive cardiac overload stress with cardiomegaly via cardiomyocyte proliferation. Using our zebrafish model, we hope to identify specific gene pathways that act as the "switch" that "turns on" efficient cardiomyocyte proliferation. We have determined that after 2 weeks of forced overload stress, zebrafish cardiomyocytes begin to actively proliferate. Not only is it important to evaluate which key genes are transcribed during proliferation —it is also important to evaluate the gene products, i.e. protein expression. Because of their involvement in cell division signaling, we have targeted IGF2, YAP1, and phosphorylated-YAP1 as proliferation switch candidates. We have designed Immunohistochemical staining experiments, Dot Blot Immunoassays, and Western Blot Immunoassays to examine the expression levels of these specific proteins in cardiac tissues of zebrafish. Tissue samples and protein extracts were taken from zebrafish that were put through a rigorous exercise program for 4 weeks to

promote excessive cardiac overload stress and cardiomyocyte proliferation. Four time point samples were collected throughout the exercise trial. The focus of these experiments will be to determine if IGF2, YAP1, or phosporylated-YAP1 are highly expressed during the period of active proliferation. If so, they may have potential to be used as a "therapeutic switch" to turn on cardiomyocyte proliferation in human hearts.

Supported by Barry University Faculty Incentive Grant, B. Schoffstall and ASCB MAC Linkage Fellowship, B. Schoffstall.

20. *Polycomb 2 (cbx4)* expression patterns during zebrafish (*Danio rerio*) development: *in situ* hybridization study.

Bertina Telusma, Chuco Glen, Stephanie Bingham, Gerhild Packert, Y-W. Peter Lin, and Teresa Petrino (Department of Biology, Barry University, Miami Shores, FL)

The *Polycomb* group genes (*Pc*) along with *trithorax* group genes (trx) were identified in many species as the group of genes that maintains transcription patterns during development. Recent findings have determined that *Pc* genes function as repressors by binding to specific response element regions of the DNA, thus modifying the chromatin state and silencing targeted genes. Our lab has previously cloned several *Danio rerio* (zebrafish) polycomb genes. The central hypothesis is that *Pc* genes are expressed at different times during embryonic development since the Polycomb proteins are used as epigenetic molecules that define the pattern of expression of embryonic genes. The objective of this study is to determine the spatial and temporal expression patterns of *Pc2* (cbx4) in the zebrafish embryo by synthesizing antisense RNA probes to carry out *in situ* hybridization experiments. Zebrafish is an ideal model organism to investigate *Pc* gene expression in vertebrates because fertilization is external and the embryo is translucent. To collect the embryos at different stages, the zebrafish (maintained in light-dark cycle of 14h:10h) were placed in breeding tanks with a ratio of 2:2 female-male. The plasmid DNA containing the isolated *Pc2* gene was retrieved through nutrient agar-ampicillin culture and plasmid miniprep. Plasmid linearized with restriction enzymes are used as templates in transcription reactions with digoxygenin-labeled UTPs to obtain sense and antisense probes for *in situ* hybridization analysis at various stages of development. The presence of *Pc* genes will help in understanding the mechanism by which these genes regulate the fate of cells during embryonic development and their own expression.

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21. A knockdown approach to studying *Pc1* gene function.

Van Williams, Susana Chan, Stephanie Bingham, Y-W. Peter Lin, and Teresa Petrino (Department of Biology, Barry University, Miami Shores, FL)

Polycomb (Pc) Group proteins are chromatin factors that repress the activity of developmentally regulated genes during embryonic cell differentiation. Based on in situ hybridization analysis, we are able to conclude that Pc1 (cbx2) is expressed as early as the 4-cell stage in zebrafish. This result indicates that the transcript is maternally derived thereby suggesting that this gene may be critical in early development. It is therefore the goal of this study to examine the phenotypes associated with disrupting the expression and function of Pc1 in the zebrafish embryo. To this end, we are using reagents known as morpholinos (MOs) to prevent translation of Pc1 in a targeted manner in the early (1–4-cell) stage embryo. Morpholinos function by binding mRNA and sterically hindering the translation initiation complex. Morpholinos for this study were designed to target a 25-bp region upstream of the translation initiation site of Pc1. As a first step, we are performing a dose-response study to determine the morpholino concentration at which

specific phenotypes can be observed without toxicity. Co-injection with a morpholino targeting p53MO will also be carried out in order to verify the specificity of the observed phenotypes as it is well established that morpholinos may sometimes produce off-target effects through p53 activation.

Supported by Department of Biology, Barry University.

22. CAK may play important role in PMA-induced differentiation of TF-1a leukemia cells.

Daria Vasilyeva, Pairat Dolinsky, Victoria Gonzalez, Palenzuela Iliana, and Tang Hu (Department of Biology, Barry University, Miami Shores, FL)

In mammals the cell cycle is controlled by a group of proteins termed cyclin-dependent kinases (CDKs). Full activation of the CDKs requires phosphorylation at threonine or serine residue within T-loop. CDK-activating kinase (CAK) is an enzyme complex that is capable of phosphorylating CDKs at the T-loop and is essential for CDK activities. The CAK is composed of CDK7, cyclin H, and Mat 1. Cyclin H is a regulatory subunit of CDK7 and Mat 1 is an assembly factor for the complex and regulates CAK substrate specificity. The effect of phorbol 12-myristate 13-acetate (PMA) on cell differentiation in several human myeloid leukemia cell lines have been reported. We have demonstrated that PMA induces macrophage-like differentiation of TF-1a leukemia cells, which involves in prolonged activation of MAPK (1). In this study, we investigated whether CAK plays any role in PMA-induced differentiation of TF-1a cells and involves in TGFβ signaling. TF-1a cells treated with PMA showed low levels of CDK7 as compared with the cells treated with DMSO (control solvent). This effect was dose dependent with 50%, and 100% reduction of CDK7 being observed 48 h after 10^{-6} M and 10^{-5}M of PMA were added to the cells, respectively. However, PMA had no effect on the expression of MAT1 (another subunit of CAK). Interestingly, TGFβ, which had no effect on differentiation of TF-1a cells, significantly upregulated CDK7 and MAT1, whereas in TF-1 cells TGFβ inhibits significantly proliferation of the cells without causing alternation in the levels of CDK7 and MAT1. As a negative control, all the cells expressed about the same levels of β-actin. Our data suggest CAK may play important role in PMA-induced differentiation of TF-1a cells.

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Department of Mathematics and Computer Sciences

23. Embedded image steganography

Lukas Bijaminas[1] and James Haralambides[1] ([1]Department of Mathematics and Computer Science, Barry University, Miami Shores, FL)

Image Steganography is the process of concealing an image within another, larger image and is considered an encryption technique. Generalized versions of the application enable the encryption of various forms of data such as text messages, files, and images. Extensive research has been done to produce encrypted data that withstand advanced cryptanalysis. We have implemented an embedded design for the image steganography problem on a Spartan-6 programmable device. Embedded systems are dedicated to specific tasks and can be optimized to reduce the size and cost of the product and increase its reliability and performance. The design is coded in the VHDL hardware description language and the software platform used is Xilinx Webpack. During the encryption process, least significant pixels bits of the host image are replaced by pixels of the hosted image. Decryption follows the reverse process. Replacement of the least significant pixel bits yields images that are undetectable to the naked eye. Pixel

mapping is performed randomly to prevent detection during cryptanalysis. A Fibonacci LFSR (linear-feedback shift register) produces the desired non-repeating random number sequence. Images are stored in DDR (double data rate) memory of the programmable device. To decrease latency, read operations for the host image are performed in bursts of four pixels while write operations for the hosted image are performed in bursts of 64 pixels with the use of DDR command, read, and write FIFOs. The entire process is tested on an Atlys Spartan-6 FPGA board. Data are transmitted from/to the FPGA using a simplified USB protocol at an approximate rate of 48 Mbps. A graphical user interface allows for FPGA board initialization and programming as well as the selection of images, encryption/decryption, and bi-directional data transmission.

24. Gradual admittance into network groups (GANG) protocol.

Lukas Bijaminas[1] and Ricardo Jimenez[1] ([1]Department of Mathematics and Computer Science, Barry University, Miami Shores, FL)

Attempts to secure existing Mobile Ad-hoc Networks (MANETs) on demand routing protocols through the use of cryptography, inherently stifles the mobility of the network. Cryptography limits the useable topology of the network to the domain of trusted nodes which hold keys. Since a MANET may encounter un-trusted nodes that have resources and reach to other networks that it may need; it is of paramount importance that MANET routing protocols have security mechanisms which allow un-trusted nodes that it may encounter to participate in the network. This work will make contributions to the ongoing effort to enhance MANETs security by developing a new approach that extends mobility to existing routing protocols that incorporate cryptography. This will be done through the creation and implementation of a unique trust management paradigm. An Analytical model of a new protocol, the Gradual Admittance into Network Groups (GANG) will be developed. This protocol extends the on-demand ad hoc routing protocol Secure Routing Protocol (SRP) which is a cryptography based extension to the on-demand routing protocol Dynamic Source Routing (DSR). In particular, the protocol will address issues pertaining to network availability and delivery, by allowing un-trusted nodes to participate in the network while still maintaining security. Furthermore, GANG will address two security threats namely black hole- and flooding- attacks which are currently not fully addressed by DSR and SRP. The current research develops and tests through NS3 simulations the GANG protocol to achieve a practical and implementable extension to DSR and SRP while taxing an acceptable penalty on network performance while adapting to changes in the environment, such as network topology and resource limitations of nodes. Furthermore, through simulations the work explores security strengths and weakness of the protocol for future work.

25. Mathematical Methods in Risk Theory and Applications to Automobile Liability Insurance.

Leo Lok Hin Law (Department of Mathematics and Computer Sciences, Barry University, Miami Shores, FL)

Actuarial science became a formal mathematical discipline in the late 17th century when E. Halley's famous mortality table permitted the mathematical analysis and calculations required in Life Insurance and Annuities. The underlying model is called the classical actuarial mathematics, or traditional life insurance model. In the 1930s and 1940s actuarial mathematics took a new orientation due to the powerful advances in probability theory and stochastic processes. The new actuarial model enabled the analysis of more complex problems by using a stochastic approach instead of the deterministic one as used in the past.

I underline the probability theory required in the Risk Theory within the context of their applications: stochastic processes with independent increments, Markov processes, compound Poisson process and the

risk process. The theoretical models developed in risk theory are applied to the calculation of risk, collective, credibility premiums in the automobile liability insurance. The mathematical statistics estimations of the theoretical parameters of the model are also discussed.

Advisor: Dr. Adina Oprisan, Department of Mathematics and Computer Science, Barry University.

26. Function visualization: 3D plotting of continuous functions and their derivatives using OpenGL.

Aarti Ragoonath[1], Hussein Allehyani[1], and James Haralambides[1] ([1]Department of Mathematics and Computer Science, Barry University, Miami Shores, FL)

Scientific visualization is very important in supporting data modelling and management, as well as computer-aided instruction for better understanding of abstract mathematical structures. We have created a tool that visualizes continuous functions of the form $z = f(x, y)$ and their partial derivatives $\frac{\partial f}{\partial x}$ and $\frac{\partial f}{\partial y}$ in three-dimensional space. The prototype utilizes library routines of the OpenGL system for rendering and transformational purposes. Functions are user-defined and employ a variety of common arithmetic operators and trigonometric functions. Operators include: addition, subtraction, multiplication, division, and the unary minus. Functions include: sine, cosine, tangent, square root, and logarithm. Functional expressions undergo multiple stages of scanning and parsing to check for errors (such as incorrect sequence of operands, unbalanced parentheses, unknown operands, extra white space, etc.). A binary expression tree T_f is constructed for a correct functional expression. T_f is traversed and differentiation rules are used to construct binary trees $T_{\partial f/\partial x}$ and $T_{\partial f/\partial x}$ that realize the corresponding partial function derivatives. Optimization techniques eliminate redundant expressions in the derivative trees. A recursive in-order traversal of each tree is used to evaluate the underlying expression and produce a collection of three-dimensional points $P(x, y, z)$. Re-evaluation of expressions occurs only on newly defined functions, thus reducing execution time substantially. An OpenGl framework allows for the definition of the system's viewpoint, bounding volume, surface characteristics, and projection method. The system uses orthographic projection. Transformation options include scaling and 3-dimensional rotation. Functions and partial derivatives may be plotted together for comparison purposes or separately for clarity. The system is capable of dynamic, speed-controlled plotting through motion. A frame per second rate is displayed to measure the efficiency of the algorithm.

Department of Physical Sciences

27. A python graphical interface to study star simulation data

Wesam Azaizeh, Maurizio Giannotti, and Michael Wise (Department of Physical Sciences, Barry University, Miami Shores, FL);

We present a python graphical and dynamical interface to study the output of stellar evolutionary data from the MESA code in a friendly and economical way. MESA is an open source code used by thousands of scientists worldwide. The interface collects and analyzes data from hundreds of output files and provides thousands of possible publishable-quality plots and several tools to study the results.

28. Metal – organic assemblies of tetrasubstituted porphyrins

Miriam Basden, Meghan Knol, and Tamara D. Hamilton (Department of Physical Sciences, Barry University, Miami Shores, FL)

Metal-organic polyhedra (MOPs) are 3-D structures consisting of organic ligands as the edges or faces and metal ions as the corners, able to encapsulate molecules and be hosts for chemical reactions. Porphyrins are large, naturally occurring, aromatic macrocycles perfect for building the faces of MOPs. This research seeks to synthesize MOPs, in particular metal-organic cubes (MOCs) using substituted porphyrins as the faces of the cube. So far, tetrakis(3-pyridyl)porphyrin (3PP), tetrakis(2-pyridyl)porphyrin (2PP), and tetrakis (2,3-dimethoxy)porphyrin (2,3-diOMeP) have been synthesized and purified, and synthesis of tetrakis(3,4-dimethoxy)porphyrin is underway. In the case of the latter two products, the methoxy groups will be demethylated to yield tetrakis(2,3-dihydroxy)porphyrin (2,3-diOHP) and tetrakis(3,4-dihydroxy) porphyrin (3,4-diOHP). These tetrasubstituted porphyrins were chosen because of their ability to act as exo-dentate ligands, meaning they can bind to a metal on the porphyrin exterior. Several attempts at self-assembly of 3PP with metal salts have been made with varying metals, anions, reacting and precipitating solvents, and reaction conditions, and here we will present one result where crystals suitable for X-Ray crystallography were obtained. Although not an MOP, this structure is promising for the future isolation of porphyrin – containing MOPs.

29. Preparation and optimization of organic-silica hybrid monoliths and characterization by capillary liquid chromatography

Denae Britsch[1], Gabriela Soto[1], Deepa Gharbharan[1], Anna-Marie Weed[1], Frantisek Svec[2], and Zuzana Zajickova[1] (Department of Physical Sciences, Barry University, Miami Shores, FL; [2]The Molecular Foundry, Lawrence Berkeley National Laboratory, Berkeley, CA)

In recent years, the development of highly permeable separation media, called monolithic columns, has immerged in capillary liquid chromatography. Monolithic stationary phases allow for high flow velocity due to low backpressure, therefore providing reduced analysis time. Traditional materials include silica-based and polymer-based monoliths. In our research, organic-silica hybrids have been prepared *in-situ* using [**3-methacryloyloxy)propyl]trimethoxysilane** as a monomer. The presence of an aqueous hydrochloric acid catalyst was necessary for hydrolysis and polycondensation of the trimethoxysilane functionality. The thermal polymerization of the methacrylate functionality was initiated using azobisisobutyronitrile. To achieve the optimum column performance, reaction conditions such as the temperature, time and concentration of catalyst and porogen (toluene) were adjusted. Progress of polymerization was followed by Fourier transform infrared spectroscopy and capillary liquid chromatography. Percent conversion of monomer to monolith was evaluated to provide information about the extent of the reaction completion. For the optimized column the following parameters were measured: efficiency of the column, selectivity for polar analytes, alkylbenzenes and steric compounds, and the degree of permeability. Minimum plate height of 9 μm (112,000 plates/m) was achieved for ethylbenzene at the optimal flow velocity of 0.27 mm/s. Methylene selectivity has been calculated as 1.28 ± 0.002, silanol selectivity as 0.13 ± 0.001, and steric selectivity as 1.70 ± 0.01. Consequently, the prepared hybrid monolithic columns prove to be efficient separation media in reversed-phase chromatography. In addition, inverse-size exclusion chromatography indicates the suitability of these monoliths towards separation of large analytes, such as proteins, and therefore possible application in proteomics.

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30. D-aspartic acid in oysters

Travis Comnick, Ramon Gutierrez, and George Fisher (Department of Physical Sciences, Barry University, Miami Shores, FL)

D-Aspartic acid (D-Asp) is an endogenous amino acid found in the nervous and endocrine systems of many vertebrates and invertebrates, where D-Asp has physiological importance as a neurotransmitter and a hormone regulator. Previous researchers found D-Asp in marine bivalves such as clams and mussels. We found and quantified D-Asp in oysters, traditionally thought to possess aphrodisiac properties. D- and L-Asp isolated from homogenized oysters were derivatized with *o*-phthaldialdehyde (OPA) and *N*-acetyl-L-cysteine (NAC) to form chiral diastereomers that were then separated by high performance liquid chromatography (HPLC) on a reversed phase C-18 column, eluted isocratically with sodium citrate-methanol (NaCit-MeOH) buffer, and fluorescence detection. Preliminary analyses show that approximately 3-4% of the total aspartic acid in oysters exists as the D-Asp enantiomer.

31. Synthesis of bitopic muscarinic antagonists and functionally selective agonists for pharmaceutical applications

Hangny Dao[1], John Boulos[1], and Jan Jakubik[2] ([1]Department of Physical Sciences, Barry University, Miami Shores, FL; [2]The Czech Academy of Sciences, Czech Republic)

The long-term research objective is to develop selective M_1/M_5 muscarinic agonists and M_2 antagonists for Alzheimer's, M_1/M_4 agonists for Schizophrenia, and M_1/M_4 antagonists for Parkinson's disease. In addition, selective M_2 and M_3 muscarinic antagonists are targets for chronic obstructive pulmonary disease (COPD), asthma and overactive bladder syndrome. We set to test the hypothesis that structural analogs of a newly synthesized bitopic antagonist and two potent and functional selective partial agonists would have improved receptor binding affinity and selectivity. The proposed hypothesis is partly based on a Shulman's model of drug-receptor interaction combined with chemical motifs known to achieve muscarinic receptor selectivity. This approach is quite innovative and has thus led to the discovery of the three muscarinic ligands mentioned above. The bitopic antagonist was shown to significantly slow down both the dissociation of N-methyl scopolamine (NMS) and acetyl choline in kinetics experiments while other two partial muscarinic agonists were shown to be potent, efficacious and functionally selective on muscarinic cell lines. The project's hypothesis will be addressed in experiments of three specific aims. Specific aim 1 proposes to synthesize structural analogs of the bitopic antagonist by repositioning and varying the alkoxy group of the phenyl hydrophobic linker whereas the nitrogenous head group will be substituted with other five and six-membered heterocyclic moieties. Specific aim 2 proposes to synthesize structural analogs of the two partial agonists by repositioning and varying the substituent bonded to the thiophene ring while.the nitrogenous head group will be substituted with other five and six-membered heterocyclic moieties. And finally, specific aim 3 proposes to assay all synthesized compounds on cell lines stably transfected with the genes of human variants of muscarinic receptors. We anticipate that this study will lead to the discovery of both agonists and antagonists with much improved binding affinity and selectivity profiles.

32. Novel nanomaterial for cancer diagnosis and treatment

Ayelet Delascagigas (Department of Physical Sciences, Barry University Miami Shores, FL)

There has been an emerging interest in the use the nanoparticles for cancer diagnosis and treatment in recent years. Linking the nanoparticles with biomolecules allows for sooner diagnosis and better drug delivery with higher affinity and specificity. But in order to do so two important factors were studied, fabrication and treatment. Nanoparticles provide a new class of biocompatible contrast agents that has great potential for cancer imaging and therapy. There are two types of nanoparticles most commonly used in studies today, polymeric and inorganic nanoparticles. Polymeric nanoparticles are flexible and biodegradable, and therefore can be used for controlled release of encapsulated biomolecules such as anticancer drugs. These in turn may be counteracted by an immune response nullify therapy. Polymeric,

these nanoparticles tend to be of relatively large size and not be ideal for drug delivery to solid tumors and metastatic sites. Inorganic nanoparticles, on the other hand, can be produced in the range of 5–30nm, which allows for a more suitable circulation in the bloodstream and into tumor tissues. More specifically the use of bio-nanocomposites (bio-NCP) and magneto-electric nanoparticles (MENs), allows for a controlled high-specificity drug delivery to ovarian and breast cancer tumor cells. Research has proven that bio-NCP and MENs provide the desired physical properties an promising material for early diagnosis and treatment. In the future, nanoparticle technology will have a dramatic impact on various disciplines ranging from biological research to oncology.

33. Synthesis and characterization of creatine ascorbate

Megan Henneberry, and Tony Wallner (Department of Physical Sciences, Barry University, Miami Shores, FL)

Creatine is most commonly used to enhance exercise performance and the use of creatine supplements has become widespread among athletes and exercise enthusiasts. Creatine also has been used in various studies as a potential therapeutic agent for neuromuscular and neurodegenerative diseases. Due to creatine's moderate solubility in water, the synthesis of a creatine salt with an increased solubility and stability are the goals of this research. More specifically, the optimized successful synthesis of creatine ascorbate is the main focus. Various stoichiometries of creatine and ascorbic acid were reacted using different solvents and reaction time in order to improve the product yield. Temperature and reaction time were also examined throughout the experiment to optimize the reaction. The synthesized products were examined and characterized with NMR. Additionally, the solubility of the new product was also determined.

34. Instrumental methods to quantitatively and qualitatively detect and recognize chemical warfare agents

Alexander J. Higa (Department of Physical Sciences, Barry University, Miami Shores, FL)

Chemical warfare agents (CWAs) have had an unfortunate negative impact on humankind over this past century. CWA attacks have been done utilizing organophosphates, which can be categorized into the G-series compounds composed of Sarin, Tabun and Soman. V-series class compounds are more potent, such as VX gas. These compounds have high levels of toxicity and exposure to CWAs can have detrimental effects on the central nervous system. Because their presence can affect humans at different ranges and their persistence in surfaces can pose a threat due to their long half-lives, the usage of chemical instrumentation in the detection of CWAs is crucial. Neutralization of CWAs using enzymes (e.g. diisopropyl fluorophosphatase from the squid *Loligo vulgaris* catalyzes the hydrolysis of several CWAs.) There are innovative methods to distinguish nerve agents by utilizing Gas Chromatography/Mass Spectrometry (GC/MS) and Nuclear Magnetic Resonance (NMR) to identify the phosphorus groups and the compound's derivatives. These studies provide detection for recognition purposes (qualitative), as well as determining how much of the CWA is present on a site (quantitative) for prevention of any accidental or malicious usage of CWAs.

35. Purification of porphyrins from mechanochemistry using entrainment sublimation

Victoria S. Hoelscher and Tamara D. Hamilton (Department of Physical Sciences, Barry University, Miami Shores, FL)

Mechanochemistry allows isolation of many products with less waste and cost than traditional solvent-based chemistry, making it ideal for development of synthetic protocols for industrial applications. Solvent-free synthesis using mechanochemistry has proven successful for a series of tetra-substituted porphyrins. Purification however, remains a problem since it is generally done by column chromatography, requiring copious amounts of solvent. Benzaldehyde and pyrrole are ball-milled in the presence of an acid catalyst to produce a mixture of cyclized precursors to a porphyrin. This mixture is then oxidized to produce tetraphenylporphyrin (TPP). However, after oxidation, the reaction mixture also contains a number of undesirable by-products. This project focuses on the purification of TPP from this mechanochemical reaction mixture by entrainment sublimation with either air or nitrogen as a carrier gas. Since TPP sublimes at around 400 °C, the purfication process will be carried out and optimized around this temperature using a tube furnace with a Pyrex processing tube. Results including purified yields from these experiments at varying heating times, and gas flow rates, and substituted benzaldehyde starting material will be reported.

36. Kinetic and solubility analysis comparing di-creatine citrate and tri-creatine citrate to creatine-monohydrate

Jason Llaneras and Tony Wallner (Department of Physical Sciences, Barry University, Miami Shores, FL)

Creatine is very well known within the nutrition and fitness industries as a nutritional supplement for increasing muscle size, endurance, and performance. Based on numerous initial studies, creatine has additionally shown an ability of reducing symptoms of neuromuscular and neurodegenerative diseases as well as improving cognitive function and treatment of depression. The most common form of creatine is creatine monohydrate and has moderate water solubility (16 mg/mL). Salt derivatives of creatine have become increasingly popular due to their potential to increase the solubility of creatine, thus increasing its bioavailability. Di-creatine citrate and tri-creatine citrate salt derivatives of creatine monohydrate were synthesized using citric acid in combination with creatine monohydrate under various conditions to maximize yields. ^{1}H NMR (Nuclear Magnetic Resonance) was used along with elemental analysis to confirm the chemical formulas of the synthesized compounds. A kinetic degradation study of the synthesized creatine salts was conducted using NMR. The solubility of each compound was measured and compared to that of creatine monohydrate to determine the usefulness of the synthesized products. This test showed that the water solubility of the two products is over 50 times that of creatine monohydrate. The effect of pH and temperature on the degradation of the creatine salts was also examined.

37. Optimization of mechanochemical oxidation of porphyrins

Taylor Sabol, Hannah Shy, and Tamara Hamilton (Department of Physical Sciences, Barry University, Miami Shores, FL)

Solvent-free chemistry is becoming crucial due to the demand for waste reduction and cost efficiency. Porphyrins are synthesized by a process in which large amounts of solvent for an acid-catalyzed condensation of pyrrole and aldehyde are used. This is then oxidized to create the porphyrin. We are investigating a solvent-free approach using a ball mill in order to produce the cyclized product, which is then analyzed by UV-VIS spectroscopy to confirm the presence of the porphyrin. We have been successful using a solvent-free approach for the condensation step and preliminary success with oxone as the mechanochemical oxidizing agent. Here we will present an optimization study, where we varied factors such as length of grinding time, size of grinding balls, presence of different grinding agents, molar ratio and tested various oxidizing agents including oxone, hydrogen peroxide, and hydrogen peroxide with the presence of sodium dodecyl sulfate.

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38. Pore surface modification of organo-silica hybrid monolithic columns through thiol-ene chemistry for application in micro-flow liquid chromatography

Gabriela Soto[1], Denae Britsch[1], Deepa Gharbharan[1], Anna-Marie Weed[1], Frantisek Svec[2], and Zuzana Zajickova[1] (Department of Physical Sciences, Barry University, Miami Shores, FL; [2]The Molecular Foundry, Lawrence Berkeley National Laboratory, Berkeley, CA)

In high performance liquid chromatography (HPLC), monolithic columns allow for the use of higher flow rates and resulting faster analysis due to the highly porous nature of the stationary phase. Hybrid monoliths possess a unique combination of both inorganic and organic functionalities on the pore surface. In this study, thermally-polymerized organo-silica parent monoliths were modified using thiol-ene click chemistry with the purpose of increasing surface hydrophobicity. Initially, the parent monoliths were prepared by polymerization of a monomer, 3-(trimethoxysilyl)propyl methacrylate (MPTMS), an aqueous hydrochloric acid catalyst, an initiator azobisisobutyronitrile (AIBN), and porogen, toluene directly inside of a 100 μ.m. I.D. UV-transparent fused silica capillaries at 80 °C. Subsequent was the on-column reaction of 1-octadecanethiol dissolved in toluene with exposed and easily accessible methacrylate functional groups at 80 °C on the pore surface of the organo-silica hybrid monolith. The reaction time was varied from 3 to 24 hours. The retention of benzene was used to monitor the changes in the surface hydrophobicity. The retention factor was calculated using retention time of benzene and unretained marker (thiourea). The mixture containing both analytes was separated under isocratic conditions using a mobile phase of 50% aqueous acetonitrile at a flow rate of 0.5 μL/min. The trend of an increased retention factor with extended reaction time has been observed until steady value was achieved. This proves successful pore surface modification and suggests the resulting application towards better retention of nonpolar analytes in reversed-phase chromatography.

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39. Comparison of different approaches in extraction of a parameter in a Linear Fit

Daria Vasilyeva, Maurizio Giannotti, and John Goehl (Department of Physical Sciences, Barry University, Miami Shores, FL);

We discuss some aspects of the linear fit analysis and show that the same data set may give different results for a physical parameter, depending on how the parameter is extracted. In particular, we focus on exchanging the axes and powering the data and discuss the question of compatibility between different approaches.

Department of Psychology

40. A Positive youth development model among Hispanic

Danielle Fair [1], Sabrina E. Des Rosiers[1], Seth J. Schwartz[2], Jennifer B. Unger[3], Lourdes Baezcondi-Garbanati[3], Juan Villamar[4], Daniel Soto[3], Monica Pattarroyo[3], and José Szapocznik[2] ([1]Department of Psychology, Barry University, Miami Shores, FL; [2], University of Miami, Miami, FL, [3], University of Southern California, Los Angeles, CA[3]; Northwestern University[4])

Positive youth development (PYD) model focuses on the skills fostered through personal strengths (Lerner, 2005). Research indicates that prosocial tendencies (PT)- the affinity for helping others as well as

Selection, Optimization, and Compensation (SOC) - a set of skills that are associated with adjustment are factors related to PYD. Several studies demonstrated the role of SOC and PT on adolescents' outcomes individually, but few have examined these patterns among immigrant youth. This study sought to identify a relationship between prosocial tendencies and SOC, and determine whether prosocial tendencies are differentially related to each domain of SOC in recent Hispanic immigrant adolescents. The study consisted of 302 Hispanic adolescents (M =14.51 (SD = 0.88) and 53% boys). The Prosocial Tendencies Measure (Carlo & Randall, 2002) and the Selection, Optimization, and Compensation (SOC) Questionnaire (Freund & Baltes, 2002) were used to measure PT and SOC respectively. Results from regression analyses indicated that PT accounted for 5% of the variance in selection, $F(6, 300) = 2.56$, $p = .020$, 8.8% of the variance in optimization, $F(6, 300) = 4.73$, $p = .000$, and 5.1% of the variance in compensation, $F(6, 300) = 2.62$, $p = .017$. Subtypes of PT predicted separate domains of SOC. Complaint prosocial behavior predicted selection ($\beta = -.25$, $p = .002$); public prosocial behavior predicted optimization ($\beta = .26$, $p = .003$); and complaint predicted compensation ($\beta = -.23$, $p = .005$). This study elucidates adjustment patterns associated with psychological well-being in recent immigrant youth.

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41. Self-esteem and acculturation

Esther Garcia [1], Sabrina E. Des Rosiers[1], Seth J. Schwartz[2], Jennifer B. Unger[3], Lourdes Baezcondi-Garbanati[3] , Juan Villamar[4], Daniel Soto[3], Monica Pattarroyo[3], and José Szapocznik[2] ([1]Department of Psychology, Barry University, Miami Shores, FL; [2], University of Miami, Miami, FL, [3], University of Southern California, Los Angeles, CA[3]; Northwestern University[4])

Many studies have shown that acculturation orientations are significantly related to adjustment outcomes among Hispanic immigrants (e.g., Des Rosiers et al., 2013). In general, a stronger orientation toward Americanism is associated with poorer adjustment compared to a stronger orientation toward retention of Hispanic culture. In the last decades, Hispanics have become the fastest growing immigrant population in the United States. Hispanic immigrants are likely to be youthful. About 20% are between the ages of 10 and 24 (PEW Hispanic Research Center, 2010). In addition, self –esteem has been shown to be an important individual characteristic during adolescence that influences a number of developmental outcomes. Despite the fact, many studies have identified relationships between acculturation and self-esteem, pattern of this association is not well known among recent immigrants. Consequently, the present study examined the relationship between acculturation patterns and self-esteem. The sample was drawn from a longitudinal study of Hispanic families that recently immigrated to Miami and Los Angeles. The sample consisted of 302 adolescents with a mean age of 14.51 (SD = .88) of whom 53% were boys. The Bicultural Involvement Questionnaire (Szapoznick et al., 1980) assessed adolescent's orientations toward Hispanicism and Americanism and the Rosenberg Self-Esteem Scale (Rosenberg et al., 1965) measured self-esteem. Results showed that Hispaniscism accounted for 24 % of the variance such that higher levels of self-esteem was significantly associated with Hispanic bicultural orientation $B = .25$, $P < .05$. The current findings have implications for understanding cultural processes that are associated with adolescent development.

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42. Adult perception of personality change in grandmothers

Maria C. Martinez and David Feldman (Psychology Department, Barry University, Miami Shores, Florida)

This research was conducted to examine the perception of personality change in elders; 65 years old and older, from the point of view of their grandchildren and how this affects the closeness of their relationship. This research tested the idea that Hispanic and African American college students will be closer and notice more the change of personality of their grandmothers than other ethnicities. Participants consisted of undergraduate students (N = 17) that completed an anonymous online survey in which they describe the personality and closeness of the relationship with their grandmothers at the present time and seven years ago. A two-way analysis of variance was conducted in each of the Big Five personality traits (Extraversion, Agreeableness, Emotional Stability, Conscientiousness, and Intellect) to evaluate the effect that personality change and race had on closeness. We found that there is a significant interaction between race and Extraversion, race and Agreeableness, and race and Emotion Stability on closeness seven years ago; As well as, race and Emotional Stability in the present. Follow-up tests were conducted, and found that there are significant differences between Hispanics and Caucasians, and African Americans and Caucasians in the way they perceive their grandmothers. Surprisingly, contrary to expected Hispanics rated their grandmother as more introverted and more disagreeable compare to Caucasians.

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43. Position dependency in object recognition

Maria C. Martinez[1] and George A. Alvarez[2] ([1]Department of Psychology, Barry University, Miami Shores, Florida; [2]SROH, Psychology Department, Harvard University, Cambridge, MA)

It was previously assumed that object recognition was independent from location, because there are neurons in human brain that respond to objects wherever they are located within the field of view. However, new research suggests that object recognition does in fact show position-dependency, suggesting that object representations are linked to specific positions in the field of view. The purpose of the present study is to substantiate that object recognition is constrained by position, and to examine whether this finding generalizes to all object categories. To test for position dependency, we present observers with a large set of objects (about 200), which are briefly presented and then "masked". Subsequently, we presented the same objects a second time, without the knowledge of the participants. The focal question is whether participants are faster at noticing the object the second time, and whether this trend is true only if the object appears in the exact same location. If so, it would suggest that the "object representation" that is being re-activated is linked to a specific position. Otherwise, if the benefit for seeing the same object again occurs for any location, it would imply that the reactivated object representation was position invariant. Preliminary analysis of the pilot study suggests that the results are in accordance with previous studies, showing that object recognition is position dependent. A follow-up experiment will focus on the kind of objects presented to subjects in order to determine if different types of objects (e.g., faces, scenes, etc.) will be processed differently.

Supported by SROH, Psychology Department, Harvard University

44. Using photovoice as a reflection tool in a psychology service-learning course.

David Tio, Eve Jacobson, Alante Simpson, and Dr. Pamela Hall (Department of Psychology, Barry University, Miami Shores, FL

Current research on service-learning places emphasis on the integrative and complementary role reflection plays in connecting community experience and academic learning. The present study seeks to evaluate photovoice as a pedagogical tool of reflection in a service-learning course. Photovoice is a community-based participatory research method whereby people identify, represent, and enhance their community through use of abstract photographs. People use cameras to record and represent their everyday realities. Their pictures promote group discussion about personal and community issues. The photographs used in photovoice serve as vehicles by which some aspect of the experience or situation reaches those who are exposed to the photographs. Twenty-two psychology students from a diverse private university were asked to use photovoice to answer the following questions: (1) what did you learn about yourself during the project? and (2) what did you learn about the children/adolescents you served during the project? The students participated in a ten-week project capturing pictures of their experiences. At the end of the project the investigator collected the pictures and analyzed them for common themes. For question one, 33 pictures were taken and the following themes emerged: a) academic enhancement, b) civic responsibility, and c) personal growth. Of the 33 pictures taken; 63% depicted civic responsibility, 39% depicted academic enhancement, and 81% depicted personal growth. For question two, 26 pictures were taken and the following themes emerged: a) community, b) self, and c) interpersonal relations. Of the 26 pictures taken; 58% depicted self, 50% depicted interpersonal relations, and 8% depicted the community. Photovoice proved to be an appropriate reflection tool to help instructors understand how students and recipients of service-learning are benefitting.

BARRY UNIVERSITY - COLLEGE OF HEALTH SCIENCES

45. Evaluation of histological staining techniques for visualization of bacterial biofilm in skin.

Daniel Packert, Sumera Ackbarali, Stephen Dunham, and Gerhild Packert. (College of Health Sciences, Barry University, Miami Shores, Fl)

Biofilms play a significant role in histopathology and are complex structures consisting of bacterial cells embedded in an extracellular matrix that contains polysaccharides, proteins and DNA. The biofilm matrix limits the effectiveness of topical antibiotic treatment in infected wounds and impedes wound healing and immune responses. The purpose of this study was to visualize the biofilm associated extracellular matrix utilizing standard histological techniques. The commercially available MatTek epidermal full thickness skin tissue model (EFT-400) was injured and infected for 24 hours with biofilm forming Staphyloccus aureus. Tissue for paraffin sections was fixed in formalin, microwave-processed and embedded in paraffin. Serial sections cut to 5 microns were stained with Periodic acid Schiff reagent, Calcofluor, modified Congo red/Ziehl carbol fuchsin stain and Feulgen reaction. Stained tissues were evaluated using light and fluorescent microscopy. A detailed analysis of the application of the different staining techniques in demonstration of biofilm associated extracellular matrix revealed that both, carbohydrates and DNA were present. Discussion of the value of each staining technique will be presented.

Supported by DARPA grant BAA 10-55 to G. Packert, Barry University.

46. Nitrite quantification and viability testing of MatTek Epidermal Full Thickness stem cell grown skin models post nitric oxide treatment.

Daniel Packert, Nathan Solomon, and Gerhild Packert. (College of Health Sciences, Barry University, Miami Shores, Fl)

Nitric Oxide (NO) has been shown to have multiple effects in the human body ranging from vasodilatation to antimicrobial properties. Recent studies suggest that NO may have potential use for

wound healing. NO, however has a very short half life, and must be indirectly measured using nitrites and nitrates as the source of measurement. Multiple ways of detecting nitrites have been developed, including colorimetric assays and chemilluminesence. The purpose of this study was to use MatTek Epidermal Full Thickness (EFT400) stem cell grown skin models for exposure to NO. The tissues were tested post exposure to quantify nitrites, measure the pH and evaluate tissue viability. Post exposure, one set of tissues was homogenized and samples were tested using Griess reagent test kits, and chemilluminsence to determine amounts of nitrites. This provides information of NO present in tissues post treatment. Separate samples were used in Viability assays (MTT) to determine the effect of the NO treatment on the tissue. The possible effects NO has on tissue viability are discussed.

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ST. THOMAS UNIVERSITY

School of Science, Technology, and Engineering Management

47. Primary neuronal cultures from the brainstem of adult zebrafish: a novel *in vitro* tool to study axonal growth across inhibitory chondroitin sulfate proteoglycans.

D. Diaz Martin[1], Isaac Chacon Rivero[1], Francelethia Shabazz[1], Katarina Vajn[2], Alexis Tapanes-Castillo[1], Martin Oudega[2], and Jeffery Plunkett[1] ([1]St. Thomas University, Miami Gardens, FL; [2]University of Pittsburgh School of Medicine, Pittsburgh, PA)

In the mammalian central nervous system (CNS), axons fail to regenerate after injury due to the presence of inhibitory molecules such as chondroitin sulfate proteoglycans (CSPGs). In contrast, adult zebrafish (*Danio rerio*) are capable of CNS axon regeneration. Specific populations of brainstem neurons can regenerate axons beyond a spinal cord lesion despite the presence of CSPGs. To investigate the axonal growth response of zebrafish brainstem neurons to CSPGs, we developed and characterized a novel, primary culture system. We hypothesized that brainstem neurons would respond distinctively to CSPGs *in vitro*. Our data demonstrate that one population was inhibited, while others had the ability to grow on or extend neurite-like processes across CSPGs. To further explore genes involved in overcoming inhibitory environments, we have also begun to investigate the effects of a pre-conditioning spinal cord injury (SCI) on axonal outgrowth *in vitro.* Data indicate that pre-conditioned brainstem neurons extend neurites into CSPG areas at a higher frequency than control neurons from uninjured fish. We hypothesize that a pre-conditioning SCI lesion enriches the expression profiles of genes involved in overcoming inhibitory environments within descending brainstem neurons. Our hypothesis is that the ability or disability of a neurite to grow across CSPGs is intrinsic to the neuron and likely involves unique sets of axon growth-related genes. Furthermore, we have extended our studies through the use of cell culture well inserts to further exploit our versatile model system. These inserts, which effectively isolate cell bodies from neuronal processes, will allow us to compare gene expression in axons versus cell bodies of brainstem neurons cultured under different conditions, including exposure to different substrates, cell types, and pre-conditioning injuries.

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48. *In vivo* and *in vitro* molecular biology techniques to study central nervous system axon growth and regeneration in adult zebrafish.

A. Lorenzo Gonzalez[1], A. Valls[1], Francelethia Shabazz[1], Katarina Vajn[2], Alexis Tapanes-Castillo[1], Martin Oudega[2], and Jeffery Plunkett[1] ([1]St. Thomas University, Miami Gardens, FL; [2]University of Pittsburgh School of Medicine, Pittsburgh, PA)

It has been well documented that the adult zebrafish (*Danio rerio*) can regenerate axons after injury to the central nervous system (CNS). Through applied molecular biology and *in vivo* as well as *in vitro* studies, we are investigating the mechanisms underlying this phenomenon. We established a primary culture system of adult zebrafish brainstem cells to analyze how brainstem neurons respond to chondroitin sulfate proteoglycans (CSPGs), and the CSPG Neurocan in particular. In mammals, CSPGs are axon growth inhibitory proteins. We cloned, transfected, and are currently purifying a Myc-tagged zebrafish Neurocan B (Myc-NcanB) protein to use as a substrate in our brainstem culture system. Preliminary data indicated that neurons responded similarly to CSPGs and Myc-NcanB. One population of neurons was inhibited by the CSPG or Myc-NcanB substrate, while other populations had the ability to grow on or extend neurite-like processes across the CSPG or Myc-NcanB substrate. In addition, we are performing gene expression knockdown studies, utilizing antisense morpholinos, to study the effect of the putative CSPG receptor, protein tyrosine phosphatase PTPsigma (Ptprsa), on neurite outgrowth. We hypothesize that decreasing Ptprsa levels will increase the number of neurites growing on or crossing into CSPG areas. Lastly, we are performing gene expression studies *in vivo* to evaluate the role of stem cells in spinal cord injury repair. We are preparing a mRNA DIG-labeled probe against the stem cell marker, nestin, to examine nestin gene expression pre- and post-spinal cord injury in the adult brainstem. These data will then be compared to immunocytochemistry data obtained with a Nestin antibody.

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49. The role of central nervous system stem cells in adult zebrafish neuron axon regeneration.

A. Badillo[1], A. Hernandez[1], A. Lorenzo Gonzalez[1], A. Cartagena[1], L. Yut[1], Francelethia Shabazz[1], Katarina Vajn[2], Alexis Tapanes-Castillo[1], Martin Oudega[2], and Jeffery Plunkett[1] ([1]St. Thomas University, Miami Gardens, FL; [2]University of Pittsburgh School of Medicine, Pittsburgh, PA)

Although post-embryonic neurogenesis is limited in the mammalian brain, zebrafish (*Danio rerio*) retain multiple proliferative neurogenic and stem cell niches throughout adult life. The focus of our research is to study how CNS injury affects the induction of neurogenic progenitor cell fates in the adult zebrafish brain. It has been well documented that in contrast to mammals, adult zebrafish recover functionally from a complete spinal cord transection injury. Damaged axons deriving from specific neuronal populations within the brainstem are able to regenerate across and beyond a spinal cord transection site. We hypothesize that spinal cord injury will induce an endogenous, quiescent population of brainstem progenitor cells that act to integrate and enable the regenerative response seen following spinal cord injury in the fish. We are currently examining regenerative brainstem regions for nestin stem cell marker expression pre- and post-injury. Prior to injury, nestin immunoreactivity was observed near ventricular areas, as well as in ventral brainstem regions, which contain nuclei from descending brainstem projection neurons. An increase in Nestin immunoreactivity was also observed in similar brainstem regions following spinal cord injury. We are currently analyzing Nestin expression surrounding retrograde labeled descending brainstem neurons capable of axon regeneration following spinal cord injury. Furthermore, we are preparing a nestin mRNA DIG-labeled probe to validate the nestin antibody data. Data from these Nestin expression studies will allow us to better study the role of stem cells in CNS axon regeneration.

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50. Quantification of zebrafish swimming behavior in order to determine the effect of gene knockdown on functional recovery after spinal cord injury.

H. Torres[1], I. Liser[1], Francelethia Shabazz[1], Katarina Vajn[2], Alexis Tapanes-Castillo[1], Martin Oudega[2], and Jeffery Plunkett[1] ([1]St. Thomas University, Miami Gardens, FL; [2]University of Pittsburgh School of Medicine, Pittsburgh, PA)

Unlike mammals, adult zebrafish (*Danio rerio*) recover from a complete spinal cord injury (SCI). The absence of functional restoration in the injured mammalian spinal cord is partly due to the chondroitin sulfate proteoglycan (CSPG) family of axon growth inhibitory molecules. Recently, a receptor for CSPG proteins, called receptor-type protein tyrosine phosphatase sigma (PTPσ), was identified. Mammalian studies have demonstrated that reducing PTPσ levels increases the growth response of axons neurons *in vivo* and *in vitro*. We have previously shown that PTPσ mRNA is present in adult zebrafish CNS. Our objective is to determine the effect of reducing PTPσ levels on functional recovery after spinal cord injury in adult zebrafish. We hypothesize that knocking down PTPσ protein levels, through the use of antisense morpholinos, will improve recovery. To determine the extent of functional recovery in PTPσ and control morpholino treated animals, we are performing a quantitative analysis of the distance swam by zebrafish using video-tracking and Image J computer software. We have obtained baseline measurements on the distance swam by injured, wild-type adult zebrafish at different time points after spinal cord injury. We are beginning to analyze the distance swam by injured, morpholino treated zebrafish.

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