

ERROR LIMITS, PADE APPROXIMANTS, AND POLE EXTRACTION FOR POWER SERIES. Joseph D. Rudmin, Dept. of Integrated Sci. and Tech., James Madison Univ., VA 22807. George Edgar Parker and James Sochacki have shown how to find, to any order, the Taylor Series approximation to a system of differential equations, if it exists. They and Paul and Debra Warne and David Carothers derived concise closed-form absolute error limits for this method. For most practical applications, a Padé approximant derived from the Taylor Series provides better fit than the Taylor Series. However, both Taylor Series and Padé Approximants have difficulty modeling poles in the solution. Often one can best model a pole by a change of variable, where the variable explicitly contains the pole. The change of variable can be found from the differential equations by eliminating the highest order feedback loop in the Parker Sochacki approximation, thus simplifying those equations.

TESTING A NEW METHOD FOR ESTIMATING BLACK HOLE MASS IN LOW LUMINOSITY ACTIVE GALACTIC NUCLEI. [Christina L. Hughes](#) & Mario Gliozzi, Dept. of Physics & Astronomy, George Mason Univ., VA 22030444. Black holes have recently become a primary topic in astrophysics as it is now known that each galaxy harbors one supermassive black hole at its center. Finding the mass of these black holes is critical to understanding the physical conditions around black holes and to shed light on the cosmological evolution of galaxies. Among techniques devised to determine black hole mass, dynamical methods are considered the most reliable. Unfortunately, such methods have severe limitations (they can be applied only on nearby galaxies) and have led astronomers to seek more far-reaching, universal methods. One recently proposed method relies on the ubiquity of the X-ray radiation in black hole systems produced by the Comptonization process and on the analogy between stellar and supermassive black holes. Recently, this method has been successfully applied to black hole systems accreting at a high rate, but has yet to be tested on low-accreting systems. This project explores the limitations of this X-ray based method by applying it to sizeable sample of low accreting black holes whose mass is known via dynamical methods and which possess high-quality X-ray data. In addition to preliminary research in astrophysics literature, this project encompasses data reduction of archival Chandra satellite observations as well as the analysis of spatial and spectral properties of each object. Both analytical and statistical techniques, applied in conjunction with an understanding of the physical processes at work, were used to ascertain calculated masses. The results of this project will provide a general understanding of the applicability and/or limitations of this method.

Biology **with Microbiology and Molecular Biology**

CONTRIBUTIONS OF CELL DIVISION AND CELL DEATH TO GROWTH AND METAMORPHOSIS OF PHARYNGEAL ARCH CARTILAGES IN THE FROG *XENOPUS LAEVIS*. W. T. Koch, V. K. Horne, & C. S. Rose, Department of Biology, James Madison University, Harrisonburg VA 22801. The pharyngeal arch cartilages of *Xenopus laevis*, including the lower jaw or Meckel's cartilage (MC) and the ceratohyal (CH), grow isometrically at tadpole stages and undergo radical shape

changes during metamorphosis. The cell behaviors that underlie the growth and shape changes of these tissues are largely unknown. We addressed this issue by mapping the patterns of cell death and cell division in frontal sections through the center of Meckel's cartilage and the ceratohyal in specimens ranging from early tadpole stages through the end of climax metamorphosis. Dying cells were identified under UV light by DAPI staining that indicated the blebbing or fragmentation of nuclei. Dividing cells were identified under phase contrast microscopy on the same sections based on the proximity and shape of nuclei, and the thickness and shape of the surrounding extracellular matrix. From these "maps", the distributions and frequencies of dying and dividing cells within cartilages were determined. Dividing and dying cells were found in both cartilages at all stages greater than NF 53. Cell division occurred at much higher frequency than cell death at all stages, and increased from NF 48 to 65 in both cartilages, as did cell death frequency in CH. There was no obvious change in the frequencies of either cell behavior at the transition from tadpole growth to metamorphic shape change for either cartilage. MC had generally higher cell division frequencies than CH (25-50% versus 10-35%), and CH had generally higher cell death frequencies than MC (0-20% versus 0-1%).

QUANTIFYING CELL CYCLE LENGTH DURING THE GROWTH AND METAMORPHOSIS OF PHARYNGEAL ARCH CARTILAGES IN THE FROG *XENOPUS LAEVIS*. V. K. Horne, W. T. Koch & C. S. Rose, Department of Biology, James Madison University, Harrisonburg VA 22807-2012. The pharyngeal arch cartilages of *Xenopus laevis*, including the Meckel's cartilage (MC) and ceratohyal (CH), grow isometrically at tadpole stages and undergo radical shape changes during metamorphosis. Questions of how frequently cells divide to contribute to the growth and shape changes of these tissues and their progression through the cell cycle were addressed by pulse labeling tadpoles at Nieukoop-Faber (NF) stages 55-56 and 58-59 with BrdU and following the labeled cells through to the end of mitosis. Tadpoles were sampled at 1-5 and 18 days after the injections and frontal sections through the center of each cartilage were made and photographed. Five categories of BrdU labeled cells throughout mitosis were recognized and counted. Interestingly, at least 60% of cells labeled in the S phase in MC and 45% of these cells in CH did not appear to advance to the start of mitosis. Of the cells that advanced through mitosis, those which completed cell division were seen at one day in MC and after two days in CH. The first to complete a second mitosis were seen three days after the pulse in MC; none were observed in CH. These and previous data support a model of cartilage growth wherein a percentage (5-30%) of cells in both cartilages enter the S phase at every tadpole and metamorphic stage, but only a small percentage of these advance to complete mitosis and second mitoses, and another small percentage stall at stages between the S phase and end of mitosis. BrdU appears to not be a good indicator of cell division in frog MC and CH.

CONFOCAL MICROSCOPY STUDY OF THE EMBRYONIC DEVELOPMENT OF THE VIVIPAROUS HOPLONEMERTEAN *PROSORHOCHMUS AMERICANUS*. Steven T Spindle & James M Turbeville, Dept. of Biol., Va. Commonwealth Univ., Richmond VA 23284. Recent studies of hoplonemertean planuliform larvae have greatly clarified their development and provided insight into larval evolution within the

phylum. However, an assessment of viviparous development using modern techniques is lacking. To help facilitate a comprehensive comparative evaluation of developmental diversity within hoplonemertans, we have initiated a confocal laser scanning microscopy investigation of the development in *Prosorhochmus americanus*, one of the few nemertean species that is both hermaphroditic and viviparous. Phalloidin staining reveals that the foregut, midgut, proboscis, and body wall musculature form early in development. These results are consistent with those for planktonic hoplonemertean larvae. The cerebral organs form from paired invaginations near the anterior end of the embryo as described for some hoplonemertean planuliform larvae. Acetylated tubulin antibody labeling shows that late stage embryos are uniformly ciliated, and in some specimens, a caudal ciliary cirrus is present, which is characteristic of species with planktonic larvae. The caudal cirrus may be interpreted as a vestigial structure in the non-swimming *P. americanus* embryos. Our preliminary observations provide no evidence for a transitory larval epidermis during the development of this species, but analysis of additional stages will be necessary to verify its absence. Analysis of the development of the nervous system in this species is ongoing, and both phalloidin staining and acetylated tubulin antibody labeling indicate that the cerebral ganglia and lateral nerve chords are present in early-stage embryos.

COURTSHIP SONG STRUCTURE AND MECHANISM OF A PARASITIC WASP. Justin P. Bredlau, Yasha J. Mohajer, Micheal L. Fine & Karen M. Kester, Dept. of Biology, Virginia Commonwealth University, Richmond VA 23284. Insects display a wide range of acoustic signals used in species recognition and courtship. Highly diverse parasitic wasps utilize wing movement or fanning to produce male courtship songs. Although courtship songs have been characterized for several parasitic wasp species, the mechanism of sound production has not been examined. We describe the male courtship song of *Cotesia congregata* (Hymenoptera: Braconidae) and examine the mechanism behind the production of its high amplitude component with high-speed videography (2,000 fps) and synchronized audio recording. The song consists of wing fanning followed by a series of pulsatile-sounding high amplitude “boings.” Boings are not produced by inter-wing contact or contact with the substrate or abdomen, and maximal sound amplitude does not occur during maximal velocity of wing motion, thereby ruling out a typical mechanical mechanism of sound generation. Instead, boings are generated at the termination of the wing down stroke when displacement is maximal but wing velocity is zero. Calculations rule out a whip-like action caused by rapid acceleration of the wing tip to supersonic speeds. Therefore, the sound is likely created by aerodynamic vortices produced by the sudden change in wing direction at the bottom of the stroke. Funding was provided by the Thomas F. Jeffress & Kate Miller Jeffress Memorial Trust.

GENOMIC COMPARISONS OF *BACILLUS* BACTERIOPHAGES: THE SEARCH FOR A *BACILLUS* PHAGE ANTI-RECEPTOR. Zein Al-Atrache & Lynn O. Lewis, Dept. of Biol., Univ. of Mary Washington, Fredericksburg VA 22401. Members of the *Bacillus cereus* group including *Bacillus cereus* (*B.c.*), *Bacillus thuringiensis* (*B.t.*), and *Bacillus anthracis* (*B.a.*) are gram positive bacteria with roles ranging from biotechnology to biowarfare. Much research regarding the taxonomy of these species has determined a less than 1% difference in their 16S rDNA sequences, suggesting

similar genomic sequences. Little is known about bacteriophages capable of infecting these bacterial hosts. Three novel *B.t.* subsp. Kurstaki bacteriophages were purified from soil samples throughout Virginia and characterized using restriction enzyme analyses. An observed characteristic of these bacteriophages is their ability to cross-infect hosts, including *B.t.* subsp. Al Hakam and *B.a.* subsp. Delta Sterne. The genomes of the three phages were sequenced and annotated to earn a more comprehensive understanding of the similarities and differences among their genomic sequences. Genomic comparisons revealed 96% coverage between the sequences of Phage Hakuna and Phage Megatron and only 5% coverage between the sequences of Phage JPB9 and Phages Hakuna and Megatron. Potential bacteriophage anti-receptors that confer host-phage specificity were also annotated in each of the phages' genomes. In the future, this knowledge gained will hopefully serve to elucidate the processes involved in bacteriophage infection of *Bacillus* hosts.

ANTIMICROBIAL PROPERTIES OF HONEY FROM ENVIRONMENTAL STUDIES ON THE PIEDMONT. Abdulla Hafid, Biology, Dr. Barney Bishop, Biochemistry & Dr. Thomas C. Wood, New Century College. George Mason University. Fairfax, VA. At Dr. Bishop's laboratory, honey collected from Environmental Studies on the Piedmont, was examined for the bactericidal impact on *E.coli* (*Escherichia*) *k9*. Honey showed to be effective in inhibiting bacterial growth and had greater rates of inhibition with higher honey concentrations. Analysis of the honey indicates the agent is water- soluble and size structure usually associated with that of a small carbohydrate or peptide. From this information, methods to introduce honey products in places of critical need can significantly increase the quality of life. However additional research must be done to precisely indicate the specific agent to be used effectively.

NOVEL ARCHITECTURE OF COSTAL CARTILAGE, IMPLICATIONS IN CHEST WALL DEFORMITIES. A Asmar, Stacey M^{1,2}, Fecteau A⁴, Werner A⁴, Kelly R Jr⁵. ¹Frank Reidy Research Center for Bioelectrics, Old Dominion University, Norfolk, VA, USA. ²Department of Pediatrics, Eastern Virginia Medical School, Norfolk, VA, USA. ³Division of General Surgery, Hospital for Sick Children, Toronto, Canada. ⁴Dept of Pathology, Eastern Virginia Medical School and Med Director of Laboratories, Children's Hospital of The King's Daughters, Norfolk VA, USA. ⁵Department of Surgery, Eastern Virginia Medical School and Pediatric Surgery Division, Children's Hospital of the King's Daughters, Norfolk VA, USA. Costal cartilage is a type of hyaline cartilage that is relatively uncharacterized in comparison to load-bearing cartilage. Abnormal formation of costal cartilage is associated with the congenital chest wall deformities pectus excavatum and carinatum. Our present study is part of a larger ongoing project in characterizing the ultrastructural biology of costal cartilage. Using immunohistochemistry, we analyzed the distribution of two important proteoglycans, Biglycan and Decorin. These proteoglycans, also known as small leucine-rich proteoglycans (SLRPs), play important roles in collagen fibril formation and organization. Our results showed that localization of pro-Biglycan, mature Biglycan, and mature Decorin were mainly in the territorial matrix, whereas pro-Decorin localized in the chondrocytes. The difference in functional properties of pro- and

mature forms are not well understood, and further investigation is needed to determine the functional impact of these results.

EXPLORING THE ROLE OF A NOVEL ZINC-BED DOMAIN CONTAINING TRANSCRIPTION FACTOR, CG3995, IN MEDIATING CYTOSKELETAL ARCHITECTURE DURING DENDRITE MORPHOGENESIS. V. Thota, S. Prakash, L. Sullivan, Y. Lau, M. Garland, S. C. Iyer, E. P. R. Iyer & D. N. Cox, Department of Molecular and Microbiology, George Mason University, Fairfax, VA 22030. Neurons are highly complex, polarized cells that come in an astonishing number of shapes and sizes, attributable largely to their elaborate dendritic branching patterns. As dendrites are primarily specialized to receive/process neuronal inputs, the specific morphology of the dendrite can govern neuronal function, signal integration, and circuit assembly. *Drosophila* dendritic arborization (da) sensory neurons have emerged as an exceptional model for dissecting the molecular mechanisms regulating class-specific dendrite development. Investigations using da neurons as a model system have revealed important roles for a broad range of biological processes including transcriptional regulation, cytoskeletal regulation, cell signaling and cell-cell interactions in mediating class specific dendritic architecture. Intriguingly, transcriptional regulation has been demonstrated to mediate da neuron dendritic morphology via modulation of the actin and microtubule cytoskeletons. We have recently identified and characterized a novel zinc-BED domain containing transcription factor, CG3995, which was found to be critical for the development of higher-order dendritic branches. To explore the potential functional mechanism via which CG3995 exerts control over dendritogenesis, we examined how changes in the levels of CG3995 expression may impact cytoskeletal architecture. To facilitate these studies, we have developed a unique transgenic *Drosophila* strain that enables simultaneous confocal imaging of F-actin and β -tubulin in live *Drosophila* da neurons *in vivo*. This approach has provided new molecular insight into the action of the CG3995 transcription factor in mediating cytoskeletal changes that ultimately result in class-specific dendritic patterning.

INVESTIGATING ABIOTIC AND BIOTIC INFLUENCES ON SPACE USE OF TWO SMALL MAMMALS IN SOUTHEASTERN VIRGINIA. Sarah A. Crawford & Robert K. Rose, Department of Biological Sciences, Old Dominion University, Norfolk VA 23529. Relatively little is known about the relationship between syntopic *Sigmodon hispidus* (hispid cotton rat) and *Reithrodontomys sp.* (harvest mouse). Literature on the associations between these two species suggests that competition may exist, particularly in southern populations; however both seem to be greatly affected by environmental and seasonal changes, making it difficult to draw any definite conclusions. Analysis of data from 34 consecutive months of live trapping on a former agricultural field in secondary succession in Southeastern Virginia revealed space use by both species to be more influenced by microhabitat preference than interspecific density. Hispid cotton rats were most influenced by ground elevation ($r_s = 0.45$, $p=0.0002$, $N=64$) and eastern harvest mice were most influenced by tree size and abundance ($r_s=0.52$, $p=0.00001$, $N=64$). As favorable habitat diminished through succession, hispid cotton rats stayed closer to food sources at the cost of moving to wetter, less appealing habitat, whereas eastern harvest mice were less affected, their distribution similar until the study site was abandoned. Hispid cotton rat and eastern

harvest mouse captures were not found to be significantly correlated. A finer examination over an extended study period may yield stronger associations but overall these two species appear to coexist, not compete, in Southeastern Virginia.

THE EPIGENETIC EFFECTS OF ENVIRONMENTAL POLLUTION ON AQUATIC SPECIES. Shawn Mitchell & Lisa Horth Dept. of Biol. Old Dominion Univ. Norfolk VA 23529. This is an ongoing study gathering data on epigenetics in *Micropterus salmoides*, *Callinectes sapidus*, *Fundulus heteroclitus*, and *Littorina littorea*. Methylation patterns may be globally reversed if many tumor suppressor genes are switched on which leads to observing the genomes of these species to see if their methylation rates may be reversed when extracted from a dirty environment and placed in a clean environment. Various tanks were set up in a controlled environment in our lab to simulate different measurable conditions of pollution. These species then had their genomes extracted and quantified. These genomes are currently be used to determine their rates of methylation. Once the rates of methylation have been determined, we will analyze our data to find a correlation between epigenetics and environmental pollution among species of the Chesapeake Bay watershed.

Posters

THE IMPACT OF LEAD TOXICITY ON CAENORHABDITIS ELEGANS POPULATION. Hunfa Asghar, Gita Sudama, Danial Khan, Anima Adhikari & Dr. Willet, School of Systems Biology, George Mason University, Manassas, VA 20110. The nematode, *Caenorhabditis elegans* (*C. elegans*) is an excellent model system in which to study fundamental biological processes of eukaryotic multicellular organisms. They provide ideal models for determining mechanisms of neurotoxins, such as the environmental contaminant, lead (Pb). The impact of lead toxicity (0, 250, 500 and 1000 parts per million [ppm]) on *C. elegans* populations was monitored. It was carried out by direct observations of the treated nematodes populations under the light microscope, over a period of nine (9) days. Life stages counts and live/dead ratios were recorded for each population. Population profile changes were observed with lead dose treatment. Increases in dauer *C.elegans* were observed with lead treatment. Decreases in population size and distribution were seen with lead treated *C. elegans* populations. The number of dead nematodes increased with lead concentration, over time.

A SYSTEMS GENOMICS APPROACH FINDS CANDIDATE GENES FOR NON-INSULIN DEPENDENT DIABETES MELLITUS. J. James, L. Jones, B.L. Sayre and G.C. Harris, Department of Biology, Virginia State University. Non-insulin dependent diabetes mellitus (NIDDM) is one of the most significant chronic human diseases, affecting over 20 million people in the United States (7% of the population). NIDDM is associated with obesity and characterized primarily by insulin resistance and impaired insulin production. In this study a multiple SNP (single nucleotide polymorphism) analysis was conducted to identify potential candidates for the disease. Mouse SNP data was mined from databases that included strains for accepted NIDDM models Tallyho (NIDDM model) and SWR/J (wild type). Expressed genes were captured in the form of mRNA, converted to cDNA, and analyzed for differential expression in 4 different tissues (fat, skeletal muscle, liver and pancreas). The results

show several of the gene candidates tested were found to be differentially expressed (DE) in various tissue samples; *Alpk1*, *Hfe2*, *Manba*, *Slc22a15*, *Slc30a7* and *Tchh11*. Future efforts can now focus attention and resources on these likely candidates for NIDDM with obesity phenotypes. These efforts confirm the significant impact a systems genomics approach can have on identifying potential candidates for multigenic inherited human diseases such as NIDDM.

THE EFFECTS OF POSITIVE AND NEGATIVE STRESSORS ON THE RATIO OF URIC ACID TO XANTHINE IN *CAENORHABDITIS ELEGANS*. Anima Adhikari, Gita Sudama, Danial Khan, Hunfa Asghar, Neeraja Podugu, Jenifer Isbister & James D. Willett, Department of Systems Biology, George Mason University, Manassas VA 20110. *Caenorhabditis elegans* (*C. elegans*) serve as an excellent model system in which to study fundamental biological processes of eukaryotic multicellular organisms. They are ideal models for determining mechanisms of action of neurotoxins, reactive oxidative stress and aging. In this study *C. elegans* populations were separated into three different cohorts; cohort 1 (young < 20 μm), cohort 2 (middle aged 20 - 35 μm) and cohort 3 (adult > 35 μm) to examine how each cohort react to positive (vitamins C and E) and negative (lead acetate and aging) stressors. Uric acid (uric) and xanthine (metabolites of the purine degradative pathways) concentrations were measured by the application of high performance liquid chromatography coupled to electrochemical detection (Coularray/HPLC). The ratio of uric/xanthine was calculated to demonstrate change in flux of the formation of uric acid, from xanthine, on exposure to the stressors. In one experiment, the *C. elegans* populations were age separated (into cohorts 1, 2, and 3) and then treated with lead acetate (0, 250, 500, 1000 ppm) for 2.5 hours. The Uric/Xanthine ratio increased with dose treatment for each cohort. In a second experiment, the *C. elegans* populations were grown with vitamin C [C] (0, 0.1, 0.5, 1.0 mg/ml), vitamin E [E] (0, 0.1, 0.5, 1 mg/ml) and vitamin C & E ([0.1 C + 0.1 E], [0.5 C + 0.1 E], [0.5 C + 0.5 E], [0.1 C + 0.5 E]) and age separated. The Uric/Xanthine ratio fluctuated with dose treatment for each cohort, due to the antioxidant properties of the vitamins.

INDUCTION OF APOPTOSIS BY NONSTRUCTURAL PROTEINS OF THE SINDBIS VIRUS IN *XENOPUS LAEVIS* EMBRYOS. Kaitlyn Childs, Jacob Graham, Kevin M. Myles, Ph.D. & Carla V. Finkielstein, Ph.D., Virginia Polytechnic Institute of Virginia, Blacksburg VA 24060. Viral infections are of interest as is related to virotherapy and host-virus interaction studies for mechanism construction that may result in knockout therapies to reduce or diminish virulence. *Sindbis* virus, an Alphavirus of the *Togaviridae* family, is transmitted by mosquitos; the endogenous strain is opportunistic, infecting immunocompromised individuals, elders and young children. The viral genome is divided into nonstructural (ns) and structural open reading frames. The nonstructural region encodes a polyprotein consisting of nsP1, nsP2, nsP3, and nsP4. In this study, we have found that the nonstructural protein nsP3 has the ability to induce apoptosis in an heterologous system and that this property is restricted to its N-terminus domain. Interestingly, whereas the nonstructural polyprotein is able to trigger apoptosis in *Xenopus* embryos when a polycistronic mRNA is injected in one-cell stage, neither nsP1, 2 nor 4 was able to accomplish this form of cell death on their own. Apoptosis was confirmed by measuring caspase-3 activity, visualization

of membrane blebbing, and cleavage of cyclin A2, a known caspase substrate in *Xenopus* early embryogenesis. Next, we investigated whether nsp3 pro-apoptotic activity was inhibited by interacting with anti-apoptotic members of the Bcl-2 family. Thus, *Xenopus laevis* embryos were injected with various nsp3 constructs in one-cell, collected at different times before MBT and will be analyzed for binding by immunoprecipitation. We hope to elucidate the mechanism behind apoptosis induction of the Sindbis virus in *Xenopus laevis* embryos in this way.

POINT MUTATIONS THAT DRIVE PIGMENTATION DIFFERENCES IN MOSQUITOFISH. Hampus A. Engstroem & Lisa Horth, Dept. of Biol., Old Dominion University, Norfolk, VA 23529. Melanic pigmentation is widespread in nature, and plays a crucial role in species fitness. The eastern mosquitofish (*Gambusia holbrooki*) melanic male morph is found in natural populations at frequencies typically maintained below 0.05 of the male population. A single albino western mosquitofish (*Gambusia affinis*) was found in nature and bred in captivity. To elucidate the genetic mechanisms that contribute to these two very rare phenotypes the genes encoding the melanocortin-1 receptor (MC1R) and enzyme tyrosinase (TYR) were analyzed for nucleotide sequence variation and gene expression differences. Sequence analyses of both genes revealed non-synonymous mutations in melanics and albinos as compared to the wild-types. Expression of both genes was higher in melanics compared to wild-types, and lower in albinos for both genes. These results demonstrate for the first time a correlation between gene expression differences and unique genotypes in the melanin biosynthesis pathway. (Supported by National Science Foundation, and Jeffress Memorial Trust.).

EFFECTS OF VITAMIN A METABOLITES ON APOPTOSIS. Anne M. Campbell and Rosemary Barra, Dept. of Biol. Sci., Univ. of Mary Washington, Fredericksburg, VA. 22401. Retinol is a metabolite of vitamin A found in all mammalian cells. Retinol must be converted to retinoic acid in order to be used by cells and the cis and trans isomers are common metabolites. Studies suggest that retinoic acid affects cellular growth and differentiation in a dose dependent manner. The objective of this experiment was to determine the effects of retinoic acid on CRL 1790 colon epithelial cells. The cells were incubated with 1mM, 10 mM and 100 mM of 13-cis retinoic acid and all-trans retinoic acid (ATRA). Cell viability was determined using the MTT assay and the concentration of CD95 was determined following an ELISA procedure. The 100mM concentration of both 13-cis retinoic acid and ATRA decreased cell viability to 60.37% and 69.16% of control, respectively. The 1 mM and 10 mM concentrations did not significantly affect cell viability. The results of the ELISA showed that cells treated with 100µM 13-cis retinoic acid and 10µM all-trans retinoic acid had increased concentrations of human CD95 suggesting that they increase apoptosis in CRL1790 epithelial cells. Further experimentation should be done to confirm these results.

THE EFFECT OF CLIMATE CHANGE AND OIL SPILL ON BLACK-SPOTTED MOSQUITOFISH GENOTYPE Iordanka N. Panayotova, Department of Mathematics and Statistics, Ann Creasy & Lisa Horth, Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529. In this work, we use a numerical model, and employ a suite of empirically derived relative fitness values, to simulate the change in

frequency of two color-pattern morphs over time in a large population of conspecific fish (*Gambusia holbrooki*). Numerical simulations are employed to model dynamics of the black-spotted mosquitofish genotype. It is shown that the climate change may have a devastating effect on the mosquitofish resulting in extinction of the mottled-black eastern mosquitofish genotype. In contrast, if an oil spill happens and kills 80% of the population of mosquitofish, the remaining 20% are enough for the population to return to the equilibrium observed in nature however it may take over 100 year for the stabilization to occur.

ATTACHMENT AND BIOFILM FORMATION OF *CITROBACTER RODENTIUM* IN THE MOUSE INTESTINE. Michael W. Canfarotta & Andrew J. Fabich, Department of Biology and Chemistry, Liberty University, Lynchburg VA 24502. *Citrobacter rodentium* DBS100 is an enteric murine pathogen similar to enterohemorrhagic *Escherichia coli* (EHEC) and enteropathogenic *Escherichia coli* (EPEC) as it forms an attaching and effacing (A/E) lesion during gastrointestinal infection. Attachment has been previously shown to be an important initial step in both colonization and biofilm formation. However, the mechanism of colonization of *C. rodentium* in the mouse intestine is poorly understood. *In vitro* biofilm formation assays were performed comparing the ability of pathogenic DBS100 and commensal *E. coli* strain MG1655 to form a biofilm on a polystyrene surface while grown in media containing specific carbon sources commonly found in intestinal mucosa. *In vitro*, pathogenic DBS100 demonstrates increased biofilm formation when grown on fucose and mannose whereas commensal MG1655 forms greater biofilms when grown on ribose. The data suggests that pathogens may form a biofilm during gastrointestinal infection by utilizing sugars that are not important in the biofilm formation of commensal intestinal microbiota.

Microbiology and Molecular Biology

NF- κ B DEPENDENT FAS SIGNALING IN HEPATOCYTES. Quoc Tran^{1,2}, Rohini Mehta², Aybike Birerdinc², & Ancha Baranova^{1,2}, ¹Biology Department, George Mason University, ²Betty and Guy Beatty Center for Integrated Research, Inova Health System. The high prevalence and substantial morbidity and mortality accompanying Non alcoholic fatty liver disease (NAFLD) makes it imminent to understand the mechanistic basis of this disorder. Hepatocyte apoptosis, is an important mechanism in the pathogenesis and progression of NAFLD with Fas signaling being an important player. The focus of the current study was to determine whether chemokines CCL21 and CCL4 induce Fas ligation and, if so, whether the Fas signaling activates downstream NF- κ B mediated inflammatory pathway, or caspase-3 dependent apoptosis. HepG2 cell line was used as in vitro model for the current study. For qPCR, cells were subjected to varying concentration of individual chemokines in a time course experiment of 2, 4, 6 and 8 hours. mRNA was extracted from the cells using Qiagen RNeasy kit (Qiagen, USA) according to manufacturer's protocol. cDNA synthesis was carried out using First strand synthesis kit (Qiagen). qPCR was performed on 18 genes specific to Fas signaling and NF- κ B inflammatory pathway. For ELISA, cells were subjected to varying concentration of individual chemokines in a time course experiment of 6, 8, 12 and 24 hours. After stimulation, supernatants were collected and

subjected to ELISA using custom Multi-analyte ELISArray kits from Qiagen (Fredericks, MD) according to the manufacturer's suggestions. Apoptosis was evaluated by detection of caspase-3/7 activity using Caspase-Glo 3/7 Assay (Promega). Apoptosis results were in agreement with the apoptotic and anti-apoptotic gene expression pattern.

IDENTIFYING TARGET GENES IN THE ERR GAMMA PATHWAY RESULTING FROM BPA EXPOSURE IN BREAST CANCER. K.L. Voss & DA O'Dell, Dept Biological Sciences, Univ Mary Wash., Fredericksburg, VA, 22401. Bisphenol A (BPA binding) to the Estrogen Related Receptor Gamma (ERR- γ) was studied to determine whether it can lead to alterations in the cell cycle by affecting the activity of oncogenes and tumor suppressor genes. Normal breast epithelial cells were divided into 3 groups; control and 2 experimental groups. Estrogen and Androgen receptors were blocked in experimental groups using fulvestrant and p-p'-DDE. ERR- γ activity was blocked with 5 μ M 5-Hydroxytamoxifen. $\frac{1}{2}$ of each group was exposed to 5 μ M of BPA in normal culture medium for 72 hours. Total RNA was extracted and assayed using RT-PCR and a commercially available microarray (SABioSciences). Twenty-one genes were found to be upregulated and 7 were down regulated in response to BPA exposure in cells with no receptors blocked. When the ERR- γ receptor was active, only 4 genes were upregulated and 7 were down regulated. When the ERR- γ receptor was blocked, 3 genes were up regulated and 4 showed downregulation. The results show that BPA does alter gene activity which could lead to changes in the cell cycle leading to a cancerous state. The results also indicate that there is yet another receptor through which BPA can exert its effects. This work was supported by an Undergraduate Research Grant (UMW) to KV and a Mary Louise Trust Award (VAS) to DAO.

GENE EXPRESSION IN HUMAN GLIOBLASTOMA CELLS POST CELL PHONE RADIATION EXPOSURE. K.M Meyer & DA O'Dell, Dept Biol. Sci., Univ. Mary Wash. Fredericksburg, VA 22401. The effects of cell phone radiation on gene expression in human glioblastoma cells was studied to determine whether EMF exposure could lead to changes in genes which regulate the cell cycle. Human glioblastoma cells were cultured to a G1 arrested state after which they were exposed to 25 min of cell phone radiation (Avg 57.3 mW/m²). Total RNA was extracted at 0min -20 min-24 hour time intervals after exposure. RT-PCR using a commercially available microarray (SABiosciences) for 84 oncogenes and tumor suppressor genes was used to analyze the changes in gene activity. Cells responded immediately after exposure by upregulating 45 genes, many of them tumor suppressor genes. After 24 hours, the number of gene upregulated increased to 71, with more tumor progression genes (oncogenes and transcription factors) activated. Two genes promoting cell death (CASP8 and FHIT) showed significant changes after 20 min while after 24 hours, significant changes were seen in 3 genes, one of which was JUND, a transcription factor and oncogene. The results show that cells respond to cell phone radiation exposure by activating genes which promote tumor suppression initially followed by genes which are involved in tumor promotion. More work to establish long term effects of cell phone radiation on gene activity in cells is needed to determine the role of cell phone radiation in promoting cancer. This work was supported by an Undergraduate Research Grant (UMW) to KM.

ADAR FACILITATED RNA EDITING IN HUMAN PLASMACYTOID DENDRITIC CELLS (PDC). A. Sharma¹, Lamya Alomair¹, Katherine Doyle¹, Patrick Gillevet³, Masoumeh Sikaroodi³, Aybike Birerdinc^{1,2}, & Ancha Baranova^{1,2,3}, ¹School of Systems Biology, George Mason University, Fairfax VA 22030, ²Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church, VA 22042, ³Microbiome Analysis Center, George Mason University, Manassas, VA 20100, ⁴National Institute of Health, Bethesda, MD 20892. Adenosine (A) to Inosine (I) RNA editing is facilitated by enzymes known as ADAR (Adenosine Deaminase that Act on RNA). ADARs specifically recognize double stranded RNA structures or RNA duplex structures as their substrates. Inosine is translated as Guanosine, since most enzymes recognize Inosine as Guanosine. Examples of physiological ADAR editing are edits to neuronal Glutamate and Serotonin receptor transcripts. Here we set to find out whether ADAR-editing in human PDCs (Plasmacytoid Dendritic Cell) is limited to TLR7, or whether it covers other known ADAR targets, including other TLR receptors, FLNA, IGFBP7, KCNA1, GABRA3, and CYFIP2. Site specific primers around previously known edited sites were designed using NCBI primer blast and then tested on cDNA derived from universal RNA and adipose tissue. Purified cDNA from PDC cells was used as templates for PCR amplification, tagged, purified, and subjected to Multitagged (MTPS) pyrosequencing on Roche GS-FLX instrument. The pyrosequencing data was assembled using Lasergene's Seqman Pro to assemble all the contigs.

USE OF CO-EXPRESSION PATTERNS FOR FUNCTIONAL ANALYSIS OF HUMAN GENES (KCNRG & KCTD7) WITH UNCLEAR CELLULAR ROLES. Sarath Babu Krishna Murthy¹, Hannah Choi² & Ancha Baranova^{1, 2, 3}, ¹School of Systems Biology, George Mason University, Manassas, VA, ²Biology Department, George Mason University, Fairfax, VA, ³Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA. KCNRG is a soluble protein with characteristics suggesting it forms hetero-tetramers with voltage-gated K⁺ channels and inhibits their function. The ONCOMINE database is an online collection of microarrays from various sources, usually cancer-related, and contains many "multi-arrays". The KCTD7 gene is a paralog of the KCNRG gene that also binds to cullin 3. We analyzed 10 different datasets containing 100 different genes each for common overlapping co-expressed genes of KCNRG, using multiple cancer studies within the Oncomine database, focusing here predominantly upon brain and CNS cancer studies. Meta-analysis result, with frequency of 3 or more, for KCNRG yielded 95 hits and was further assessed for ontology and full gene names. This genelist was used as input file for an advanced analysis using MetacoreTM, an integrated software suite for pathways and network analysis of OMICs data. GeneGo Pathways Maps show that top scored map (map with the the lowest p value) based on the enrichment distribution sorted by 'Statistically significant Maps' set is Transport RAN regulation pathway. Analysis of co-correlations is a powerful tool that allows one to get a glimpse into function of genes with no known function. Cancer-related Oncomine database is a suitable input for analysis of co-correlations.

MDM2 IS AN UBIQUITIN E3 LIGASE MEDIATING PROTEASOME-DEPENDENT DEGRADATION OF CIRCADIAN RHYTHM PERIOD 2. Jingjing Liu, Dept. of Biol., Virginia Tech., Blacksburg VA. 24061. The circadian rhythm and cell cycle are the two main oscillatory systems in cells. How cells sense time and decide what is the best time for growing, dividing or die? One possibility is that there are crosstalks between these two systems. Based on the fact that Period 2 (Per2) also plays essential role in DNA damage response (*Fu, L., and Lee, C. C.* 2002), Per2 is supposed to connect circadian rhythm and cell cycle, which makes Per2 work as a tumor suppressor. We found Per2 regulating p53 pathway but little is known about how Per2 itself is regulated. One interesting finding is that independent of transcriptional regulation, overexpressed Per2 protein also oscillating, this implies posttranslational modifications are essential for sustaining Per2 protein oscillation. Per2 binds to Mdm2, a well-known E3 ubiquitin ligase, both *in vitro* and *in vivo*. Mdm2 induces Per2 ubiquitination *in vitro*, but further experiments are needed to verify Mdm2 is an E3 ligase for Per2 *in vivo*.

GLOBAL CHARACTERIZATION OF DNA METHYLATION PATTERNS IN AN ALZHEIMER'S DISEASE MODEL. Courtney A. McKenzie, Rebecca C. Garrett, Noor M. Taher & Gary D. Isaacs, Dept. of Biology and Chemistry, Liberty University, Lynchburg VA 24502-2269. Epigenetics play a role in regulating transcription through gene silencing by DNA methylation. Epigenetics have been implicated in multiple diseases. This experiment consisted of a genomic scale analysis of neuronal cells to determine the methylation patterns associated with models for mature neurons, cancer, and Alzheimer's disease (AD). The restriction endonucleases MspI and HpaII were utilized because they both cleave methylation sensitive regions, but HpaII only cleaves these sequences when they are not methylated. To determine genomic methylation patterns, DNA from each model was differentially digested with the enzymes and hybridized with fluorescent markers to a microarray. Analysis revealed global differences in methylation levels between cancer, mature neuron, and AD models. Regions have been identified where a gene's methylation status is different in the AD model than it is in the mature neuron model. These regions represent genes that were either turned on by hypomethylation or turned off by hypermethylation as a result of AD pathogenesis. Gene specific studies to determine the affected cellular processes are currently in progress.

EXPRESSION OF THE PROPANOYL-COA METABOLIC PATHWAY FROM *T. fusca* IN *E. coli*. Allison Yaguchi & Dr. Stephen Fong, Dept. of Chemical and Life Sciences Engineering, Virginia Commonwealth University. The objective of this project was to successfully express a metabolic pathway found in *Thermobifida fusca*, a thermophilic, cellulolytic actinobacteria, in the model organism, *Escherichia coli*. A potentially novel method for biologically producing 1-propanol was found in an engineered strain of *T. fusca*. Direct confirmation of the novel pathway's activity is difficult in *T. fusca*, thus expression of the target pathway in *E. coli* would provide a direct means of testing the novel pathway. The *T. fusca* gene, Tfu_2395, was transformed into *E. coli* and positive transformants were confirmed with blue/white screening and DNA sequencing. Secretion of 1-propanol by the engineered strain of *E. coli* would functionally demonstrate the activity of novel metabolic pathway for

production of 1-propanol found in *T. fusca* and secondary confirmation will be achieved using molecular measurements such as real-time PCR of pathway genes. This research was funded by the Virginia Academy of Science and Virginia Commonwealth University.

ROLE OF *RETINOIC ACID INDUCED-1 (RAI1)* DOSAGE IN *XENOPUS* EMBRYOGENESIS AND THE FORMATION OF CRANIAL NEURAL CREST DERIVATIVES. R.Tahir¹, A.J.Dickinson², & S.H.Elsea³, ¹Center of the Study of Biological Complexity, ²Dept. of Biology and ³Depts. of Pediatrics and Human & Molecular Genetics, Virginia Commonwealth University. Haploinsufficiency of transcription factor *Retinoic acid induced-1 (RAI1)* is the primary cause of Smith-Magenis Syndrome (SMS), a rare congenital disease marked by mental retardation, craniofacial abnormalities, obesity, and an inverted circadian rhythm. In the present study, we characterize the expression of *Rai1* during embryonic development of *Xenopus* using whole-mount *in situ* hybridization. Furthermore, we reduce the dosage of *Rai1* during development using an antisense morpholino and analyze the resulting abnormalities. Our work demonstrates that *Rai1* is highly expressed in facial and dorsal regions of the developing embryo, with *Rai1* expression in maxillary and nasal prominences. In addition, expression is localized to a region that appears to be migrating neural crest. A disturbance in *Rai1* dosage during development can lead to significant craniofacial abnormalities, including abnormal formation of cartilage and cranial nerves, two important cranial neural crest derivatives. This study was funded in part by Howard Hughes Medical Institute and Virginia Academy of Science.

SUPPRESSION OF THE MATURATION AND ACTIVATION OF THE DENDRITIC CELL LINE DC2.4 BY MELANOMA-DERIVED FACTORS. Kristian M. Hargadon, Osric A. Forrest, & Pranay R. Reddy, Dept. of Biol., Hampden-Sydney College, Hampden-Sydney VA 23943. Dendritic cells play important roles in both innate and adaptive immunity, and their numerous functions are tightly linked to their maturation and activation status. Many tumors have been shown to induce anti-tumor immune dysfunction, but the basis for this dysfunction is often unclear. Here, we characterize the influence of melanoma-derived factors on the maturation and activation of the murine dendritic cell line DC2.4. Exposure of DC2.4 cells to the Toll-like receptor ligand lipopolysaccharide induces both maturation and activation of these cells, characterized by upregulation of costimulatory molecule expression and proinflammatory cytokine/chemokine production. This maturation and activation is suppressed by soluble factors derived from both the highly tumorigenic B16-F1 and the poorly tumorigenic D5.1G4 murine melanoma cell lines. Interestingly, the extent of DC2.4 immunosuppression by these melanomas correlates with their tumorigenicity. The impact of this suppression on the quality of T cell responses elicited by tumor-altered dendritic cells points to a critical role for tumor cell/dendritic cell interactions in regulating the quality of anti-tumor immune responses. (Supported by: Virginia Academy of Science Jeffress Research Grant, Virginia Foundation for Independent Colleges Mednick Memorial Fellowship, Virginia Foundation for Independent Colleges Undergraduate Science Research Fellowship, Sigma Xi Grant-in-Aid of Research, and Hampden-Sydney College Research Grant from the Arthur Vining Davis endowment).

CELL FUSION AND THE GROWTH FACTOR IGF IN MYOCARDIAL REPAIR. Syeda S. Baksh, Dept. of Biol., The University of Mary Washington., Fredericksburg VA, 22401. Traditionally, the myocardium has been considered terminally differentiated tissue due to the incapability of cardiomyocytes to regenerate in adult life. Therefore, these cells are not able to compensate for the cell loss as a result of myocardial infarction. However, in the past couple of years, there has been significant evidence that the heart does have regenerative potential. This evidence suggests that in response to growth or injury, the myocardium recruits stem cells/progenitor cells to repair and regenerate. One mechanism possibly used to differentiate the stem cells of the heart into cardiomyocytes is known as cell-cell fusion. In order to enhance fusion, previous studies have employed insulin-like growth factor (IGF), and the results illustrated that the addition of IGF was successful in skeletal muscle cells. Our goal in this study is to show whether or not stem cells fuse with cardiomyocytes, if IGF promotes this fusion, and if fusion stimulates the cardiomyocytes to reenter the cell cycle. Cardiac stem cells and cardiomyocytes were isolated from newborn rat pups and adult rats (respectively) using the Worthington Biochemical Corp. Neonatal Cardiomyocyte Isolation System. After fluorescently labeling the stem cells with a Qtracker® Cell Labeling Kit, they were co-cultured with the cardiomyocytes for four days. IGF was added to half of the cultures. After four days, fusion was assessed and was observed in the culture with IGF, but was not observed in the culture without IGF, indicating that IGF successfully enhanced fusion of cardiac stem cells with cardiomyocytes. Whether fusion stimulates cardiomyocytes to reenter the cell cycle could not be investigated due to a shortage of time.

THE EFFECTS OF EXERCISE TRAINING AND ESTROGEN ON THE ATHEROSCLEROTIC PLAQUE SIZE AND COMPOSITION. Leslie N. Valenzuela & Kathryn E. Loesser, Dept. of Biol., University of Mary Washington, Fredericksburg VA 22401. Atherosclerosis and cardiovascular disease are the leading cause of death in the western world. Twelve normal mice and twelve Apolipoprotein E deficient mice were used to investigate the effects of exercise training and estrogen on the atherosclerotic plaque size and composition in atherosclerosis-prone mice. Half of the mice were labeled as sedentary and the other half were under the exercise protocol. In the beginning of the study, the mice were fed a normal chow diet. At six weeks of age, the mice were switched to a high fat diet. At six weeks of age, twelve mice that were under the exercise protocol began their exercise, which was swimming. The mice swam for a total of six weeks. At the start of a new week, 4 minutes were added to the swimming protocol. A program called ImageJ was used to measure the blood vessel wall thickness of the mice. An Estradiol EIA Kit was used to measure the estradiol levels in the mice. A student's t-test with unequal variance was used to find statistical differences for the estradiol levels between the males and females. There was no statistical difference between the levels of estradiol in the males and females of either normal or Apolipoprotein E deficient mice. For the blood vessel wall thickness, all the mice that swam had a smaller blood vessel wall thickness than the mice that were sedentary highlighting that exercise leads to having healthier arteries. It appears that exercise is more important than either gender or predisposition to atherosclerosis in preventing vessel wall thickening.

CRM1-INDEPENDENT NUCLEAR EXPORT OF THE THYROID HORMONE RECEPTOR IS MEDIATED BY EXPORTIN 5. K. S. Subrmanian, H. N. Nelson, & L. A. Allison, Biology Department, College of William and Mary, Williamsburg VA 23185. Thyroid hormone receptors (TR α 1 and TR β 1) bind to thyroid hormone to regulate target genes involved in metabolism, growth, and development. Although primarily found in the nucleus, TRs rapidly shuttle in and out of the nucleus through the nuclear pore complex. Previously, we showed that TR nuclear export is not completely blocked when the export factor CRM1 is inhibited, suggesting that TR can also exit the nucleus by a CRM1-independent pathway. To determine which export factors are involved in the CRM1-independent pathway, RNA interference was used to knockdown gene expression of several different export factors. The effect of knockdown on the shuttling kinetics of GFP-tagged TR (α 1 and β 1) was assessed in live HeLa cells using FRAP. Knockdown of exportin 5 altered TRs nuclear export dynamics; recovery was markedly slower in photobleached nuclei, indicating that nuclear export was inhibited. To determine whether increased nuclear export had an impact on TR-mediated gene expression, we co-expressed TR, exportin 5, and a thyroid hormone response element (TRE)-mediated CAT reporter gene. CAT ELISA showed a decrease of TRE-mediated CAT reporter gene expression when increased amounts of exportin 5 were present. Further, we showed that when exportin 5 is over-expressed, the distribution of TR shifts to a more cytoplasmic localization. Taken together, our data suggest that TR nuclear export is mediated, in part, by exportin 5, and that disrupting the fine balance between nuclear import and export can lead to changes in TR-mediated gene expression. (Supported by: NIH #2R15DK058028-03 to LAA)

EARLY MARKERS OF CYTOGENETIC ANOMALIES IN THE INTERPHASE NUCLEI. Tatiana Glazko, Nanobiotechnology Centre of Russian State Agrarian University – MTAA named after K.A. Timiryazev, Moscow, Russia. Cytogenetic anomalies are common in both premalignant and malignant cells. Formation of the cells with these anomalies is a multifactorial process that is disturbed as a result of the disruption or insufficiency in the chromosomal arrangement in interphase nuclei. Our observations indicate that, in a normally functioning nucleus, the paternal and maternal haploid chromosome sets behave relatively independently of each other. Here we present the data supporting this hypothesis and derived from the study of the following models: metaphase plates in the peripheral blood lymphocytes of the cattle and polytene chromosomes in the salivary glands of *Chironomus thummi* larvae. Close contacts between haploid chromosome sets enhance the stability of the cell's genome, while its relative dissociation leads to an increase in the frequency of aneuploidy.

CHERNOBYL LESSONS IN GENETICS: AN ADAPTATION OF MAMMALIAN POPULATIONS TO THE EXTREME ECOLOGICAL STRESS. Valery Glazko, Nanobiotechnology Centre of Russian State Agrarian University – MTAA named after K.A. Timiryazev, Moscow, Russia. In 1986, Chernobyl disaster forever changed our understanding of the place of human kind within the Earth's environment. Twenty five years later, we still continue to derive important scientific insights from the consequences of this global catastrophe. Here we present the data collected using three animal models chronically exposed to substantially elevated levels of irradiation: laboratory mice, natural populations of various species of voles (*Microtus arvalis*,

Microtus oeconomus, *Clethrionomys glareolus*) and experimental herd of cattle. In cattle, the dramatic decrease in the fertility was observed subsequent to irradiation. This observation was compatible with preferential elimination of embryos with radiosensitive genotypes. In dairy breed Holstein cows, there was a change in the structure of population toward the loss of the breed specialization and reversal to genetic characteristics of the primitive breed, thus, confirming Shmalgausen's Rule of the preferable reproduction of the least specialized forms in case of dramatic environmental change. In voles, the spread of radioresistant genotypes/phenotypes through entire populations took approximately 26-30 generations. Thus, the main consequence of the Chernobyl disaster was observed at the level of populations rather than individuals and manifests through the change in the genetic structure of population due to an increase in the level of the genomic instability.

Posters

DETERMINATION OF GENOME-WIDE METHYLATION IN NEURONS TREATED WITH AMYLOID- β . Rebecca Garrett, Courtney McKenzie, Noor Taher, & Gary Isaacs, Department of Biology and Chemistry, Liberty University, Lynchburg VA 24502-2269. Alzheimer's disease (AD) is a form of dementia characterized by the formation of neurofibrillary tangles and amyloid- β plaques. Known causes of AD cannot account for the large number of AD cases, so the connection between AD and the epigenome, specifically CCGG genomic loci, is being studied. Several factors make the case for an epigenetic basis for AD. A global change of DNA methylation levels is shown in AD subjects relative to control groups, and several AD-associated genes are regulated by DNA methylation. To study the differing levels of methylation in a disease vs. non-disease state, isoschizomer enzymes MspI and HpaII (which cut CCGG regions) were used. HpaII can only cleave if the CCGG is unmethylated, allowing for determination of methylation status. DNA samples from undifferentiated IMR32 cells (cancerous state), differentiated IMR32 cells (normal state) and amyloid- β treated differentiated IMR32 cells (AD-like state) were digested using MspI and HpaII. Samples were then concentrated using the HELP assay, fluorescently labeled, and hybridized to a microarray. Genome-wide increases and decreases in methylation of CCGG regions were observed between the cancerous, normal, and AD-like states. Microarray data was used to pinpoint specific genomic regions where the methylation status changed; studies of CCGG regions of these specific genes are currently being conducted.

INVESTIGATION AND ANALYSIS OF THE MOLECULAR BIOLOGY AND EVOLUTION OF THE APPETITE REGULATING HORMONE AGOUTI-RELATED PROTEIN (AgRP). C. Gerner^{1,2}, A. Birerdinc^{1,2}, Z. Younossi^{1,2,3}, A. Baranova^{1,2}, & M. Estep¹, ¹Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA, ²Center for the Study of Genomics in Liver Diseases, Molecular and Microbiology Department, George Mason University, Fairfax, VA and ³Center for Liver Diseases, Inova Fairfax Hospital. Agouti-Related Protein (AgRP) is an orexigenic peptide hormone that suppresses metabolism. Its role in appetite, metabolism, dyslipidemia, inflammation, and melanogenesis makes the dysregulation of AgRP a likely contributor to metabolic disease. The aim of this research is to

compare AgRP DNA and protein sequences, gene structure, and genetic neighborhood across several species in an attempt to identify important regulatory motifs and possibly develop hypotheses regarding their specific function. Bioinformatic comparisons using clustering and alignment tools will be used to assess consensus and divergence. Preliminary results have already identified two conserved putative miRNA binding sites; the putative binding site for hsa-miR-554 is highly conserved in the primates examined, while the putative binding site for hsa-miR-375 is conserved among all mammals examined but not other species. Our study could offer insights into intervention targets for the regulation of AgRP.

FUNCTIONAL ANALYSIS OF PUTATIVE TUMOR SUPPRESSOR GENES KCNRG AND KCTD7. Hannah Choi¹, S. Krishnamurthy¹ & A. Baranova^{1,2}, ¹Biology Department, George Mason University, Fairfax VA, ²Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA. KCNRG is a soluble protein with characteristics suggesting it forms hetero-tetramers with voltage-gated K⁺ channels (K_v) and inhibits their function. However, KCNRG related proteins do not bind (K_v) but are associated with ubiquitin ligase cullin 3, suggesting that the function of KCNRG may be different from that hypothesized before. Cullin 3 ubiquitination is suspected to directly modify the activities of K_v. KCTD7 gene is a paralog of the KCNRG gene that also binds to cullin 3. The Oncomine database is an online collection of microarrays that profile various types of human cancer samples. Hundreds of tumor samples are described as a single, co-processed multi-array study to allow analyses of co-expression patterns. Separate analyses of ten different Oncomine datasets for co-expression patterns for the top 100 genes co-correlating with KCNRG and KCTD7 were performed with CNS samples. The meta-analysis with genes found in 3 or more of the datasets yielded 95 gene hits for KCNRG and 37 for KCTD7. This data was used as input for an advanced analysis using Metacore, an integrated software suite for pathways and network analysis of OMICs data. The “Analyze Single Experiment” workflow in Metacore was employed for the meta-analysis of the data using 650 Canonical Pathways maps. This analysis showed that the top scored map based on the enrichment distribution for genes co-expressed with KCNRG is “Transport_RAN regulation pathway”. The top score map for KCTD7 revealed to be “Cadherin mediated cell adhesion”. Further research is currently in progress.

EFFECTS OF NEUROTRANSMITTERS ON THE LOCOMOTION OF TERRESTRIAL SLUG, LIMAX MAXIMUS. Jamie P. Warrick, April C. Nivens, and Brett G. Szymik. Longwood University, Farmville, VA 23909. This project investigates the putative role of various neurotransmitters on the locomotory behavior of the terrestrial slug *Limax maximus*. Time-lapse videography and still photography were used to determine the behavioral effects of administration of serotonin, dopamine, and ergometrine neurotransmitters, as well as a saline control. Pedal wave number and speed as well as overall animal speed were measured after neurotransmitter injection into the body cavity. Serotonin decreased the average number of pedal waves but increased pedal wave speed. Dopamine increased the inter-wave length while decreasing wave speed. Overall, this project begins to hone-in on the neurotransmitters that may be endogenously used to modulate locomotory speed in the terrestrial slug *Limax*.

ROLE OF THYMOSIN BETA4 IN EPITHELIAL TO MESENCHYMAL TRANSITION IN IDIOPATHIC PULMONARY FIBROSIS. C Collins¹, M Leema^{1,2}, E McLaughlin¹, S. Nathan² and G Grant¹. ¹SSB GMU, Manassas VA, ²IHVI Inova Fairfax, Falls Church, VA. The transformation of epithelial cells to mesenchymal cells (EMT) is an important, normal cellular process. However, EMT can also play a role in diseases as seen in metastatic cancer and fibrotic diseases such as Idiopathic Pulmonary Fibrosis (IPF). IPF is a fatal scarring disease of the lung involving deregulated tissue repair. IPF is exacerbated by an over population of fibroblasts and EMT is believed to contributed to this overabundance. Thymosin beta-4 (Tβ4) is a small (4.9kDa) protein, predominantly involved in the actin cytoskeleton assembly. However, recently additional roles have been attributed to this protein including migration, pro-survival/anti-apoptosis and the ability to initiate EMT. We have recently discovered that Tβ4 is over expressed in IPF fibroblasts. Therefore, here we investigated the potential of the protein Tβ4 to induce EMT in lung alveolar cells and thereby contribute to IPF. The model alveolar type II cell line A549 was employed. These cells were exposed to 0, 5, 10ng/ml Tβ4 at various serum concentrations over a 7, 24, and 72-hour period. In addition stable Tβ4 over-expressing transfected A549 cell lines were derived to investigate the effect of endogenously over expressed Tβ4. Cells exposed to 0, 5, 10 ng/ml transforming growth factor- beta (TGF-β) at 1% and 10% serum concentrations served as a positive control. EMT was tracked by quantitative real-time PCR (qRT-PCR) and western blotting using markers of EMT such as E-cadherin. (Supported by: The Jeffress Memorial Trust)

DEVELOPING A SINGLE STEP DETECTION OF ANTIGEN-ANTIBODY INTERACTIONS IN SOLUTIONS. Ekaterina Marakasova^{1,2}, Alexei Shevelev² & Ancha Baranova¹, ¹School of Systems Biology, George Mason University, Manassas, VA 20110, USA, and ²Department of Virology, Moscow State Academy of Veterinary Medicine and Biotechnology, 23 Akademika Skryabina, Moscow 109472, Russia. Instant immunodetection of relevant chemical compounds performed by pocket-size devices may be useful in clinical assays as well as in customs and security service, in the product quality control and in environmental monitoring. Moreover, a quality of medical care can be substantially elevated if serological tests for inflectional diseases could be carried out in several minutes, not days. The same advantages are commonly applicable in veterinary practice as well. We propose a detection system based on antigen-induced molecular rearrangements in C2h and Ch3 domains of IgG. This system consists of several artificially designed proteins containing fluorescent moieties. These proteins are readily compatible with any types of available antibodies against infectious agents or other chemical compounds. When a specific molecule is recognized, a fluorescent signal may be detected by eye or by a special mobile device.

UNKNOWN BACTERIAL STRAINS IDENTIFIED THAT PRODUCE INHIBITORY PRODUCTS. Grant Waldrop and Dr. Michaela Gazdik, Dept. of Natural Sciences, Ferrum College, Ferrum VA. Three unknown bacterial strains exhibited inhibitory capabilities towards gram-negative and gram-positive bacterial strains through contamination in the lab. Through differential media, metabolic characteristics, stains, and 16 S gene rDNA sequencing the unknown bacterial strains were identified as three different strains of *Paenibacillus polymyxa*. The antimicrobials produced by these

bacterial strains had an effect on gram-negative and gram-positive bacteria including: *Mycobacterium smegmatis*, *Escherichia coli*, *Leifsonia shinshuensis*, *Proteus vulgaris*, *Staphylococcus epidermidis*, *Pseudomonas fluorescens*, *Bacillus cereus*, *Staphylococcus typhomurium*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*, although effectiveness varied. *Mycobacterium smegmatis* being a model organism for *Mycobacterium tuberculosis* research became a target organism of interest. The stability of the unknown antimicrobial/antimicrobials inhibitory factor collected in the TSB broth of cultures was exposed to varying temperatures and its effectiveness was tested on *Escherichia coli*. The determination of the most viable culture incubation time period for the most effective antimicrobial/antimicrobials was determined as well. Further details of the unknown antimicrobial/antimicrobials will be eluded through future work along with molecular and genomic origins. NIAID grant number 1R15AI084058-01

LONGEVITY AND NEURONS: MAKING *DROSOPHILA* LIVE LONGER THROUGH ELECTRON TRANSPORT CHAIN RNAI IN SPECIFIC NEURONAL SUBTYPES. Bethany J. Johnson, Charise J. Garber, & Jeffrey M. Copeland, Eastern Mennonite University, Department of Biology. Over the past few years several genetic screens have isolated genes important for determining lifespan. Genes for the electron transport chain can dictate lifespan when partially inhibited in neurons and various other tissues. We have conducted tissue specific genetic inhibition of the ETC, and have observed that ETC inhibition in motor neurons is sufficient for lifespan extension. Importantly, ETC inhibition specific to intestines and glutamatergic neurons fail to extend lifespan. These results point to an important role of motor neurons in longevity.

A *DROSOPHILA* MUTANT RESISTANT TO OXIDATIVE STRESS. Charise J. Garber & Jeffrey M. Copeland, Eastern Mennonite University, Department of Biology. While oxidative damage is known to play an important role in the aging process and the re-oxygenation after an ischemic stroke, the molecular mechanisms are still poorly known. To better understand the cellular response to oxidative stress, we have conducted an X chromosome screen in *Drosophila* to find mutants resistant to elevated oxygen levels. The mitochondrial gene *CG7772* showed increased resistance to hyperoxia, but not to paraquat, another reactive oxygen species generator. Mutants for *CG7772* do not confer resistance to general stressors as *CG7772* females do not resist starvation.

THE PSEUDOPHOSPHATASE MK-STYX ROLE IN NEURONAL DIFFERENTIATION. K. E. Wong & S. D. Hinton, Dept. of Biology, College of William and Mary. The pseudophosphatase MK-STYX [MAPK (mitogen-activated protein kinase) phosphoserine/threonine/tyrosine-binding protein] has been previously implicated to cause neuronal differentiation. MK-STYX is structurally similar to the MAPK protein family, whose proteins are involved in pathways regulating cell proliferation and differentiation. We hypothesized that MK-STYX plays a direct role in neuronal differentiation. To determine if MK-STYX has a role in neuronal differentiation, PC12 cells were transfected with pMT2, MK-STYX-FLAG, and pEGFP vectors. Cells were examined and scored 5 days post-transfection. Here, we show that MK-STYX is endogenously expressed in PC12 cells. Furthermore, over-

expression of MK-STYX encourages neurite production. Neurite expression is seen in the presence and absence of NGF, nerve growth factor. Finally, MK-STYX can induce neurite outgrowth when MEK is inhibited. Together, these data are significant because they provide more insight into MK-STYX's potential role in neuronal differentiation. Future directions should explore other proliferation and differentiation pathways to determine MK-STYX's role.

Biomedical and General Engineering

VARIABLE CRACKING PRESSURE SWING CHECK VALVE. Cameron J. Grover, Samantha L. Leach, Graham S. Kelly, Stephen J. Warren, Charles E. Taylor & Gerald E. Miller, Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA. 23220. Aortic Valve Sclerosis is a heart condition affecting up to thirty percent of the population over the age of sixty-five. It is characterized by a calcification of the aortic valve leaflets. If the condition is left unchecked, it can lead to aortic valve sclerosis, which may significantly impede blood flow to the heart. This leads to an increased left ventricular load and an increased pulse pressure, both of which may cause complications and undue stress on the body. This project models aortic valve sclerosis by developing a variable cracking pressure swing check valve. Using laser printed acrylic of quarter-inch thickness, a casing for the valve was built with openings comparable to that of a sclerotic valve. The valve was built with sixteenth-inch thick acrylic and pivots on a pin hinge. The elastic material Thera-band Silver was affixed to the valve and attached to a linear-actuator. The linear-actuator pulls the strip of Thera-band, making it more difficult for the valve to open. In future studies, this model can be used in mock circulatory loops to test left ventricular assist devices interaction with pathological valve states.

IN VITRO STEREOSCOPIC FLOW INVESTIGATION OF A TILTING DISC VALVE AT AN AORTIC ROOT MODEL. Stephen J. Warren, Graham S. Kelly, Charles E. Taylor, Gerald E. Miller., Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA. 23220. Currently, bench top experimental fluid mechanics study for biomedical applications require physiologically accurate flow and geometries. Because of this, it was necessary to include working anatomical models in the mock circulatory systems being used to simulate cardiovascular hemodynamics. The first step in this process was a rigid model of the aortic root, which would serve as accurate exit geometry from the aortic valve. An acrylic aortic model was created from cryoslice data from the National Library of Medicines Visible Human project. The model was implemented into an automated mock circulatory loop that would provide the downstream resistance and compliance to create relevant flow patterns. A tilting disc valve (Bjork-Shiley®) was used in this experiment to display the effects of a central occluder on the exit flow of the valve into the aortic root. Stereoscopic Particle Image Velocimetry was included to allow for three velocity components to be taken in to account at once rather than a multi-planar comparison. The studies concluded that the large central occluder design confirms the presence of large low flow regions in the sinuses of the aortic root. These flow patterns could result in thrombosis formation in coronary sinuses, which could result in myocardial infarction if coronary flow becomes interrupted.