# Proceedings of the 90th Annual Meeting Virginia Academy of Science Norfolk State University, Norfolk Virginia May 23-25, 2012

### **Aeronautical and Aerospace Sciences**

A HYBRID AIRSHIP CONCEPT M. Leroy Spearman Langley Research Center, Hampton, VA. An airship differs from an airplane in that the lift required for flight is provided by a lighter-than-air gas contained within the airship hull. For an airplane the lift required for flight is generated by moving air over an airfoil shaped wing. The lift for an airship is called buoyant lift and it requires no forward motion. The lift for an airplane is called kinetic lift and requires forward movement of the vehicle through the air to produce the lift. A large airship is capable of lifting very heavy loads but is limited in speed and range. A hybrid airship is one in which buoyant lift is combined with kinetic lift. The proposed concept would be a twin-hull airship with a connecting inboard wing. The use of twin hulls would provide the required amount of lighter-thanair gas to be contained in a length half that of a single hull. Jet engines would be attached to the wing to provide for the forward flight that would create the kinetic lift. Particular designs can be developed by manipulating such factors as the volume of gas, the wing area, the airfoil contour and the engine thrust. Designs could be made that varied from small, lightweight systems to very large, heavy-load systems. Such a vehicle would also permit access to almost any remote location because of the vertical landing and take-off capability provided by buoyancy. Such a vehicle would be suitable for military logistic support missions. In commercial use the vehicle could serve small airports, could reduce community noise, could relieve air traffic delays, and could improve fuel efficiency.

SOME LESSONS LEARNED WITH NACA/NASA WIND TUNNELS. M. Lerov Spearman. Langley Research Center, Hampton, VA & Robert W. Heath, Canon of Virginia, Newport News, VA. The NASA Langley Research Center in Hampton, VA has been engaged in improving our nations aircraft for over 92 years. Named after aviation pioneer, Samuel Pierpont Langley, the Langley Memorial Aeronautical Laboratory was the first laboratory of the National Advisory Committee on Aeronautics (NACA). When the first of many wind tunnels went into operation at Langley in the early 1920's the typical aircraft was a low-speed, propeller-driven airplane Early research at Langley led to airfoil shapes that maximized the lift and minimized the drag. Other drag reduction led to the development of engine cowling for propeller-driven airplanes, the covering of the open cockpit, the cantilevered monoplane wing, and the retractable landing gear. When jet-propulsion was developed for aircraft the aerodynamic research was extended to transonic, supersonic and hypersonic speeds. New techniques and new wind tunnels were developed at Langley to address the problems at speeds beyond the speed of sound. One of the models tested at Langley, the Bell XS-1, was the first airplane to fly through the sound barrier. Essentially every supersonic aircraft built in the US has been influenced by Langley wind tunnel results. When further results lead to shapes that permitted access to space the NACA was replaced by the National Aeronautics and Space Administration (NASA) and research related to both aircraft and spacecraft continues at the Langley Research Center.

VARIABLE GEOMETRY AIRCRAFT. M. Leroy Spearman. NASA-Langley Research Center. Hampton, VA . A given aircraft design is generally a fixed shape that fits the mission requirement and different requirements require different shapes. Since the early days of manned flight aircraft have generally had a lift-producing wing that was aligned normal to the airstream direction. As the air moves over the wing surface the air begins to compress and with increased speed a point is reached where the air cannot be moved any further and increased flight speed is not possible. By sweeping the leading edge of the wing back at an angle to the flight direction the air flow over the wing travels a greater distance from the wing leading edge to the trailing edge and this apparent slimming delayed the onset of compressibility. While swept wing designs did permit high speeds they had poor stability at low speeds that required translating the wing fore and aft as the sweep angle was changed. NASA Langley resolved the problem with a design in which only a portion of the wing was swept. This lead to the concept that a wing with variable sweep could combine the speed advantage of high sweep with the good stability characteristics of low sweep. In 1961 the U.S. Secretary of Defense, Robert McNamara initiated the Transonic Experimental Fighter (TFX) program to develop a common airframe suitable for the Navy and the Air Force. The General Dynamics F-111 airplane was produced that met the mission requirements but the Navy cancelled out because the airplane exceeded weight and size limits imposed by aircraft carriers. Other airplanes that have made use of variable sweep in the U.S. are the Navy Grumman F-14 and the Air Force Rockwell B-1.

### Astronomy, Mathematics and Physics / Materials Science

AN INEXPENSIVE RADIO TELESCOPE IN A COLLEGE PHYSICS LAB. <u>T.C.</u> <u>Mosca III</u>, Dept. of Mathematics & C. Crook Dept. of Chemistry and Physics, Rappahannock Community College. An amateur radio telescope was established on the Glenns campus of Rappahannock Community College. The components included a commercial amateur radio receiver, an antenna designed to operate in the 15-30 MHz range, and a laptop computer running a freely available software package. Data were collected in 24-hour increments at 20.100 MHz, which is a frequency reserved for radiotelescopy. Numerous events were recorded, several of which were simultaneously observed at sites up to 3000 km away, indicating that these events originated from extraterrestrial sources. Physics students experienced a tangible application of electromagnetism principles, and learned that extraterrestrial objects are radio emitters. They also learned that real and valid research can be conducted without sophisticated and expensive equipment. In the future, we hope that students can be more directly involved in the collection and analysis of data, and that data can possibly be collected from sources other than our sun.

THE ACCELERATING JET OF 3C 279. S. D. Bloom<sup>1</sup>, C. M. Fromm<sup>2</sup>, and E. Ros<sup>3</sup>, <sup>1</sup>Department of Physics & Astronomy, Hampden-Sydney College, <sup>2</sup>Max Planck

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Institute for Radio Astronomy, <sup>3</sup>University of Valencia. Analysis of the proper motions of the sub-parsec scale jet of the quasar 3C 279 at 15 GHz with the VLBA shows significant accelerations in four of nine superluminal features. Analysis of these motions is combined with the analysis of flux density light curves to constrain values of Lorentz factor and viewing angle (and their derivatives) for each component. The data for each of these components is consistent with no changes to the Lorentz factor, but significant changes to the viewing angle and azimuthal angle, suggesting jet bending with minimal changes in speed. We see that for these observed components Lorentz factors are in the range of 10<sup>-36</sup>, and for viewing angles of 0.1°-4.6°, and intrinsic (source frame) flux density, F=(7 x 10<sup>-8</sup> - 1.6 x 10<sup>-4</sup>) Jy. Considering individual components, the viewing angles vary in time from 1.2 to 1.4 in the case of C1 (the least extreme example) and vary from 0.06° to 1.4° in the case of C6 (the most extreme example). The intrinsic flux density varies by factors from 1.6 for C8 and 240 for C5. Theoretical analysis of the accelerations also indicates potential jet bending. In addition, for one component, C5, polarization measurements also set limits to the trajectory of the jet.

FORMATION OF RELATIVISTIC OUTFLOWS IN ADAF DISKS WITH SHOCKS. <u>Truong V. Le<sup>1</sup></u>, Cassandra Brown<sup>1</sup>, Peter A. Becker<sup>2</sup>, and Santabra Das<sup>3</sup>, <sup>1</sup>Dept. of Physics, Governor's School for Science and Technology, <sup>2</sup>School of Physics, Astronomy & Computational Science, George Mason University, and <sup>3</sup>Dept. of Physics & Astronomy, Indian Institute of Technology Guwahati, India. We have developed a new self-consistent theory for the production of the relativistic outflows observed from radio-loud black hole candidates and active galaxies as a result of particle acceleration in hot, inviscid accretion disks containing standing, centrifugally supported isothermal shocks. Utilizing this model, we estimate the jet locations of 13 low-power radio-loud AGNs using the associated observed jet powers and their inferred mass accretions. Our results suggest that there is a direct correlation between the jet power and the Bondi accretion power, and that the jets location increases further away from the event horizon as the mass accretion rates and jets powers increases.

ON THE REDSHIFT DISTRIBUTION OF GAMMA RAY BURSTS IN THE SWIFT ERA. <u>Truong V. Le<sup>1</sup></u> and Charles Dermer<sup>2</sup>, <sup>1</sup>Dept. of Physics, Governor's School for Science and Technology, and <sup>2</sup>E. O. Hulburt Center for Space Research Naval Research Laboratory. A simple physical model for long-duration gamma ray bursts (GRBs) is used to fit the redshift (z) and the jet opening-angle distributions measured with earlier GRB missions and with Swift. The effect of different sensitivities for GRB triggering is sufficient to explain the difference in the z distributions of the pre-Swift and Swift samples, with mean redshifts of  $\langle z \rangle \sim = 1.5$  and  $\langle z \rangle \sim = 2.7$ , respectively. Assuming that the emission properties of GRBs do not change with time, we find that the data can only be fitted if the comoving rate-density of GRB sources exhibits positive evolution to  $z \approx 3-5$ .

SEARCHING FOR INTERMEDIATE MASS BLACK HOLES WITH X-RAYS. <u>Insuk Jang<sup>1</sup></u>, Mario Gliozzi<sup>1</sup>, Lev Titarchuk<sup>1</sup> & Shobita Satyapal<sup>1</sup>, <sup>1</sup>School of Physics, Astronomy, and Computational Data Science, George Mason Univ., Fairfax VA. 22030. The black hole mass is a crucial parameter to shed light on the physics of accretion. While the presence of stellar mass black holes (sMBHs) in binary systems and supermassive black holes (SMBHs) in the center of galaxies is widely accepted, the very existence of intermediate mass black holes (IMBHs) is still a matter of debate. It has been suggested that this type of black holes within the mass range of  $M_{BH} = 10^2$  $-10^5 M_s$  may reside in Ultraluminous X-ray sources (ULXs) which are very bright off nuclear X-ray sources. Recently, a new method to constrain the mass of BH systems, based solely on X-ray data, was successfully used for sMBHs and SMBHs. Since the X-ray emission is thought to be produced by the same process (Comptonization) in all accretion objects, in principle, this method can be applied to estimate the mass of black holes in ULXs. We have carried out a systematic analysis of a sample of 43 ULXs with multiple X-ray observations and applied this novel method. Our preliminary results suggest that ~70% of the sample harbor IMBHs and indicate a good agreement between the values of BH mass obtained with this technique and those derived with different methods present in the literature.

STATUS OF NSU'S RAPID REPONSE ROBOTIC TELESCOPE AT FAN MOUNTAIN, VA. Carlos W. Salgado, Dept. of Physics, Norfolk State Univ., Norfolk, VA 23504. The Norfolk State University (NSU), 60 cm, Ritchey-Chretien, Rapid Response Robotic Telescope (RRRT) is located at Fan Mountain Observatory, Covesville, VA, about 15 miles south of Charlottesville, VA. It is a fully automatic and remotely controllable telescope, initially equipped for UBVRI photometric studies. The RRRT is used for observational study of compact stars (and their progenitors), and in particular, the early photometry and polarimetry of Gamma-ray Optical Afterglows (GRB-OA). This telescope is part of the Swift follow-up team. The RRRT is a low inertia telescope (fast slew), dedicated to the study of fast transient phenomena, with rapid imaging polarimetry capabilities. The RRRT is also a central part of our educational and public outreach projects. For example, the Back Bay Amateur Astronomers use the telescope, and helped to build it. Furthermore, the RRRT and its data are available to NSU students and other students from the Tidewater area of Virginia. Such users include those at UVa, GWU, ODU, CNU, and JMU.

WHAT WE CAN LEARN ABOUT QUANTUM CHROMODYNAMICS (QCD) STUDYING THE MESON SPECTRUM. Carlos W. Salgado, Dept. of Physics, Norfolk State Univ., Norfolk, VA 23504. The study of the hadron spectrum led more than forty years ago to the development of the quark model, where baryons and mesons are described as bound (by a strong interaction) systems of three quarks and of a quarkantiquark pair, respectively. As a component of the current "Standard Model" of particle physics, Quantum Chromodynamics (QCD) is the accepted theory of the strong interactions. While much progress has been made in understanding QCD at high energies, strong interactions at the energy regime where hadrons are bounded (confinement) have remained obscure. A clear understanding of this regime is essential, since it is where the strong interactions are dominant (quark are confine into hadrons). While the original quark model still holds and has been proven to reproduce many features of the hadron spectrum, now we know that there are many more aspects (as hadron masses and spins) that cannot be explained only in terms of quarks. The dynamics of strong interaction fields (the QCD's gluons) plays a crucial role. Mesons, being made by the minimum possible aggregate of quarks (a quark - anti-quark pair),

are the simplest quark bounded system and therefore an ideal bench to study the interactions between quarks, to understand the role of gluons, and to investigate the mechanism of confinement. In this talk, we will outline how the modern study of the light-mass meson spectrum can provide information on those topics. New meson spectroscopy experiments planned for the future upgrade of Jefferson Lab will be also described.

ASSESSMENT OF DELIVERY METHODS IN TEACHING AN UNDERGRADUATE STATISTICS BY DISTANCE LEARNING. T.C. Mosca III, Dept. of Mathematics, Rappahannock Community College. Distance learning is sometimes criticized as being less effective than traditional methods because the teacher is not physically present in the room with the students. In the fall semester of 2011 I taught three sections of MTH 240 Statistics that were nearly identical. The classes differed only in the amount of face-to-face time. One class section was online; there was no face-to-face time. The others were two sections of a hybrid class. The hybrid class met once per week by two-way interactive audio and video connection. One section met face to face three times per month, and the other class section met face to face one time per month. Class meetings and additional lessons were recorded. Students in the online class were provided both sets of recordings. All students received exactly the same instruction, with the only difference being the amount of face-to-face time for each section. The proportions of students who were successful vs. the proportions who failed or received a grade of "D" in each section were compared using a Chi-squared multiple comparison routine. Numerical grades were compared using ANOVA. The hypothesis that face-to-face time is a determining factor in student success was not supported.

MEASUREMENTS OF STELLAR POLARIZATION USING THE VMI OPTICAL POLARIMETER. <u>Gregory A. Topasna</u> & Daniela M. Topasna, Department of Physics and Astronomy, Virginia Military Institute, Lexington, VA 24450. We have designed and built an optical polarimeter for use with the Virginia Military Institute's 0.5 meter telescope. The polarimeter uses an achromatic half-wave plate and Wollaston prism to image the ordinary and extraordinary rays onto a CCD camera after the light passes through a B, V, R, or I filter. Aperture photometry is performed to determine the flux for the ordinary and extraordinary stellar images and the normalized Stokes parameters with their associated uncertainties. Observations of unpolarized stars are used to determine instrumental polarization and highly polarized stars are used to determine the instrumental zero-point reference. We present an analysis of the polarimeter and its performance characteristics.

COMPARATIVE STUDY OF SELF-ASSEMBLED POLYMER THIN FILMS. <u>D.</u> <u>M. Topasna</u>, A. R. Firehammer, & G. A. Topasna, Department of Physics and Astronomy, Virginia Military Institute, Lexington, VA 24450. We fabricated multiple layer polymer thin films using a layer-by-layer self-assembly method by automated and manual fabrication. The films were characterized by UV-VIS-NIR and SEM measurements for a range of thickness values. The characterization results demonstrate that both methods yield films with similar optical and physical properties. 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 *XENPUS 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 hoplonenemertean planuliform larvae have greatly clarified their development and provided insight into larval evolution within the

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phylum. However, an assessment of viviparous development using modern techniques is lacking. To help facilitate a comprehensive comparative evaluation of developmental diversity within hoplonemerteans, 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. <u>Zein Al-Atrache</u> & 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. <u>A Asmar</u>, Stacey M<sup>1,2</sup>, Fecteau A<sup>4</sup>, Werner A<sup>4</sup>, Kelly R Jr<sup>5</sup>. <sup>1</sup>Frank Reidy Research Center for Bioelectrics, Old Dominion University, Norfolk, VA, USA. <sup>2</sup>Department of Pediatrics, Eastern Virginia Medical School, Norfolk, VA, USA. <sup>3</sup>Division of General Surgery, Hospital for Sick Children, Toronto, Canada. <sup>4</sup>Dept of Pathology, Eastern Virginia Medical School and Med Director of Laboratories, Children's Hospital of The King's Daughters, Norfolk VA, USA. <sup>5</sup>Department 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  $\beta$ -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<sub>s</sub>=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. <u>Shawn Mitchell</u> & 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. <u>Hunfa Asghar</u>, 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 Tchhl1. 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  $\leq 20 \ \mu m$ ), cohort 2(middle aged 20 - 35  $\mu m$ ) and cohort 3(adult > 35  $\mu$ m) 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

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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 immunoprecitation. 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 wildtypes. 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 motted-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-kB DEPENDENT FAS SIGNALING IN HEPATOCYTES. Quoc Tran<sup>1,2</sup>, Rohini Mehta<sup>2</sup>, Aybike Birerdinc<sup>2</sup>, & Ancha Baranova<sup>1,2</sup>, <sup>1</sup>Biology Department, George Mason University, <sup>2</sup>Betty 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-kB 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 (Oiagen, 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-kB 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-y) 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- $\gamma$  activity was blocked with 5 uM 5-Hydroxytamoxifen.  $\frac{1}{2}$  of each group was exposed to 5  $\mu$ 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-y receptor was active, only 4genes were upregulated and 7 were down regulated. When the ERR- $\gamma$  receptor was blocked, 3genes 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/m<sup>2</sup>). 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 Birerdinc1,2, & Ancha Baranova1,2, 1, School of Systems Biology, George Mason University, Fairfax VA 22030, 2Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church, VA 22042,3 Microbiome 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 Murthy<sup>1</sup>, Hannah Choi<sup>2</sup> & Ancha Baranova<sup>1, 2, 3</sup>, <sup>1</sup>School of Systems Biology, George Mason University, Manassas, VA,<sup>2</sup>Biology Department, George Mason University, Fairfax, VA,<sup>3</sup>Betty 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 coexpressed genes of KCNRG, using multiple cancer studies within the Oncomine database, focusing here predominantly upon brain and cns cancer studies. Metaanalysis 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 Metacore<sup>™</sup>, an integrated software suite for pathways and network analysis of OMICs data. GeneGo Pathways Maps show that top scored map (map with the lowest p value) based on the enrichment distribution sorted by 'Statistically significant Maps' set is Transport RAN reglation pathway. Analysis of cocorrelations 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*. <u>Allison Yaguchi</u> & 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

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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. <u>R.Tahir<sup>1</sup></u>, A.J.Dickinson<sup>2</sup>, & S.H.Elsea<sup>3</sup>, <sup>1</sup>Center of the Study of Biological Complexity, <sup>2</sup>Dept. of Biology and <sup>3</sup>Depts. 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 Rail 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 *Rail* is highly expressed in facial and dorsal regions of the developing embryo, with Rail expression in maxillary and nasal prominences. In addition, expression is localized to a region that appears to be migrating neural crest. A disturbance in Rail 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 Tolllike 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 tumoraltered 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 Fellowhsip, 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 PLAOUE SIZE AND COMPOSITON. 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 $\alpha$ 1 and TR $\beta$ 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 ( $\alpha 1$  and  $\beta 1$ ) 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 OG CYTOGENETIC ANOMALIES IN THE INTERPHASE NUCLEI. <u>Tatiana Glazko</u>, 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 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 normally functioning nucleus, the paternal and maternal haploid chromosome sets behave relatively independent 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 polythene chromosomes in the salivary glands of *Chironomus thummi* larvae. Close contacts between haploid chromosome sets enhance the stability of the cell' 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. <u>Valery Glazko</u>, 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' environment. Twenty five years later, we still continue to derive important scientific insights form 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. <u>Rebecca Garrett</u>, 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-ß 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- $\beta$ 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). <u>C. Gerner<sup>1, 2</sup></u>, A. Birerdinc<sup>1, 2</sup>, Z. Younossi<sup>1, 2, 3</sup>, A. Baranova<sup>1, 2</sup>, & M. Estep<sup>1</sup>, <sup>1</sup>Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA, <sup>2</sup>Center for the Study of Genomics in Liver Diseases, Molecular and Microbiology Department, George Mason University, Fairfax, VA and <sup>3</sup>Center 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 Choi<sup>1</sup>, S. Krishnamurthy<sup>1</sup> & A. Baranova<sup>1,2</sup>, <sup>1</sup>Biology Department, George Mason University, Fairfax VA, <sup>2</sup>Betty 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<sub>v</sub>) 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 coexpression 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 Collins<sup>1</sup>. M Leema<sup>1,2</sup>. E McLaughlin<sup>1</sup>, S. Nathan<sup>2</sup> and G Grant<sup>1</sup>. <sup>1</sup>SSB GMU, Manassas VA, <sup>2</sup>IHVI 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, prosurvival/anti-apoptosis and the ability to initiate EMT. We have recently discovered that T $\beta$ 4 is over expressed in IPF fibroblasts. Therefore, here we investigated the potential of the protein T $\beta$ 4 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, 10 ng/ml T $\beta$ 4 at various serum concentrations over a 7, 24, and 72-hour period. In addition stable T $\beta$ 4 over-expressing transfected A549 cell lines were derived to investigate the effect of endogenously over expressed T $\beta$ 4. Cells exposed to 0, 5, 10 ng/ml transforming growth factor- beta (TGF-B) 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<sup>1,2</sup>, Alexei Shevelev<sup>2</sup> & Ancha Baranova<sup>1</sup>, <sup>1</sup>School of Systems Biology, George Mason University, Manassas, VA 20110, USA, and <sup>2</sup> Department of Virology, Moscow State Academy of Veterinary Medicine and Biotechnology, 23 Academika 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. <u>Grant Waldrop</u> 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 where identified as three different strains of *Paenibacillus polymyxa*. The antimicrobials produced by these

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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. <u>Bethany J. Johnson</u>, 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. <u>Charise J.</u> <u>Garber</u> & 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. <u>K. E. Wong</u> & 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.

PHOSPHOLEMMAN IS A NEGATIVE FEED-FORWARD REGULATOR OF CA2+ IN B-ADRENERGIC SIGNALING, ACCELERATING B-ADRENERGIC Jason H. Yang & Jeffrey J. Saucerman, Dept. of Biomedical INOTROPY. Engineering, University of Virginia, Charlottesville VA 22903. Sympathetic stimulation enhances cardiac contractility by stimulating  $\beta$ -adrenergic signaling and protein kinase A (PKA). Recently, phospholemman (PLM) has emerged as an important PKA substrate capable of regulating cytosolic Ca<sup>2+</sup> transients. However, it remains unclear how PLM contributes to β-adrenergic inotropy. Here we developed a computational model to clarify PLM's role in the β-adrenergic signaling response. Simulating Na<sup>+</sup> and sarcoplasmic reticulum (SR) Ca<sup>2+</sup> clamps, we identify an effect of PLM phosphorylation on SR unloading as the key mechanism by which PLM confers cytosolic  $Ca^{2+}$  adaptation to long-term  $\beta$ -adrenergic receptor ( $\beta$ -AR) stimulation. Moreover, we show phospholamban (PLB) opposes and overtakes these actions on SR load, forming a negative feed-forward loop in the  $\beta$ -adrenergic signaling cascade. This network motif dominates the negative feedback conferred by B-AR desensitization and accelerates  $\beta$ -AR-induced inotropy. Model analysis therefore unmasks key actions of PLM phosphorylation during  $\beta$ -adrenergic signaling, indicating that PLM is a critical component of the fight-or-flight response.

SUCCESS OF FLATFOOT SURGICAL CORRECTION ON PLANTAR PRESSURE DISTRIBUTION. Erika A Matheis, Edward M Spratley, Charles W Hayes, Robert S Adelaar & Jennifer S Wayne, Virginia Commonwealth University, Richmond VA. Adult Acquired Flatfoot Deformity (AAFD) is a progressive disease affecting the soft tissue structures of the foot in which joint alignment degenerates and significant dysfunction results. This pre-operative/post-operative study focused on the plantar pressure foot patterns of both stance and walking of AAFD participants to determine the effectiveness of surgical correction. The pressure pattern was divided into nine regions, from toes to heel and medial to lateral, with peak pressure and % body weight loading in each region calculated. Arch index was also determined to quantify flatness as the area of midfoot loading relative to the entire foot. Additionally, participants completed two health questionnaires (SF-36 and FAOS surveys). A lateral shift in pressure loading was evident post-operatively in both the forefoot and midfoot regions. Scores on both surveys increased post-operatively. Arch index was inconclusive. Clinically, plantar pressures as well as surveys may be useful to assess the success of the surgical technique for AAFD.

SEARCH ENGINE OF PHYSIOLOGICAL CONDITIONS IN THE PHYSIOBANK DATABASE. <u>Nitin S. Panwar</u>, Graham S. Kelly, Charles E. Taylor & Gerald E. Miller, Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA. 23220. PhysioBank is an open source database where patient information from multiple sources is collected. The patient data of interest to this laboratory is the blood pressure waveform data and physician annotations pertaining to pathological events that occurred during data collection. However, the size of the library (27000 files) makes it difficult to locate files of interest. This Matlab program has been implemented to sort and categorize these files to make relevant data easily accessible. A search of the databases was performed to obtain a list of files that contain blood pressure waveform data. This list is then compared against every annotation (57)

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in the PhysioBank approved annotation list to filter for records that contain pathological events; yielding 3700 records. The list is filtered to include only the databases of interest (Mimic I, Mimic II, MGH/MF, Fantasia, SLPDB); as these databases have the most complete waveform data. Finally, the records are narrowed to create a unique download list of individual files each with a physiological annotation (1990 records). This method provides a faster alternative to the Waveform Database (WFDB) toolbox for Matlab, as it accesses the database information directly from the website. It delivers functionality not seen in the WFDB tools through the identification of blood pressure waveform records containing specified annotations. This projects assists the current work of identifying gold standards for pathological event effects on blood pressure waveforms.

FINITE ELEMENT ANALYSIS OF TRANSVERSE MEDIAL MALLEOLAR FRACTURE FIXATION. Ruchi D. Chande, John R. Owen & Jennifer S. Wayne, Virginia Commonwealth University, Richmond VA. Fracture of the medial malleolus, or distal end of the tibia, can occur in pronation loading scenarios. If such ankle injury is left untreated, more severe conditions such as osteoarthritis can result. Via various devices, open reduction/internal fixation (ORIF) may be utilized to secure the malleolar fragment to the proximal tibia. In this study, finite element analysis (FEA) was employed to investigate transverse fracture fixation by two cancellous screws or a relatively newer fixation device known as the Medial Malleolar Sled<sup>™</sup>. The performance of these two devices in both tension and torsion was first assessed during cadaveric testing. Following experimentation, SolidWorks was used to develop a computer model of the study, and this model was then validated against experimental results by performing FEA. Force, torque, and displacement results demonstrated the validity of the models, and stress analyses were successful in predicting regions of failure corresponding to those observed during experimentation. Such results illustrate the general utility of computational modeling for the investigation of biomechanical systems.

DESIGN OF A COMPUTATIONAL MODEL FOR ELBOW JOINT BIOMECHANICS. C. A. Woodcock, E. M. Spratley & J. S. Wayne, Orthopaedic Research Laboratory, Virginia Commonwealth University, Richmond VA 23284. Computational modeling is an effective but underutilized method to study the biomechanics of joints. It provides a high degree of adaptivity and reproducibility, while enabling the investigator to study parameters such as stress, strain and forces that are not easily measured otherwise. Using computed topography scans of a cadaver specimen and Mimics<sup>TM</sup> (Materialise) software, three-dimensional representations of bony anatomy were created using masking and remeshing tools. A functional computational model of the elbow was then developed using the commercially available software package SolidWorks<sup>TM</sup>. Soft tissue constraints were defined and modeled using osteoarticular surfaces and in situ strains. The model was validated through an abbreviated reproduction of the cadaver study Hull et al (2005), which explored the effects of coronoid process resections on varus stability of the elbow. Thus, it was shown that the model accurately represented elbow joint biomechanics via physiological movement and quantitative constraining loads during applied perturbations throughout a range of motion. Further expansion upon the computational

model will focus on radial head contributions to stability, as its effects are under researched despite being the secondary constraint against valgus instability.

REAL-TIME DIGITAL SIGNAL PROCESSING OF MOCK CIRCULATORY LOOP PRESSURE SENSOR DATA USING AN XPC TARGET SOLUTION. Robert B. Thompson, Charles E. Taylor & Gerald E. Miller, Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA. 23220. Real time analysis and control of a mock circulatory loop is highly desirable. Mock Circulatory loop experimentation involves extensive analysis of the cause and effect relationship between many parameters, including pressure at determined points in the mock circulatory loop. Real time analysis allows for dynamic experimentation; high frequency iterative acquisition of parallel sensor data with automatic or host operator controlled response of control of mock circulatory loop parameters. A National Instruments Embedded Controller, operating a parallel Input/Output device, under the LabVIEW Real-Time Kernel is proposed as an xPC target to run a LabVIEW Virtual Instrument (VI) to accomplish dynamic mock circulatory loop experimentation. A VI consisting of an eight Hertz timed loop and network published variables is created to acquire pressure sensor data from a mock circulatory loop. Pressure sensor voltage, acquired at 512Hz, is mathematically manipulated as a dynamic data type on the xPC target to determine real time measurements of cardiac output and total peripheral resistance. The network published variables are read and displayed by a host computer, allowing for operator monitoring and control. An xPC target will perform mock circulatory loop signal processing and control outside of a PC operating system environment, enabling high frequency sampling, up to 1MHz, and unloading the processor requirements of the PC.

APPLICATION OF SIMULINK CODER™ TO A COMPUTATIONAL MODEL OF A SYSTEMIC MOCK CIRCULATORY LOOP. Charles E. Taylor & Gerald E. Miller. Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA 23220. Mock circulatory loops provide an essential in vitro assessment tool for evaluating cardiovascular devices through their operation as a hydraulic analog to the human circulatory system. A computational model of this physical system enables experimental settings to be pre-determined, which assists the efficiency of in vitro experimentation. The development of computational models has been traditionally an intensive programming effort. Utilization of Simulink® Simscape<sup>™</sup> toolboxes expedites the model construction process and yields a high fidelity computer model. However, these pre-constructed physical modeling components are computationally intensive. The simulation times for large models may take longer than the experimental run time in the mock circulatory loop. Simulink Coder™ can accelerate these models by compiling the code into more efficient C code, which can be executed more efficiently outside of the Matlab® runtime environment. Various levels of the Simulink Coder<sup>TM</sup> (Accelerator, Rapid Accelerator, Rapid Accelerator Standalone) provide improvements in execution, and the approach of each level in code packaging will be discussed. The impact of the acceleration on a computational model of a mock circulatory loop will be discussed with a focus on the topology of the model and what sections prevented simulation time reduction.

DESIGN OF AUTOMATED DETECTION OF INCOMPLETE EMPTYING FROM VENTILATOR GRAPHICS: EVALUATING THRESHOLD VALUES. Nvimas Y. Isti Arief, Curtis N. Sessler, Paul A. Wetzel, and Mary Jo E. Grap, Dept. of Biomedical Engineering, Virginia Commonwealth University. Auto-PEEP stemming from incomplete emptying of breath inhalation is an undesired excessive pressure in the lungs of patients on invasive mechanical ventilator has been a hidden presence that hinders optimal care. Noninvasive indicator for the presence of auto-PEEP is known to those who specialize in ventilator graphics whom are very rare. Incomplete emptying of the breath indicative of auto-PEEP can be automatically detected through a computerized algorithm. Two distinct algorithms have been developed; the first being dependent upon pressure waveform and the second is evaluating flow waveform independent from pressure. Both algorithms are dependent upon varying threshold values. For one criterion of the threshold, the first algorithm yields a sensitivity of 84.7% and specificity of 92.6%, and the second algorithm yields sensitivity of 90.1% and specificity of 82%. Thresholds are adjustable for finding the optimum rate of detection for incomplete emptying that can eventually be used for an automated detection of auto-PEEP.

HIGH-LEVEL GPU COMPUTING IN MATLAB®: TWO CASE STUDIES OF ACCELEREYES JACKET IN BIOMEDICAL ENGINEERING APPLICATIONS. Graham S. Kelly, Charles E. Taylor & Gerald E. Miller, Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA. 23220. Graphical Processing Units, or GPUs, have emerged as important scientific computing tools due to their ability to handle computationally intensive algorithms in parallel, providing drastic reductions in execution time. As the demand for GPU acceleration has increased, higher-level computing languages have emerged to streamline the process of memory transfer to and from the device and executing custom kernels (e.g. CUDA, OpenCL). AccelerEves Jacket, a GPU computing environment for MATLAB®, provides the very-high-level functionality of MATLAB® with speed and overhead superior to the native MATLAB® Parallel Computing Toolbox™. We illuminate the usefulness of Jacket through two differently structured Particle Image Velocimetry algorithms, which track particle displacement in moving fluid. These algorithms show marked improvements in execution time with minimal changes in code between the CPU and GPU variants.

### **Botany**

THE EFFECT BY HYDROLOGIC REGIMES AND SAHDE ON ATLANTIC WHITE CEDAR (*CHAMAECYPARIS THYOIDES*) GROWTH IN THE CAVALIER WILDLIFE MANAGEMENT AREA IN CHESAPEAKE, VIRGINIA. Justin L. Weiser, Jackie Roquemore, & Robert B. Atkinson, Department of Organismal and Environmental Biology, Christopher Newport University, Newport News VA. 23606. The Virginia Department of Game Inland Fisheries began restoring a 1538-ha Atlantic white cedar (AWC) Swamp in Chesapeake, Virginia in 2007. Reestablishment of AWC is critical in restoration of this globally- threatened ecosystem. The purpose of this study is to compare morphometric parameters of two tree planting types, propagated seedlings and rooted cuttings, at two locations characterized by hydric and mesic

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hydrologic regimes, which were assigned via prevalence index of wetland indicator status for non-cedar vegetation. In August 2010 and 2011, field crews quantified survivorship and growth (estimated by height, canopy diameter and stem diameter), and shade intensity at each location. Data were analyzed using t-tests and linear regressions. Growth was significantly greater (p<0.05) for rooted cuttings than for propagated seedlings over one growing season and shade negatively impacted all growth indices. Growth in hydric hydrologic regime for all three morphometric parameters was less than in mesic plots (p < 0.05). Rooted cuttings had greater mean growth than propagated seedlings and shade is greater in the hydric plots. Reestablishment of AWC may be effective in sites that have a prevalence index of between 2.5 and 3.5; however, mesic sites might lack the self- maintenance capacity in that seeds could be destroyed by fire.

THE FLORA OF VIRGINIA PROJECT: A 2011-2012 UPDATE. <u>Marion B. Lobstein</u>, (Retired) Dept. of Biology, Northern Virginia C.C., Manassas, VA 22205. Virginia, for its landmass, has the most diversity of vascular plant species of any state in the United States. The colony of Virginia had the first flora, of any of the 13 colonies, the *Flora Virginica* in 1739 and last updated 250 years ago in 1762, yet Virginia does not have a modern *flora*. The Virginia Academy of Science for over eighty years has supported efforts to produce a modern *Flora of Virginia*. In 2001 the Foundation of the Flora of Virginia (FFVP), Inc, was formed and by May 2002 received 501(c) 3 status. Since 2001 both the Academy and the Fellows have generously given both financial and moral support to FFVP in the development of a modern *Flora of Virginia*. The publication date of the *Flora of Virginia* by the FFVP is December 2012. BRIT (Botanical Research Institute of Texas) Press will be the publisher. The *Flora of Virginia* will be a 1,500 page manual describing and aiding in the identification of the 3,200 species of vascular plants that are native or naturalized in Virginia.

SOIL COMPOSITION IN THE GREAT DISMAL SWAMP: BEFORE AND AFTER FIRE. <u>Kristina M. Kowalski</u>, Jackie Roquemore & Robert B. Atkinson, Christopher Newport University. The physical and chemical properties of peat soils are influenced by fire and may effect regeneration of native species, such as Atlantic white cedar (AWC) in the Great Dismal Swamp National Wildlife Refuge (GDSNWR). This study was conducted to determine soils changes associated with the 2008 South One Fire. In 1999 soil samples were collected from GDSNWR AWC stands that had not burned in at least several decades including 27 plots in 3 stands. In the summer of 2011, samples were collected from 21 plots in 5 AWC stands that burned in the 2008 South One Fire and 2 stands containing 7 plots that were unburned. Soil samples for all years and plots were collected at 10 cm depth and AWC needle samples were also collected from the 2011 plots. Grand mean bulk density increased from 0.148 g cc<sup>-1</sup> to 0.214 g cc<sup>-1</sup> in burned, and 0.173 g cc<sup>-1</sup> in unburned stands. Grand mean carbon remained near 47.5% across all treatments. Total nitrogen increased, but in both burned and unburned stands. There were no clear trends in AWC tissue total nitrogen content.

ATLANTIC WHITE CEDAR REGENERATION IN THE GREAT DISMAL SWAMP. <u>Shawn J. Wurst</u>, R. B. Atkinson & J. D. Roquemore, Department of Organismal and Environmental Biology, Christopher Newport University, Newport News VA 23606. A peat-based seed bank underlies many East Coast Atlantic white

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cedar (AWC) swamps, and this globally-threatened ecosystem exhibits selfmaintenance through high rates of natural regeneration after a stand-clearing fire. AWC stands in the Great Dismal Swamp have been in decline for approximately 200 years at least in part due to the draining of water by ditches. In this study we report the amount of regeneration of AWC in the Great Dismal Swamp after the South One Fire of 2008. For regeneration in 8-m<sup>2</sup> plots during 2009 and 2010, regenerants were counted in 143 and 41 plots, respectively. Mean regeneration in 2009 (26,500 ± 23,800 stems ha-1) was not significantly different than in 2010 (29,300 ± 38,000 stems ha-1, Paired t-Test *P*=0.315). Regeneration rates suggest that most regeneration occurred in the first year after the fire. Regeneration rates reported in the literature were much higher for natural cedar swamps and the lower regeneration rate in the current study might be the result of low water tables coincidental with the 2008 fire.

ANTIOXIDANT ANALYSIS OF SPICES FROM THE APIACEAE. R. A. McNeive and M. H. Renfroe, Dept. of Biology, James Madison Univ., Harrisonburg VA 22801. Antioxidants in one's diet are an important component of protection from cellular oxidative damage, effects of overall aging, and certain diseases such as some forms of cancer. Spices from plants have been shown not only to have antimicrobial properties, but also to contain potent concentrations of antioxidants. One plant family, the Apiaceae, has provided more spices to the human diet than perhaps any other single plant family. Therefore we analyzed the antioxidant content of seven of the spices from Apiaceae to establish an intrafamilial comparison. Results indicated these spices contained greater quantities of hydrophilic antioxidants than lipophilic antioxidants. Fennel, cumin and dill contained the greatest quantities of hydrophilic antioxidants, while celery contained the greatest quantity of lipophilic antioxidants. Fennel, cumin and dill contained significantly greater concentrations of total antioxidants compared to celery, anise, which were not statistically different from one another, but which were significantly greater than caraway, which was significantly greater than coriander. Analysis of these spices will increase our understanding of the intrafamilial variation in antioxidant production by plants and also provide valuable information for dieticians interested in considering antioxidants as a dietary component. From this, there were no large differences between fresh weight measurements and dry weight measurements of the spices. Celery was found to be no different than anise regarding antioxidants. Cumin was found to be equal to dill and fennel while caraway and coriander were each different from the other spices.

INTERACTIVE EFFECTS OF ALLELOPATHY, PHOTOPERIOD, AND TEMPERATURE ON THE GROWTH OF *CUCUMIS SATIVUS*. Catherine Daniels & Mary E. Lehman, Dept. of Biological and Environmental Sciences, Longwood University, Farmville VA 23909. Allelopathy involves the interaction of plants through the release of chemicals into the environment, often negatively affecting the growth of surrounding plants. Little is known about how variability in environmental factors interacts with allelopathy. Cucumber seedlings were grown in nutrient culture systems containing 0-0.8 mM ferulic, *p*-coumaric or salicylic acid, three common allelopathic chemicals. The seedlings were also exposed to either a 6- or 12-hour photoperiod and to either variable or constant temperatures. Some significant interactions were seen, but were not consistent across all allelochemicals and environmental conditions. The

strongest effects were seen with the reduction in allelopathic effects of salicylic acid under lower temperatures and shorter photoperiods, even when light intensity was adjusted to maintain equivalent total energy supply. Additional experiments suggest that the significant interaction is only between allelopathy and temperature.

APOMIXIS IN A PISTILLATE CLONE OF FLUEGGEA SUFFRUTICOSA (PHYLLANTHACEAE). Jessica Kelly & W. John Hayden, Department of Biology, University of Richmond, Richmond, VA 23173. Pistillate specimens of Flueggea suffruticosa, dioecious shrubs native to temperate Asia, produce viable seeds in the absence of pollen. First (spring) flowers abort shortly after anthesis but some later (summer) flowers form fruits with apomictic seeds. Summer flowers, fruits, and seeds from plants cultivated in central Virginia were studied via light microscopy to document development of apomictic seeds. Ovules are hemitropous, bitegmic, crassinucellate, with nucellar beaks and obturators. Embryo sacs abort shortly after anthesis; nevertheless, many ovaries and ovules persist and grow despite absence of embryos. The hypostase region is responsible for some post-anthesis growth of unfertilized ovules but no apomictic embryoids were observed in this region; approximately 90% of these abort after enlarging three times greater than their size at anthesis. In about 10 % of post-anthesis ovules studied, adventitious embryony initiates ca two weeks post anthesis via mitotic proliferation of nucellus cells located in the general vicinity of the former egg apparatus. Early apomictic embryoids are irregular masses of cells; ordinary basal cells and suspensors cannot be distinguished. Mature apomictic seeds possess bi-layered sclerified testa and straight dicotyledonous embryos surrounded by relatively empty nucellus cells; only one apomictic seed studied possessed densely cytoplasmic nucellus cells. This is the first report of apomixis in Phyllanthaceae.

POLLINATOR PREFERENCE BASED UPON ULTRAVIOLET CUES. L. Abbott & L. Horth, Dept of Biology, Old Dominion University, Norfolk, Va. 23529. Pollinators are responsible for 1/3 of all the food that humans consume. This accounts for 15 billion dollars in food crops annually. Ultraviolet Cues are a way to attach pollinators to these crops. Ultraviolet cues are ray that are emitted from the sun and absorbed by the petals of the plant producing a black ring. Two experiments were preformed one on *Rdubekia hirta* and one on *Ranunculus* sp. In both of these experiments bees were monitored for there preference between large UV cues and small UV cues. In the *R. hirta* experiment three flower types were used: an enhanced flower (90% of the flower petal had an ultraviolet cue), a cut and paste control flower (roughly 45% cue), and an unmanipulated flower (roughly 45% cue). This same manipulation was done in one of the subset experiments for the buttercup. The other subset of the *Runculus* experiment was monitoring natural UV cues to see if the small margins in UV had an effect on what flowers bees chose. In all experiments the pollinators showed preference to the large Ultraviolet cues. Pollinators chose the large ultraviolet cues in; wild R. hirta, cultivated R. hirta, unmanipulated Ranunculus and enhances Ranunculus. Regardless of the species and treatment of the plant when the flower had a larger Ultraviolet cue pollinators chose this cue more often than a smaller UV cue. Due to the recent decline in bees, knowing what bees chose to pollinate can be a solution to sustain more crops.

ANTIOXIDANT ANALYSIS OF COMMERCIAL SPICES. D. Killeen and M. H. Renfroe, Dept. of Biology, James Madison Univ., Harrisonburg VA 22801. There is a growing awareness of the role of antioxidants as an important component of human health. Antioxidants have been implicated in preventing degenerative diseases such as cancer, cardiovascular and neurological diseases, and reducing the effects of aging. Spices have long been part of the human diet and are known to have antimicrobial properties. We investigated eleven spices from around the Mediterranean, southern and southeastern Asia, and the West Indies. Cloves (823 µmol TE/g dw) and cinnamon (387 µmol TE/g dw) contained the greatest hydrophilic antioxidant content. Sage (68 µmol TE/g dw) and rosemary (60 µmol TE/g dw) contained the greatest lipophilic antioxidant content. Overall, cloves (835 µmol TE/g dw) and cinnamon (397 µmol TE/g dw) contained the greatest total antioxidant content. Other spices analyzed were oregano, mint, marjoram, allspice, ginger, allspice, and turmeric. Results indicated that spices are very potent sources of antioxidants and can complement one another to provide a variety of antioxidant molecules. Spices can complement other dietary components such as fruits and vegetables to greatly increase available dietary antioxidants.

REHABILITATION OF RARE SPECIES POPULATIONS. Erin Gillin<sup>1</sup>, Stephen W. Fuller<sup>1</sup> & Phillip Sheridan<sup>2</sup>, <sup>1</sup>University of Mary Washington, <sup>2</sup>Meadowview Biological Research Station. Cell tissue culture of plant seeds has been used in previous research for horticultural and commercial purposes. This study focuses on successful seed propagation for the restoration of a variety of rare and endangered species. A standard tetrazolium test was used to determine the viability of the seeds. Platanthera blephariglottis (Willdenow) Lindley, white-fringed orchid, and Tetragonotheca helianthoides L., pinelands nerve-ray, seeds were used for both cell tissue culture and soil planting propagation. In cell tissue culture the seeds were sown aseptically on nutrient full media and stored in sealed sterile bags. Orchid protocorm development was expected, but only one vessel withstood contamination long enough to see some growth before also becoming contaminated. Failure in the aseptic processes caused loss of results for the project. Additionally, no growth was seen in the soil flats. This procedure has been successful in previous research, so we suggest further development of the methods to decrease contamination growth. With attention paid to sterile technique, plantlet development should be attainable in tissue culture for application for large scale production and wild population restoration. Further work with seed stratification and soil types is also recommended.

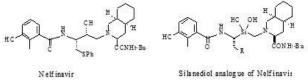
### Chemistry

ASSESSMENT OF GUIDED INQUIRY IN GENERAL CHEMISTRY LABS: A THREE YEAR PROJECT. <u>Heather N. Anthony</u> & Jack K.Steehler, Department of Chemistry, Roanoke College, Salem VA 24153.This project investigates student improvements in chemistry laboratories when guided inquiry experiments are added. The project investigated student anxiety levels, content learning, and student perceptions of instruction. Three years of General Chemistry 111-112 were analyzed; the first year studied included mostly cookbook experiments, while years two and three

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included a mixture of cookbook and guided inquiry experiments. Lab anxiety decreased after completing the course. The years in which guided inquiry experiments were incorporated saw a smaller decrease in anxiety about time availability. Students gained content knowledge during each experiment, regardless of pedagogy. For most experiments in which pedagogy changed, student content learning did not change significantly when the guided inquiry format was used. Students perceived *designing experiments yourself* as least helpful to their learning; however, as guided inquiry was added, their perception of helpfulness on this topic increased. This study was funded by the National Science Foundation.

COMPUTATIONAL EVALUATION OF SILICON-BASED HIV PROTEASE INHIBITORS, <u>Chynna Blaker</u>, Latricia Bowman, Victoria Parker & Wondwossen Arasho, Dept. of Chemistry and the Center for Biotechnology and Biomedical Sciences, Norfolk State University, 700 Park Avenue, Norfolk, VA 23504. There is no known cure for HIV/AIDS and it is imperative to develop a drug that will prevent the replication of the virus, without the stressing concern for resistance. Scientists across the globe struggle to create a mechanism that will slow down the mutations of the virus and suppress viral load in the infected human. This study focuses on HIV-1 inhibition therapy and utilizes the drug Nelfinavir (Viracept®) as a starting platform for preparation of a newly improved silicon-based drug. SYBYL software was used to test and compare the effectiveness of Nelfinavir against the silanediol analogue and other modified silanediol analogues. The original silanediol analogue (4.48p/l) proved to more effective than Nelfinavir (2.68p/l) according to the Surflex dock calculations from the SYBYL software.



THERMODYNAMICS OF THE SN2 VERSUS E2 COMPETITION. <u>Charles M.</u> <u>Bump</u>, Department of Chemistry, Hampton University, Hampton, VA 23668. Reactions between alkyl halides and strong bases give a mixture of both substitution  $(S_N 2)$  and elimination (E2) products. The major product is usually discerned by students of organic chemistry on the basis of rote memorization of generalizations such as "2° halide plus strong base = elimination". The "answer" is technically correct, but an analysis of the factors that influence the major product is frequently sacrificed in favor of a quick answer to a complex question. Thermodynamics and kinetics are two major driving forces for organic reactions. We will describe how calculated thermodynamic properties (Gaussian 98, B3LYP) predict whether substitution or elimination is favored in these reactions.

THERMAL DECOMPOSITION OF METAL HYDROXIDES, A.Bagley, G.Berman & <u>T.C. DeVore</u>, Dep. of Chemistry and Biochemistry James Madison University, Harrisonburg VA 22897. Serious concerns about the quality of decomposition dynamics measurements obtained using standard thermal analysis methods have been

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expressed over the past 25 years. The thermal decomposition of Al(OH)<sub>3</sub> has been investigated under a variety of experimental conditions to further explore the cause of these concerns. The results of these investigations indicate that while analysis using standard techniques such as those developed by Ozawa, Kessinger, and others appear to give describe the dynamics well, they give different answers as experimental conditions change. For example, the apparent activation energy increases by ~ 10 kJ mol<sup>-1</sup> is decreased from 10 mg to 0 mg (determined by measuring the dynamics at several sample masses and extrapolating to zero mass). These results clearly indicate that this decomposition does not follow a one step mechanism and clearly indicate the need to use several sets of experimental conditions to determine the reaction dynamics for the process.

3,4:3',4'-BISBENZO[B]THIOPHENE AND RELATED DERIVATIVES: SYNTHESES AND INVESTIGATION OF THEIR ELECTRO-OPTICAL, CHELATING, AND BIOLOGICAL PROPERTIES. Marissa L. Estep, Diego S. Suarez Boscan, Matthew VanTil, Zachary R. Rhodenizer, Garth McGibbon & Michael R. Korn, Dept. of Biol. & Chem., Liberty Univ., Lynchburg, VA 24502. 3,4:3',4'-Bisbenzo[b]thiophene (BBT) was prepared following a published patent protocol. Molecular modeling computations of BBTcalculated itsbandgap (i.e. the energy difference between the HOMO and of LUMO) to be 3.61 eV (343 nm) (DFT, B3LYP, 6-31G\*); to test for the accuracy of these computations, UV-vis spectroscopy of BBT was performed. The resultingspectrum (in dichloromethane) showed two maxima at approximately 383 nm and 365 nm, and two smaller peaks at 349 nm and 332 nm (shoulder); the UV-vis cut-off wavelength was at 396 nm (3.13 eV). BBT was tested for intercalation into DNA because of its planar structure. Agarose gels were run in the presence of BBT and a DNA ladder (0.5-10 kb); however, no intercalation of BBT was observed as determined by the absence of fluorescence when gels were exposed to wavelengths of 254 and 365 nm; additional experiments are planned to further investigate the interaction of BBT with agarose gels as well as with DNA. BBT was also tested for antibacterial activity because of its two sulfur atoms; two strains of Pseudomonas aeruginosa were investigated which were grown in the presence of small paper discs soaked with BBT (from water and from DMSO). No growth inhibition was observed. Chelating properties are still under investigation as well as improved synthetic pathways to increase yields and reduce overall synthetic steps. (Supported in part by a \$500 award from the Virginia Academy of Science).

A LUMINESCENCE DEMONSTRATION REACTION. <u>Susan Hannegan</u> & H. Alan Rowe, Department of Chemistry, Norfolk State University, Norfolk, VA 23504.Luminol ( $C_8H_7N_3O_2$ ), also known as o-aminophthalylhydrazide, 5-amino-2,3dihydro-1,4-phthalazinedione,and 3-aminophthatichydrazide, is used in popular luminescent chemical demonstration reactions as well as in forensic science for the detection of blood. Luminol added to a metal catalyst in a basic solution is oxidized resulting in the release of energy as light (maximum wavelength of 424 nm). Light from the oxidation of luminol is the basis for a sensitive method for the detection of blood. A popular chemistry luminescent lecture demonstration involves the addition of two solutions in the dark creating a dramatic glow. In one popular version, the solutions are noted as "A-1", consisting of luminol, in a complex salt solution, and

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"A-2" a hydrogen peroxide solution. To establish the optimum conditions for this reaction each component wasvaried and the response evaluated using a computerinterfaced light sensor. De-oxygenation of both solutions resulted in an increase in both intensity and half-life presumably due to the elimination of quenching. Inclusion of cobalt instead of copper as the metal catalyst increased bothparameters. Alteration of the hydrogen peroxide concentration did not significantly improve the results, while exclusion of some components had a reciprocal effect: increasing intensity and decreasing half-life (sodium bicarbonate) and vice versa (sodium carbonate).

OYSTER RESTORATION AND WATER QUALITY ANALYSIS IN AN URBAN SUB-ESTUARY. Melinda Hopper, Matthew Boyce, & Dr. Maury Howard, Chemistry Department, Virginia Wesleyan College, 1584 Wesleyan Drive #B314, Norfolk, VA 23502. This study was conducted to monitor a local body of water and to determine the effect of oysters on water quality. The purpose is to determine whether oyster restoration can help the quality of the water and whether this site is an ideal location for restoration. Water samples were collected over a year long period at 3 different Each sample was tested for various water quality indicators, including sites. chlorophyll a, total phosphorus, temperature, pH, BOD and salinity. Sediment samples and oyster tissue samples were also collected and run through an ICP-MS to identify the toxic metals found within them. High concentrations of lead, mercury, arsenic, and other heavy metals were some of the main components. The water contained very low levels of metals due to the tidal influence. However, the sediments and oyster tissue contained high concentrations of many of them, showing the effects of bioaccumulation. It also shows the value of the ovsters as filters for the system, removing toxins and nutrients from the ecosystem.

NEW SULFONE-DERIVATIVE PHENYLENEVINYLENE-BASED CONJUGATED POLYMERS FOR OPTOELECTRONIC APPLICATIONS. Thuong H. Nguyen & Sam-Shajing Sun, Department of Chemistry., Norfolk State University., Norfolk VA. 23504. A series of stable, processable, and end functionalizable sulfone-derivatized phenylenevinylene based conjugated polymers (SFPVs) containing different donor type co-monomers have been synthesized and characterized. The polymer main chains consist of a sulfone-phenylene electron accepting unit coupled with an electron donating unit derived from one of the dialdehyde co-monomers based on benzene, thiophene, and pyrrole. The solution optical energy gaps and the electrical energy gap (thin films) of these polymers are in the range of 1.94 - 2.45 eV and 2.04 - 2.51 eV respectively. The lowest energy gap was obtained from the polymer containing pyrrole unit due to the smallest resonance energy. The vinylene bounds on the polymer main chain are still chemically stable to survive strong basic conditions as compared to the S,S-dioxo-thiophene based PTV polymers developed earlier. These polymers also have very good thermal stability (onset decomposition temp, in N<sub>2</sub> gases, >270 °C). The lower energy gap P(Pyrrole-SFPV) exhibited ten times better photoelectric power conversion efficiency than the higher energy gap P(TV-SFPV). The diphosphonate ends functionalized and frontier energy level engineered of these conjugated polymers are very attractive in development of supramolecular block copolymers for the next generation of optoelectronic devices particularly in solar energy conversion applications.

BLUING OF HYDRANGEA MACROPHYLLA SEPALS BY INTRODUCING ALUMINUM IONS THROUGH ROOTS, CUT STEMS, AND SEPAL SURFACES. Henry D. Schreiber, Corinne M. Lariviere, Andrew H. Jones, Kelly M. Mayhew & Judith B. Cain, Department of Chemistry, Virginia Military Institute, Lexington, VA 24450. Sepals of many Hydrangea macrophylla are red when grown in basic to neutral soils, blue in acidic soils, and shades of purple in soils of intermediate pH. Delphinidin-3-glucoside that provides the red color to the sepals is transformed to a blue complex in the presence of molar Al(III). Al(III) is only mobile in acidic soils so the roots can only assimilate aluminum as a citrate complex into the plants under acidic soil conditions. The threshold Al(III) content for bluing is about 40-80  $\mu$ g/g fresh sepal. If the Al(III)-citrate complex is introduced into cut stems of red hydrangea inflorescences, several orders of magnitude more Al(III) must be distributed to the sepals before bluing occurs. In addition, the bluing is not homogeneous throughout the sepals, but instead results in unique patterns in which the blue color advances from the center and outer edges. Evidently, the diffusion of Al(III) to the reaction centers, or pigment location, in the cell vacuoles is slow. An aqueous spray of a pH-adjusted Al(III)-citrate solution was also directly applied to the sepals over a period of time. The rapidity of the bluing was related to the pigment concentration of the sepals; the cultivars with the lighter sepals and less pigment blued faster as less Al(III) was required for a molar excess. The threshold Al(III) contents were about the same as when obtained through the roots.

HETEROGENEOUS OPENING OF EPOXIDES AND EPISULFIDES WITH AMINO ACIDS – AN APPROACH TO THE SYNTHESIS OF THIOLS. <u>Marc-Antoine</u> <u>Tremblay, Eric P. Ginsburg</u>, Chelsea L. Brown, Ellen R. Simmons, Dylan J. Jamieson & Jeffrey M. Carney, Department of Molecular Biology and Chemistry, Christopher Newport University, Newport News, VA 23606. Cystinuria is a metabolic disorder that results in the accumulation of insoluble and reoccurring cystine stones in the kidneys. Only a few methods for the treatment of cystinuria exist and surgery to remove the stones is frequently required. Further research is necessary to determine how to best prevent and treat this painful disorder. One known treatment involves the use of thiols to break up cystine and increase solubility of the stones through disulfide exchange. Our vision is to develop a small library of thiol compounds via the nucleophilic opening of episulfides with amino acid derivatives. Epoxide ring-opening with amino acids represents a simpler model system, but it has also not been extensively studied under heterogeneous conditions. We report our progress in the development of both epoxide- and episulfide-ring opening using amino acids and a solid catalyst.

MEASURING STUDENT COMPREHENSION OF LEARNING OBJECTIVES IN ORGANIC CHEMISTRY COMPUTATIONS EXPERIMENTS, <u>M.K. Waddell</u>, C. Bump, E. Ndip, and G. Nwokogu, Dept. Of Chemistry, Hampton University, Hampton, VA 23668. The incorporation of computation macromodels into several organic chemistry laboratory experiments has been an ongoing endeavor. As a result, the measurement of student comprehension of these computational experiments has been assessed. Students were surveyed on their perceptions of the learning objectives of an organic chemistry computation experiment. Pre and post surveys were administered through the BlackboardTM course webpage. Results were compared to identify trends and key areas of learning deficiencies.

## **Computer Science**

ALERT: AN ARCHITECTURE FOR THE EMERGENCY RETASKING OF WIRELESS SENSOR NETWORKS. Sved R. Rizvi, Stephan Olariu, & Michele C. Weigle, Dept. of Computer Science, Old Dominion University, Norfolk, VA 23529. When an emergency or disaster strikes, first responders work as part of a complex emergency management network that calls upon many functions, resources, and capabilities. The objective of our research is to design a real-time information system to improve emergency-response functions by bringing together information to respond to a terrorist attack, natural disaster or other small or large-scale emergency. We call this system ALERT: An Architecture for the Emergency Retasking of Wireless Sensor *Networks*. The novel contribution of this research to the emergency response strategies is the seamless integration of various wireless sensor networks by retasking them with explicit missions involving a dynamically changing situation. Preliminary results have shown that retasking sensor networks for emergency response is a promising new paradigm that can not only promote a wider adoption of sensor network systems in support of guarding our national infrastructure and public safety, but can also provide invaluable help with disaster management and search-and-rescue operations.

DENSE UNSTRUCTURED AND STRUCTURED MATRIX COMPUTATIONS USING MPI. Stephen V. Providence, Dept. of Computer Science, Hampton University, Hampton, Virginia 23668. Computations with dense unstructured general matrices requires  $O(n^3)$  operations and  $O(n^2)$  words of storage for  $n \ge n$  input matrices. Such matrices are encountered in applications to solving integral equations. The best algorithms involving computations with dense structured matrices require  $O(n \log^2 n)$ operations and O(n) words of storage with small overhead constants. MPI or the message passing interface binds to the C programming language and is used to implement parallel algorithms. The time and space complexity estimates above are for sequential algorithms involving matrix computations. We have interest in the complexity estimates for parallel implementation of the sequential versions. For p processors where p < < n, straightforward complexity estimates are proportional to  $O(n^3)/p$  operations and  $O(n^2)/p$  words for parallel implementation of dense unstructured general matrix computations. Analogously, computations with dense structured matrices require  $O(n \log^2 n)/p$  operations and O(n)/p words for parallel implementation. We conduct experiments on a high-performance computing cluster computer system to obtain the hidden constants in the O-notation of the estimates given.

# Education

THE IMPROVING GROUNDS EXHIBITION. E. G. Maurakis, R. Conti, and D. Hagan. Science Museum of Virginia. Objectives of the Improving Grounds exhibition project are to create exhibits, programs, audio and video media for mass communication, and web-based curriculum materials on how to improve health and fitness. The overarching theme is a science perspective on understanding, testing, and measuring self-improvement in health and fitness. Exhibits and programming will be

based on the 21<sup>st</sup> century learning and innovation skills (critical thinking, problem solving, creativity, innovation, communication, collaboration, visual literacy, scientific and numerical literacy, and cross disciplinary thinking) of the Institute of Museums and Library Services (IMLS), which are consistent with the Point 1 Virginia Science Standards of Learning. A curriculum integrated web portal will contain a data bank of health and nutrition facts that students, teachers and the general public can use to track their own health and fitness improvements over time and compare them with like populations. Funded in part by Health Diagnostics Laboratory, Inc.

INCORPORATING A JOURNAL CLUB EXPERIENCE IN THE UNDERGRADUATE BIOCHEMISTRY CURRICULUM. Lisa S. Webb, Department of Molecular Biology and Chemistry, Christopher Newport University, Newport News, VA. In an effort to increase student exposure to the primary literature and improve critical thinking skills, a journal club experience was incorporated into the Introductory Biochemistry courses. Students were expected to read the assigned article, conduct relevant background research necessary to understand it and place it in context, and discuss it in class. A rubric for evaluating classroom participation in the discussions was presented. In addition to discussing the articles in class, there was also a midterm and final quiz for the journal club portion of the grade. The quiz questions varied in level, with the first questions lower on Bloom's Taxonomy (knowledge, comprehension) and later questions much higher (application, analysis, evaluation). Anonymous student responses to the journal club were mixed. Many students commented on how much they learned from (and, in a few cases, enjoyed) the experience, but several complained about how hard they had to work to understand the material. Overall, it was a positive experience that will be repeated.

SO MANY PLANTS - SO LITTLE TIME. E. G. Maurakis, University of Richmond. Objectives of this project are to heighten awareness of indigenous knowledge of plants and their uses, and archive the interplay of culture, plants, and animals in the US Virgin Islands. Studies over three years yielded a children's book, Musical Seeds and Medicine Leaves and a 27-min film documentary, So Many Plants - So Little Time. In the children's book, a teenager from New York City discovers the natural treasures that the island of St. John USVI has to offer through unexpected circumstance, curiosity, and exploration which lead to discover of the "old ways" that have been forgotten. He then comes to realize how much fun he can have "unplugged" while still learning about his heritage and the environment (Flesch Kincaid reading ease=87.2; Flesch Kincaid reading level=3.3 and up; with an accompanying DVD of plants encountered in the book). The documentary, So Many Plants - So Little Time, explores the relationship between peoples and local flora and fauna of St. John USVI, and how the loss of the knowledge base of plants and their uses are related to the changing island culture. Funded by the Kantner Family, University of Richmond, and the Virginia Academy of Science.

RENEWABLE ENERGY CURRICULUM MATERIALS USING MULTIPLE MEDIA FORMATS. David B. Hagan, Science Museum of Virginia, 2500 W. Broad Street, Richmond, VA 23220. This project arose from a challenge to present curriculum content on energy sources and consumption in Virginia, made possible by

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a grant from the Dominion Foundation. Science Museum of Virginia is a center of informal science education, generally defined as "science teaching and learning that occurs outside of the formal school curriculum in places such as museums, the media, and community-based programs." (NSTA Position Statement: Informal Science Education). The museum presents different content components in several forms of media focused on alternative and conventional energy sources. These include: a 2' x 3' original, detailed poster map of Virginia (*Energy Virginia*) showing sites of sources of energy and delivery mechanisms, including alternative and conventional sources. *Question Power* is an original six-minute video presenting the challenge of discovering clean, abundant energy sources. Eight one-minute videos display advantages and disadvantages of each of the major energy sources (wind, water, solar, geothermal, uranium, coal, oil, natural gas). The *Watt Wall* is a large computer-driven100 square-foot display showing global energy demand and consumption. In addition, there is a K-12 Teacher Guide for these curriculum materials.

SERVICE LEARNING AND SUBSEQUENT COMMUNITY ENGAGEMENT IN A FIELD MAPPING CLASS. Dr Julia A. Nord, Atmospheric, Oceanic & Earth Sciences, & Dr Thomas C. Wood, New Century College. George Mason University. Fairfax, VA. Student feedback on experiential learning (EL) activities in a Mason, upper division Field Mapping Techniques course, provided insight in line with literature confirming the value of EL. This course was revised using service learning, and community learning pedagogies to improve student knowledge and affect domains. We surpass our traditional field oriented activities, provide services to the partners, and engage students through active learning projects and reflection. Teams create maps working with various techniques and equipment from GPS, GIS, and transits to pace-and-compass. All projects include field reconnaissance, mission planning, equipment check, data collection and processing followed by the creation of the map. The final project entails meeting with the Director of Environmental Studies on the Piedmont to discuss spatial information needs. Students design and develop projects that enable them to collect data and present usable maps and suggestions back to the Director. We utilize Kolb based EL to engage current, real world issues with repetitive, reflective practice. Students work at the upper levels of Bloom's revised Taxonomy and use at least seven of Gardner's multiple intelligences. We hypothesize, students will increase interest, capacity and ability to apply their knowledge and consequently improve. The NSF developed Student Assessment of Learning Gains instrument will measure these outcomes.

#### **Environmental Science**

COMPARISON OF THE NI RIVER AND MASSAPONAX CREEK IN SPOTSYLVANIA, VA. D. Gutierrez, M. Recta, and M.L. Bass, Department of Earth and Environmental Sciences, UMW, Fredericksburg, VA 22401. The purpose of this research was to compare the water quality of the Ni River and Massaponax Creek, in Spotsylvania County. The Ni River is located in the more rural areas of the County while Massaponax Creek flows through more urbanized areas. Planned development of the Ni Village community will be occurring around the Ni River in the next few years and we expect the stream to be impacted like Massaponax Creek. Three different

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sites were sampled along the Ni River and for Massaponax Creek. In order to determine the current water quality, parameters were measured both in the field and in the laboratory. Macrobenthic samples were gathered from each sampling site, then separated the aquatic insects into the orders: Ephemeroptera, Plecoptera, Trichoptera, Diptera, Megaloptera, Odonata, Coleoptera, and Miscellaneous. Using the macrobenthic data we measured %EPT and the Family Biotic Index. we measured parameters in the field such as Dissolved Oxygen, Temperature, Conductivity, and pH. Water samples were collected to analyze Nitrate, Phosphate, Ammonia, Alkalinity, and Hardness in the laboratory using LaMotte test kits. Water samples were also used to determine the Total Suspended Solids(TSS). Whirlpak bags were used to collect water at each site to determine the fecal coliform levels. The Phosphate, Nitrate, and Total Suspended Solids (TSS), pH, and fecal coliform levels are important EPA parameters for TMDL's. Our results showed that %EPT was higher in Massaponax Creek however; there was more variety of Ephemeroptera, Plecoptera, and Trichoptera in the Ni River. The order Trichoptera dominated the %EPT in Massaponax Creek and yielded a high percentage. The magnitude of the physical and chemical parameters measured in the field was not dramatic in either stream. Nitrate and Phosphate levels were found higher in the Ni River which is possibly due to the rural farms located around the stream. The TSS were found in higher amounts in the Massaponax Creek which may be caused by the urbanization. Fecal Coliform was found in high amounts at site 1 of Massaponax Creek but all other sites had lower levels. In conclusion, the urbanization around Massaponax Creek may have had a greater impact in the past; however the stream is recovering from stabilization of the riparian area along the stream. Further research will determine if the planned development around the Ni River will show the same pattern of impairment then recovery as Massaponax Creek.

RAIN GARDEN REMEDIATION EFFECTIVENESS OF TWO NATIVE PLANT SPECIES. Emma Wallace and Dr. Barbara B. Kreutzer; Marymount University. This study examined the effectiveness of *Panicum virgatum* and *Lobelia cardinalis* in reducing phosphate and nitrate levels in rainwater runoff. Both plant species are commonly implemented in rain gardens, landscaping tools that remediate rainwater runoff. Testing the phosphate and nitrate concentration in each pot-grown plant's leachate determined both species are similarly effective in reducing phosphate and nitrate levels.

SUMMARY OF WATER QUALITY ANALYSIS DATA FROM THE STORM WATER MANAGEMENT PONDS OF THE CENTRAL PARK DEVELOPMENT, FREDERICKSBURG, VA. M.L. Bass, M.Recta, and D. Gutierrez, Department of Earth and Environmental Sciences, UMW, Fredericksburg, VA 22401. The purpose of this research was to assess the impact of commercial and residential development on nearby streams. The Chesapeake Bay area is undergoing policy change in which there will be stricter regulations on the amount of material that can be emptied into the Bay. Limits on the Total Maximum Daily Load (TMDL) will cause the Chesapeake Bay community to be more conscious of land management practices. The area of study during the summer of 2011 included 6 sampling stations on England Run and an unnamed tributary that have been affected by the construction of Celebrate Virginia North located in Stafford County, Virginia. Methodology for the water quality study

includes on-site field water quality analysis using YSI models 85 and YSI ProPlus meters that measured temperature, dissolved oxygen, conductivity, ammonia, nitrate, and pH. Water samples were analyzed for levels of nitrate, phosphate, hardness, ammonia, and alkalinity using LaMotte test kits. Total suspended solids(TSS) were also studied to determine the impact of construction causing an increase in sedimentation and turbidity. Fecal coliform presence was determined. Water quality analysis showed that the levels of nitrates, phosphates, TSS, and fecal coliform were below EPA standards. In addition, the macrobenthic community was also studied. The insects sampled within the stream were identified and analyzed using various biological community evaluation metrics such as percent Ephemeroptera Plecoptera Trichoptera (%EPT), Family Biotic Index (FBI), and Hilsenhoff Biotic Index (HBI) that indicate the health of the stream based on the ratios of pollution sensitive to pollution tolerant insects within a given sampling station. The data collected during the summer of 2011 was compared to baseline data from the pre-development and early development stages of Celebrate Virginia North. During the early development stages of the Celebrate Virginia North project, many of the ecological habitats were negatively impacted by the development. Recent research using the biological assessment of the streams shows that the macrobenthic community is showing improvement as a result of stabilization with vegetation of previously scoured and erosion-prone areas. Further research continues in order to determine the impact of development on these streams.

OYSTER RESTORATION AND WATER QUALITY ANALYSIS IN AN URBAN SUB-ESTUARY. Melinda Hopper<sup>1</sup>, Matthew Boyce, and Maury Howard. Chemistry Depart., Virginia Wesleyan College. This study was conducted to monitor a local body of water and to determine the effect of oysters on water quality. The purpose is to determine whether oyster restoration can help the quality of the water and whether this site is an ideal location for restoration. Water samples were collected over a year long period at 3 different sites. Each sample was tested for various water quality indicators, including chlorophyll a, total phosphorus, temperature, pH, BOD and salinity. Sediment samples and oyster tissue samples were also collected and run through an ICP-MS to identify the toxic metals found within them. High concentrations of lead, mercury, arsenic, and other heavy metals were some of the main components. The water contained very low levels of metals due to the tidal influence. However, the sediments and oyster tissue contained high concentrations of many of them, showing the effects of bioaccumulation. It also shows the value of the oysters as filters for the system, removing toxins and nutrients from the ecosystem.

ORGANOPHOSPHATE PESTICIDE CONCENTRATIONS IN TOMATOES FROM THE HARRISONBURG AREA. <u>K. Åkerson<sup>1</sup></u> and Doug G. Neufeld, Dept. of Biology, Eastern Mennonite University. We wanted to combine an extraction method and a detection assay, and to discover the pesticide concentration difference between large farm and small farm organic tomatoes. First, the QuEChERS extraction method and enzyme-based detection assay was combined to successfully detect organophosphates/carbamate pesticides (OP/C) at concentration levels of at least one part per billion (ppb) or greater. From this, we concluded that QuEChERS and the

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enzyme-based assay work in combination, and even reproduce precise results at an average 19.4% relative percent difference (RPD). Second, because OP/C causes harmful health effects at high doses, we investigated pesticide concentration of farmers' market tomatoes compared to supermarket tomatoes. Our results suggested OP/C in three samples: 2 out of 10 farmers' market tomato samples and 1 out of 5 supermarket tomato samples. The detection of OP/C surprised us, though, because locations claimed organic production. One explanation of this could be that sources other than the farmers contaminated the samples.

## **Medical Science**

α7 nAChR NEGATIVE ALLOSTERISM: A PROMISING APPROACH FOR COGNITIVE DISORDERS. O. I. Alwassil<sup>1</sup>, G. Abdrakhmanova<sup>2</sup>, and M. Dukat<sup>1</sup>, <sup>1</sup>Department of Medicinal Chemistry and <sup>2</sup>Department of Pharmacology & Toxicology, Virginia Commonwealth University, Richmond VA 23298. Progression of Alzheimer's disease (AD) is associated with an increase of  $\beta$ -amyloid peptide 1-42  $(A\beta_{1,42})$  neurotoxic interactions with  $\alpha$ 7 neuronal nicotinic acetylcholine receptors (nAChRs) leading to tau protein hyperphosphorylation and neuron deficit. Through their neuroprotective abilities against toxic  $\alpha$ 7 nAChR-A $\beta_{1-42}$  interactions,  $\alpha$ 7 nAChR ligands might represent promising targets for AD symptomatic therapy. We have identified *meta*-chlorophenylguanidine (mCPG;  $IC_{50} = 8.0 \mu M$ ) as the first small-molecule negative allosteric modulator (NAM) at a7 nAChRs. mCPG might serve as a lead in developing structure–activity relationships for NAMs at  $\alpha$ 7 nAChRs. Several analogs of mCPG were synthesized and evaluated in whole-cell patch-clamp assay. Introduction of a methyl group on the aniline nitrogen atom of mCPG resulted in a more potent  $\alpha$ 7 NAM (IC<sub>50</sub> = 1.3  $\mu$ M) than *m*CPG. Since the exact interaction site(s) and mechanism by which the  $\alpha$ 7 nAChR NAMs work has not been yet fully described, we developed 3D models of the extracellular domain (ECD) of human MBOL97/f"Symbol"/s107 nAChRs. Modeling studies resulted in the identification of two out of five binding sites in the ECD that are supported by empirical data. The different docking solutions are consistent with functional data. Supported in part by the Virginia Center on Aging (Award No. 12-2).

THE INTERACTION BETWEEN WIN55,212-2 AND RADIATION ON INHIBITING THE GROWTH OF BREAST CANCER CELLS. <u>S. M. Emery</u><sup>1</sup>, E. T. Sumner, Q. Tao, A. H. Lichtman & D. A. Gewirtz<sup>1</sup>, <sup>1</sup>Department of Pharmacology and Toxicology, Virginia Commonwealth University, Richmond VA 23298. Win55,212-2 (WIN2) is classified as a full agonist for cannabinoid receptors CB1 and CB2, as well as an agonist for known off targets of the endocannabinoid system including TRPV1 and PPAR $\alpha$ - $\gamma$ . Recent reports have shown that WIN2 has anti-proliferative effects on cancer, but no studies have been performed to evaluate potential interactions between WIN2 and ionizing radiation (IR) used in cancer treatment. We have shown that WIN2 has the capacity to significantly enhance the anti-proliferative effects of IR in the MCF-7 breast tumor cell line. This effect has been reproduced in MDA-MB-231 and 4T1

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breast tumor cells and in the syngeneic tumor model of 4T1 cells in Balb/c mice; of importance for selectivity, this combination effect does not transfer to the MCF-10a model of normal breast epithelial cells. Interestingly, in MCF-7 cells this enhancement of sensitivity is not observed for other drugs of the cannabinoid class, including CP 55,940, Methanandamide, Cannabidiol, Nabilone and  $\Delta^9$ -Tetrahyrdocannabinol. RT-PCR demonstrates that in MCF-7 cells neither CB1 nor CB2 are expressed and selective antagonists for each receptor failed to antagonize WIN2's actions. Although both TRPV1 and PPAR $\gamma$  are expressed in MCF-7 cells, pharmacological studies using Capsazepine(TRPV1 antagonist), Capsaicin(TRPV1 agonist), GW9662(PPAR $\gamma$ antagonist), and Pioglitazone(PPAR $\gamma$  agonist) demonstrate that neither TRPV1 nor PPAR $\gamma$  are likely mediating the radiosensitizing effects of WIN2. Supported by a predoctoral training grant from the DOD to Sean Emery.

INVOLVEMENT OF CCR5 IN MORPHINE AND TAT-MEDIATED NEURODEGENERATION. E.M. Podhaizer<sup>1</sup>, Y. Zhang<sup>2</sup>, P.E. Knapp<sup>1,3</sup>, K.F. Hauser<sup>1</sup>. Depts. of <sup>1</sup>Pharmacol. & Toxicol., <sup>2</sup>Med. Chem., <sup>3</sup>Anat. & Neurobiol., Virginia Commonwealth Univ., Richmond, VA. Studies have shown that dual morphine & Tat treatment produce synergistic neurotoxicity in neuron-glial co-cultures. We chose to examine CCR5's role in this effect due to its interactions with the µ-opioid receptor, and its ligand's involvement in glial activation. We hypothesized that blockade/deletion of CCR5 would prevent morphine and Tat interactive neurodegeneration. A CCR5 antagonist, maraviroc (MVC), was used first to assess CCR5's role in morphine & Tat toxicity. While MVC had no effect on morphine or Tat toxicity alone, it blocked the enhanced effect of the combined treatment. Use of CCR5 knockout (KO) glia with wild-type (WT) neurons showed a delay in the onset of neurotoxicity in morphine, Tat, or dual-treated cells and the overall toxicity of the combined treatment was reduced. When WT glia and CCR5 KO neurons were used, morphine & Tat toxicity was significantly reduced, while morphine toxicity was enhanced. However, deletion of CCR5 in both neurons and glia showed significant toxicity under basal conditions and suggests that CCR5, per se, is neuroprotective. To determine if the glial component of the CCR5 response could be due to beta-chemokine release, CCR5's endogenous ligands, MIP-1 $\alpha$ , MIP-1 $\beta$ , and RANTES were applied to co-cultured cells. While the highest concentrations did show significant neurotoxicity, physiologically relevant levels did not produce an effect, suggesting that elevated levels of CCR5 agonists were not responsible for enhanced morphine & Tat toxicity and suggest an alternative mechanism. Funding sources: R01 DA019398, T32 DA007027, F31 DA033203.

AGOUTI-RELATED PROTEIN (AGRP) RNA EXPRESSION IN THE VISCERAL ADIPOSE TISSUE OF NON-ALCOHOLIC FATTY LIVER DISEASE (NAFLD) PATIENTS. David Van Natta<sup>1,3</sup>, <u>Christopher Gerner</u><sup>1,3</sup>, Ancha Baranova<sup>1,2,3</sup>, J. Michael Estep<sup>1,3</sup>, 1, Biology Department, College of Science, George Mason University 2. School of Systems Biology, College of Science, George Mason University3. Beatty Liver and Obesity Research Project, Institute of Research, Inova Health System. AgRP is an appetite regulating peptide that may play roles in inflammation and calcium homeostasis. Recently it was demonstrated that hypothalamic AgRP expression is regulated by FFA. This pilot study measures AgRP expression in the VAT of morbidly

obese patients in the context of metabolic disease. Of the 21 patients analyzed to date, 15 (71%) were female, 6 (60%), and 11 (52%) had non-alcoholic steatohepatitis (NASH) while the other 10 had non-NASH NAFLD (47%). Importantly, Patients with a diagnosis of NASH expressed AgRP significantly less than those with a diagnosis of non-NASH NAFLD (FD=-3.95, P=0.01).

PHARMACOLOGY OF ALPHA5\* NICOTINIC ACETYLCHOLINE RECEPTORS IN MEDIATING ETHANOL- RESPONSIVE BEHAVIORS. A. J. Dawson<sup>1</sup>, M.F. Miles<sup>1</sup>, M.I. Damaj<sup>1</sup>. Dept. of Pharmacology & Toxicology, Virginia Commonwealth University, Richmond VA 23298. Nicotine and Alcohol (ethanol) are two of the most widely abused drugs in society. Because of the high co-morbidity of addiction to these drugs, it is thought that nicotinic acetylcholine receptors (nAChRs), the principle mediators of nicotine addiction, may also contribute to alcohol addiction. Recent human genetic evidence suggests that the CHRNA5/A3/B4 gene coding region, which codes for  $\alpha 5^*$  nAChRs (\* denotes the presence of additional subunits), is associated with both nicotine and alcohol dependence. While behavioral evidence exists that support this finding in nicotine-responsive behaviors in animals, there are virtually no published studies that have investigated the effects of a5\* nAChRs on ethanolresponsive behaviors in vivo. We sought to determine the effect of  $\alpha$ 5 nAChR gene removal on the effects of acute ethanol exposure and chronic drinking behavior using high-ethanol preferring C57BL/6J null mutant mice. These a5 Knockout (KO) mice were tested for changes in acute ethanol-responsive behaviors including loss of righting reflex, hypothermia, and anxiolysis, as well as for modulation of drinking behavior. Results revealed that while a5 KO mice showed a general enhancement in their response to ethanol's acute effects, there was no difference in their drinking behavior. Furthermore, because  $\alpha 5^*$  nAChRs must necessarily co-assemble with  $\alpha 4\beta 2^*$  or  $\alpha 3\beta 4^*$ nAChRs, we tested for the involvement these nAChR subtypes in the previously mentioned measures using pharmacologically and genetically manipulated C57BL/6J mice. Preliminary results showed that antagonism of these receptors differentially modulated some acute ethanol responses. Taken together, these data suggest a real, albeit, complex role for  $\alpha 5^*$  nAChR subtypes in mediating initial sensitivity to some of the effects of acute ethanol exposure.

THE DINUCLEAR PLATINUM AGENT, BBR3610-DACH, INDUCES G1/S AND G2/M CELL CYCLE ARREST THROUGH DIFFERENT ROUTES IN HUMAN COLORECTAL CANCER HCT116 CELLS. <u>Vijay Menon<sup>1</sup></u>, E. Peterson<sup>2</sup>, L.F.Povirk<sup>1</sup>, N. Farrell<sup>1,2</sup>, <sup>1</sup>Dept. of Pharmacology and Toxicology, and <sup>2</sup>Dept. of Chemistry, Virginia Commonwealth University, Richmond, VA. BBR3610-DACH is a long-chain, bifunctional dinuclear Pt (II) complex shown previously to be resistant to metabolic decomposition by sulfur-containing nucleophiles. Initial observations utilizing the comet assay indicated that both BBR3610 and BBR3610-DACH formed interstrand crosslinks. However, from our subsequent studies, it is seen that both drugs induced different cellular effects with BBR3610-DACH causing a significant G1/S and G2/M cell cycle arrest in HCT116 cells in a time dependent manner with depletion in the S phase cells. The G1/S arrest was accompanied by a stabilization of p53 between 24 to 48 h and a concomitant increase in the cyclin dependent kinase inhibitors, p21 and p27.

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An early decrease in the cyclin A level suggested a G2/M arrest. Studies with synchronized cells showed that BBR3610-DACH specifically inhibited the progression of G1 arrested cells into the S phase. Also, utilizing isogenic p53 and p21 null cell lines, it was seen that both p53 and p21 determine the sensitivity of BBR3610-DACH induced growth arrest. BBR3610-DACH also induced an early apoptosis along with an increase in the levels of cleaved PARP-1. Together these findings suggest that BBR3610-DACH is a major cell cycle inhibitory DNA binding platinum agent that could be developed further as a major chemotherapeutic.

IDENTIFYING GENES THAT INFLUENCE ACUTE ETHANOL RESPONSE BEHAVIORS IN Caenorhabditis elegans. J.T. Alaimo, A.G. Davies, J.C. Bettinger, Department of Pharmacology and Toxicology, Virginia Commonwealth University, Richmond, VA 23298. Alcohol dependence is a complex disorder that is strongly influenced by genetic factors. An individual's initial response to acute ethanol is a strong indicator of future abuse, but little is known about how acute ethanol exposure leads to changes in neuronal activity and behavioral responses. Previous studies have shown that acute ethanol treatment induces changes in expression of many genes that are important in ethanol responsive behaviors. Using the model organism *Caenorhabditis elegans*, we plan to survey gene expression changes induced by ethanol and construct gene networks to identify candidate functional loci. We predict that transcriptional changes that occur during and subsequent to the acute ethanol response are critical for changes in behavior. However, we hypothesized that acute responsive genes may be needed to observe the behavior. We asked if transcription of acute response causing genes is required for the behavior acute functional tolerance (AFT). We tested this by inhibiting transcription during acute ethanol exposure using actinomycin D and observing the development of AFT in two strains, wild type N2 and a mutant strain that rapidly develops AFT due to the lack of a functional neuropeptide Y receptor (*npr-1*). Both N2 and *npr-1* mutant animals retained the ability to develop AFT when transcription was inhibited, suggesting that transcription of the acute response genes is not required for AFT. We hope that uncovering ethanol responsive genes and their networks by transcriptional profiling will uncover important genes involved in ethanol responses.

MITOCHONDRIAL DNA AS A QUANTITATIVE MARKER OF BROWN ADIPOSE ACTIVITY. <u>K. Doyle<sup>2</sup></u>, A.Neupane<sup>1, 2</sup>, R. Mehta<sup>1, 2</sup>, Z. Younossi<sup>1, 2, 3</sup>, A. Birerdinc<sup>1, 2</sup>, & A. Baranova<sup>1, 2</sup>, <sup>1</sup>Betty and Guy Beatty Center for Integrated Research, Inova Health System, <sup>2</sup>School of Systems Biology, GMU, and <sup>3</sup>Center for Liver Diseases, Inova Fairfax Hospital. Obesity is one of the biggest health problems in the United States and is linked to a number of degenerative diseases. Recent data suggests that brown adipose activity may play a role in both adult body weight homeostasis and the regulatory mechanisms of obesity. Unfortunately, brown adipose tissue related genes can remain dormant under certain conditions making it difficult to use gene expression studies to determine if it is present. However, a large number of mitochondria are found in this cell type and so we are using both mitochondria specific DNA and genomic DNA primers to quantify the amount of each type of DNA using qPCR. This data will be used to obtain a ratio that indicates the total amount of brown adipose tissue in the samples of subjects with and without NASH.

GENE EXPRESSION PATTERN ANALYSIS IN GASTRIC TISSUE OF PATIENTS WITH NON-ALCOHOLIC FATTY LIVER DISEASE. E. Stolworthy<sup>1</sup>, R. Mehta<sup>1,2,3</sup>, L. Wang<sup>1,2,3</sup>, A. Baranova<sup>1,2,3</sup>, Z. Younossi<sup>1,2,3</sup>, & A. Birerdinc<sup>2,3</sup>, <sup>1</sup>Biology Department of Science, George Mason University, Fairfax Virginia 22030. <sup>2</sup>School of Systems biology, College of Science, George Mason University, Fairfax, VA 22030. <sup>3</sup>Betty and Guy Beatty Center for Integrated Research, Inova Health Systems. Over the past few decades, obesity is accompanied with increasing incidence of Non-alcoholic fatty liver disease (NAFLD). As a result, there is great interest in identifying which genes differentially expressed in NAFLD. Gene expression in gastric tissue was evaluated by qPCR. Normalized data was subjected to group comparision, correlation analysis and regression analysis. Group comparison between NASH and no NASH cohort showed CCL1 (1.23, p=0.05), CCR3 (3.45, p=0.04), CCR9 (2.97, p=0.04), CXCL12 (0.65, p=0.05), IL1RN (3.96, p=0.01), IL5 (3.14, p=0.05), IL8RA (15.05, p=0.04), IL8RB (3.14, p=0.05), and IL9 (9.49, p=0.01) to be significantly differentially expressed. Similarly CXCL14 (1.77, p=0.03), IL1F10 (2.25, p=0.04), and IL8RB (1.57, p=0.02) were differentially expressed between Steatosis and no Steatosis cohort. Group comparison between Mild Inflammation and Advanced Inflammation found that CCL4 (0.43, p=0.03), CXCL2 (0.35, p=0.02), CXCL6 (0.23, p=0.03), IFNA2 (0.26, p=0.02), ILF19 (0.25, p=0.02), IL1F8 (.025, p=0.02), and IL8 (0.21, p=0.02) to be differentially expressed.. These inflammatory genes may be helpful in understanding the gastric liver axis in the pathogenesis of Non-Alcoholic Fatty Liver Disease.

A NOVEL DIAGNOSTIC APPROACH FOR CHRONIC SYSTEMATIC DISEASE: DISTANCE MEASURE OF GENE EXPRESSION PROFILE. Lei Wang<sup>1,2</sup>, Ganiraju Manyam<sup>3</sup>, Boris Veytsman<sup>1</sup> & Ancha Baranova<sup>1,2</sup>, <sup>1</sup>School of System Biol., George Mason Univ., Fairfax, VA. <sup>2</sup>Ctr. of Liver Diseases, Inova Fairfax Hosp., Falls Church, VA. <sup>3</sup>Betty and Guy Beatty Ctr. for Integrated Research, Inova Health System, Falls Church, VA. <sup>3</sup>Dept. of Bioinformatics & Comp. Biol., The UT MD Anderson Cancer Ctr., Houston, TX. Protein or RNA biomarkers are widely used for diagnosis. However, identification of novel biomarkers usually suffers from either low specificity or unsatisfactory reproducibility. Here we propose a novel approach that measures global distance between entire gene express profiles. We use psoriasis, as model disease to demonstrate the feasibility of our new concept. We analyzed microarray data sets and used Mahalanobis distance as an estimation of the degree of differentiation. The effectiveness of global signatures was compared with that of traditional specific signatures. In conclusion, the global distances of gene expression profiles can serve as reliable good classifiers as specific signatures, and therefore it is worth considering using them as diagnostic markers.

ETHANOL-INDUCED DEFECTS IN OLFACTORY NEURON CELL FATE DECISIONS IN CAENORHABDITIS ELEGANS. <u>L.M. Kondo</u>, R.C. Raabe, K.S. Kauv, M.H. Bolling, A.G. Davies, & J.C. Bettinger, Department of Pharmacology and Toxicology, Virginia Commonwealth University, Richmond VA 23298. Fetal alcohol syndrome (FAS) is the leading preventable cause of mental retardation, but the underlying molecular mechanisms are not well understood. We have taken a genetic approach to studying the effect of ethanol on a discreet cell fate decision occurring during embryogenesis in the nematode, *C. elegans*. AWC cells are a pair of olfactory

neurons that allow *C.elegans* to discriminate between volatile attractive odorants in chemotaxis and odorant discrimination assays. Early in development, AWC neurons make an activity dependent cell fate decision, and subsequently, particular groups of G protein-coupled receptors are asymmetrically expressed. A GFP tagged receptor STR-2 (STR-2::GFP) allows us to monitor cell fate decisions between AWC neurons. SLO-1, a voltage-gated potassium channel is expressed in these neurons, and activation of this channel can lead to both AWCs adopting the same cell fate. Previous studies from our lab have shown that SLO-1 is a major molecular target of ethanol and mediates ethanol sensitivity. We tested ethanol exposure during embryogenesis and hypothesize that the interaction between ethanol and the SLO-1 channel alters AWC cell fate. Furthermore, by altering the lipid composition of the cell membrane, we can render this cell fate decision resistant to the effects of ethanol.

SULFATIDE REGULATION OF PARANODAL FORMATION IN THE PERIPHERAL NERVOUS SYSTEM. H.F. Herman<sup>1,2</sup>, E.L. Kwong<sup>1</sup>, and J.L. Dupree<sup>1</sup>, <sup>2</sup>, <sup>1</sup>Dept. of Anatomy and Neurobiology and <sup>2</sup>Molecular Biology and Genetics, Virginia Commonwealth University, Richmond, VA 23298. Sulfatide is a galactolipid and a major lipid component of the myelin sheath. Its production is catalyzed by the enzyme cerebroside sulfotransferase (CST). To determine the functions of sulfatide, the gene encoding CST was genetically disrupted resulting in mice incapable of sulfatide synthesis. We have employed a combination of immunocytochemistry and confocal microscopic analysis of the CST null mice to determine the role of sulfatide in protein cluster onset and maintenance in the PNS by quantifying the number of protein clusters in WT and CST KO mice of ages ranging from 4 days to 10 months. Neuronal voltagegated sodium channels were decreased in the CST KO at 4 days of age, while clusters of the glial protein neurofascin 155 were decreased in the CST KO at 7 days of age, though both of these deficits were righted by 15 days of age. Qualitative ultrastructural studies were also undertaken to analyze the stability of the nodal and paranodal regions at these ages. Together, our results indicate that sulfatide plays a role in the onset of protein clustering in the node and paranode as well as in the formation and maintenance of the node and particularly the paranode in young mice, as transverse bands are rarely observed in the CST KO mice.

"BUBBLES" AND "BATH SALTS" AND DOPAMINE TRANSPORTERS. <u>R.</u> <u>Kolanos<sup>1</sup></u>, K. N. Cameron<sup>2</sup>, L. J. De Felice<sup>2</sup>& R. A. Glennon<sup>1</sup>. <sup>1</sup>Department of Medicinal Chemistry and <sup>2</sup>Department of Physiology and Biophysics, Virginia Commonwealth University, Richmond VA 23298. In the last few years there has been a worldwide increase in the abuse of psychoactive *bath salts*. Recent analyses indicate that the active ingredients in many brands of *bath salt* products contain mephedrone (a. k. a. "bubbles") and methylenedioxypyrovalerone (MDPV), sometimes in combination with other synthetic cathinones such as methylone, methedrone and flephedrone. Surprisingly, very little is known about the mechanism of action of these synthetic cathinone derivatives despite an increasing number of *bath salts*-related hospitalizations and deaths. The limited studies on mephedrone and MDPV, along with the structural resemblance to methcathinone, suggested that their behavioral effects might have a dopaminergic mechanism similar to dopamine-releasing agents. In an effort to better understand the effect of synthetic cathinones on the human dopamine transporter (hDAT) we synthesized some of the most common ingredients of *bath salts* and recorded currents through hDAT-expressing *Xenopusleavis* oocytes using a twoelectrode voltage clamp technique. Our studies showed that mephedrone is behaving like a dopamine releaser similar to methcathinone and methamphetamine. MDPV, though containing a methcathinone core, is acting like a potent hDAT blocker similar to cocaine. In fact, the combination of mephedrone and MDPV might simultaneously increase dopamine release and inhibit dopamine reuptake, leading to an unusually high synaptic concentration of the neurotransmitter DA and, hence, the high abuse potential of *bath salt* products. Supported in part by NIH DA033930.

SULFATIDE REGUALTES MEMBRANE ASSOCIATION OF NFASC155 AND MAG DURING MYELINATION. A.R. Hackett, A.D. Pomicter, N. Purdy, J.M. DeLoyht & J.L. Dupree. Dept. of Anatomy and Neurobiology, Virginia Commonwealth University, Richmond VA 23298. Mice incapable of synthesizing the myelin lipid sulfatide form normal paranodes that deteriorate with age. Mice that lack either the neuronal paranodal proteins contactin or caspr or the myelin paranodal protein neurofascin155 (Nfasc155), the 3 proteins that comprise the junctional complex that tethers the myelin sheath to the axon, also exhibit unstable paranodes. However, since sulfatide does not cluster in the paranode, has no paranodal binding partner and since sulfatide null mice express the 3 paranodal junctional proteins, it is unclear how the lack of sulfatide results in paranode instability. Here, we test the hypothesis that sulfatide maintains paranode stability by regulating the distribution of Nfasc155 through the formation of lipid rafts. Consistent with our hypothesis, clusters of Nfasc155 are unstable in the absence of sulfatide. We also show that the membrane association of myelin-associated glycoprotein, another myelin protein that maintains myelin-axon adhesion, also requires sulfatide while the membrane associations of structural myelin proteins are sulfatide independent. These findings are significant as the levels and fatty acid composition of sulfatide are altered in both neuronal and myelin degenerative diseases indicating a potential loss of myelin-axonal adhesion in disorders not typically associated with demyelination.

#### Posters

OPIOID AND HIV-1 ASSOCIATED NEURODEGENERATION: IMPLICATIONS FOR P2X<sub>4</sub>RECEPTOR INVOLVEMENT. <u>M.E. Sorrell<sup>1</sup></u>, S. Zou<sup>2</sup>, P.E. Knapp<sup>1,2</sup>, and K.F. Hauser<sup>1</sup>. <sup>1</sup>Dept. Pharm. & Tox., <sup>2</sup>Dept. Anat. & Neurobiology., VCU, Richmond, VA. HIV-1-associated neurocognitive disorders (HAND) is seen in 50% of AIDS patients. Individuals who abuse opiates can have an increased incidence of HAND with more symptoms. To test HIV-1 and opioid-induced neurotoxicity are mediated by purinergic signaling, co-cultures of primary neurons and mixed glia were treated with combinations of Tat, morphine, and TNP-ATP, a P2X antagonist. Tat and morphine neurotoxicity was reversible by treatment with TNP-ATP. Tat + morphine decreased dendritic length, this was prevented by TNP-ATP treatment. Tat and morphine increased intracellular Ca<sup>2+</sup> levels, this was preventable with TNP-ATP present. Antagonists against the P2X<sub>1</sub>, P2X<sub>3</sub>, and P2X<sub>7</sub> receptor subtypes were screened. Findings showed that these subtypes were not involved in Tat + morphine neurotoxicity supporting that P2X<sub>4</sub> receptors are involved. Human P2X microarray data from HIV infected and non-infected individuals [courtesy NNTC] suggest that  $P2X_4$  is altering neuroAIDS and the  $P2X_4$  receptor may be a novel therapeutic target for HAND. *Support: NIH DA018633, DA028741, DA007027, and the NNTC.* 

ASSOCIATIONS BETWEEN SLEEP DIFFICULTIES AND WELL BEING IN BREAST CANCER SURVIVORS. Anthony Loria<sup>1,3</sup>, Patrice Winter<sup>1,3</sup>, Katherine Doyle<sup>1,3</sup>, Yang Wang<sup>2,3</sup>, Lynn Gerber<sup>1,2</sup>, Ancha Baranova<sup>1,3</sup>& Zobair Younossi<sup>1</sup>, 1. Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church, VA, United States 2. Center for the Study of Chronic Illness and Disability, College of Health and Human Services, George Mason University, Fairfax, VA, United States 3. George Mason University, Fairfax, VA, United States. Sleep difficulties are often documented in cancer patients following treatment. The aim of this study was explore the relationships between sleep difficulties and well being. Self report of well being (FACIT-F) and blood samples were collected from women with breast cancer who were more than 3 months past primary treatment. Measures of sleep difficulty were analyzed with respect to pro-inflammatory cytokines, C reactive protein and subscales of the FACIT-F and ascribed for significance (Mann-Whitney U tests). Forty-seven women  $[57 \pm years]$  of whom 67% reported sleep difficulties were binarily examined by 25th and 75th percentile with respect to subscales. Social, Emotional and Functional well being were all significantly different between those with and without sleep difficulties ( $p \le .032, Z \ge -2.143$ ). In the fatigue subscale, TGF-B1 (-1.899, .058) and C reactive protein (-1.833, .067) were approaching significance. Sleeping problems may be influenced by multiple domains of function and biological homeostasis in breast cancer survivors.

SUMMER ANATOMY INTERNSHIP FOR OMS-I: AN OPPORTUNITY FOR AUGMENTED TRAINING. R.P. Wyeth, K. D'Amato, I. Danelisen, J. Anstrom. VCOM Anatomy Department and the Plastination Laboratory, Via College of Osteopathic Medicine, Virginia Campus, Blacksburg VA 24060. During the 2011 summer recess an internship was offered to selected first year students based on both academic achievements and anatomical interests. The internship had two purposes: 1) provide students an opportunity to increase their anatomical knowledge in an area of special interest, and 2) develop expertly dissected specimens that could be subsequently plastinated and used as anatomy teaching aids in future years. Sixteen students, working individually or in small groups, chose a dissection project and worked under supervision of the authors. An anonymous survey was administered at the conclusion of the internship with 15 of 16 participants responding. Six five-level Likert items were included in the questionnaire, which was distributed online. Compilation of the results indicates that: self assessment of the student's anatomical knowledge increased by 87% when compared to perceived knowledge prior to the internship (p < 0.05); available facilities were adequate (73%); responsiveness of the faculty was viewed favorably (94%); available time was adequate (67%); and a majority of participants would recommend such an internship program to future students (86%). Thus, the summer anatomy internship is a valuable new adjunct to anatomy teaching that demonstrates a beneficial impact on participating students' knowledge while providing the medical community with well-prepared prosected specimens for plastination. Supported by Seed Money from the VCOM Foundation as a part of the Plastination Project.

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ANOMALY OF THE CIRCUMFLEX CORONARY ARTERY IN A CONTINUOUS SERIES OF 338 VIRGINIA CADAVERS. R.P. Wyeth, A. Santo, K. Lester, S. Wachob, A. Petty, J. Bookbinder. Via College of Osteopathic Medicine, Virginia Campus, Blacksburg VA 24060. Canonically, the left main coronary artery originates in the posterolateral left sinus of Valsalva from a single ostium while the RCA begins in the right sinus of Valsalva arising from a single ostium. The left main coronary artery then bifurcates into the left anterior descending (LAD) and circumflex arteries (LCX). The LAD courses along the anterior interventricular sulcus giving rise to a variable number of branches. The LCX typically departs the left main coronary artery turning sharply laterally and posterior terminating in obtuse marginal branch(es). Most commonly the LCX terminates immediately distal to its obtuse marginal branch(es). Infrequently the LCX continues towards the crux cordis reaching the posterior interventricular sulcus producing the posterior interventricular branch (PDA). More commonly the RCA gives rise to the PDA. These hearts are considered "right dominate". If the LCX terminates as the PDA the heart is termed "left dominate." In this current study, the LCX transverses the poster crux cordis without giving rise to the PDA. That is, arising from the opposite aortic ostium, the right ostium, following a retroaortic course. We report here the occurrence of a LCX arising from its own ostium within the right sinus of Valsalva and coursing retroaortic within the left atrioventricular sulcus to supply the diaphragmatic aspect of the heart terminating in the marginal branches. This is the first LCX variation in a continuous series of 338 human hearts (an incidence of 0.3%) studied in conjunction with the Human Anatomy Course at the Via College of Osteopathic Medicine.

GENDER AND TIME DIFFERENCES OF NON-INVASIVE BLOOD PRESSURES IN THE SIEGEL LARGE AND SMALL LINE OF CHICKEN—INFERENCE FOR A GALLIFORM MODEL OF THE METABOLIC SYNDROME. N. Sheth<sup>1</sup>, R. Clark<sup>1</sup>, L. Solis Lopez<sup>1</sup>, P.B. Siegel<sup>2</sup>, R.P. Wyeth<sup>1</sup>, 1Via College of Osteopathic Medicine, Virginia Campus. <sup>2</sup>Dept. of Animal and Poultry Sciences, Virginia Polytechnic Institute and State University, Blacksburg VA 24060. Cardiovascular disease is the leading cause of adult death. Systemic blood pressure (NIBP) is a function of cardiac power, circulating volume and vascular tonus. The current study compares NIBP to determine if weight differences and gender differences affect hypertension. A total of 16, age matched, chickens were used: 4 high weight females (HWF), 4 high weight males (HWM), 4 low weight females (LWF) and 4 low weight males (LWM). Pressures were obtained from the brachial artery. The total means of systolic, diastolic, and calculated mean of each day for each line were then averaged for class of bird and gender for type NIBP perimeter. The body mass of chickens in this study demonstrated significant differences in weight HWM and LWM and between LWF (p=0.001). This study showed gender differences between NIBP, the male blood pressures were significantly higher in diastolic (p = 0.011), systolic (p = 0.002) and calculated mean (p = 0.004). As time increased, NIBP increased in only LWM while NIBP decreased in HWM, LWF, and HWM. The inconsistencies of NIBP on Day 491 are explained by change to a female handler. The current study demonstrates that significant gender related differences in NIBP. The study also illustrates that in these lines, blood pressure decreased with time. Further research is needed to ascertain if (handler sex vs. chicken sex) gender differences plays a role in NIBP monitoring.

GENDER AND BODY MASS DIFFERENCES IN SEROTONERGIC FEMORAL ARTERIAL RESPONSES IN A GALLIFORM MODEL OF THE METABOLIC SYNDROME. N. Sheth<sup>1</sup>, R. Clark<sup>1</sup>, L. Solis Lopez<sup>1</sup>, P.B. Siegel<sup>2</sup>, R.P. Wyeth<sup>1</sup>. <sup>1</sup>Via College of Osteopathic Medicine, Virginia Campus, Blacksburg. <sup>2</sup>Dept. of Animal and Poultry Sciences, Virginia Polytechnic Institute and State University, Blacksburg VA 24060. The metabolic syndrome (MES) predisposes to atherosclerosis inducing platelet activation and degranulation releasing several factors including 5-HT. 5-HT induces vessel vasoconstriction and platelet aggregation capable of causing ischemia and infarction. The current study hypothesizes that there is no difference in 5-HT vasomotor activity with respect to body mass and gender in a galliform model of MES. This was tested in ischiatic arteries. Samples were constricted with 5-HT. Analysis of the aorta showed neointimal hyperplasia and atheroma consistent with disease process in humans. 5-HT stimulation produced dose response curves similar to those seen in human arteries. Efficacy, determined by maximum tension developed, was different in high weight females (HWF). While potency, as 50% of the concentration required to reach maximum contraction approached significance in HWF vs. low weight (LWF), and was significant in LWF vs HWF (p<0.003). Thus, a significant difference was noted in 5-HT potency between weight lines. The current study illustrates that, when comparing these high weight and low weight chickens: 1) 5-HT significantly modulates maximal contraction in HWF. 2) There is significant difference in potency  $(EC_{50})$ between HWF and LWF, and HW male (M) and LWM. These initial findings suggest that obesity and gender differentially effect 5-HT activity and are significant effectors of 5-HT induced vasospasm, sensitive to both gender and body mass in this MES model.

REAL-TIME PCR BASED APPROACHES TO THE QUANTIFICATION OF BROWN ADIPOCYTE ACTIVITY WITHIN WHITE ADIPOSE DEPOTS. A.S. Neupane<sup>1, 2</sup>, A. Baranova<sup>1, 2</sup>, A. Birerdinc<sup>1, 2</sup>& R.Mehta<sup>1, 2, 3</sup>, <sup>1</sup>Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA, <sup>2</sup>Center for the Study of Genomics in Liver Diseases, Molecular and Microbiology Department, George Mason University, Fairfax, VA and <sup>3</sup>Center for Liver Diseases, Inova Fairfax Hospital. The pandemic of obesity has been fueled by the easy access to high-calorie diet and an increasingly disengaged lifestyle brought upon by technological advancement. This not only poses a severe threat to the health of individuals, but also extends the adversity to the already overburdened global public health. Mammals, equipped with thermogenic brown adipose tissue (BAT), have evolved a unique and efficient biological mechanism to dissipate large amounts of stored energy as heat. Contrary to the energy storage function of white adipose tissue, BAT is a mitochondria rich tissue which, with the help of UCP1 protein, uncouples energy production and thus functions as an energy dissipating organ. Hence, it is highly plausible that BAT upon activation may contribute to the process of energy homeostasis. Until recently, the presence of active BAT in adult humans was highly debated. The possibility of reduced amounts or dysfunctional BAT as the underlying cause of obesity is a tantalizing and new approach in addressing the obesity epidemic. In this study, we attempt to detect the presence of BAT using novel approach. The ratio of mitochondrial DNA over nuclear DNA will be used as an indicator of the presence of mitochondria rich BAT in the visceral adipose depot of morbidly obese patients with and without NAFLD.

RT-PCR PROFILING OF MITOCHONDRIAL AND GENOMIC DNA IN VISCERAL ADIPOSE TISSUE OF NASH AND NON-NASH PATIENTS. L. Singh<sup>1,</sup> <sup>2</sup>, K. Doyle<sup>2</sup>, R. Mehta<sup>2</sup>, A. Birerdinc<sup>1, 2</sup>, A. Baranova<sup>1, 2</sup>& Z. Younossi<sup>1,3</sup>, <sup>1</sup>Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA, <sup>2</sup>Center for the Study of Genomics in Liver Diseases, School of Systems Biology, George Mason University, Fairfax, VA and <sup>3</sup>Center for Liver Diseases, Inova Fairfax Hospital. The increased prevalence of obesity has placed great emphasis on brown adipose tissue (BAT) and its role in energy homeostasis and body fat regulation. BAT characteristically has high levels of mitochondria and is distinguished by the presence of uncoupling protein 1 (UCP1) in its inner mitochondrial membrane. Data indicates an inverse relationship between body mass index (BMI) of an adult and BAT activity. In order to quantify the presence of BAT in this study, mitochondrial DNA and genomic DNA were extracted from the visceral adipose tissue of 150 morbidly obese patients with and without NASH. The resulting ratio of genomic and mitochondrial DNA served as an indicator of mitochondrial levels in NASH and non-NASH patients regardless of its activation. Custom primers were designed and specificity verified by NCBI BLAST.QIA amp DNA mini kit was used to extract the total DNA. qPCR was done to target the genomic and mitochondrial DNA. Melt curves were analyzed for product specificity. The threshold cycle  $(C_i)$  from the amplification plot was then used to calculate the mitochondrial DNA to genomic DNA expression ratio.

ASSESSMENT OF VITAMIN D LEVELS AND PARATHYROID HORMONE (PTH) LEVELS IN PATIENTS WITH NON-ALCOHOLIC FATTY LIVER DISEASE (NAFLD). Mariam Hashemi<sup>1</sup>, Massih Abawi<sup>1,2</sup>, Rohini Mehta<sup>1,2</sup>, Lei Wang<sup>1</sup>, Zobair M. Younossi<sup>2</sup>, Ancha Baranova<sup>1,2</sup> & Aybike Birerdinc<sup>1,2</sup>, <sup>1</sup>Center for the Study of Genomics in Liver Diseases, College of Science, George Mason University, Fairfax, VA <sup>2</sup>Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA. The liver has an important function in processing Vitamin D, a fat-soluble vitamin which can be obtained from sun exposure and is naturally present in foods and dietary supplement. However, patients with Nonalcoholic Fatty Liver Disease (NAFLD) are usually deficient in Vitamin D. Parathyroid hormone (PTH) increases the activity of  $1-\alpha$ -hydroxylase enzyme, which converts 25-hydroxycholecalciferol to 1,25dihydroxycholecalciferol, the active form of Vitamin D in the kidneys. The purpose of this study is to determine the correlation between the presence of Vitamin D and the presence of PTH, which regulates the body's calcium levels. Clinical samples and data were obtained from 39 morbidly obese patients during bariatric surgery. Vitamin D and PTH levels were determined in the serum using enzyme-linked immunosorbent assay (ELISA).Circulating levels of Vitamin D and PTH in the serum were inversely correlated with each other within the NASH cohort (r = -0.29, p < 0.05). However, no significant correlation was found between PTH and Vitamin D levels when comparing the NASH and non-NASH cohorts (r = -0.26, p > 0.05). In conclusion, Vitamin D and PTH levels are negatively correlated with each other significantly only in the NASH cohort.

THE ROLE OF SULFATIDE IN MEDIATING OLIGODENDROCYTE MORPHOLOGY THROUGH TAU EXPRESSION. J.M. DeLoyht & J.L. Dupree, Dept. of Anatomy and Neurobiology, Virginia Commonwealth University, Richmond

VA 23298. During development, oligodendrocyte (OL) lineage cells progress from simple bipolar cells to complex branched cells. The transition from this simple to the complex morphology temporally corresponds with the appearance of the OL sphingolipid sulfatide. Sulfatide has been identified as a potential regulator of OL differentiation and morphology. Our laboratory has reported that mice incapable of synthesizing sulfatide maintain OLs that exhibit a less complex morphology by extending fewer myelin forming processes than wild type (WT) mice. Since cellular morphology is regulated by cytoskeletal elements, we investigated the distribution and phosphorylation state of the microtubule associated protein tau. Phosphorylated tau was not observed in either OLs of 15 day old WT or sulfatide null mice. Surprisingly, accumulations of phosphorylated tau were observed in OLs of both WT and null OLs at 1 month of age. By 7 months of age, the prevalence of tau clusters was maintained in the WT cells but increased in the sulfatide null cells. Based on our data we propose that phosphorylated tau plays a role in normal OL development and myelination; however it remains to be determined how and why sulfatide depletion results in an increased accumulation of phosphorylated tau in Ols.

## Natural History & Biodiversity

DRAGONFLY PERCH SELECTION RELATED TO PERCH HEIGHT AND LOCATION. Jessica Beard & Deborah Waller, Dept. of Biological Sciences, Old Dominion Univ., Norfolk, VA 23529. A community of dragonflies (Odonata) was studied in July and August, 2011, at Hoffler Creek Wildlife Preserve, Portsmouth, VA. Male dragonflies patrol territories to secure food and mates and rest on perches. Two experiments were conducted with bamboo poles to study perch selection by adult males in relation to perch height and location, respectively. Four out of eight species present competed for the experimental perches (Pachydiplex longipennis, Brachymesia gravia, Celithemis eponia, and Libellula needhami). In the first experiment, two species used short perches (30cm) most frequently and two species used tall perches (90cm) over short perches. Perch height selection was not related to dragonfly size. Pole tops were preferred perching sites for all species but L. needhami frequently perched mid-pole. Brachymesia gravida was the dominant perching species in the beginning of the season and the least common species at the end of the season. In the second experiment, perches farther from shore (2m) were selected more frequently than those closer to shore (0.5m), regardless of perch height. Overall, species differences in perch height and seasonal use of perches could have implications in niche partitioning and competition among these species.

DEMOGRAPHY OF THE MEADOW VOLE (*MICROTUS PENNSYLVANICUS*) IN SOUTHEASTERN VIRGINIA. Jana F. Eggleston & Robert K. Rose, Dept. of Biological Sciences, Old Dominion Univ., Norfolk, VA 23529. We conducted a mark-capture-release (MCR) monitoring program of the meadow vole on the Su Tract from 2002 through 2005 and began one on the Stephens Tract in 2005. These sites are a part of the Nature Conservancy Stewardship of lands and were acquired via the Virginia Wetland Restoration Trust Fund as mitigation sites on the Northwest River drainage basin in Chesapeake, Virginia. On both tracts we established an 8 x 8 research grid, at12.5m intervals, and with two modified Fitch traps per station. We

trapped on both grids for three days each month, averaging 4600 trap nights per year. From these sites, we were interested in evaluating the life traits of these two southeastern populations. Previous analyses of the early data collected determine the growth rates and average survival rates on Su, as well as the body mass for residents versus transients, as well as found that transients were only greater than residents in spring for both sites. In this analysis, we present the data collected to date for both sites for the body mass for juveniles and adults with respect to sex ratios, the average survival rates, yearly and seasonal density variations, as well as the periods of breeding and recruitment for meadow voles.

BAIT SIZE SELECTION BY CARRION BEETLES. Jonathan M. Schmude & Deborah A. Waller, Dept. of Biol., Old Dominion Univ., Norfolk VA 23529. This work is an ongoing study on the carcass selection tendencies of carrion beetles of the family Silphidae found in Southeastern Virginia using a pitfall trap. Pitfall traps are baited with 25, 50, or 75g of a cat food/mackerel mixture and left to decay and attract beetles for one week to investigate if any Silphidae species prefer a single mass over another. The study further investigates seasonality, and mark/re-capture traveling of Silphidae. Current data suggests that all species will select any carrion found and that larger masses support greater numbers of beetles. *Nicrophorus tomentosus* significantly prefer 75g baits while *Nicrophila americana* and *Oiceotoma inaequale* prefer the 50g carrion baits. The rest of the seven carrion beetles found in southeastern Virginia resulted in either non-significant results or not enough data collected to interpret. Micro-habitat and seasonality seems to have a great affect on carrion beetle carcass selection for most species. No data has been collected yet on distance for a carrion beetle to locate a carcass in this study.

HABITAT SELECTION BY DRAGONFLY NYMPHS RELATED TO PREY DENSITY. Stefanie Snyder & Deborah Waller, Dept. of Biological Sciences, Old Dominion Univ., Norfolk, VA 23529. Dragonflies (Odonata) are important predators both as terrestrial adults and aquatic nymphs. In nature nymphs inhabit lakes, streams and ponds with a variety of bottom substrates including sand, mud, rocks and aquatic vegetation. The objective of this research was to investigate if odonate nymphs can learn to associate substrate type with food abundance. Nymphs were tested on aquarium stone and sand alone and in choice conditions to determine if they exhibited an innate preference for substrate type. Nymphs showed no color preferences among green, red, clear, or dark or light blue stones. They preferred aquarium stone over bare container bottoms, but moved randomly between substrates in choice tests with half stone/ half sand bottoms. To test nymphs for a food/substrate association, seven containers with stone bottoms and seven containers with sand substrates housed a nymph who was fed mosquito larvae for two days and then switched to the opposite substrate and starved for two days. In the following choice tests, nymphs significantly preferred the fed substrate if they had received 10 mosquito larvae/day during the trials but not if they had been fed only one larva/day. These preliminary results indicate that dragonfly nymphs can associate substrate type with food rewards.

OVIPOSITION BEHAVIOR BY DRAGONFLY SPECIES AT HOFFLER CREEK PRESERVE. Laura Campbell, Jessica Beard & Deborah Waller, Dept. of Biological

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Sciences, Old Dominion Univ, Norfolk, VA 23529. Oviposition in eight species of dragonflies (Odonata) was studied in July and August, 2011, at Hoffler Creek Wildlife Preserve, Portsmouth, VA. Females oviposited either in open water or on aquatic vegetation, and eggs were either widely scattered or clustered. Males frequently mate guarded ovipositing females by either flying in tandem with them or circling them as they lay eggs. Oviposition and mate-guarding patterns were likely related to nymphal ecology as follows: Nymphs that develop in open water must be scattered to avoid inter- and intraspecific predation, while clustered nymphs can be protected from predation in dense aquatic vegetation. Females that deposit eggs in open water (Celithemis eponina, Tramea lacerata) fly in tandem with males due to the long distances they traverse; females that lay eggs near shore in aquatic vegetation (Brachymesia gravida, Erythemis simplicicolis, Libellula needhami, Pachydiplax longipennis, Perithemis tenera, Plathemis lydia) can be monitored by hovering or perching males. Females varied in oviposition behavior related to biotic and abiotic influences. Factors that affect female oviposition patterns include seasonality (to avoid peak nymphal crowding times), time of day (to avoid male harassment), and weather (sun, rain and wind conditions).

OVIPOSITION BEHAVIOR BY DRAGONFLY SPECIES AT HOFFLER CREEK PRESERVE. Laura Campbell, Jessica Beard & Deborah Waller, Dept. of Biological Sciences, Old Dominion Univ, Norfolk, VA 23529. Oviposition in eight species of dragonflies (Odonata) was studied in July and August, 2011, at Hoffler Creek Wildlife Preserve, Portsmouth, VA. Females oviposited either in open water or on aquatic vegetation, and eggs were either widely scattered or clustered. Males frequently mate guarded ovipositing females by either flying in tandem with them or circling them as they lay eggs. Oviposition and mate-guarding patterns were likely related to nymphal ecology as follows: Nymphs that develop in open water must be scattered to avoid inter- and intraspecific predation, while clustered nymphs can be protected from predation in dense aquatic vegetation. Females that deposit eggs in open water (*Celithemis eponina, Tramea lacerata*) fly in tandem with males due to the long distances they traverse; females that lay eggs near shore in aquatic vegetation (Brachymesia gravida, Erythemis simplicicolis, Libellula needhami, Pachydiplax longipennis, Perithemis tenera, Plathemis lydia) can be monitored by hovering or perching males. Females varied in oviposition behavior related to biotic and abiotic influences. Factors that affect female oviposition patterns include seasonality (to avoid peak nymphal crowding times), time of day (to avoid male harassment), and weather (sun, rain and wind conditions).

DISRUPTIVE EFFECTS OF TROPICAL STORMS ON ALGAL BLOOMS IN THE JAMES RIVER. <u>T.A. Egerton<sup>1</sup></u>, K.C. Filippino<sup>2</sup> & H.G. Marshall<sup>1</sup>, <sup>1</sup>Dept. of Biological Sciences, ODU, and <sup>2</sup>Dept. of Ocean, Earth and Atmospheric Sciences, ODU, Norfolk, VA 23529. Algal species respond to numerous environmental variables, including pulses of nutrient concentrations. In estuarine systems, the main source of nitrogen and phosphorus is from terrestrial runoff, leading to a positive correlation between precipitation, nutrient inputs, and algal growth. This relationship can be observed in the Chesapeake Bay and its tributaries. Many cyanobacteria and dinoflagellate blooms including those in the James River occur following precipitation events. While

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precipitation/ streamflow has a positive effect on algal abundance, it significantly reduces the level of phytoplankton diversity observed annually. During September 2011, the James River watershed was subjected to hurricane Irene and tropical storm Lee. Prior to the storms, there were ongoing algal blooms dominated by cyanobacteria upstream and dinoflagellates downstream undergoing high rates of nitrogen uptake and primary productivity with relatively low levels of diversity. Following the storms, the bloom species were washed out, with decreased cell densities, nutrient uptake and productivity, and an increase in diversity as other taxa became reestablished in the wake of the blooms. The intermediate disturbance hypothesis states diversity will be highest when disruptive forces are neither too little nor too great. While the 2011 storms are an example of disturbance reducing the competitive advantage of bloom species and increasing local diversity, the long term data suggests the predicted increase in precipitation may lead to a more eutrophic, less diverse phytoplankton community. This work was supported by Virginia DEQ

POPULATION DYNAMICS AND HABITAT RESTORATION OF AMBYSTOMATIDAE SALAMANDERS IN FAUOUIER COUNTY VIRGINIA. Douglas Messier & Thomas C. Wood. New Century College, George Mason University, Fairfax, VA. Two species of Ambystoma salamanders, A. jeffersonianum (Jefferson) and A. maculatum (spotted) were investigated as part of a long-term monitoring program at Environmental Studies on the Piedmont in Warrenton, Virginia. Population monitoring in previous years indicates a relatively small Jefferson population among a large population of spotted salamanders in common breeding pools. We hypothesized interspecies competition during larval development in the spring pools would favor spotted larvae, thus resulting in a smaller adult Jefferson population. Our initial results from one common breeding pool suggest the alternative hypothesis to be correct. Despite larger numbers of egg masses and eggs laid, spotted larvae were fewer and less developed than Jefferson larvae x weeks after hatching. Jefferson larvae were more abundant (Jn=33; Sn= 16) and had a larger mean body length (Jl=24.8; Sl=16.0) at this intermediate stage of development. These results suggest that vernal pool conditions from January through May of 2012 favored Jefferson larvae development. Therefore, differences in adult population structure may be due to factors influencing other phases of the life cycle outside the breeding pools.

Continued monitoring of larvae populations in subsequent years will help us understand the dynamics of these species and additional factors that influence their reproduction and survival.

BROWNIAN PARTICLES OR BROWN LITTLE PARTICLES? RACCOON (*PROCYON LOTOR*) MOVEMENT ON THE VIRGINIA BARRIER ISLANDS. R. D. Dueser<sup>1,2</sup>, <u>N. D. Moncrief<sup>2</sup></u>, O. Keišs<sup>1,3</sup>, & J. H. Porter<sup>4,1</sup>Utah State University. <sup>2</sup>Virginia Museum of Natural History. <sup>3</sup>University of Latvia, <sup>4</sup>University of Virginia. Circumstantial evidence suggests that the distribution of raccoons (*Procyon lotor*) has increased on the Virginia barrier islands over the past 30+ years. Estimates of the timing, frequency, and trajectory of movements by individuals are among the most challenging data to obtain. We examined evidence for raccoon movements in this

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island system between 1999 and 2007. We observed inter-island movement by 5 of 203 individuals that were ear-tagged and released at the site of capture on 9 islands. We documented inter-island movement by 3 of 50 animals that were radio-collared and released at the site of capture on 8 islands. We also radio-collared and translocated 27 animals to a different island before releasing them. We observed return movements by 19 of those individuals. Finally, we examined frequency of re-colonization for 6 islands subject to biennial removal of raccoons. The 3 islands near the mainland were re-colonized repeatedly. Our translocation experiments confirm that raccoons are readily capable of crossing marshes and open water in order to move between islands. However, evidence from animals that were marked and released at the site of capture suggests that such movements are relatively rare. Except for islands very near the mainland, we suspect that island-to-island movements are more important in determining the distribution of raccoons on the Virginia barrier islands than are mainland-to-island movements.

DIAGNOSTIC TEST TO IDENTIFY FOSSILS OF THE EASTERN FOX SOUIRREL (SCIURUS NIGER) USING FLUORESCENT PROPERTIES OF BONES AND TEETH. A. C. Dooley, Jr. & N. D. Moncrief. Virginia Museum of Natural History, Martinsville, VA 24112. Remains of tree squirrels of the genus Sciurus are relatively common components of Pleistocene microvertebrate assemblages and Holocene archaeological deposits. In eastern North America, two species (the eastern gray squirrel, S. carolinensis, and the eastern fox squirrel, S. niger) are frequently reported. However, their morphological similarity and generally sympatric distributions make it difficult to distinguish between these taxa in mixed assemblages, complicating paleobiogeographic and paleoenvironmental studies. A genetic condition, congenital erythropoietic porphyria (CEP), occurs in most, if not all, living eastern fox squirrels but is absent in eastern gray squirrels and other members of the genus *Sciurus*. Due to an excess of uroporphyrin I, the bones of animals with CEP fluoresce pink under ultraviolet (UV) light, especially at approximately 400 nm. Examination of sciurid remains from several paleontological and archaeological sites has confirmed that UV fluorescence can be detected in ancient S. niger remains. This technique provides an inexpensive, non-destructive test that can definitively distinguish between S. niger and S. carolinensis in mixed fossil assemblages, and may aid in understanding how Sciurus populations repopulated eastern North America following the Wisconsinan glaciation.

THE OCCURRENCE OF THE HISPID COTTON RAT (*SIGMODON HISPIDUS*) IN A SUCCESSIONAL WETLAND IN SOUTHEASTERN VIRGINIA. <u>Meredith E.</u> <u>Bowles</u> & Jana F. Eggleston, Dept. of Biological Sciences, Old Dominion Univ., Norfolk, VA 23529. In 2005, we began a mark-capture-release (MCR) monitoring program of the small mammal community on a site known as the Stephens Tract. This site was purchased by The Nature Conservancy via the Virginia Wetland Restoration Trust Fund as a mitigation site on the Northwest River drainage basin in Chesapeake, Virginia. As such, with a rapid rate of vegetative succession indicative of the Tidewater Region, the small mammal community changed from one dominated by herbivorous old field species to those of forested wetlands, with an increase in species richness with the decline of the small mammal population, and a shift in dominance from meadow voles to hispid cotton rats. With the shift in dominance, we expected the adult female

hispid cotton rats, previously found to be residents, to show a significant increase in body weight over time. With this increase, we expected to find the females clustering on the grid, reflecting the partitioning of preferred resources. Our study site consisted of an 8 x 8 grid, at12.5m intervals, with two modified Fitch traps per station. We trapped for three days per month, averaging 4600 trap nights per year. Using ArcGIS, we analyzed the average yearly weights of females by location. Our analysis showed a steady yearly increase in weights, as well as a yearly shift in habitat use. We believe continued monitoring and the inclusion of specific preferred resources such as vegetation, as well as possible competition with the meadow voles may better resolve their occurrence on the study site.

THE OCCURRENCE OF THE MEADOW VOLE (MICROTUS PENNSYLVANICUS) IN A SUCCESSIONAL WETLAND IN SOUTHEASTERN VIRGINIA. Leah E. Thiel & Jana F. Eggleston, Dept. of Biological Sciences, Old Dominion Univ., Norfolk, VA 23529. In 2005, an ongoing monitoring program of the meadow vole population was begun on a site owned by The Nature Conservancy. Given this site is an example of the rapid rate of old field to forested wetland succession common in southeastern Virginia, it was expected that the small mammal community would change from herbivorous old field species to those of forested wetlands, as well as increase in species richness. After peaking, we expected the densities of the numerically dominant meadow vole to decline along with that of the small mammal community. Our study site consisted of an 8 x 8 grid, at12.5m intervals, and had two modified Fitch traps per station. We trapped for three days each month, averaging 4600 trap nights per year. After determining the yearly and seasonal densities for the meadow voles, we used ArcGIS to map and conduct a cluster analysis of the yearly meadow vole occurrence on the study site. With the progression of succession, this site indeed has experienced a general decline of all old field species, as well as the expected slight increase in species richness. Our densities showed a parallel decline in meadow voles both yearly and seasonally, and our cluster analysis yielded nothing significant. For our future analysis, we believe by including the hispid cotton rat data with that of dominant vegetation, we will be better able to resolve the occurrence of the meadow voles on the study site.

CONSERVATION GENETICS OF THE CRITICALLY ENDANGERED BLACK RHINOCEROS AT ADDO ELEPHANT NATIONAL PARK, SOUTH AFRICA. <u>S.</u> Josway<sup>1</sup>, C.W. Edwards<sup>2</sup>, J.M. Meyer<sup>3</sup>, R. Santymire<sup>4</sup>, & E. Freeman<sup>5</sup>, <sup>1</sup>Dept. of Environmental Science and Policy, George Mason University, Fairfax VA 22030, <sup>2</sup>College of Science, George Mason University, Fairfax VA 22030, <sup>3</sup>Freewalker Volunteer and Adventure for Charity, Port Elizabeth, Eastern Cape 6001, <sup>4</sup>Davee Center for Epidemiology and Endocrinology, Lincoln Park Zoo, Chicago IL 60614 and <sup>5</sup>New Century College, George Mason University, Fairfax VA 22030. Genetic diversity and mating strategies will be investigated in two subpopulations of wild black rhinos, *Diceros bicornis bicornis*, at Addo Elephant National Park (AENP) using appropriate molecular techniques. DNA was isolated from 76 non-invasively collected fecal samples from the Addo Section (1 male; 3 females; 13 unknown) and the Nyathi Section (8 males; 6 females; 47 unknown) of AENP. A total of 10 microsatellite loci will be screened for polymorphisms. This research will provide novel data that will

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help inform conservation strategies to maximize breeding success and genetic health in the wild populations of this critically endangered species at AENP and throughout southern Africa.

SIBLING SPECIES OR SUBSPECIES? MOLECULAR PHYLOGENETICS OF *NEOTOMA MEXICANA* FOUND SOUTH OF THE ISTHMUS OF TEHUANTEPEC. <u>M. K. Unkefer<sup>1</sup></u>, R. D. Bradley<sup>23</sup>, N. Ordonez-Garza<sup>2</sup>, & C. W. Edwards<sup>1</sup>, <sup>1</sup>College of Science, George Mason University, Fairfax VA 22030, <sup>2</sup>Department of Biological Sciences, Texas Tech University, Lubbock TX, 79409 and <sup>3</sup>Natural Science Research Laboratory, The Museum, Texas Tech University, Lubbock TX 79409. Phylogenetic relationships between members of *Neotoma mexicana* were investigated using standard molecular techniques. Mitochondrial DNA was isolated from tissue samples from 11 woodrat specimens collected from locations south of the Isthmus of Tehuantepec in southern Mexico. Sequences from the mitochondrial DNA cytochrome-*b* gene were amplified and used to elucidate the phylogenetic relationships among select *N. mexicana* subspecies and to verify the subspecific integrity of *N. mexicana* found south of the Isthmus of Tehuantepec. In the future, results from this study will be used to examine phylogenetics of the *N. mexicana* clade and to describe the historical role of the Isthmus of Tehuantepec in the evolution of rodents in North and South America.

## Psychology

ANALYSIS OF EYE TRACKING PATTERNS DURING A SIMULATED LUGGAGE-SCREENING TASK. Ivory B. Miller & Molly Liechty. Dept. of Psychology, Old Dominion University, Norfolk, VA 23508. This study assessed the impact of context in the visual scene on decision-making through a luggage-screening task. It was hypothesized that the participants would implicitly associate the distractor with the target and apply it to their visual search resulting in biased decision-making. 40 undergraduate participants performed a luggage-screening task in which spatial context was manipulated. First, participants were trained using 25 luggage images, each of which contained a target (i.e., knife) and a specific distractor (i.e., iPod). During the post-training session, participants screened 100 bags with a target base rate set at 50%. The bags contained either the distractor and the target (25 bags), the target only (25 bags), the distractor only (25 bags), or neither the distractor nor the target (25 bags). The visual search pattern was investigated through the Eyelink 1000 eye tracking system that recorded dwell time, fixation count, saccade count, saccade amplitude, and scan paths. It was found that when the spatial context of the distractor and target were relatively close, participants appeared to encode the objects together, thereby improving search efficiency. The results suggest that participants utilized relationship between objects in the visual scene to simplify the visual scene.

DOES SIMULATION STYLE MATTER? INFLUENCES OF SIMULATION STYLE ON LUGGAGE SCREENING PERFORMANCE. <u>Rachel R. Phillips</u>, Chelsea Nash & Poornima Madhavan, Dept. of Psychology, Old Dominion University, Norfolk VA 23529. Movement captures attention and, when incorporated into visual presentations, results in altered search patterns. Luggage-screening displays incorporate movement; however, many luggage-screening simulations rely on static image display.

Additionally, luggage-screening simulations tend to differ between research labs. In order to evaluate the impact of simulation style on performance, participants completed a luggage-screening task in which the stimuli were presented for 3 seconds, 9 seconds, or moving across the screen. A series of 2 (block) x 3 (condition) mixed ANOVAs revealed that participants were more likely to detect the target when image presentation times were longer (9 seconds or scrolling) and that participant perceptions of performance ability differed between conditions. These results suggest that simulation style may impact performance outcomes and should, therefore, be considered when designing experiments or comparing results between labs.

THE EFFECTS OF EMOTIONAL AND RACIAL PRIMING ON VISUAL THREAT DETECTION. Michael Padgett, Kimberly E. Culley & Poornima Madhavan, Department of Psychology, Old Dominion University, Norfolk, VA. Previous research has demonstrated that decision-making in risky situations is influenced by affect and biases associated with particular racial or ethnic groups and gender. The present study examined their combined influence in a simulate luggage-screening paradigm. The task was for participants to detect the presence of dangerous objects in x-ray images of passenger luggage. Participants were primed with one of 3 emotions — anger, fear, or sadness,- then viewed a photo of the passenger to whom each luggage item purportedly belonged before making the decision to stop or pass each of 100 bags. The results of the study revealed a significant main effect of affect for both hit rate and false alarm rate. There were no significant differences in hit rate across passengers of different races. However, there was a significant main effect of passenger race on false alarm rate. These findings indicate that affect-based heuristics exert a stronger influence on correct detections than do racial or gender biases, though decision-making performance may be degraded by both affect-based heuristics and social-cognitive biases in terms of false alarms. Generally, the results of this study indicate that affectbased heuristics and biases exert a greater influence on the decision-making processes than do social-cognitive biases relating to race or gender. Training luggage screeners to attend to task-irrelevant emotions may prevent catastrophic failures in hazard detection or chronic hindrance of security processes due to unnecessary detention of passengers.

TESTING THE VALIDITY OF THE ANTICIPATED DSM-5 INHALANT USE DISORDER DIAGNOSIS: AN ITEM RESPONSE THEORY ANALYSIS. <u>Amanda</u> <u>E. Halliburton<sup>1</sup></u>, Bethany C. Bray<sup>1</sup> & Ty A. Ridenour<sup>2</sup>, <sup>1</sup>Virginia Tech, Department of Psychology and <sup>2</sup>University of Pittsburgh, School of Pharmacy. Inhalants, which usually take the form of legal, inexpensive household chemicals and other substances, are a prevalent substance of misuse in America's preteens and teenagers. Consequences of inhalant use include cardiovascular complications, neurological damage, and kidney or liver failure. However, inhalants have not received the same attention in prevention and treatment efforts as other substances like alcohol, tobacco, and marijuana. The present study sought to compare diagnostic agreement between two measures of inhalant use disorders, the Substance Abuse Module and Schedules for Clinical Assessment in Neuropsychiatry, and to evaluate differences in the DSM-IV inhalant abuse and dependence syndromes versus the proposed DSM-5's inhalant use disorder. The community sample of 162 adolescents and young adults from St. Louis, Missouri

was 66.7% male and 83.3% Caucasian. Results indicated that diagnostic agreement between the two instruments was less than "good" based on weighted kappa. Item response theory analyses suggested that abuse and dependence diagnostic criteria lie on a single dimension, removal of the "legal problems" criterion would not alter item parameters of other criteria, and the proposed "craving" criterion is associated with severe inhalant use disorder. The findings also suggest that inhalant use is better conceptualized on one continuum (as proposed for DSM-5), than as separate abuse/dependence diagnoses (as in DSM-IV). This research was funded by grants from the National Institute on Drug Abuse (R01 15984; P50 10075; T32 017629).

EVALUATING INDIVIDUAL DIFFERENCES IN MOTION RESEARCH. Brittany N. Neilson, J. Christopher Brill, Jessica L. Habermehl & Monique S. Henderson, Department of Psychology, Old Dominion University, Norfolk VA 23529. The purpose of the present investigation is to address the importance of accounting for individual differences in susceptibility when conducting motion research. Motion sickness susceptibility is a multidimensional construct involving, but not limited to, initial sensitivity to motion, rate of adaptation to motion, and the ability to retain motion adaptation long-term (i.e., habituation or desensitization). Additionally, susceptibility to motion sickness presents differently across various forms of motion. Predictors of individual differences in susceptibility have also been identified: Women, greater than 6 years of age, Asian race, and more incidences of motion sickness among biological relatives. Previous research has assessed susceptibility factors using questionnaires to measure previous responses to sickening motion (i.e., motion sickness and simulator sickness/vection), previous responses to mild motion (i.e., sopite syndrome), and perceptual style. Another potential contributor to differences in susceptibility may be psychological processing. Individuals with high motion sickness susceptibility endorse significantly higher levels of trait anxiety. The relationship between trait anxiety and susceptibility to motion sickness should be explored further to determine if anxiety is a significant predictor. Furthermore, it is suggested that susceptibility be assessed in research using multiple measures to either categorize susceptibility groups or measure susceptibility as a covariate. If individual differences are not taken into account, then researchers may fail to find effects that are in fact present.

## **Structural Biology, Biochemistry and Biophysics**

SULFATIDES PARTITION DISABLED-2 IN RESPONSE TO PLATELET ACTIVATION. Karen E. Drahos<sup>1,2</sup>, John D. Welsh<sup>2</sup>, Julia L. Button<sup>2</sup>, Carla V. Finkielstein<sup>2</sup>, & Daniel G. S. Capelluto<sup>1</sup>. <sup>1</sup> Protein Signaling Domains Laboratory, Virginia Polytechnic Institute and State University and <sup>2</sup>Integrated Cellular Responses Laboratory, Virginia Polytechnic Institute and State University. Platelets contact each other at the site of vascular injury to stop bleeding. One negative regulator of platelet aggregation is Disabled-2 (Dab2), which is released to the extracellular surface upon platelet activation. Dab2 inhibits platelet aggregation through its phosphotyrosine-binding (PTB) domain by competing with fibrinogen for aIIbb3 integrin receptor

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binding by an unknown mechanism. Using protein-lipid overlay and liposome-binding assays, we identified that the N- terminal region of Dab2, including its PTB domain (N-PTB), specifically interacts with sulfatides. Moreover, we determined that such interaction is mediated by two conserved basic motifs with a dissociation constant (Kd) of 0.6 mM as estimated by surface plasmon resonance (SPR) analysis. In addition, liposome-binding assays combined with mass spectroscopy studies revealed that thrombin, a strong platelet agonist, cleaved N-PTB at a site located between the basic motifs, a region that becomes protected from thrombin cleavage when bound to sulfatides. Sulfatides on the platelet surface interact with coagulation proteins, playing a major role in haemostasis. Our results show that sulfatides recruit N-PTB to the platelet surface, sequestering it from integrin receptor binding during platelet activation. This is a transient recruitment that follows N-PTB internalization by an actin-dependent process.

STRUCTURAL AND FUNCTIONAL ANALYSIS OF TRYPANOSOMA CRUZI UDP-GALACTOPYRANOSE MUTASE. Jacob Ellerbrock<sup>1</sup>, Michelle Oppenheimer<sup>1</sup>, Richa Dhatwalia<sup>2</sup>, John J. Tanner<sup>2</sup>, and Pablo Sobrado<sup>1 1</sup>Virginia Tech, Department of Biochemistry, Blacksburg, VA 24060. <sup>2</sup>University of Missouri, Department of Chemistry, Columbia, MO 65211. Trypanozoma cruzi is the protozoan parasite that causes Chagas disease. Chagas disease is found predominantly in Latin America, where 8-11 million are infected and 50,000 people die from this disease every year. Galf is a sugar found on the cell surface of T. cruzi where it plays a role in host-parasite interaction. Because Galf is not found in humans, enzymes in its biosynthetic pathway are targets for drug design. UDP-galactopyranose mutase (UGM) is a flavin-containing enzyme responsible for the conversion of UDP-galactopyranose to UDPgalactofuranose (UDP-Galf), the precursor to Galf. UGM is a unique flavoprotein as the flavin must be reduced to catalyze the non-redox formation of UDP-Galf. Our group has previously shown that the flavin remains in the reduced state in order to perform approximately 1000 reactions, before it gets oxidized. Recently, the structure of T. cruzi UGM (TcUGM) was solved and several residues have been proposed to be important to stabilize the reduced form of the flavin and for substrate binding. We present the structure-function analysis of three residues important for TcUGM activity. Specifically we show that G61 and H62 play a role in maintaining the reduced state of the flavin. This work is important for understanding how TcUGM is able to maintain the reduced state and perform this unique reaction. Funding provided by NIH grant R01 GM094468.

HUMAN BETA-SYNUCLEIN FORMS A TETRAMERIC ALPHA-HELICAL COMPLEX. <u>Agatha Munyanyi</u>, Jason C. Collins & Lesley H. Greene, Department of Chemistry & Biochemistry, Old Dominion University, Norfolk VA 23529. Alpha, beta and gamma- synucleins are cytoplasmic proteins expressed primarily in the neurons. Alpha- synuclein shares 78% and 60% identity with beta- and gamma- synuclein respectively. While alpha and beta -synuclein colocalize in the presynaptic axon terminals of the central nervous system including the dopaminergic neurons of the substantia nigra, gamma-synuclein is expressed in the neurons of the peripheral nervous

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system. The N-terminal domains of the three proteins predominantly consist of several degenerate 11 amino acid repeats containing a conserved KTKEGV sequence, while the C-terminal domains are acidic and vary between the proteins. The expression of recombinant human beta-synuclein yielded a high molecular weight form which was confirmed by running analytical gel filtration chromatography. The high molecular weight form eluted at the same location as a 55.6 kDa protein marker, glutamic dehydrogenase, indicating a tetrameric complex of the 14 kDa beta-synuclein monomer. Far-UV circular dichroism spectra indicated a helical secondary structure for the beta-synuclein complex. The ultimate aim is to solve the three-dimensional structure of this important complex which may be the natural physiological and functional form of the protein in vivo.

CHARACTERIZATION OF THE OLIGOMERICSTATES OF HFQ FORM *THERMOTOGA MARITIMA* ALLUDES TO A COMPLEX REGULATORY INVOLVING TWO DISTINCT OLIGOMERIC STATES. Jennifer Patterson & Cameron Mura. University of Virginia, Charlottesville, VA 22904. RNA-based regulatory pathways and their effect on gene expression enable bacteria to rapidly adapt to various environments. For instance, mRNA levels for a particular protein depend on rate of transcription and half-life; rapid degradation of mRNAs is essential to the ability of bacteria to respond to their environments. The host factor 'Hfq' is involved in regulation of bacterial gene expression via its role as an RNA chaperone. The functional form of Hfq in *Escherichia coli* and other bacterial species have been previously determined to be a homohexamer, but our studies of the Hfq from *Thermotoga maritima* suggests that the functional form is the homohexamer and a homododecamer. Data suggests that the functional form is the homohexamer, which is able to interact with adenosine and uracil rich sequences, suggesting an additional level of regulation in this already complex system.

CAPTURING RNA-DEPENDENT PATHWAYS FOR CRYO-EM. J. Tanner<sup>1</sup>, K. Degen<sup>2</sup> & D. F. Kelly<sup>1</sup>, <sup>1</sup>Virginia Tech Carilion Research Institute, Virginia Tech, Roanoke, VA 24015 and <sup>2</sup>Department of Biomedical Engineering, University of Virginia, Charlottesville, VA 22908. Transmission Electron Microscopy (TEM) performed at cryo temperatures is the technique of choice for gathering structural details of large macromolecular complexes. To withstand the TEM vacuum system, specimens are frozen at high velocity and preserved in a thin film of vitreous ice. While this procedure is highly effective at maintaining the structural integrity of biological complexes, it also traps active assemblies in the midst of performing their duties. This results in a static snapshot of dynamic processes. Here we present a novel strategy to visualize multiple components of cellular pathways within a functionally relevant framework. We used the bacterial protein synthesis machinery as a model system in conjunction with modified EM Affinity Grids. In doing so, we were able to recruit multiple protein assemblies bound to nascent strands of mRNA. Our system reveals the native interactions of transcription-translational coupling events that control gene expression in prokaryotes. The combined use of Affinity Capture technology and single particle EM provide the basis for visualizing RNA-dependent pathways in a remarkable new way.

CRYSTALLOGRAPHIC SCREENING OF METABOLITE COCKTAIL BINDING AS A TOOL FOR DETERMINATION OF UNKNOWN PROTEIN FUNCTION. I.A. Shumilin<sup>1,3</sup>, M. Cymborowski<sup>1,3</sup>, O. Chertihin<sup>1</sup>, K.N. Jha<sup>1</sup>, J.C. Herr<sup>1</sup>, S.A. Lesley<sup>2</sup>, A. Joachimiak<sup>3</sup> & W. Minor<sup>1,3</sup>, <sup>1</sup>Univ. of Virginia, Charlottesville, VA, <sup>2</sup>Joint Center for Struct. Gen., <sup>3</sup>Midwest Center for Struct. Gen., An estimated 30-40% of sequenced bacterial genes, and an even higher fraction of archaea and eukaryotes genes, encode for proteins that lack an assigned function. Identification of unknown protein functions is a high-priority task in our efforts to understand cellular processes. We applied crystallographic screening to assess the binding of compounds in a metabolite library to proteins in two families. The two families are the YjeF N family of unknown function, represented mouse apolipoprotein-I binding protein and the N-terminal domain of Tm0922 from *Thermotoga maritima*, and the PF01256 family, previously annotated as a family of small molecule kinases, represented by YxkO from *Bacillus* subtilis and the C-terminal domain of Tm0922. Selective binding of several structurally-related compounds observed for each protein provided the basis for identification of the likely ligands. Subsequently, PF01256 proteins were shown to catalyze ATP-dependent NAD(P)H-hydrate dehydration. The YjeF N proteins interacted with an adenosine diphosphoribose-related (ADPR) substrate and most likely serve as ADPR transferases. Metabolite library screening may be used as an efficient tool for the functional studies of uncharacterized proteins. Funding provided by U01-HD060491, U54-GM74492, U54-GM074942 and R01-GM53163 (NIH).

EXPRESSION, PURIFICATION, AND PRELIMINARY CHARACTERIZATION OF MEMBERS OF THE N-HYDROXYLATING MONOOXYGENASE FAMILY. Nick Keul, Reeder Robinson & Pablo Sobrado, Department of Biochemistry, Virginia Tech, Blacksburg, 24061. Microbial N-hydroxylating monooxygenases (NMOs) are a family of flavin-containing monooxygenases that catalyze the hydroxylation of amines to be incorporated into hydroxamate-containing siderophores. This class of enzymes is relatively uncharacterized, mainly due to difficulties in the production of soluble and stable recombinant enzymes. We have identified novel NMOs that might be amenable for in vitro studies. Two selections include the putrescine monooxygenase AlcA from Bordetella pertussis and the L-lysine monooxygenase NbtG from Nocardia farcinica. These enzymes are involved in the biosynthesis of alcaligin and nocobactin, respectively. Both AlcA and NbtG were expressed as fusions to maltose-binding protein and purified to homogeneity with a bound flavin cofactor. Both were found to be active by detection of hydroxylated product and preliminary kinetic values were obtained with NbtG. The L-lysine hydroxylase NbtG was found to have  $k_{cat}$  values for oxygen consumption of  $1.06 \pm 0.02$  s<sup>-1</sup> and  $1.19 \pm 0.02$  s<sup>-1</sup> for NADPH and NADH, respectively. The  $K_{\rm m}$  values for NADPH and NADH obtained were  $257 \pm 20 \,\mu\text{M}$  and  $465 \pm 34 \mu$ M, respectively. Comparison with other studied NMOs will be presented as well. Further advancement of these studies will lead to a better understanding of the mechanism of action of the NMO family of flavin-containing monooxygenases. This research was funded by a grant from NSF (MCB-1021384).

CONVERSION OF AN ALL  $\alpha$ -HELICAL GREEK-KEY PROTEIN INTO HIGHLY ORDERED AMYLOID FIBRILS COMPOSED OF  $\beta$ -SHEET STRUCTURE. Jason C. Collins & Lesley H. Greene, Department of Chemistry & Biochemistry, Old Dominion University, Norfolk VA 23529. The transition of native proteins into amyloid fibrils is associated with many prevalent diseases. The investigation into the determinants of protein misfolding and fibril formation is an increasingly expanding and important field of research. The transformation of a functional folded protein into a fibril-like polymer composed of  $\beta$ -sheet structure is a fascinating transition. Proteins can be induced to alternatively fold into amyloid fibrils *in vitro* using a variety of factors such as primary sequence polymorphisms, agitation, temperature and solution conditions. In this talk we present the identification of a specific set of conditions required to induce the transition of the all  $\alpha$ -helical Fas-associated death domain, a nonamyloidogenic protein, into a fibril-like morphology composed of a rich  $\beta$ -sheet structure. The transition to fibrils is monitored using a broad range of spectroscopic techniques which include atomic force microscopy, fluorescence spectroscopy and circular dichroism. In summary, we show that out of ninety-five conditions attempted only one condition produced fibril-like structures which indicates, that proteins are robust against the amyloid conformation.

UBIOUITIN MODULATES TOLLIP FUNCTION IN THE ENDOCYTIC PATHWAY. <u>S Mitra<sup>§</sup></u>, A Traughber<sup>§</sup>, S Gomez<sup>§</sup>, C. V. Finkielstein<sup>⊥</sup>, L Li<sup>||</sup> & D. G. S. Capelluto<sup>§</sup>, <sup>§</sup>Protein Signaling Domains Laboratory, <sup>⊥</sup>Integrated Cellular Responses Laboratory and <sup>||</sup>Laboratory of Innate Immunity and Inflammation, Department of Biological Sciences. Mono or polyubiquitylation of proteins mediate plethora of cellular processes such as signaling, histone modification, proteosomal degradation, and receptor-endocytosis. Ubiquitin modifications on target proteins are recognized by ubiquitin binding domains that are found in several effector proteins. In this study, we describe the function of Toll-interacting protein (Tollip), which is an effector protein found in mammalian innate immune signaling pathway. We demonstrate for the first time ubiquitin and lipid binding regulate Tollip's role as an endocytic adaptor protein. We propose that ubiquitin binding to Tollip induces a conformational change leading to a closed conformation, in which it cannot interact with endosomal membrane phosphatidylinositol 3-phosphate. Release of ubiquitin from Tollip induces an open conformation suitable for endosomal membrane targeting. Overall, our findings will provide the structural and molecular basis to understand how Tollip, as an endocytic adaptor protein, is modulated by ubiquitin and determines the fate of polyubiquitinated cargo for endosomal degradation.

COLOCALIZATION OF ROBO1 AND ALCAM IN MOUSE BRAINS. <u>Caroline</u> <u>Owen</u> & Amanda G. Wright, Department of Biology and Physical Sciences, Marymount University, Arlington, VA 22207. It is shown here that Activated Leukocyte Cell Adhesion Molecule (ALCAM), a member of the immunoglobin superfamily, and ROBO1, the receptor for the SLIT family of ligands, colocalize in postnatal day 0 (P0) mouse brains. This experiment was done using coimmunofluorescence staining of cryopreserved mouse brain sections. Fluorescent microscopy, paired with Hoescht staining, was used to analyze location of the two proteins within individual cells. The results showed that ROBO1 and ALCAM do colocalize in postnatal day 0 mouse brain and suggests a likely interaction of the two

proteins to potentially regulate axon guidance. Supported by Marymount University's DISCOVER program and the Clare Boothe Luce Foundation.

A WINDOW INTO THE YEAR 2100: THE EFFECT OF HIGH pCO, ON GENE EXPRESSION IN THE CYANOBACTERIA Trichodesmium erythraeum. Nardos Sori<sup>1</sup>, Ivy Ozman<sup>2</sup>, Adele Kruger<sup>3</sup>, Margaret Mulholland<sup>2</sup>, Alice Hudder<sup>4</sup> & Lesley Greene<sup>1</sup>. <sup>1</sup>Department of Chemistry and Biochemistry, Old Dominion University, Norfolk, Va. <sup>2</sup>Department of Ocean, Earth and Atmospheric Sciences, Old Dominion University, Norfolk, Va. <sup>3</sup>School of Medicine, Wayne State University, Detroit, Michigan. <sup>4</sup>Institute of Environmental Health Sciences, Wayne State University, Detroit, Michigan. Increase in atmospheric carbon dioxide is a key factor in global climate change and can lead to alterations in ocean chemistry. It is estimated that by the year 2100 atmospheric carbon dioxide will exceed 700 ppm. Cyanobacteria are ubiquitous organisms believed to have originated about 2.7 billion years ago that can help in studying the biological response to increasing carbon dioxide. Trichodesmium erythraeum was chosen to understand the effect of global warming on cyanobacteria. Total RNA sequencing was used to quantify changes in gene expression in Trichodesmium erythraeum grown under present day and projected  $pCO_2$ concentrations for the year 2100. A bioinformatics analysis indicates that there are a substantial number of genes affected by high pCO2. The results of our studies will be presented.

CIRCADIAN MODULATION OF ESTROGEN RECEPTOR ALPHA PROMOTER ACTIVITY. Kelly A. Barford, Linda M. Villa, Marian Vila & Carla V. Finkielstein, Integrated Cellular Response Laboratory Virginia Polytechnic Institute. The circadian rhythm is an important method of controlling the body's internal clock and adapts to environmental changes. When this clock is disrupted, various diseases have been shown to arise, including breast cancer. Period 2 (PER2), a circadian protein, is the key protein that drives the circadian clock. Cancers, such as breast and ovarian, have shown a loss of PER2 function. This connection between loss of PER2 and the development of estrogen-related tumors has led to a correlation between this protein and estrogen receptor alpha (ER $\alpha$ ). ER $\alpha$  mRNA oscillates during the day, similar to PER2, but instead of peaking during the day, ER $\alpha$  peaks at night. Up-regulation of ER $\alpha$  has been shown to influence tumor growth, and if this mechanism can be determined it will lead to a better understanding of estrogen-related cancers, specifically breast cancer. The focus of this project is the effect of Per2 on ERa. This will be done through increasing the level of Per2 in MCF-7 cells and analyzing the expression of the downstream genes through qRT-PCR.

CONFORMATIONAL CHANGE OF THE RYANODINE RECEPTOR AT 10 ANGSTROMS RESOLUTIOn. <u>Tyler W.E. Steele</u> & Montserrat Samsó, Department of Physiology and Biophysics, Virginia Commonwealth University, Richmond VA 23298. The ryanodine receptor isoform 1 (RyR1) is an intracellular ion channel with an important role in depolarization-induced Ca<sup>2+</sup> release and skeletal muscle contraction. Several of RyR1's molecular partners are distantly situated from its ion gate, suggesting that long-range conformational pathways play an important role in RyR1's function. We have compared two kinds of conformational changes by cryo

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electron microscopy and analysis of the 3D reconstructions at 10 Angstroms resolution: those resulting from the transition of the closed to the open state and those resulting from the removal of FKBP12 under closed-state conditions. The vector representation indicates movement of different magnitudes and directions along the 3D structure of RyR1, delineating a long-range allosteric pathway, spanning more than 10 nm, that could couple the binding of RyR1's binding partners and the state of its ion gate.

CIRCADIAN AND CELL-CYCLE MEDIATED REGULATION OF PER2 LOCALIZATION IN COLON CANCER CELLS. Kaitlyn J. Andreano, Marian Vila-Caballer & Carla V. Finkielstein, Integrated Cellular Responses Laboratory, Department of Biological Sciences, Virginia Polytechnic Institute and State University. A key aspect of cancer research is identifying new regulatory pathways involved in proliferation and differentiation of a cancer cell. Disruption of circadian rhythm has recently emerged as a new potential risk factor in the development of cancer, pointing to the core gene *period 2 (per2)* as a tumor suppressor. Immunofluorescence studies show endogenous Per2 localizes in both cytosol and nucleus in asychronized colon cancer cells. Initial experiments aimed to evaluate whether Per2 localization was cell cycle dependent. Accordingly, HCT116 colon cancer cells were arrested by treatment with nocodazole, hydroxyurea, and thymidine. Using immunoflourescence microscopy, we visualized the localization of endogenous Per2 when cells were arrested at either S or G2 phase as monitored by flow cytometry. Results show Per2 localizes in both nuclei and cytosol when cells are arrested in S but later shuttles to the cytosol during G2 phase. Further experiments aim to elucidate the various Per2 partners that drive its localization and the function of Per2 in the various compartments and among the different cell cycle phases.

HIGH THROUGHPUT MICROARRAY STUDY OF THE EFFECT OF HEME-MEDIATED REGULATION OF PER2 STABILITY FOR GENE EXPRESSION. R. Powles<sup>1</sup>, J. Yang<sup>1</sup>, R. V. Jensen<sup>2</sup> & C. V. Finkielstein<sup>1</sup> Integrated Cellular Responses Laboratory, <sup>1,2</sup> Dept. of Biological Sciences, Va. Polytechnic Inst. & State Univ., Blacksburg VA 24061. Results from our group show that proteasome-mediated degradation of the human core circadian transcription factor period 2 (hPer2) results from heme, a metabolite that exhibits rhythmic oscillations and is crucial for maintaining cellular homeostasis. In vivo studies show heme levels directly control hPer2 stability and by extension influence the period length and phase-shifting properties of the biological clock in synchronized cells. Using microarray-based gene expression analysis of Aml-12 mouse liver cells to study the effects of hPer2 reduction either by hemin-treatment or by silencing using siPer2, we identify novel pathways regulated by hPer2 using Pathway Analysis based on Gene Ontology (GO) classifications and the biomedical literature. Multiple transport and metabolic genes are found to be differentially modulated by heme-mediated hPer2 degradation and siPer2 treatment. Many of these genes are overexpressed in cancer cells or belong to major transcription factors involved in cellular proliferation. Results reveal multiple cell-wide pathways that act together in controlling normal cell physiology and, when deregulated, lead to proliferative disorders.

IDENTIFICATION OF STRUCTURAL DETERMINANTS OF COENZYME SELECTIVITY IN EUKARYOTIC UDP-GALACTOPYRANOSE MUTASES. LuisMiguel Solano & Pablo Sobrado, Dept. of Biochemistry, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061. UDP-galactopyranose mutase (UGM) is the key enzyme in the biosynthesis of galactofuranonose (Galf), a sugar found in the cellular surface of many prokaryotic and eukaryotic pathogens including the fungus Aspergillus fumigatus. The chemical mechanism of the UGM from A. *fumigatus* has been recently elucidated indicating that the conversion of the substrate UDP-galactopyranose (UDP-Galp) to UDP-Galf, requires previous reduction of UGM enzymes by NAD(P)H. Here, we identify key amino acids in AfUGM involved in NAD(P)H binding based on structural and mechanistic studies. The role of the residues studied on coenzyme reactivity in AfUGM was determined by rapid reaction kinetics of purified mutants in a stopped-flow spectrophotometer. Measurements of the rate of reduction  $(k_{red})$  of the flavin cofactor at 452 nm, shows that mutations on residues R91, S93 and R447 produce a significant effect on the enzyme kinetics. These mutants have diminished  $k_{red}$  in comparison with wild-type AfUGM. This work gives new insights into the mechanism of AfUGM by elucidating key residues involved in NAD(P)H binding. This work was supported by NIH grant R01 GM094468.

DAB2: COMPARATIVE STUDIES BETWEEN ANTI-AGGREGATORY DRUGS. Julia L. Button<sup>1</sup>, Olutayo Sengura<sup>2</sup>, Marian Vila<sup>1</sup>, Daniel G.S. Capelluto<sup>3</sup>, & Carla V. Finkielstein<sup>1</sup>, <sup>1</sup>Integrated Cellular Responses Laboratory, Virginia Polytechnic Institute and State University, <sup>2</sup> Virginia College of Osteopathic Medicine and <sup>3</sup> Protein Signaling Domains Laboratory, Virginia Polytechnic Institute and State University. Cancer patients, especially those in the metastatic stage, have been found to have highly elevated platelet counts. The thrombocytosis (high platelet count) symptom has been associated with poor prognoses in many cancers. Platelets play a role in metastasis by forming platelet-tumor cell masses in the circulatory system preventing recognition and activation of an immune response. Anti-platelet drugs are supported to be potential cancer therapies by preventing the activation and aggregation of platelets in the bloodstream, thus preventing cancer cells from surviving and proliferating in target metastatic tissues. Disabled-2 (Dab2) is a protein that inhibits platelet aggregation by binding to sulfatides found on the outer-leaflets of the platelet cell membrane. The focus of this project is to compare the cytotoxic potential of currently prescribed antiaggregatory drugs, including aspirin and plavix, to Dab2, a sulfatide-deficient form of the protein and a recombinant peptide comprising the sulfatide binding motifs. Our results show the N-terminus portion of the Dab2 exhibits less cytotoxic effect than current anti-aggregatory drugs, while inhibiting platelet/cancer cells association. These results provide basis for the use of Dab2 as part of an adjuvant therapy to treat cancer.

THE EFFECTS OF CITRATE ON GLYOXASOMAL MALATE DEHYDROGENASE. Jacqunae Mays<sup>1</sup>, James D. Marion<sup>2</sup> & Ellis Bell<sup>1</sup>, <sup>1</sup>Laboratory for Structural Biology, Biophysics & Bioinformatics, University of Richmond, Richmond, VA 23173, <sup>2</sup>Department of Biochemistry & Molecular Biology, Virginia Commonwealth University, Richmond, VA 23298. Malate Dehydrogenase plays critical roles in the cytosol, the mitochondrion and in plants also in glyoxysomes. Mitochondrial and gyloxysomal forms of malate dehydrogenase are thought to be

subject to allosteric regulation by citrate. To explore the mechanisms of citrate effects on the enzyme we have used a combination of initial rate kinetics, site directed mutagenesis, circular dichroism (in conjunction with thermal melts) to follow effects on secondary and tertiary structure, dynamic light scattering to follow effects on quaternary structure and limited proteolysis to explore local flexibility/accessibility changes. Citrate, which binds to only one of the two otherwise identical active sites, is a partial inhibitor under all circumstances examined (varied pH, forward or reverse reaction), has no effect on the quaternary structure of the protein but induces changes in local flexibility/accessibility in a loop near the subunit interface. Several mutations at the subunit interface (V195A, L269A, E256Q, H90Q)also impact citrate inhibition. Together these results suggest that citrate exerts its effects on activity by binding to the "empty" active site in a reciprocating subunit mechanism, eliciting altered subunit interactions that contribute to overall catalysis. This work is supported by NSF Grant MCB 0448905 to EB.

ENHANCING STUDENT ENGAGEMENT IN AN ADVANCED PROTEIN STRUCTURE, FUNCTION & BIOPHYSICS CLASS. Ellis Bell, University of Richmond, Richmond, VA 23173. Junior and Senior majors in a biochemistry and molecular biology major take advanced courses in the discipline yet have often only been exposed to traditional lecture based courses. As a result they are often ill-prepared to think about foundational concepts or apply them to new situations and are not adept at the various skill sets necessary for them to compete in graduate school or future professions. To counter these effects a dynamic and active classroom engages students more fully in their own education. In an upper level protein structure, function and biophysics course which revolves around three major themes, Protein Structure and Flexibility, Energy Barriers and ways to overcome them, and Function and Regulation, a variety of active learning strategies including small group discussion and peer teaching, literature discussion and presentation, original summary synthesis of topics from the literature, challenging problem sets containing real data, and a fully integrated laboratory experience where students have to design their own experiments and interpret data replace traditional lectures and labs. Although students take a while at the start of the course to adapt to such a different teaching and learning style, pre and post testing using tools designed to assess foundational knowledge and skills shows significant gains in student understanding. This work is supported by NSF Grant MCB 0448905 to EB.

INHIBITORY MECHANISM AND REGULATORY RELEASE OF SIKE FROM THE TBK1 KINASE COMPLEX. J.D. Marion & J.K. Bell, Dept. of Biochem. and Mol. Biol., Virginia Commonwealth University, Richmond, VA 23284. The innate immune system is the body's first line of defense against infectious agents. Essential to this response are cellular mechanisms that recognize, sequester and eradicate these invading organisms. Toll-like receptor 3 (TLR3), a transmembrane receptor found in endosomes, is stimulated by the viral genomic material double stranded RNA (dsRNA), initiating a signaling cascade that leads to the production of type I interferons and proinflammatory cytokines. Critical to this pathway is a proposed kinase complex, NAP1 (NAK associated protein 1) – TBK1 (TANK Binding Kinase 1) – IKKe (IkB kinase), which upon activation, leads to the phosphorylation of IRF3/7 (interferon regulatory factor) and production of IFNb. To control this activity, SIKE (suppressor of IKKe) acts as a physiological inhibitor of IKKe and TBK1 through a previously undefined mechanism. These studies show SIKE to be a mixed-type inhibitor, regulating the kinases through both competitive and non-competitive modes of inhibition. Further studies have elucidated post-translational modifications that affect SIKE before and after dsRNA stimulation. We hypothesize that post-translational modifications mediate SIKE release from the NAP1-TBK1-IKKe complex allowing for IRF3/7 activation. Current studies are focused on 1) SIKE post-translational modifications, 2) how modifications may alter SIKE protein interactions, and 3) kinetic analysis of SIKE's effect on TBK1 activity. Funding provided by the Jeffress Memorial Trust.