dimensionless displacement S(t) of the form:

$$\frac{d S(t)}{dt} = \alpha_1 S - \alpha_2 S^3 + \widetilde{F}(t) \text{ where } \widetilde{F}(t) \text{ is purely random,}$$

stationary, Gaussian, process with zero mean, and represents the effects of the amplitude fluctuations due to the random impacts of the ambient air on the beam and

where the  $\alpha_i$  are constants dependent upon the thermal and modal bending moments, damping ratio and the phase difference between the velocity fluctuation and thermal bending moments. We solve the stochastic differential equation by constructing a symmetry breaking, bi-stable *Thermal Potential Energy Function*, a Lyapunov global stability function which permits a general investigation and analysis of the system's stability and the effects of pressure on thermally induced bending vibrations.

ASTRONOMICAL POLARIMETRY AT VIRGINIA MILITARY INSTITUTE. <u>Gregory A. Topasna</u>, Daniela M. Topasna & Gerald B. Popko, Department of Physics and Astronomy, Virginia Military Institute, Lexington, VA 24450. We present current work on the design, construction, and testing of a two-beam optical polarimeter to be used with the 20-inch telescope at the Virginia Military Institute observatory. The basic operation of the device will be discussed as well as results which demonstrate the two-beam method in the laboratory. Issues regarding automation and data handling as well as planned observations will be presented.

# **Biology**

DELAYED TREATMENT WITH SILDENAFIL ATTENUATES ISCHEMIC CARDIOMYOPATHY. V.Q. Chau, F.N. Salloum & R.C. Kukreja. Div. of Cardiology, Virginia Commonwealth Univ., Richmond, VA 23298. We previously showed that chronic inhibition of phosphodiesterase-5 (PDE-5) with sildenafil immediately after permanent occlusion of the left anterior descending coronary artery (LAD) limits myocardial infarction (MI)-induced heart failure (HF) in mice. To mimic more clinical scenarios, we hypothesized that chronic treatment with sildenafil beginning at 3 days post MI would also preserve LV function and reduce HF progression. Adult male ICR mice underwent MI by permanent ligation of the LAD after baseline echocardiography was performed. Three days post MI, a repeat echocardiography was conducted. Mice with LV fractional shortening (FS) less than 25% received sildenafil (21 mg/kg; ip; BID, Group I) or volume-matched saline (Group II) for 25 days. At the completion of 28 days following MI, the mice underwent a repeat echocardiography prior to sacrifice. Group I expressed less LV dilatation than group II, and group I showed better contractility as compared with group II. LV enddiastolic diameter (LVEDD), increased from a baseline value of  $3.4 \pm 0.1$  mm to 4.2 $\pm$  0.1 at 72 hr post MI. At 28 days post MI, LVEDD was increased to 5.2 $\pm$ 0.1 mm for group II, as compared 4.6±0.1 mm in group I (P<0.05 vs. Group II). Fractional shortening decreased from a baseline value of 47±1% to 19±1% at 72 hr following MI. At 28 days post MI, FS was  $21\pm1\%$  for group I and  $13\pm1\%$  for group II, (P<0.05 vs.

Group II). For the first time, these results show that chronic sildenafil treatment initiated at 3 days post MI attenuates ischemic cardiomyopathy by limiting LV dilatation and preserving FS. Sildenafil may be a promising therapeutic tool for prevention of HF in patients with MI.

CONTROLLED ASSEMBLY OF NANOSCALE PROTEIN/DNA "BASKETS" FOR THE IN VIVO DELIVERY OF siRNA PARTICLES. A.C. Zirzow, C.B. Smith, and A.V. Baranova, Dept. of Mol. and Microbiol., R. Couch and A.S. Patanarut, Dept. of Chemistry, George Mason Univ., Fairfax VA 22030. The purpose of this research is to develop a novel vector for the *in vivo* delivery of siRNA particles. In attempt to overcome current siRNA transfection problems of cytotoxicity, physical size of vector, instability of siRNA, and rapid clearance from the bloodstream, we developed the concept of a nanoparticle comprised of an outer cage made of protein and DNA (DNA "basket") that is capable of carrying siRNA cargo. This investigation demonstrates that protein/DNA interactions can be exploited to form DNA "baskets" with a stable mean size distribution. Derinat (Деринат), a DNA-based immunomodulator, is employed as the DNA component of these DNA "baskets". When prepared in the absence of protein, Derinat retains a stable mean size distribution of  $762.4 \pm 7.26$  nm. When this DNA is vacuum concentrated with a specific quantity of bovine serum albumin (BSA), the mean size distribution can be significantly reduced by up to 98%. Reduced mean diameters of the DNA/BSA complex may allow for more efficient in vivo cellular uptake. The next step in this investigation is to determine if a DNA/BSA complex can contain and hinder the degradation of GFP specific siRNA particles for in vivo delivery into GFP mice. The degradation of fluorescently tagged/quenched siRNAs will be monitored *in vitro* in nuclease-containing serum and *in vivo*. This novel approach to siRNA based therapy may minimize side effects, increase cellular uptake, and provide a scaffolding upon which ligands may be attached to direct siRNA to tissues of interest.

A SEARCH FOR KCNRG MUTATIONS IN MULTIPLE MYELOMA CELL LINES. Stephanie L. Coon & Aybike Birerdinc & Ancha Baranova. Dept. of Mol. and Microbiol., George Mason Univ., Fairfax VA. 22030. Deletions and or rearrangements on chromosome 13q14.3 are observed in more than half of multiple myeloma (MM) and chronic lymphocytic leukemia (CLL) cases and are also frequently seen in other hematopoietic malignancies. The minimal common deleted region (CDR) in MM cells contains candidate tumor suppressor gene KCNRG (potassium channel regulating gene), the transcript of which suppresses Kv channels associated with the proliferation of lymphocytes. KCNRG exerts growth suppressive and pro-apoptotic effects in HL-60, LnCaP and RPMI-8226 cells. In this study we sequenced KCNRG gene in three multiple myeloma cell lines. We found that RPMI-8226 cell line contains a delT mutation in the core promoter initiator element. Deletion of T decreases matrix similarity of the match from 0.945 to 0.941, and, therefore, might negatively influence expression of KCNRG in RPMI-8226 cells. This suggests that KCNRG expression may be negatively influenced in this model line. The haploinsufficiency of KCNRG might be relevant to the progression of CLL and MM at least in a subset of patients. This research was performed under NIH R1R15CA113331-01, RFFI07-04-00379-a, 07-04-12232-ofi, and 04-04-08154-ofi.

KPP: KEGG PATHWAY PAINTER. Ganiraju Manyam, Vikas Chandhoke & Ancha Baranova, Department of Molecular and Microbiology George Mason University, Fairfax, VA 22030. High-throughput technologies became common tools to decipher changes of gene expression (GE) patterns. Functional analysis of GE patterns is a daunting task as it often requires recourse to the public repositories of biological knowledge. On the other hand, in many cases researcher's inquiry can be served by a comprehensive glimpse. The KEGG PATHWAY database is a compilation of manually verified maps of biological interactions presented as a set of pathways related to signal transduction and other cellular processes. Rapid mapping of the differentially expressed genes to the KEGG pathways may assist in evaluation of the functional relevance of the results from microarrays and other high-throughput technologies. Web based graphic tool KEGG Pathway Painter (KPP) provides fast and comprehensive visualization of the changes in GE patterns by color-coding pathways from the KEGG database using user defined sets of the candidate genes accompanied by "overexpressed" or "underexpressed" marks, for example, those generated by microarrays. KPP is freely available and can be accessed at http://www.cos.gmu.edu/~gmanyam/kegg/. The study was supported by NIH 1R15CA113331-01 and Service GRA of College of Science, George Mason University.

CUG2 (C6ORF173) IS A NOVEL ONCOGENE INVOLVED IN BREAST CARCINOMA. Elizabeth D. Nohelty<sup>1</sup>, M. Skoblov<sup>2</sup>, V. Kuznetsov<sup>3</sup>, & A. Baranova<sup>1,2</sup>, <sup>1</sup>Department of Molecular and Microbiology, George Mason University, Fairfax, VA; <sup>2</sup>Russian Center for Medical Genetics, Moscow, Russia, <sup>3</sup>Bioinformatics Institute, Singapore. The genetic mechanism of the aggression of breast carcinoma has been a topic of research efforts since its discovery in the human population. Previous studies showed that an increase of C6ORF173 expression is associated with shorter survival after breast carcinoma diagnosis (Ivshina et al., 2007). Using Real-Time PCR ready TissueScan Cancer qPCR Arrays comprised of normalized cDNA prepared from pathologist-verified breast carcinoma samples we demonstrated that expression levels of C6ORF173 are significantly (P < 0.013) higher in Grade 3 breast carcinoma tumors as compared to Grade 2. C6ORF173 has been cloned into the pCDNA3.1 expression vector and stably transfected into HCC2157, NM2C5 and MCF-10A breast carcinoma cell lines. Analysis of transfected NM2C5 cells demonstrated a statistically significant increase of proliferation after 48 hrs of incubation with BrDu (P < 0.00016) as well as an increase in migration and invasion, while apoptosis ability of NM2C5 was not changed. Weak homology of C6ORF173 to a known downregulator of transcription (DR1) suggests its involvement in gene expression regulation in a broad sense. If the C6ORF173 gene indeed plays a large role in the aggression of breast carcinoma, it is possible that a genetic screen can be implemented to further improve the predictive diagnostic and treatment of breast carcinoma.

FRANCISELLA NOVICIDA FORMS IN VITRO BIOFILMS MEDIATED BY AN ORPHAN RESPONSE REGULATOR. <u>Meghan W. Durham-Colleran</u>, Anne Brooks Verhoeven, & Monique L. van Hoek, Department of Molecular and Microbiology, National Center for Biodefense and Infectious Diseases, George Mason University, Manassas, VA 20110. *Francisella tularensis* is associated with water and waterways,

Virginia Journal of Science, Vol. 60, No. 2, 2009

and infects many species of animals, insects, and protists. The mechanism Francisella utilizes to persist in the environment and in tick vectors is currently unknown. We have demonstrated for the first time that Francisella novicida, a model organism of F. tularensis, forms a biofilm in vitro. Selected F. novicida transposon mutants were tested for their ability to form biofilm compared to the wildtype F. novicida strain. Mutation of the putative qseB gene led to an impairment in the ability to form biofilm with no impairment in bacterial growth. A gseC mutant had impaired growth, but demonstrated a marked impairment in biofilm production. Mutation in *capC* affected both bacterial growth and biofilm formation, but no biofilm production impairment was seen with *capB* or *pilE* mutants. A deletion mutant in the orphan response regulator FTN 1465, which we propose is the putative QseB, formed significantly less biofilm than the wildtype. When FTN 1465 was complemented back into the deletion mutant, biofilm formation was restored. Thus, the orphan response regulator FTN 1465 is an important factor in biofilm production in vitro in F. novicida. These results demonstrate that Francisella species are able to form biofilms in vitro, suggesting that biofilm formation may be important for the life cycle of this organism in the environment or possibly in the tick vector.

TWO MINOR SPECIES AS DOMINANTS IN AN OLDFIELD RODENT COMMUNITY. Robert K. Rose, Dept of Biol. Sci., Old Dominion Univ., Norfolk, Virginia 23529-0266. Oldfields are early stages in secondary succession dominated by herbivores, including three species of common rodents (meadow voles, cotton rats, marsh rice rats). In two oldfield community studies in eastern Virginia, these species comprise >90% of captures, but all were absent in another oldfield habitat, created after logging of a pine forest, clearing, and mechanical planting of pines. There the grasses, sedges, and spikerushes provided habitat to support populations of two minor herbivorous rodents, southern bog lemming (*Synaptomys cooperi*) and woodland vole (*Microtus pinetorum*), which reached densities of 15/ha and 35/ha, respectively, across an 18-month capture-mark-release study. Besides the >450 captures of these two dominants, 7 captures of harvest mice were recorded, but no meadow voles, cotton rats, or marsh rice rats. Some studies conducted elsewhere suggest that southern bog lemmings lose in competition with meadow voles, but the reasons for the presence or absence of a species cannot be determined without experimentation.

BODY SIZE AND GROWTH PATTERNS OF *MICROTUS PENNSYLVANICUS* (ORD.) IN CHESAPEAKE, VIRGINIA. Sara E. Bell & Robert K. Rose, Dept. of Biol., Old Dominion Univ., Norfolk VA 23529-0266. From Dec 2002-Feb 2008, we did a capture-mark-release study on 2 Chesapeake, VA populations of meadow voles (*Microtus pennsylvanicus*). The study sites were effectively 1 ha grids in oldfields. We put 2 modified live Fitch-type traps at 12.5 m intervals and trapped on the grids in monthly 3-day sessions. In northern North America, voles experience autumn and winter weight loss, demonstrate delayed growth and sexual maturation in autumn-born young, have lifespans under 15 weeks, and typically weigh no more than 55 g. Chesapeake voles experienced no seasonal weight loss, exhibited no delayed growth or sexual maturation, lived over 20 weeks, and nearly 20% weighed over 70 g. The longest-lived vole was an 80-week-old male. The heaviest individual voles were over

90 g and present in late autumn and winter at both sites. Breeding occurred year-round. Thus, meadow voles in eastern Virginia contrast sharply with more northerly populations in many aspects of their biology.

NATURAL GENETIC VARIATION IN METABOLIC RATE AND ACTIVITY IN WHITE-FOOTED MICE (PEROMYSCUS LEUCOPUS) IN RELATION TO GENETIC VARIATION IN REPRODUCTIVE PHOTORESPONSIVENESS. Madelyn G. Crowell<sup>1</sup>, Paul Kaseloo<sup>1</sup>, and Paul Heideman<sup>2</sup>, <sup>1</sup>Dept. of Biol., Va. State Univ. Petersburg VA 23806 and <sup>2</sup>The Coll. of William and Mary, Williamsburg VA 23185. A naturally-variable life history trait with underlying physiological variation is the photoperiodic response of many temperate zone rodents, including white-footed mice (Peromyscus leucopus). Male P. leucopus were obtained from a short photoperiod responsive (R) line, selected for reproductive suppression in short-day conditions (SD) and a non-responsive (NR) line selected for reproductive maturity in SD. NR mice consume  $\sim 50\%$  more food than R mice, but have no significant difference in body mass. We quantified differences in the energy budgets of these lines through respirometric measurements at thermoneutral temperature. Basal metabolic rate (BMR) was significantly greater in NR than R mice. In addition, NR mice engaged in significantly more daily activity. No significant difference in mass of major metabolic organs or dry mass digestibility of food was found between lines. The increased BMR and sustained metabolic rate in NR mice was correlated with testis size, but not with major central organs. The genetic difference in intake requirements between lines was great enough to be reasonably attributable to selection on the natural genetic variation in BMR and activity in the wild source population. These findings are consistent with differences in thyroid-related hormone activity which recent findings suggest mediate the response to photoperiodic reproduction. This study was funded in part by Howard Hughes Medical Institute.

HABITAT AVAILABILITY AND SPECIES-AREA RELATIONSHIPS OF INDO-PACIFIC SHORE BIOTA. Jonnell C. Sanciangco & Kent E. Carpenter, Department of Biological Sciences, Old Dominion University, Norfolk VA 23529. In marine biogeography, the measure of available habitat is a key factor in identifying the distribution patterns of species richness. The species-area relationship (SAR) has been widely used to infer this correlation of species richness to available habitat. While several studies have shown that larger habitat areas account for a higher number of species, the factors influencing the species richness and the amount of variation have yet to be identified. In this study, the SAR of Indo-Pacific shore biota was tested using the habitat diversity index (H) and coastal length (CL) as functions of area. The H was calculated using the Shannon-Weiner formula with areas of coral reefs, seagrasses, mangroves, and soft bottom as the parameters. Species distribution maps of 6830 marine shore biota (fishes, molluscs, and crustaceans) were created using Geographic Information System (GIS). In addition, multiple GIS tools, extensions and scripts were used to create a 200 meter bathymetry shapefile which was divided into three scale sizes of equal area sections (100, 500, and 1000) to minimize area effect. Values of H, CL, and species richness (S) were identified in each section. Linear regression analyses were performed for S vs H, S vs CL, and S vs H + CL. Results showed significant

68

differences (<0.001) for all relationships in all scales. H accounts for more variation (14.3-19.3%) than CL (7.6-13.2%), suggesting H as a better predictor of the species richness. Results are portrayed spatially using GIS, where species distribution of marine biota can be easily identified in the map. These results are used to assess the conservation status of marine species and to identify priorities for management.

SEASONAL PHYTOPLANKTON POPULATIONS IN BACK BAY, VIRGINIA Nathan Bowman, Todd Egerton & Harold Marshall, Department of Biology, Old Dominion University Norfolk, Virginia 23529. Back Bay is a flat-bottomed, shallow water ecosystem separated from the Atlantic Ocean by a narrow zone of marshlands, dunes, and residential development. Water depth in the Bay is influenced by the prevailing northeast winds, which may alter the depth in near shore regions by as much as 1.0 m. Presently, the only salt water entry to Back Bay is wind forced, passing into the Bay through a narrow channel from a large sound to the south. Back Bay is classified as a temperate, oligonaline estuary containing salinity ranges from 1.0 - 1.9, and has gained regional interest and concern by state and federal agencies regarding changes to its ecological status. A specific objective of the Back Bay National Wildlife Refuge is to reduce the impact of various environmental factors such as nutrient loading and high turbidity levels that would deteriorate its natural setting. One of the most sensitive components within this habitat to environmental changes is the phytoplankton, which may be used as an ecological indicator of Back Bay's eutrophic status. During the course of one year, the freshwater reaches of the Back Bay oligohaline estuary were sampled bimonthly at a series of six stations comprising the entirety of the bay. The goal of this study is to determine if the specific water quality conditions in this habitat are associated with seasonal changes in the abundance and dominance of specific phytoplankton components, including a changing seasonal flora and phytoplankton categories that occurred between September 2006 and September 2007.

THE BIOLOGICAL ACTIONS OF HYDROXY-CIS-TERPENONES. Tristan A Hayes, Lin Zhang, Qibing Zhou, Ghislaine Mayer & Jennifer Stewart, Virginia Commonwealth University. Hydroxy-cis-terpenone (HCT) was synthesized by Dr. Qibing Zhou in the VCU Chemistry Department. HCT is converted to oxidized HCT (OHCT) in aqueous media. Previous studies demonstrated that micromolar concentrations of HCT and oxidized HCT (OHCT) protect human liver cells from aflatoxin. Additionally, Dr. Ghislaine Mayer in the VCU Biology Department found that nanomolar concentrations of OHCT kill all blood stages of Plasmodium falciparum, the parasite responsible for most cases of human malaria. The goal of this project was to investigate mechanisms of HCT actions. Because binding of aflatoxin to microsomal proteins is needed for activation of aflatoxin, we investigated effects of HCT on binding of <sup>3</sup>H-labeled aflatoxin to human liver HepG2 cell membranes and human liver microsome proteins. Effects of various concentrations of HCT on protein binding of <sup>3</sup>H-labeled aflatoxin were measured at various times from 30 sec to 10 min. The data indicated that HCT at 20 - 40 uM decreased binding of <sup>3</sup>H-labeled aflatoxin to proteins within 30 sec. Binding was not ATP-dependent. Low concentrations of HCT (<10 uM) did not affect binding to cellular proteins. These findings suggest HCT may reduce aflatoxin toxicity by reducing aflatoxin binding to liver cell proteins. This

Virginia Journal of Science, Vol. 60, No. 2, 2009

70

work was supported by the Jeffress Memorial Trust J-849, NSF Grant MCB-013149, and the Advisory Committee for Undergraduate Research and Creative Scholarship.

INVESTIGATION OF DUAL PHENOTYPE GABA/GLUTAMATE NEURONS IN ZEBRAFISH. Lauren P. Bell & Dianne M. Baker, Dept. of Bio. Sci., Univ. Mary Washington, Fredericksburg, VA 22401. The purpose of this research was to investigate the presence of neurons expressing both the inhibitor neurotransmitter, GABA, and the excitatory neurotransmitter, glutamate, in zebrafish. This novel class of neurons has been recently identified in rodents, and increasing evidence suggests they play a role in GnRH signaling. To determine the presence of dual phenotype neurons, we developed *in situ* hybridization (ISH) probes for mRNA encoding proteins involved in GABA synthesis (GAD67) and glutamate transport (VGLUT 2.1 and 2.2). To test theses probes, we performed single-label ISH on whole-mount larva and on adult brain sections. These tests provided consistent evidence that the GAD67 riboprobe is functional in both larval and adult brain tissue. However, the results of ISH using the VGLUT 2.1 and 2.2 riboprobes were inconsistent. Further optimization of ISH conditions is necessary before dual-label ISH can be used to test for presence of dual phenotype GABA/glutamate neurons in zebrafish.

CHARACTERIZATION OF THE EXPRESSION OF CARCINOEMBRYONIC ANTIGEN-RELATED CELL ADHESION MOLECULE 1 IN ZEBRAFISH. Colby S. Croft & Dianne M. Baker, Dept. of Bio. Sci., Univ. Mary Washington, Fredericksburg, VA 22401. The objective of this research was to characterize the spatial and temporal expression of carcinoembryonic antigen-related cell adhesion molecule 1 (CEACAM1) in zebrafish, Danio rerio. To assess temporal expression, we first cloned four fragments of the coding region of the zebrafish CEACAM1 gene into Escherichia coli so that the CEACAM1 sequence in our strain of zebrafish could be determined. Then, real time primers and probes were designed based on this sequence. Real time PCR was performed on cDNA synthesized from mRNA isolated from embryonic and larval zebrafish at 24, 48, and 72 hours post-fertilization (hpf), and one week post-fertilization. We found a progressive increase in CEACAM1 mRNA expression over a one week period, with levels significantly higher at one week postfertilization than at 24 hpf (p < 0.05). To characterize the spatial expression of CEACAM1 in zebrafish, we synthesized sense and antisense DIG-labeled RNA probes for in situ hybridization (ISH). The results of the ISH did not reveal a tissue-specific pattern of expression, as both the sense and antisense probes bound nonspecifically in embryos and larvae.

THE CLONING AND CHARACTERIZATION OF A PUTATIVE TYPE VI SECRETED CONSERVED PROTEIN (PA0083) FROM *Pseudomonas aeruginosa*. <u>Nasira M. Rushdan<sup>1</sup></u>, William B. McVaugh<sup>1</sup>, Thomas M. Kerkering<sup>2</sup>, & Jayasimha Rao<sup>1,2</sup>, <sup>1</sup>Biomedical Sciences Department, Jefferson College of Health Sciences, Roanoke, VA 24013, <sup>2</sup>Infectious Diseases, Virginia Tech Carilion School of Medicine, Roanoke, VA 24013. Differentially expressed proteins from *Pseudomonas aeruginosa* have been identified based on a two-dimensional (2-D) gel electrophoresis analysis. The pattern was compared between two genetically similar but phenotypically distinct

*P. aeruginosa* strains, non-mucoid 383 and mucoid 2192, which were isolated from the same CF patient. In this study, a protein spot was cored based on its elevated expression pattern in the non-mucoid 383 strain. The cored spot was subjected to tandem-mass spectrometry, and the identified peptide sequences were classified as PA0083, an unknown hypothetical protein from the PAO1 genome. Bioinformatics analysis predicted that PA0083 has IcmF-associated homologous protein-related loci with a type VI secretion system. PA0083 was cloned into the 6x his-tagged expression system using the Gateway cloning method and recombinant PA0083 protein was produced in *Escherichia coli*. Studies are underway to determine whether PA0083 protein does have a role in pathogenesis.

CLONING AND CHARACTERIZATION OF PUTATIVE SECRETORY HYPOTHETICAL PROTEIN (PA0460) FROM PSEUDOMONAS AERUGINOSA CLINICAL ISOLATES FROM A CYSTIC FIBROSIS PATIENT. Joel R. Saul<sup>1</sup>, William B. McVaugh<sup>1</sup>, Thomas M. Kerkering<sup>2</sup>, & Jayasimha Rao<sup>1, 2</sup>, <sup>1</sup>Biomedical Sciences Program, Jefferson College of Health Sciences, Roanoke, VA 24013, and <sup>2</sup> Infectious Diseases, Virginia Tech Carilion School of Medicine, Roanoke, VA 24013. Pseudomonas aeruginosa is an important pathogen causing chronic lung infections in patients with cystic fibrosis (CF). Differentially expressed proteins in P. aeruginosa have been identified based on two-dimensional (2-D) gel electrophoresis analysis. The pattern was compared between two genetically similar but phenotypically distinct P. aeruginosa strains, non-mucoid 383 and mucoid 2192, which were isolated from the same CF patient. In this study, a protein spot was selected based on the expression pattern, which was seen only in the mucoid 2192 strain. The cored spot was subjected to tandem-mass spectrometry or mass mapping, and peptide sequences were classified as PA0460, an unknown hypothetical protein from the PAO1 genome. Bioinformatics analysis showed that PA0460 has a putative signal sequence with a cleavage site after 22 amino acids, suggesting that PA0460 could be a secretory protein. PA0460 was cloned into the 6x His-tag expression system using the Gateway cloning method, and recombinant PA0460 protein was produced in Escherichia coli.

EFFECTS OF HYPOXIA ON MOUSE CARDIAC MUSCLE MORPHOLOGY. <u>Quincey Garcia<sup>1</sup></u>, Lei Xi<sup>2</sup> & Kathryn E. Loesser-Casey<sup>1</sup>,<sup>1</sup> Dept. of Biol., Univ. of Mary Washington, Fredericksburg, VA, 22401 and <sup>2</sup>Dept. of Cardiology, Virginia Commonwealth Univ., Richmond, VA 23298. Systemic hypoxia (SH) can be caused naturally by high altitudes or as a result of a disease process such as sleep apnea or heart failure. Regardless of the initial cause, SH can interfere with a person's oxygen supply resulting in the cells' inability to make ATP by oxidative phosphorylation. However, studies have shown that SH may also have beneficial effects, such as a lower incidence of heart attacks. The exact mechanism of this protection has not been clearly defined and few morphologic studies have been done to study the effect of SH on cardiomyocytes. The goal of this study was to begin characterizing the effect. Three ICR mice were subjected to 2 cycles of systemic hypoxia using a normobaric plexiglass chamber with 10% oxygen. After 4 hours of hypoxia, the animals were allowed to recover for 24 hours and the cycle repeated. The hearts were perfusion fixed, embedded in wax, sectioned and stained. At least 3 sections from each of 3 control mice and the 3 SH mice were photographed and the areas and diameters of the cells were measured using Image J. The mean diameter of the cardiomyocytes decreased by 16% following SH compared to the control cells. Although there appeared to be a similar decrease in area, a Student's T-test determined that the means of the control and treated groups were not different (p=0.056). Other cells, such as neurons, have been shown to decrease in size after SH but whether the mechanism is similar in cardiomyoctes is unknown.

THE EFFECT OF GLIMEPERIDE AND GLIPIZIDE ON MYOCARDIAL PROTECTION IN STEM CELLS. Jessice R. Themak & Kathryn E. Loesser-Casey, Dept. of Biol., Univ. of Mary Washington, Fredericksburg, VA, 22401 Sulfonylureas are hypoglycemic drugs often used to treat patients suffering from diabetes mellitus. They work by binding to and blocking ATP sensitive potassium channels in  $\beta$ -cells of the pancreas thus regulating the release of insulin. These K<sup>+</sup> ATP channels are also present on the membranes of cardiomyocytes and the opening of these channels can have a cardioprotective effect. However sulfonylureas may ameliorate the beneficial effects of KATP channels openers and thus prevent myocardial preconditioning, increase infarct size, and reduce time before ischemic contracture develops. A newer sulfonylurea, glimepiride may be more effective in treating diabetes mellitus due to its lower binding affinity for KATP channels in cardiac cells which suggests that ischemic preconditioning can be maintained with pre-treatment of this drug. To further investigate the effects of glimepiride on myocardial preconditioning, brown adipose tissue-derived stem cells were pretreated with glimepiride and glipizide and then exposed to hypoxia for a period of 18 hours. The mean number of surviving cells appeared to be greater in those cells pre-treated with glipizide when compared to the control. However, statistical analysis revealed that glipizide or glimepiride had no effect on myocardial preconditioning. Further study should be conducted to look at the effects of sulfonylureas on myocardial metabolism as well as action potentials which also seem to play an important role in preconditioning.

#### **Biomedical and General Engineering**

(No Abstracts Submitted)

#### **Botany**

PHYLOGENY OF THE LEGUME GENUS *ARACHIS* USING NUCLEAR AND PLASTID SEQUENCE INFORMATION. <u>S.A. Friend<sup>1</sup></u>, D. Quandt<sup>2</sup>, & K.W. Hilu<sup>1</sup>. <sup>1</sup>Dept. of Biological Sciences, Virginia Tech., Blacksburg, VA 24061 and <sup>2</sup>Rheinische Friedrich-Wilhelms-Universität, Nees-Institut für Biodiversität der Pflanzen, Meckenheimer Allee 170, D-53115, Bonn, Germany. The peanut genus *Arachis* L. (Fabaceae) contains 80 species and is native to South America. Krapovicaks and Gregory (1994) divided *Arachis* into nine sections based on morphology, geographic distribution and cytogenetics: *Arachis, Caulorrhizae, Erectoides, Extranervosae, Heteranthae, Procumbentes, Rhizomatosae, Trierectoides*, and *Triseminatae*. The largest of these, section *Arachis*, has been further subdivided into three genomes (A, B, and D) based on cytogentics. While this genus contains the crop peanut, a

72