designed and constructed at Virginia Military Institute. We discuss the transition from laboratory testing and design to the construction of a portable working prototype currently being evaluated at the 20-inch telescope at the VMI observatory. Current results and capabilities of the instrument, as well as planned observations, will be presented

EXPLORING "THE GAME OF LIFE" IN SMALL WORLDS. Richard L. Bowman, Dept. of Physics, Bridgewater College, Bridgewater, VA 22812. While Conway's "Game of Life," an example of 2-D cellular automata, has been shown to have parallels with the biological world, researchers debate the role of boundary conditions in large universes on the patterns and behaviors observed there. This paper examines the effects of various boundary conditions on small worlds, 25 X 25 cells or smaller, and illustrates the dramatically different behavior resulting from an identical starting arrangement of cells (seed), the R-pentamino. The boundary conditions are referred to by their geometric analogs: torus, box, loop, and Möbius strip.

WHEN STEVEN HAWKING'S ALIENS ATTACK, A MATHEMATICAL MODEL. Yooryeon "Eddy" Jeon and Kristopher M. Kalish, Dept. of Mathematics and Statistics, James Madison University, Harrisonburg, VA 22807. This project is a model of a hypothetical extraterrestrial biological attack on the planet earth. The model used is a modified Kermack-McKendrick S-I-R model for infectious diseases. The model itself tracks the progression of a zombie outbreak throughout the population of the Earth. It takes into account three primary factors: susceptible humans in the population, the number of zombies created in relation to the population, and the process of reanimation. Each equation also has several factors that are used to judge the spread of infection throughout the population: a birth rate that is consistent with the population, natural death, reanimation, and contact with an infected individual. A Zombie attack presented several challenges due to the fact that there is no known natural immunity or cure, and that the members of the population who die either naturally or by contact with a zombie, will reanimate and become a zombie. The method used to solve the S-I-R differential equations is the Parker-Sochacki method, developed by Dr. Edgar Parker and Dr. James S. Sochacki of James Madison University's Dept. of Mathematics and Statistics. The software programs Maple 13 and MatLab were used to model the progression of the attack through the population once the equations were set up. This was a group project between the presenters' Math 441 class at James Madison University.

Biology (including Microbiology and Molecular Biology)

CORRELATION OF CHRONIC DISEASES WITH THE PRESENCE OF *TROPHERYMA WHIPPLEI* DNA IN SALIVA. <u>Muhammed Faizan Casim</u> & Lynn O. Lewis, Department of Biological Sciences, University of Mary Washington, Fredericksburg VA 22401. Whipple's disease is a rare systemic infection caused by the

bacterium Tropheryma whipplei. The disease is mainly known for its non-specific symptoms, including mal-absorption, diarrhea, and weight loss. T. whipplei is in the family Actinomycetes, which are environmental microbes usually found in soil and water, so a population with a poor diet and limited healthcare access could have a higher chance of having T. whipplei. In this study, we looked for the presence of T. whipplei DNA in human saliva and a correlation with any symptoms subjects might have. We tested two different populations; a population from a local free clinic with limited healthcare access and a poor diet and a population from the university with better healthcare access and a better diet available. We tested 95 subjects from the clinic and 60 students from the university. We collected saliva samples in sterile tubes and then extracted nucleic acid by using a Qiagen DNA MiniPrep extraction kit. The DNA was amplified through PCR by using Promega Master Mix with specific T. whipplei primers TW27F/TW182R. The medical history of the subjects was confidentially recorded for later use to make correlations. We found no positives in students or clinic populations therefore we were not able to make any correlation of chronic diseases with the presence of T. whipplei. Our study detected no difference in T. whipplei incidence between these two populations.

ENDOGENOUS REFERENCE GENE VALIDATION FOR ORT-PCR STUDIES ON HUMAN VISCERAL ADIPOSE TISSUE. Rohini Mehta^{1,2}, Aybike Birerdinc^{1,2}, Noreen Hossain^{2,3}, Arian Afendi^{2,3}, Vikas Chandhoke^{1,2}, Zobair Younossi^{1,2,3} & Ancha Baranova^{1,2}, ¹Molecular and Microbiology Dept. and Center for the Study of Genomics in Liver Diseases, George Mason Univ., Fairfax, VA 22030. ²Translational Research Institute, Inova Health System, Falls Church, VA 22042. ³Center for Liver Diseases, Inova Fairfax Hospital, Falls Church, VA 22042. Real-time PCR (qRT-PCR) is the standard method for studying changes in relative gene expression in different tissues and experimental conditions. However, variations in amount of starting material, enzymatic efficiency and presence of inhibitors can lead to quantification errors; therefore the need for accurate data normalization is vital. Among several known strategies for data normalization, the use of reference genes as an internal control is the most common approach. Recent studies have shown that both obesity and presence of insulin resistance influence expression of commonly used reference genes in omental fat. In this study we validated expression stability of eight selected candidate reference genes using visceral adipose samples from obese (n=10) and lean (n=9) individuals. To determine the expression stability, RNA expression levels were measured in 19 visceral adipose tissue samples and cross-validated using three popular algorithms, GeNorm, NormFinder and BestKeeper. We recommend ACTB and RPII as stable reference genes most suitable for gene expression studies of human visceral adipose tissue.

MECHANISMS OF NEUROPEPTIDE AF-INDUCED ANOREXIA IN CHICKENS. Brandon A Newmyer¹, Paul B Siegel², and Mark A Cline¹. ¹Department of Biology, Radford University, Radford, VA,24141. ²Department of Animal and Poultry Science, Virginia Tech, Blacksburg, VA, 24060. Recently, we demonstrated the anorectic effect of neuropeptide AF (NPAF) when intracerebroventricularly (ICV) administered to

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Cobb-500 chicks, which was associated with changes in hypothalamic chemistry. The purpose of the present study was to better elucidate the pathways to NPAF-induced anorexia and to test NPAF in models of hypo- and hyperphagia. In Exp 1-3, we selectively antagonized the mu, kappa, and delta subtypes of opioid receptors and coinjected NPAF in order to if NPAF'S anorectic effect was mediated through either of these subtypes; our results demonstrate that NPAF's anorectic effect is mediated through the mu and kappa but not delta subtypes of opioid receptors. In Exp 4-5, we cannulated both divisions of the paraventricular nucleus and administered microinjections of NPAF directly to these nuclei in order to determine which nucleus, if either, was the primary site of action in the NPAF pathway to anorexia. Our results demonstrated that NPAF did not affect food intake in chicks with cannulas targeting either nucleus, suggesting that neither region is the primary site of action in this pathway. In Exp 6-7, we ICV administered NPAF to chicks selected for low or high body weight and examined their food intake and behavior. We demonstrated that NPAF causes differential effects in these chicks and these effects are not associated with any behaviors competitive with appetite.

GASTRIN-RELEASING PEPTIDE CAUSES PRIMARY ANOREXIGENIC EFFECT IN CHICKENS. <u>Collette R. Dougherty</u> & Mark A. Cline, Radford Univ., Radford, VA 24012. The 27 residue gastrin-releasing peptide (GRP) is a secretagogue vital to digestion. We studied the effect of GRP on appetite. In Experiment 1, chicks that were injected intracerebroventricularly (i.c.v.) with GRP reduced both food and water intake in a dose-dependent manner. In Experiment 2, food-restricted chicks did not reduce water intake in response to i.c.v. GRP. Thus, the effect on water intake in Experiment 1 was prandial. GRP-treated chicks did not have reduced alimentary canal transit time for a gavaged red marker in Experiment 3, thus the effect in Experiment 1 may not be gut in origin. GRP-treated chicks had fewer feeding pecks. Behaviors including exploratory pecks, defecations, drinks, jumps and time spent standing, sitting, perching, and preening were not affected by GRP injection, indicating that the effect of GRP on appetite regulation is likely a primary effect. Thus, it is possible that GRP could be a sufficient appetite suppressant in humans.

DISPERSION AND ORIENTATION OF EGG CASES OF THE CHINESE PRAYING MANTIS ON WAX MYRTLE TREES. <u>Heidi Etter</u> & Robert K. Rose, Department of Biol. Sci., Old Dominion University, Norfolk, Virginia 23529-0266. The Chinese praying mantis, *Tenodera aridifolia sinensis*, was introduced to North America in 1896 near Philadelphia, Pa. A generalist insect predator inhabiting temperate successional old growth fields, this mantis is univoltine and can be iteroparous or semelparous depending on the region it inhabits. The study site for our project was a 7-year-old successional old field with volunteer wax myrtle trees located in Chesapeake, Va. One objective of the study was to determine if early laid egg cases hatch earlier than later laid egg cases. Preliminary results support this hypothesis. A second objective sought to learn if as the season progresses, egg cases are laid higher in trees and laid in a more southerly compass orientation. (An earlier study conducted

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near our study site found the majority of egg cases to be oriented in a southerly direction and often high in trees. In late autumn, a southern orientation at heights increases the body temperature of the female mantis and thus her ability to convert food into eggs.) A third objective was to determine if the mean weight of the egg cases decreases throughout the season as food becomes more limited. Preliminary results support the latter study objectives.

THE CHANGE IN SMALL-SCALE SPATIAL DISTRIBUTION OF A SMALL MAMMAL COMMUNITY THROUGH OLDFIELD SUCCESSION. Jay P. Kiser & Robert K. Rose, Dept of Biol. Sci., Old Dominion University, Norfolk, VA 23529-0266. The distribution of a community of small mammals was evaluated during the succession of an oldfield habitat into a young pine forest, From 2002 through 2009 a capture-mark-release (CMR) study was conducted on a one-hectare grid at the Su Tract of Chesapeake Virginia. Over this time the small mammal community had variably consisted of cotton rats (Sigmodon hispidus), meadow voles (Microtus pennsylvanicus), marsh rice rats (Oryzomys palustris), eastern harvest mice (Reithrodontomys humulis), house mice (Mus musculus) and, towards the end, golden mice (Ochrotomys nuttalli). During the course of the CMR study, the change in succession was defined by the presence and density of the loblolly pine trees (Pinus taeda) found throughout the study area. Measurements of all the loblolly pines were taken in the winters of 2005 and 2008 and used to calculate the living basal areas throughout the grid. Associations were examined through regression analysis of the basal areas and captures of each small mammal species. House mice and meadow voles were found to be significantly negatively correlated with the living basal area, whereas rice rats showed no correlation. Cotton rats and harvest mice were significantly positively correlated with basal area up to a moderate level of pine growth and then negatively correlated when pines were taller.

GROWTH AND SURVIVAL OF VOLUNTEER LOBLOLLY PINE (*PINUS TAEDA*) TREES IN AN OLD FIELD IN EASTERN VIRGINIA. <u>Robyn M. Nadolny</u> & Robert K. Rose, Department of Biol. Sci., Old Dominion Univ., Norfolk VA 23529-0266. Old field succession to mixed hardwood forest is often prefaced by the invasion of open spaces by fast-growing loblolly pines (*Pinus taeda*). We examined growth rates, sources of mortality and demographics of volunteer loblolly pine trees within a 1.23 ha oldfield study grid in Chesapeake, Virginia. In the winter of 2005 we learned that 15.1% of 15,675 pine trees in a 5-year-old oldfield had been killed by girdling, and a further 50.0% partially girdled, by a high density of hispid cotton rats (*Sigmodon hispidus*). Three years later we examined sources of mortality after the rodent population had dropped to near zero. During the winter of 2008 the 7-year-old pine forest lost 138 trees (0.8%) to girdling but 23.0% (3,846 stems) to natural mortality, among 16,766 stems >0.8 m tall on a 1.23 ha grid. We measured the trees again in the winter of 2009-2010 to examine the rates of growth (basal area) from period to period, and to evaluate the effects of stem density on growth and survival. We conclude that

there was a drastic shift in mortality source between 2005 and 2008-10, and that high tree density predicts higher rates of mortality in all stages of succession.

INIHIBTION OF METHACILLIN RESISTANT STAPHLYCOCCUS AUREUS (MRSA) BY UNIQUELY SYNTHESIZED IMIDAZOLE BASED ANTIMICROBIALS. Caitlin J. Hubbard, Randall D. Hubbard, & A. Garth McGibbon, Dept. Of Biology and Chemisty, Liberty University, Lynchburg VA. 24502. Substituted benzene rings containing a primary amine were selected. Diazitization of the amine was carried out using NaNO₃ at low pH and -10°C. This allowed for the formation of an unstable azynyl intermediate. Imidazole, an aromatic nitrogen containing, five membered ring, was subject to deprotonation at N1 at pH 14.5 using 50/50 base. Resonance created a nucleophile on the 2C, which in turn attacked the benzene azynyl group; this resonates to create an azo bond between the two ring systems. Azo coupling completion was visualized by color change due to change in absorbency properties; acidification of the solution led to protonation and precipitation of the product, which was repeatedly recrystalized to ensure purity. Compounds synthesized were (E)-(2-chlorophenyl)-(1H-imidazol-2-yl)diazene, (E)-(3chlorophenyl)-(1H-imidazol-2-yl)diazene, (E)-(4-chlorophenyl)-(1H-imidazol-2yl)diazene, 4-[(E)-1H-imidazol-2-ylazo]phenol, and (E)-1H-imidazol-2-yl-(4methoxyphenyl)diazene. Compounds were dissolved in 10% DMSO with 5% acetone. Kirby-Bauer disk diffusion technique was used to evaluate compound inhibition against community and clinically acquired MRSA strains. Compounds 2 & 5 were most effective for clinical MRSA strain, while compounds 1 & 5 were most effective for community MRSA strain. The addition and placement of large functional groups to the benzene ring increases the MRSA inhibitory properties of the imidazole derivatives.

THE ROLE OF A DEFENSIN PROTEIN IN ARBUSCULAR MYCORRHIZAL DEVELOPMENT. <u>Barbara B. Kreutzer</u>, Oscar R. Rocha & Rajendra P. Kandel, Dept. of Biol. and Phys. Sci., Marymount University, Arlington, VA 22207. Defensin proteins are found in many organisms and act as an efficient part of innate immune antifungal response. We hypothesized that some herbaceous plants express a brassica defensin protein which prevents symbiosis with mycorrhizal fungi. The defensin gene was found in nonhosts but not in hosts. SDS-PAGE indicates the gene was expressed in *Brassica rapa* exposed and not exposed to AM inoculum. Implications for AM development and plant and animal pathogenesis were discussed.

THE ISOLATION AND IDENTIFICATION OF A NEW *CAENORHABDITIS* SPECIES TO BE USED IN COMPARATIVE GENOMICS. <u>Ann Wang</u>, Erika Baardsen, & Theresa Grana. University of Mary Washington.

Nematodes are the most abundant multicellular organisms on the planet. To date, the most important nematode species to the scientific community is *Caenorhabditis*

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elegans. C. elegans is commonly used in developmental, neurobiological, and genetics research because it has a defined cell lineage, is easily maintained in a laboratory, and reproduces quickly. It was also the first multicellular organism to have its genome sequenced. Many researchers have benefited from comparative studies between C. elegans and C. briggsae. These species share many of the same characteristics, however, there are important morphological differences between the two nematodes. Researchers now agree that a more closely related species that exhibits a greater degree of morphological and molecular similarity will be isolated. The discovery of a new sister species would be beneficial to the C. elegans research community. Four nematode species have been isolated from Fredericksburg, Virginia. 4D imaging suggests that their internal structures differ from C. elegans. However, they also demonstrate some morphological and behavioral similarity to C. elegans. The 1A, 6A, 4D, and 2G species have been sequenced and aligned with the C. elegans 18S ribosomal sequence. These sequences differ significantly, leading us to conclude that these species are not sister species of C. elegans and would be of little use in comparative genomic studies for this project.

ANNOTATION OF THE TERMITE METATRANSCRIPTOME. Neerja Katiyar², Natalie Fedorova¹, Alan Lax¹ Gennady Denisov² & William C. Nierman³, George Mason University², Fairfax, Virginia, 22030 and J. Craig Venter Institute^{1,2,3}, Rockville, Maryland, 20850. The sequencing of mRNA transcripts from the Formosan subterranean termite, Coptotermes formosanus, resulted in 131,637 Sanger reads and 6,942,682 Illumina reads. Sanger reads were assembled into 25,939 unigenes, which represent up to 60% of the species transcriptome. Phylogenetic analysis showed that over 50% of unigenes exhibited no sequence similarity to other proteins or PFAM and TIGRFAM domains and may, thus, represent termite-species specific genes or noncoding RNAs. Fifty percent of the other unigenes shared similarity with other insect genomes and 25% with Trichomonada, suggesting the presence of protozoan endosymbionts (parabasalids). Pathway analysis suggested that the majority of carbohydrate, glycan, and xenobiotic pathways in the Metabiome were contributed by endosymbiotic parabasalids. In addition, the CAZy system was used to classify enzymes involved in the carbohydrate metabolism. Expression levels of the 20% most abundant transcripts from one termite cast, Cf4, were captured using RNA-Seq technology. The most abundant unigenes included two Drosophila homologs: Tequila, which is essential for information processing in, and myofilin, which is required for filament assembly in all muscles.

CARDIAC STEM CELLS: MIGRATION ASSAY UTILIZING VEGF AND STATINS. <u>Stacey Rickard</u> & Kathryn E. Loesser-Casey, Dept. of Biol., Univ. of Mary Washington, Fredericksburg, VA, 22401. Conventional treatments of heart disease do not result in myocardial restoration. However, recently discovered cardiac stem cells have been found to proliferate and differentiate into functional cardiomyocytes, giving researchers hope for use in treatment of heart disease. Understanding the factors that

influence the migration of these cardiac stem cells is paramount and therefore, my study examined the in vitro chemotaxic capabilities of 50 ng/ml VEGF and 0.1 µM rosuvastatin as well as the effect of age on cardiac stem cell migration. Cardiac stem cells were isolated from the hearts of newborn, 6-7 week old and 27 week old Sprague Dawley rats. Migration assays were performed on cells cultured from the newborn and 6-7 week old rats. Cell morphology indicated that both of the younger two age groups of rat cells appeared to be cardiac stem cells but the 27 week old rat cells appeared to be adipocytes. This indicates that the younger cardiac stem cells may be better suited for myocardial regeneration. The data from the migration assays produced no significant results when comparing the migration factors used, however, the newborn rat cells had significantly less migrated cells compared with the 6-7 week old rats. However, there was a second, unintentional variable. This study found that at the tested concentrations, VEGF and rosuvastatin did not induce cardiac stem cell migration. This study also supports the idea that younger cardiac stem cells are better suited to proliferate and differentiate into cardiomyocytes but age impacting the migration of cardiac stem cells cannot be concluded.

AGE-RELATED CHANGES IN STEM CELL MARKERS. Dana Hunt & Kathryn E. Loesser-Casey, Dept. of Biol., Univ. of Mary Washington, Fredericksburg, VA, 22401 The use of stem cells to promote endogenous repair of cardiac tissue provides hope for millions of Americans suffering from cardiovascular disease. Adipose tissue may provide an easy, noncontroversial supply of stem cells. However, the origin of the fat, culturing conditions, and media formulations can influence the proliferation rate and differentiation capacity of these stem cells. Mesenchymal stem cells are reported to express CD105 and lack the hematopoietic lineage markers including CD34. However, studies have demonstrated that CD34 shows high levels of expression early in passage. This study was conducted to examine whether CD34 and CD105 expression varied due to location of fat and/or the number of days the cells were cultured. Brown fat was isolated from male laboratory mice from two locations, between the scapulae and in the groin. Isolated cells were grown on slides and CD34 and CD105 were localized using immunocytochemistry. The total number of positive cells were counted and a one-way ANOVA was performed.. CD105-positive cells were found to be statistically more abundant in adipose isolated from both between the scapulae and in the groin than CD34 labeled cells or control. Control slides (no antibody) did however, show some staining indicating that we need to repeat the experiments to reduce nonspecific binding of the antibodies. Our study supports the loss of CD34 markers with cell age and the presence of CD105-positive stem cells in adipose from several locations in the mouse.

Heat Shock Protein 60 Detection by Enzyme-Linked Immunosorbent Assay in House Sparrows (*Passer domesticus*). <u>W. Humayon</u>, A. Dolby, & D. O'Dell, Dept. of Biol., Univ. of Mary Washington., Fredericksburg, VA 22401. Stress is caused by everchanging environmental conditions that affect every organism. The stress response, which includes both release of stress hormones and production of heat shock proteins

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(HSPs), protects animals from such biological challenges. Furthermore, not only are HSPs being applied to avian stress research on a limited basis, methods currently being employed by ornithologists to measure them do not allow them to be precisely quantified. These methods, based on colorimetric protein detection, only permit subjective comparisons to be made among samples. The objective of this project was to determine the efficacy of an alternative colorimetric HSP60 detection method that allows objective quantification, which would allow more meaningful analyses to be carried out of the factors that contribute to stress. Blood samples were collected from 16 House Sparrows at the University of Mary Washington campus during the spring of 2008 and 2009. Indirect and trap Enzyme-linked immunosorbent assays (ELISA) were used to measure HSP60 protein levels in them. Both methods detected HSP 60 proteins in the samples, but the trap ELISA was found to be more sensitive and showed less variability than the indirect ELISA. Funds were provided by Mrs. Thyra Valade Memorial Fund and UMW Undergraduate Research Fund. Thanks to V. Zimmermann, K. McAndrew & A. Dougherty for collecting samples.

BISPHENOL A INTERACTS WITH ESTROGEN RELATED RECEPTOR GAMMA TO REGULATE PRODUCTION OF C-FOS AND PS2 GENE PRODUCTS. Shannon Tucker. Department of Biology, University of Mary Washington, Fredericksburg, VA 22401. Bisphenol A (BPA) has been linked to breast carcinoma in humans for over 20 years, yet the mechanism by which BPA causes breast cancer has yet to be determined. We propose a novel mechanism by which bisphenol A acts through the estrogen related receptor γ (ERR γ), to upregulate potentially cancerous proto-oncogenes such as pS2 and c-fos. Our results show that blocking either the estrogen receptors (ER) or ERR γ , (by use of fulvestrant and 4-hydroxytamoxifin, respectively) in human breast cells reduces the levels of c-fos protein and pS2 in cells exposed to carcinogenic levels of BPA, with the greatest reduction occurring in the ERR γ blocked cells. Moreover, when both of these receptors are blocked, our results showed even lower amounts of c-fos (72.4% lower than control) and levels of pS2 proteins were undetectable. With further research, these results could finally explain the positive correlation between levels of BPA and breast cancer.

Biomedical and General Engineering

LACK OF EFFECT ON CELL-MEDIATED IMMUNITY FOLLOWING IN VIVO EXPOSURE TO ELECTROSPUN POLYCAPROLACTONE. C.E. McLoughlin1, M.J. Smith2, G.L. Bowlin1, & K.L. White, Jr.2 1Department of Biomedical Engineering, Virginia Commonwealth University, Richmond, VA 23284, 2Department of Pharmacology and Toxicology, Virginia Commonwealth University, Richmond, VA 23284. Studies in our laboratory have focused on the effects on the immune system following in vivo exposure to electrospun polycaprolactone (EPCL). We are investigating the role of fiber diameter of EPCL, specifically comparing EPCL with average fiber diameter of 1.7um ("microfibrous") compared to 225nm ("nanofibrous").