Abstracts of the

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Southwestern Oklahoma State University - Weatherford

PSEUDOMONAS SYRINGAE MOTILITY APPENDAGES: ROLES IN SURFACE COLONIZATION AND SURVIVAL

Jennifer L. Absire and Regina S. McGrane, Southwestern Oklahoma State University

Outstanding Undergraduate Poster

Pseudomonas syringae is a bacterial phytopathogen that causes disease in economically important crops. P. syringae is found in many different environments, each with varying levels of moisture. Therefore, the appendage it relies on for motility varies. The removal of genes encoding for these motility factors significantly impairs pathogenicity. The objective of this project was to evaluate the impact of P. syringae motility related appendages on colonization of varying environments. To characterize the role of flagella and pili, movement of deletion mutants was observed in saturated, liquid, and natural surface environments. Flagella mutants had significantly reduced motility in both saturated and liquid conditions, whereas pili mutants had significantly reduced motility in saturated conditions only. This indicates that flagella are the most relied upon appendage for motility and that pili have minor impacts. Similarly, on simulated natural surfaces flagella were the primary movement appendage. To characterize the impact of flagella and pili on colonization of plants, attachment to leaves and seeds as well as growth on leaves were evaluated. Mutants lacking flagella or both flagella and pili were impaired in leaf colonization but showed seed attachment similar to the wild type. However, mutants lacking both the flagella and pili had greater levels of attachment to leaves. To characterize the impact of flagella and pili on antibiotic sensitivity, mutants and the wild type were evaluated using a Kirby-Bauer sensitivity assay. Mutants were more sensitive to ciprofloxacin than the wild type, which is opposite to observations made in a different P. syringae strain. These results indicate that removal of the genes encoding for motility factors such as pili and flagella, directly impacts pathogenicity. Limiting the motility of P. syringae could halt or decrease the symptoms associated with this disease-causing pathogen and increase crop yields of host plants.

IOT SEMI-TRUCK MONITORING & SAFETY SYSTEMS

Mohamed Afify, Mohamed Keblwei, and Nesreen Alsbou, University of Central Oklahoma

Outstanding Undergraduate Paper in Engineering Science Section

The Trucks accidents are deadly and cost the companies a lot of money each year. We are designing a system to prevent the Truck accidents by design a mechanical device to enhance the safety system of the trucks by implementing a blind spot system and LIDAR system to detect the obstacles and alert the driver. We also installing a system that can detect the sudden braking of the vehicle in front of the truck to except any distance a few seconds before. We designing a medical vest also to give readings of the driver health conditions to prevent accidents as heart attacks or the driver falling in sleep.

ASSESSMENT OF IRON OXIDE NANOPARTICLES AS CHEMICAL TRACERS IN OKLAHOMA GROUNDWATER

Kelsey H. Anderson, Brian Bigelow, Rabeka Lashbrook, and Randall D. Maples, East Central University

It is important to be able to assess the risks of contamination of groundwater associated with the aquifer recharge process. Nanoparticles such as magnetite (Fe3O4) may offer several advantages over traditional chemical tracers including stability in undesirable conditions, detection at low concentrations, and ability to functionalize to suit a variety of different uses. This study begins assessing the potential of iron oxide nanoparticles as chemical tracers in groundwater using columns packed with glass beads, limestone, sandstone and dolostone from the local area and used water from local sources including tap water, creek water and the Arbuckle-Simpson aquifer as small scale environmental simulations. We then measured the recovery of the nanoparticles after flowing through the columns.

USING GROUNDWATER FAUNA TO DETERMINE SUBTERRANEAN HYDRAULIC CONNECTIONS IN KARSTIC AQUIFERS: A CASE STUDY FROM THE ARBUCKLE MOUNTAINS

Kevin Blackwood, Justin Harris, Laramie Edens, Kay Woodring, and Stacy Gantt-Blackwood, East Central University

Geology plays a significant role in both the storage and transport of groundwater, as well as the distribution and diversification of subterranean and aquatic organisms. The effect that stratigraphic and structural features might have on hydrogeology is by acting as either conduits or barriers to groundwater flow. Determining whether hydrologic connections exist between two terranes can be difficult, especially when time and hydraulic gradients might not be sufficient for the injection of chemical tracers or pumping tests might not be practical. By using endemic groundwater fauna (such as cave amphipods) as biological tracers, hydrogeological parameters may be deduced by comparing genotypic and phenotypic relationships between populations on either side of potential groundwater barriers due to allopatric isolation. Cave amphipods and groundwater isopods both occur in relative abundance within the study area, but cave amphipods are the most reliable for determining groundwater connections due to their restriction to the phreatic environment. Groundwater isopods have been documented traversing the vadose environment over films of water and may be capable of navigating around potential groundwater barriers. Therefore, groundwater isopods may be unreliable as biological tracers. Looking at examples from the Arbuckle Mountains of southern Oklahoma, we examine genotypic and phenotypic relationships of endemic groundwater fauna, using DNA barcoding and taxonomic methods. By employing these techniques, we can assess the effectiveness of various types of stratigraphic and structural features as either pathways or as barriers in a region heavily altered by structural deformation and modified by karst processes.

EVALUATION OF DNA EXTRACTION PROTOCOLS FUNCTIONALITY FROM DRIED TESTUDINE BONE

Mariah Ewy, Alisha Howard, and Kenneth Andrews, East Central University

There are numerous papers written about DNA extraction out of bones and animal tissue. The variety of DNA extraction protocols and the lack of performance consistency across different organisms argues for a lack of robustness. A total of 3 different protocols have been evaluated attempting to extract DNA from dried testudine bone, or shell. The first evaluated protocol is the Armed Forces DNA Identification Laboratory (AFDIL) protocol. This protocol was written to obtain DNA from skeletal remains for identification. The second evaluated protocol was written by Cold Spring Harbor Laboratory and titled Using DNA Barcodes to Identify and Classify Living Things. This protocol was created to isolate DNA from plant, fungal, or animal samples. The third protocol evaluated was the Nature Protocol titled Ancient DNA Extraction from Bones and Teeth. The goal of this protocol is to maximize recovery of PCR-amplifiable DNA from bone and teeth, while limiting the amount of contaminants that can inhibit PCR. All protocols were altered slightly to fit the resources available in the lab where the experiments were conducted. Each extraction protocol was unique in its methodology. Polymerase Chain Reaction (PCR) was performed with all DNA extraction products to amplify specific single segments of DNA.

HEIRLOOM MICROBES: THE HISTORY AND LEGACY OF ANCIENT DAIRYING BACTERIA

Shannon Fulton and Paul Lawson, University of Oklahoma

Soninkhishig Tsolmon, Mongolian Dietetic Association

Christina Warinner, Jessica Hendy, Matthäus Rest, Sanjeet Kumar, and Bjorn Reichhardt, Max Planck Institute for the Science of Human History

Human communities have utilized microbes for thousands of years as exemplified by the development of dairy products such as yoghurts and cheeses. The adoption of dairy foods into the adult human diet, and its consequent effect on the human genome, is a clear example of gene-culture co-evolution. The mechanisms of this process are not well understood, although we know that milk must have been heavily processed in order to create digestible products, with microbes likely playing a key role. Modern dairy production methods use microbial strains that are highly regulated in order to maintain hygiene standards and reproducibility. Subsequently, we remain unaware of the vast microbial diversity involved in ancient food preparation, and the impact this microbial diversity may have had on flavors and textures. With food globalization and industrialization, traditional methods of dairying and their unique microbial cultures are now being rapidly lost. It is known that lactic acid bacteria (LAB) are primarily responsible for the natural fermentation of animal milk and its by-products. Therefore, this study focuses on the screening of LAB in traditional dairy products from Central Asia (Mongolia) using culture dependent isolation of representation organisms to explore their genetic identification and deposit in microbial culture collections. Various dairy products, including raw and boiled animal milk, were collected from nomadic herding communities in Mongolia. A series of enrichments were designed to enable characterization of the LAB communities of the cow and yak derived Mongolian dairy samples. Two growth mediums, MRS and M17, containing different carbohydrate sources and nutritional components were selected for incubation in a reduced oxygen environment at 27.9°C and 45°C in order to select for mesophilic and thermophilic LAB, particularly Streptococcus thermophilus. Species belonging to the genera Lactobacillus, Streptococcus, Enterococcus, Lactococcus, Pediococcus, Leuconostoc, Weissella, Carnobacterium, Gluconobacter, Lelliottia, Bacillus, Anoxybacillus and Brevibacillus were recovered.

EXTRACTION AND CHARACTERIZATION OF DIHYDROXYACETONE FROM SUGAR CANE

William Gathright and Amanda J. Nichols, Oklahoma Christian University

Best Undergraduate Paper of the Academy and Outstanding Undergraduate Paper in Physical Sciences Section

Dihydroxyacetone is a chemical interest because of its ability to react with the amino acids in the outer layer of skin, giving the skin a bronzed look. As a result of its behavior, dihydroxyacetone is the typical active ingredient in self-tanners. Industry usually synthetically makes the compound, but it is naturally found in sugar beet and sugar cane. No published extraction procedure for dihydroxyacetone from sugar cane or sugar beets could be found. Preliminary methods were developed in order to isolate and characterize dihydroxyacetone from sugar cane and commercial sugar beet deer feed. Reflux boiling in ethanol was used as the extraction technique. Characterization techniques included high-performance liquid chromatography (HPLC) analysis and Fourier-transform infrared spectroscopy (FTIR) analysis. A preliminary characterization method was developed that used gelatin to mimic the self-tanning process on skin. Color changes of the gelatin 'skin' was observed between the extracted dihydroxyacetone and commercial self-tanners. One research area of chemical education focuses on the development of experiments appropriate for an undergraduate laboratory. These experimental methods can be used in a personal care products-themed lab component of an introductory chemistry course.

UNDERSTANDING GENE EXPRESSION REGULATION THROUGH CHARACTERIZATION OF TRANSCRIPTION START SITES IN *DROSOPHILA ELEGANS*

Amy Giemza, Sidney Wilkins and Lindsey J. Long, Oklahoma Christian University

The Genomics Education Partnership (GEP), Washington University in St. Louis

Outstanding Undergraduate Paper in Biological Science-Zoology Section

Every living organism is composed of genes. The position of these genes on a chromosome is important for proper regulation of gene expression. Alteration of positioning or gene regulatory elements can lead to diseases such as cancer. Understanding the mechanisms of gene regulation can help explain why genes are misregulated in diseases and, furthermore, how to abrogate this misregulation. Because highly important DNA sequences are often conserved throughout evolution, our strategy was to identify gene regulatory elements in various species related to Drosophila melanogaster as the genome of this species is fully annotated. Transcription start sites (TSSs) are where transcription of DNA into RNA begins which ultimately leads to the production of proteins. Our specific research focused on identifying TSS positions for several genes and compiling the data to distinguish trends for the regulatory elements that surround the TSS of genes that are classically "on" or "off." To pinpoint the location of the putative TSS for the D. melanogaster unc-13 ortholog in D. elegans, we aligned the D. melanogaster unc-13 sequence to the D. elegans genome to search for homology. The identified search region was further scrutinized using lines of evidence derived from experimentation to identify the exact location of the TSS. Core promoter motifs in the region were also evaluated to investigate the possibility of alternative TSSs. Of particular interest, unc-13 had low sequence homology with the D. melanogaster ortholog and supplementary evidence was unsuccessful in difinitively identifying the TSS for all isoforms of the gene. Ultimately, the data supported that only a few of the isoforms of unc-13 were conserved in the evolution of this species.

MNIST DATASET ANALYSIS

Reid Kinder, East Central University

Outstanding Undergraduate Paper in Math, Statistics, & Computer Science Section

Teaching a computer to classify data accurately through multi-layer neural network processing is known as deep learning. The MNIST dataset was used to explore and compare machine learning processes to deep learning through packages such as SKLearn, and Tensorflow. Through SKLearn, different dimensional reduction techniques were used to manipulate the dataset, such as Principal Component Analysis (PCA) and T-Distributed Stochastic Neighbor Embedding (t-SNE). PCA and t-SNE were used to reduce the number of dimensions of the dataset, while conserving certain characteristics of the data. Finally, K-Nearest Neighbors (KNN) was used to classify the data after dimensional reduction. After this classification, a graphical representation of the data was presented. An accuracy greater than 85\% on the test set was achieved through this method. Tensorflow was also successfully applied to the data set. Through Tensorflow, we reached a result of greater than 95\%.

CLONING AND EXPRESSION OF *CHLAMYDIA TRACHOMATIS* INCLUSION MEMBRANE PROTEINS

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Outstanding Graduate Poster

Chlamydia trachomatis is an obligate intracellular human pathogen that resides inside host cells within a parasitophorous vacuole called an inclusion. In order to replicate and grow Chlamydia must usurp host cell proteins from within this vacuole. To do this, Chlamydia produce and secrete proteins, termed inclusion membrane proteins (Incs), that insert into the inclusion membrane with the N- and C- terminus facing the host cytosol. Currently, C. trachomatis is predicted to have 50 Incs, however very few of these have known functions. Very little knowledge about their function can be gained via bioinformatics analysis since they lack similarity to any proteins outside of *Chlamydia*. This has made characterizing Incs or identifying possible functions very difficult. The goal of this project is to clone and express the C-terminus of certain Incs which will be used to produce purified protein for future crystallography studies. This study focuses on the CT229-CT224 operon which is only found in human pathogens. Here, we present the cloning strategy of each Inc into the expression plasmid pET28a which will generate a C-terminus Inc fusion to a 6X His tag. To date CT226, CT227 and CT228 have been successfully cloned, verified by sequencing, and transformed into BL21 for expression studies. Protein production with these constructs has been induced with IPTG. Progress with the CT226 construct has proceeded to successful solubilization after expression, and re-folding is evident from the defined elution from size exclusion chromatography as an apparent trimer, in addition to strong amide bond signals in Fourier-transform infrared spectroscopy indicating approximately half of the protein adopts a helical conformation. By assessing their structures, insights may be gained as to possible functions based on similarity to other characterized proteins.

OKN RELIEVES OXIDATIVE STRESS INDUCED BY SEPSIS-ASSOCIATED ENCEPHALOPATHY

Tyler McKenzie, Redlands Community College,

Debra Saunders, Nataliya Smith, Shania Do, William Towler, Marvin Cruz, and Rheal Towner, Oklahoma Medical Research Foundation

Introduction: Sepsis-associated encephalopathy (SAE) is an oxidative stress-related disease of the brain caused by the introduction of bacteria, and it causes an increased permeability of the blood brain barrier (BBB). Neuroinflammation can lead to cognitive impairment which is linked with age. Methods: Lipopolysaccharide (LPS) induced rat models were used to compare OKN treated, untreated, and saline injected negative controls. OKN is a spin trapping compound for reactive oxygen and nitrogen species that prevents further damage to tissue from free radicals. Various Magnetic Resonance Imaging (MRI) techniques such as contrast enhancement, MRI perfusion, and MR spectroscopy were used to scan the brain for signs of a compromised BBB, vascular alterations, and metabolite changes, respectively. Results: MRI contrast enhancement showed that the OKN treatment was lowering MRI intensity within the cortex, hippocampus, thalamus, and peri-rhinal cortex. LPS-injected untreated rats had a greater MRI intensity post-contrast injection for week one. It was observed with MR spectroscopy that metabolites within the brain had also stabilized with the OKN treatment indicating the reduction of damaged tissue. Low perfusion rates were examined during post-LPS injection of both week one and three, which signaled constriction of the vessels within the brain. Conclusion: We can conclude the BBB and vascularity is negatively affected longterm by LPS. SAE promotes neuroinflammation which affects BBB permeability, vasoconstriction, and decreased brain metabolites. OKN proved to be an effective treatment with the rat models for minimizing the effects of SAE within the brain. Funding: OK-INBRE and NIH RO1NS092454.

STRUCTURE OF POXVIRUS A6 PROTEIN REVEALS A MECHANISM FOR STABILIZING OPEN-ENDED CRESCENT MEMBRANE

Prabhat Kumar Pathak, Shuxia Peng, and Junpeng Deng, Oklahoma State University

Best Graduate Paper of the Academy

Cellular membranes are maintained as closed compartments, broken up only transiently during membrane reorganization or lipid transportation. However, open-ended membranes, likely derived from scissions of the endoplasmic reticulum, persist in vaccinia virus-infected cells during the assembly of the viral envelope. A group of viral membrane assembly proteins (VMAPs) were identified as essential for this process. To understand the mechanism of VMAPs, we determined the 2.2-Å crystal structure of the largest member, named A6, which is a soluble protein with two distinct domains. The structure of A6 displays a novel protein fold composed mainly of alpha helices. The larger C-terminal domain forms a unique cage that encloses multiple glycerophospholipids with a lipid bilayer-like configuration. The smaller N-terminal domain does not bind lipid but negatively affects lipid binding by A6. Mutations of key hydrophobic residues lining the lipid-binding cage disrupt lipid binding and abolish viral replication. Our results reveal a protein modality for enclosing the lipid bilayer and provide molecular insight into a viral machinery involved in generating and/or stabilizing open-ended membranes.

TIMING OF AVIAN MIGRATION ONSET THROUGH THE OKLAHOMA CITY AREA 1995-2017 USING NOAA WEATHER DATA

Jennifer Prophet and Zach Jones, Southwestern Oklahoma State University

Outstanding Undergraduate Paper in Applied Ecology and Conservation Section

Timing and duration of avian migration patterns may be linked to large-scale climate patterns and reflect long-term shifts in average regional temperatures. The National Centers for Environmental Information (NCEI) provides a publicly-available archive of National Oceanic and Atmospheric Administration (NOAA) weather radar data scans occurring every ten minutes dating back to 1995. Our primary data of interest are 1) the measure of reflectivity: the amount of power returning to a radar after hitting water, and 2) radial velocity: the movement of water relative to radar position. Aerial density and flight direction will be collected annually and compared to global and regional temperatures to determine effects on seasonal migrations. For the current study, we used KTLX (Oklahoma City) weather radar data to determine time, duration, intensity and direction of fall avian migration. Our analysis currently includes data from 1995-2017, with the fall migrations beginning in September and the spring migrations beginning in March. The global average temperature change over this time range was 2.358°C. More data will be analyzed within this range in order to develop more accurate results regarding the effect of temperature change on fall and spring migratory onset. In addition to migratory information, we have observed fine lines of insects caught in outflow air masses as well as regular sunrise takeoffs of waterfowl overwintering near Lake Hefner. We also have encountered radar signals caused by wind turbines, but this has not noticeably affected avian flight behavior or our data collection on the scale and altitudes being sampled.

COMPUTATIONAL STUDY OF VOLATILE ALUMINUM HYDROXIDE

Uendi Pustina and Dwight L. Myers, East Central University

Reactivity and compatibility of oxides with other materials and with each other plays a significant role in choice of materials for developing Thermal Barrier Coatings (TBCs) or Environmental Barrier Coatings (EBCs) for use in combustion environments. We are performing a computational study of the gas phase molecule aluminum hydroxide. The ultimate goal of this study is to obtain a reliable value of the enthalpy of formation of aluminum(III) hydroxide. The software we are using is the GAMESS ab initio package. Presently we are to the stage of optimizing the geometry of the molecule. Results to date will be presented.

EFFECT OF STORAGE TEMPERATURE ON BIOACTIVITY OF A COMMERCIAL SACCHAROMYCES BOULARDII PROBIOTIC FORMULATION

Reid Reding and Jonathan Hunt, Oklahoma Christian University

Outstanding Undergraduate Paper in Microbiology Section

The commercial probiotic industry is rapidly expanding and gaining traction among the common population. Many people see advertisements for probiotics making claims that they will contribute to better overall health and potentially help restore health after dealing with gastrointestinal issues, etc. Probiotics have helped many infections such as *C. difficile* and ulcerative colitis by restoring the gut microbiota after antibiotics have killed many of the beneficial microorganisms. Probiotics do this by competing with the pathogen for resources necessary to sustain life and by interfering with important metabolic pathway steps. Furthermore, the probiotics promote a healthy gut flora in general, which helps to eliminate symptoms caused by pathogenic species. Many manufacturing companies state that the probiotics need to be stored in a refrigerated area to maintain their efficacy; however, often times consumers forget to keep them in a refrigerator. The experiment at hand measured the viability of a commercial preparation of *Saccharomyces boulardii* probiotic under varied storage conditions (4°C, 23°C, and 30°C) once a week over a four-week period to assess the necessity of specific storage conditions.

RAD4 IS REQUIRED FOR MAINTENANCE OF DNA INTEGRITY

Brandon Reed, Whitney Bohannan, and Lindsey Long, Oklahoma Christian University

Outstanding Undergraduate Paper in Biomedical Science Section

UV radiation exposure can cause bulky adducts (such as cross-linked thymine bases) in DNA. The cell uses the Nucleotide Excision Repair (NER) pathway as the primary mechanism to repair these adducts. Normally, these adducts are repaired; however, when not repaired, these adducts lead to diseases such as xeroderma pigmentosum and an increased risk of skin cancer. In S. cerevisiae, RAD4 encodes for protein that is a component of the NER pathway. Rad4 forms a heterodimer complex with Rad23 to form Nuclear Excision Repair Factor 2 (NEF2), which is necessary for the recognition of the damaged bases. After DNA damage recognition, the remaining repair machinery is recruited to remove the bulky adduct and restore the DNA integrity. It has been shown that deletion of the RAD4 gene in S. cerevisiae leads to increased UV sensitivity when compared to the wild type (wt) strain, due to the inability of the machinery to recognize and repair lesions caused by UV radiation. We also showed that the Δrad4 strain showed significantly lower viability after UV radiation exposure when compared to the wt strain that was exposed to the same UV dosage. To investigate as to whether this reduction in viability was related to unrepaired DNA mutations, we used a reverse mutation canavanine assay to calculate DNA mutation frequency. Δrad4 cells showed a higher DNA mutation frequency when compared to the wild type strain. These results indicate that RAD4 is critical for DNA damage repair after UV exposure, and failure to repair DNA damage results in DNA mutation and cell death.

BACTERIOPHAGE--A POTENTIAL REPLACEMENT FOR ANTIBIOTICS

Abbey Renner and Madison Snow, Oklahoma City University

Bacteriophage ("phage") are viruses that infect and replicate in bacteria. Since phage infect bacteria very specifically, there is considerable interest in exploiting them as antibacterial agents. Phage are found in natural environments such as soil, where there are complex bacterial ecosystems. We extracted phage from nutrient-rich soil by mixing the soil with a buffer (SM buffer), and filtering out the large particles and living organisms using a 0.45 micron syringe filtration system. Phage were added to cultured bacteria (*Bacillus cereus* or *Serratia marcesens*), mixed with top agar, and plated on LB-media. After incubation, the plates were checked for zones of clearing ("plaques"), indicating the presence of specific bacteriophage.

CLONING, SEQUENCING, AND IDENTIFICATION OF PHAGE P13, AN UNKNOWN SALMONELLA OR EHEC (ENTEROHEMORRHAGIC E. COLI) BACTERIOPHAGE

Ryan Sloan, W.J. Reddig, and Earl L. Blewett, Oklahoma State University-Center for Health Sciences

Divya Jaroni, Oklahoma State University-College of Agriculture

Bacteriophage have been isolated from the environment that specifically infect bacteria responsible for food poisoning. These bacterial viruses kill *Salmonella* and Enterohemorrhagic *E. coli* and may be useful in the food industry to reduce bacterial food contamination. We cloned and sequenced fragments of genomic DNA from one of these bacteriophage, P13. Bioinformatic analyses showed this bacterial virus was most similar to phage from the *Yersinia* genus and not that similar to phage from the *Salmonella* group.

USE OF ENCLOSURE SPACE BY LONG-TAILED MACAQUES AT MINDY'S MEMORY PRIMATE SANCTUARY

Madison A. Snow, Tesa J. Martin, Kyle J. Copp, and Laurie Kauffman, Oklahoma City University

Huyen Tran and Tephillah Jeyaraj-Powell, University of Central Oklahoma

In terms of behavioral benefits, naturalistic environments have shown promise for primates in captivity. With the goal of providing data which supports the use of particular structures to stimulate and encourage natural behaviors, we investigated how 11 male long-tailed macaques (*Macaca fascicularis*), located at Mindy's Memory Primate Sanctuary in Newcastle, Oklahoma, use their enclosure space. Data was collected on usage of the ground, platform, firehose, stumps, warm house, and enclosure wall space. We also compared the patterns of enclosure space use of former pet macaques versus former lab macaques. Data collection included use of a video camera to record and narrate for 15 minute periods. Our initial hypothesis was that there would be no differences in use of enclosure space or structures. We found no significant difference between former pet and lab monkeys with use of a 2x7 mixed ANOVA. However, there was significant difference is structure use although not as we had hypothesized. We will communicate our results with the primary sanctuary in order to provide an environment which will enhance the macaque's overall health.