Thirteenth Annual

Fall Research Symposium

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September 25, 2011
St. Thomas Campus
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St. Thomas Campus, U.S. Virgin Islands

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Fall 2011 Research Symposium Presenters

The role of the non-canonical (NC) NF-kB pathway in canine DLBCL.

Frazly Alexander
Dr. Nicola Mason and Dr. Anita Gaurnier-Hausser (mentors)
University of Pennsylvania

NF-kB is a highly conserved family of transcription factors, that control innate and adaptive immune responses. NF-kB activation is highly regulated through either the canonical or the non-canonical pathway. The canonical pathway is activated by hundreds of immune and inflammatory stimuli and regulates over three hundred genes involved in innate and adaptive immune responses. The non-canonical (NC) NF-kB pathway is activated by a very restricted set of stimuli and regulates the expression of cytokines and chemokines that play a role in lymphoid organogenesis, B-cell maturation, proliferation and B cell survival. In health, the NC NF-kB pathway is highly regulated only being activated by B cell growth factors and inflammatory stimuli. However, this pathway has been shown to be constitutively active in certain cancers of humans including multiple myeloma, Chronic Lymphocytic Leukaemia (CCL) and B and T cell lymphomas. This constitutively active pathway is believed to promote survival and proliferation of neoplastic plasma cells and lymphomas and inhibiting this pathway could have beneficial effects in the treatment of lymphoma. Cells from the lymph nodes of dogs with Diffuse Large B-cell Lymphoma (DLBCL) were tested for the presence of constitutively active NC NF-kB activity by qRT-PCR and Western Blot analysis (WB). The cell lines Gl-1 a B-cell leukaemia cell line that has been around for about 10 years, CLBL-1 a recently characterized cell line that came from a canine malignant lymph node and the 17-71 cell line that was derived from the peripheral blood of a dog with multicentric lymphoma were also interrogated by WB for NC NF-kB pathway activity in order to examine the significance of the pathway in lymphoma development and survival. We found that canine DLBCL cells had evidence of constitutively active non-canonical NF-kB pathway activity based on the over-expression of non-canonical regulated genes and the presence of increased p52:p100 ratio. Our resuts suggest that this pathway may be involved in lymphomagenesis in canine patients and that the dog may serve as an important spontaneous model for human DLBCL subtypes exhibiting constitutive non canonical NF-kB activity.

This research was supported by NIH MARC Grant # 5T34GM008422.

Diversity of bacteria associated with *Montastraea spp*. across sea water quality gradient in the United States Virgin Islands

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Hollings Marine Laboratory, University of South Carolina

The objective of this study was to describe and quantify the microbial community composition associated with the mucus of the *Montastraea annularis* species complex, which are the major framework building species (50-80% of total coral cover) of corals in the U.S. Virgin Islands region. Coral reefs in the region are greatly impacted by various kinds of anthropogenic stress and there is a trend of decreasing coral cover and health across a nearshore to offshore stress gradient. The microbial community found in coral mucus forms the primary line of defence against pathogenic invasion and it has been suggested that changes in the physical and chemical properties of sea water can cause changes in their populations, potentially making corals more susceptible to diseases. To understand whether the mucus-associated microbial community composition changed with the proposed water quality gradient, mucus samples were collected from corals in nearshore, shallow (<30 m) and highly impacted reefs as well as offshore, shallow and relatively pristine reefs. Samples were analyzed using 16S rDNA technique for patterns of change in microbial composition across the above mentioned gradient.

Preliminary results suggest that the microbial population is dominated by members of the phylum Proteobacteria with Beta Proteobacteria being the most abundant in all the samples. Soil related bacteria and pathogenic bacteria including *Vibrio* and *Clostridium* were isolated from the nearshore impacted reefs but not offshore pristine reefs. Further statistical analysis will help in understanding whether the microbial community composition changes as one moves across this proposed water quality gradient and whether this change is associated with coral health.

A special thanks to N. Kimes, Dr. P.J. Morris and Maria I. Vizcaino at Hollings Marine Laboratory, University of South Carolina. This project was funded by an incubator grant awarded to M. Brandt by the NSF Virgin Islands EPSCoR program and a scholarship awarded to S. Arora by the Lana Vento Charitable Trust.

Activation of the UNC-82 Kinase

Hema Balkaran
Dr. Pamela Hoppe (mentor)
Western Michigan University

One specific kinase is the unc-82 kinase found in *C. elegans.* UNC-82 is homologous to ARK5 (NUAK1) and SNARK (NUAK2), which are human proteins involved in metastasis of cancer. The *C. elegans* unc-82 mutant has muscle organization defects during embryogenesis. The main question addressed by this experiment is how the unc-82 kinase is activated. To go about answering this question, two minor questions were asked. The first question is whether the *unc-82* is present in the pharynx muscle and to go about answering this question, the unc-82 promoter was made and transformed several times without success. The second question is if specific phosphorylation sites in the UNC-82 protein are required for *unc-82* activation for normal muscle. To carry out this experiment, the approach was to mutagenize *C. elegans* in vitro by changing cloned DNA to test for function in striated muscle. The transformation was unsuccessful. Future work is needed to get the transformation to work then continue the approach. If this works, we may even be able to use it in cancer research.

NIH MBRS-RISE Grant Award No. GM061325 and Western Michigan University NSF-REU funded grant

Coral Disease Prevalence of Coconut Island: a Temporal Study Between 2007-08 and 2011

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Coral reefs, while they cover less than .1% of the ocean surface area, harbor up to 25% of all marine fish species and have been estimated to be associated with 1-9 million species of organisms. This ecosystem provides humans services valued at approximately $US375 billion worldwide. Global warming, ocean acidification, over fishing, eutrophication, and coastal development threaten the health and prosperity of coral reefs. Synergism of these coral hazards has caused the loss of an estimated 27% of coral reefs in 2002. Since then multiple events have devastated coral diversity and cover in the Caribbean as well as many other coral hotspots and it is believed that coral disease outbreaks are increasing in number.

The Pacific/Indo-Pacific when compared to the Atlantic Ocean has fewer coral species affected by disease and greater species diversity. Hawaii, due to its geographically isolated location, exhibits low coral species diversity. Thus the coral ecosystem could be severely impacted by coral disease.

In 2007-2008 Williams *et al*. (2010) conducted a disease abundance survey on the reefs of Coconut Island, Oahu, Hawaii. Since then the area has been subjected to many disease outbreaks including a recent outbreak of *Montipora* white syndrome, which led to the death of many *Montipora capitata* in 2010. In the summer of 2011, we conducted an identical abundance survey to that of Williams *et al.* under the assumption that coral community structure has changed in response to increased disease outbreaks. A total of 8 sites and 40 transects were surveyed for coral lesions including predation, *Porites* tissue loss, *Montipora* white syndrome, *Porites* and *Montipora* growth anomalies, *Porites* bleaching with tissue loss, and *Porites* trematodiasis.

In all, 6291 coral colonies were observed in an 800 m^2 area from July-August, 2011. Of these, 2654 (42%) had one of the above lesion types (961 predation, 53 bleaching, 10 bleaching with tissue loss, 140 *Montipora* white syndrome, 301 *Porites* growth anomaly, 120 *Porites* tissue loss, 1064 *Porites* trematodiasis, 5 *Montipora* growth anomaly). In this study, changes in coral species composition in relationship to disease prevalence are investigated using uni- and multivariate statistical tools.

Due to the low coral species diversity in Hawaii, disease can have great implications for benthic composition/community structure, consequently affecting the productivity of the coral reef ecosystem. A change in this composition can mean a decrease in rugosity or habitat space necessary for other species' survival.

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Genetic Variants in CYP4F2 Gene Affect Maximal Diastolic Blood Pressure and BMI in Response to Exercise Training

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High blood pressure is a major risk factor for heart disease, stroke and kidney disease. Cardiovascular diseases (CVD) and its risk factors are highly heritable and prospective research has suggested that they have a strong genetic basis. In addition, there is evidence that suggests that CVD and its response to exercise training are also heritable. Our goal was to investigate the association of genetic variants of the cytochrome P450 (CYP4F2) gene with hypertensive characteristics and other CVD risk factors in response to exercise training. We focused our analysis on a 1347 G/A polymorphism (rs2108622) of the CYP4F2 gene, that was previously associated with hypertension, with carriers of the A allele being at higher risk relative to non-carriers. One hundred and eighty four sedentary, normotensive and dietary stabilized; older Caucasian men and women were involved in the study. DNA was extracted before and after a six month endurance exercise training program and the genotypes were determined by PCR-RFLP technique. The PCR products were subjected to sequencing to confirm genotypes. Data were analyzed using SPSS software to test for statistical significance for the hypertensive phenotypes before and after training. The A allele associated significantly with body mass index and maximal diastolic blood pressure ($p=0.036$ and 0.044 respectively) in response to exercise training. Overall, our finding suggests that CYP4F2 variation explains the inter-individual differences in the response of hypertensive phenotypes to exercise training. Further work is still being done to genotype more individuals. In conclusion, these data contribute to the growing understanding of CYP4F2 as a candidate gene for hypertension and other CVD.

This work was supported by NIH AR059913, UMD Kinesiology GRIF (KCJ) and NIH MBRS-RISE Grant Award No. GM061325.

DNA extractions of white mangroves: The enzymes RNase & proteinase K produced the purest DNA

Lavida Brooks and Gemel Joseph
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Mangroves are known for their high salt tolerance and ability to provide shelter to many aquatic and non-aquatic organisms. They protect coastal lands from erosion by trapping sediment runoff from flowing into oceans and other bodies of water. Molecular data are currently not available for the white mangrove species, *Laguncularia racemosa,* which is one of four species of mangroves found in the Virgin Islands. This is because the DNA extracted in the past by scientists of the VI often was not clean enough to allow for further examination of its genetic variation. In this study, we investigated the question "What method of DNA extraction produces the highest quality DNA sample for *L. racemosa* for use in genetic testing?" There were four DNA extraction methods: the CTAB protocol, the double chloroform (CTAB) protocol and two DNA extraction kits, Epicentre and Ultraclean. After extracting the DNA, we measured the purity and quantity of the sample using UV spectrometry. We cleaned the least pure samples using 3 enzymes: proteinase K, phosphatase and RNAse. After which we amplified the DNA samples and analyzed the DNA bands (under UV light) after they were separated in an agarose gel. The use of enzymes proteinase K and RNase produced the best samples with AAA_{260}: A_{280} ratios of 1.85 and 1.86 respectively. The outcome from amplifying and analyzing the DNA bands is inconclusive. The results indicate that we are well on our way to finding the ideal extraction method which produces high quality DNA for further genetic testing.

This research was funded by NSF HBCU-UP grant # HRD – 0506096 and NSF VI EPSCoR.

Study of intramembrane protease IMP1 gene functions in knockout mouse and mammalian cells models

Charnele Burton
Ilya Alexandrov, Anastasia Grigorenko, and Evgeny Rogaev (mentors)
University of Massachusetts Medical School

(This abstract is intentionally withheld until results are professionally published in a referred journal.)

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Comparison of Angle Resolved Photoemission and Scanning Tunneling Microscopy on High Tc Superconductors Using the Yang-Rice-Zhang Model

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Using the Yang-Rice-Zhang model, this experiment sought to monitor the behavior of electrons in a sample of $Bi_2Sr_2CaCu_2O_8$ for pseudo and superconductor images. Two experimental techniques are known to perform such a task. Scanning Tunneling Microscopy (STM), which determines electron behavior after some considerable data processing, and angle resolved photoemission spectroscopy (ARPES), a more direct measurement, appear to show behaviors for high-temperature cuprate superconductors. While the Yang-Rice-Zhang model appears to work for ARPES, it must now be tested against the STM data. The main thrust of this project sought to determine the energy range the model agrees with the STM data, if at all. In order to accomplish this, fermi surface images were first created. The Gwidyon software was then used to create auto correlation images. The dispersion image was then used, as it relates to the energy versus momentum in the crystal, to formulate the q-vectors. It was observed that by creating two equal rectangles with widths equal to that of the pseudo-gaps that the Pythagorean theorem could then be used to create equations to solve for the q-vectors. Once this was calculated, a script was then written in MATLAB and effectively ran to formulate the dimensions of the q-vectors for binding energies -105 to 105. Where high density states were visible darks spots were apparent and the vectors lined up evenly. They fit perfectly on the edges of the Fermi surface images, as well as on the auto correlation images. One of the major findings includes the fact that the graphs are not symmetric, like other researchers believed. Being one of the first to plot this graph from the negative binding energies we were able to see the intire graph, along with be able to see other specifics such as the fact that the q-vectors are asymmetric in binding energies and the effects of the pseudo gaps and where they open. Though this study started off determining q-vectors manually, it was able to be automated to some extent and is still in the process of evolving as far as simplifying the calculation of the q vectors.

Special thanks to National Science Foundation for sponsoring this research, as well as Brookhaven National Lab for providing the venue with which the experiment was performed. This work was supported by NSF HBCU-UP grant number HRD – 0506096

Machine Learning to Classify *E. coli* Promoters into Functional Categories Is More Accurate When Using Tetranucleotide Frequencies Rather than Di- or Trinucleotide Frequencies

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Shenelle Dore and Dr. Stuart Ketcham (mentor)
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A promoter is a DNA segment that serves as the initiation site for expression or transcription of a gene. A DNA segment consists of a series of chemical subunits that are represented by a linear sequence of letters. Transcription is the copying of information from a gene by an RNA polymerase enzyme. In *E. coli* and other bacteria, the part of the RNA polymerase that recognizes a promoter is a subunit called sigma (σ). There are different versions of σ that are active under different environmental conditions and recognize different sets of promoters. So the σ's play a key role in regulating bacterial gene expression. Machine learning may be a relatively inexpensive way to identify candidates of different types of promoters compared to molecular biology experiments. The purpose of our research is to discover whether or not machine learning algorithms can accurately classify promoters into classes recognized by different σ's and which features are most useful for classification. We compared classifications using the 3 following alternative types of features: the frequencies of each of the 16 possible dinucleotides in the DNA segment; the frequencies of the 64 possible trinucleotides, and the frequencies of the 256 possible tetranucleotides. We used the programming language PERL to develop scripts to calculate these frequencies in promoters known to be specifically recognized by each of the 6 types of σ's: σ70, σ54, σ38, σ32, σ28, and σ24. With these frequencies, we used the J.48 tree and Naïve Bayes algorithms in the machine learning package WEKA to classify promoters into one of two σ classes (for example: σ70 vs. σ54). Applying these methods, we rejected the null hypotheses of no difference between the distribution of correct and incorrect sequence instances classified between classifications using dinucleotide features vs. those using trinucleotides (p value = <.0001) and trinucleotides vs. tetranucleotides (p value = 0.0024). The % correctly classified increased from 65% to 78% to 83% for the classifications using di-, tri- and tetranucleotides, respectively. The J.48 tree classifications used only a subset of the initial features to construct the trees. In general, reclassification using only those features resulted in higher classification accuracies. These results suggest that machine learning may be useful in identifying important features of different types of promoters and recognizing previously unknown promoters.

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The promoters used in this research were retrieved from the RegulonDB Database (http://regulondb.ccg.unam.mx/).

Temperature and feeding affect Caribbean corkscrew sea anemones' stinging temperament

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University of the Virgin Islands

Sea anemones are known to use mechanical and chemical stimuli to determine when to fire their cnidocytes, cells on their tentacles that fire a type of toxic filled harpoon for defense or for prey capture, but other factors may influence the degree of the firing of these cells. The common corkscrew Caribbean sea anemone, *Bartholomea annulata,* is usually quite aggressive yet sometimes seems to be very docile in our holding tanks. We explored several factors which may lead to changes in sea anemones' stinging temperament including temperature and feeding habits. We measured anemone temperament for stinging with a gloved finger poke on a scale of 0-3. We conducted field experiments to explore the effect of feeding on anemone stinging temperament, by comparing temperament before feeding, during feeding and post-feeding (~45min after feeding). *B annulata* was more aggressive during feeding than before or after feeding. To explore the role of temperature, we compared the temperament of anemones in warm (34^{o}C), or cold water (20^{o}C) to their temperament in normal seawater (28^{o}C). We found that anemones in cold water were more aggressive than those in normal or warm water. The changes in anemone aggressiveness may have implications for the cleaner shrimp that inhabit these anemones, such that the shrimp may have to alter their behavior during cooler times and when anemones are feeding. The increase in anemone aggressiveness in colder water may be due to increased oxygen supply in colder water, though we have not tested this directly yet. Furthermore, a large die off was noted during these experiments which coincided with an increase in the acidity of the holding tanks' seawater, suggesting we should further investigate the role of seawater pH in relation to the health and aggressiveness of the corkscrew anemone.

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Chemical Synthesis of Phosphonate Compounds

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The work that was performed will help with the understanding of naturally produced phosphonate compounds that have not been fully studied. These compounds are similar in structure to phosphate esters but instead of having an oxygen-phosphorus bond, they have a stable carbon-phosphorus bond. The importance of the synthesized compounds lies in the properties that they hold. Some of these compounds are known for their ability to be herbicides, pesticides, or antibiotics. By being able to synthesize these compounds we can study the properties that they may hold, which will be of a great interest. Rhizocticin A is toxic to fungi, but not to humans, whereas dehydrophos is known for its wide range of effectiveness against gram-positive and gram-negative bacteria. The first molecule to be used was from leucine, a common amino acid found in the body. The leucine was first methylated, and then the free amine was protected. The methyl ester was then replaced with an amide, and then it was coupled with dimethyl acetylphosphonate. The resulting compound is a derivative of a member of the dehydrophos family. A hydrated derivative of Rhizocticin A will be the second phosphonate that will be synthesized. After these compounds are made, they can be tested by others in the laboratory.

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Preliminary Results: Measurement of Sediment Production and Particulate Organic Material in Small Subtropical Watersheds on the East End of St. Croix, USVI

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University of the Virgin Islands

Ongoing research in the US Virgin Islands is being conducted in an effort to better understand the relationship between increased terrestrial sediment inputs and the degradation of receiving marine ecosystems. Degredation of near-shore coastal habitats, resulting from sediment layden runoff, continues to be one of the main nonpoint sources of pollution contaminating surrounding waters in US Virgin Islands. Land use changes that decrease vegetation cover, increase the potential for soil erosion and therefore, sedimentation into coastal marine environments. Increased development, unpaved roads and a lack of effective erosion control practices have massively contributed to increased sediment loading rates in the territory. Increased erosion rates effectively decrease organic content in surficial soils by simple soil loss. Particulate organic material is critical for nutrient and moisture holding capacities in soils and for productive plant growth. Productive plant growth stabilizes soil structure and reduces erosion. This study was designed to quantify and compare sediment production rates from natural and anthropogenic sources of sediment within the subtropical environment of the East End and Boiler Bay watersheds. Sediment production rates are being measured from undisturbed, vegetated hillslopes, and disturbed areas, represented by old unpaved roads that are currently used as foot trails. The main objectives of this project are: 1) to compare erosion rates between the two types of surfaces (i.e., trail vs hillslope); and 2) to quantify the particulate organic material composition of the collected sediment samples from both source types. Monitoring sites have been established throughout the watersheds by the installation of 21 sediment traps that collect material from trail, hillslope and cliff surfaces. Material collected from each of the sediment traps during the 2010 study period were analyzed and correlated with rainfall data, slope and vegetation cover. Subsamples taken from the material collected from the sediment traps were analyzed for organic content, using the Loss on Ignition (LOI) method. Preliminary results are indicating that erosion rates on trail surfaces are higher than undisturbed hillslopes. In addition, the ranges of particulate organic material present in the soil samples are showing that samples taken from trail surfaces are generally lower than those taken from undisturbed hillslopes.

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Using paper chromatography to deliver paper spray ion source to an ion mobility spectrometer

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University of the Virgin Islands and New Mexico State University[2]

A fairly new technique called Paper Spray Ionization is being innovated in Dr.G.A.Eiceman's chemistry lab at NMSU. This technique uses paper chromatography to deliver sample ions sources to an Ion Mobility Spectrometer for detection. The main purpose of developing this technique to ion mobility spectrometry is that the use of paper is cheaper and more convenient since it is easier to obtain than using other sample delivering techniques such as electrospray and mass spectrometry. The development of paper spray could be beneficial to security agencies, government agencies and organizations such as the Port Authority, and to other commercial agencies that rely on the use of chemicals. The question asked was, *"What is a quick and easy way to introduce the sample to the ion mobility spectrometer using paper?"* In the experiments atmospheric pressure ionization using novel paper spray ionization technique has been demonstrated with ion mobility spectrometry. Cellulose in the paper provide the tortuous path for the analytes to reach the tip for the triangular paper tip where the taylor cone is formed when high voltage is applied. Whatman filter paper with 11μm pore size has been used to check the response of solvent and other chemicals using paper chromatography. Paper chromatography is a separation technique used to separate compounds in colored substances (eg: dyes or colored substances that have color added to them). Separation is based on the relative rates of compound migration through a special filter paper. Colors associated with different compounds will separate on the paper showing bands or trails of its component colors at different heights along the strip. Factors affecting the mobility of molecules through the chromatographic paper include the size of the molecule and the concentration of the solvent. The concentration of the solvent refers to the proportion of methanol in water in this experiment. The solvent concentration can be manipulated in order to use the technique for applying paper spray with IMS Technology. Findings from the experiment showed that 80% methanol was best suited for the type of chromatographic paper that is used in the paper spray ionization technique. At a concentration of 80% there is clear separation of the compounds that are needed for detection by an ion mobility spectrometer. Other solvents can also be used at their respective concentrations. A few chemicals were able to be tested, of which clear distinctions were seen on the graphs produced by the ion mobility spectrometer. The use of Paper Spray is however, still in the developing stage and no definite conclusions can be made yet.

Ion Mobility Spectrometry is an analytical technique used to separate and identify ions of a gaseous compound based on their mobility through an electric field against a counter flowing neutral gas. Via the use of ion mobility spectrometers, identifying the types of ions made from proton transfer or electron capture reactions with neutral compounds that are present in a given ambient atmosphere can be accomplished with both natural and artificial conditions. Temperature and pressure are often altered so that the properties of the samples are changed to suit a desired purpose. The IMS "inhales" the sample (gases could be drawn, pumped or injected depending on the ionization source being used or the physical state of the sample), separates the ions, and produces a mobility spectrum (a display) showing unique arrival time responses of the ions formed from the compound when the gaseous compound was "inhaled" and ionized. This system is very efficient in that it can make accurate determinations using the minutest samples found in the atmosphere or from a surface. It is also inexpensive, easy to use (commercial models), and fast because one duty cycle (the time over which the experiment takes place) happens within a matter of milliseconds. A number of different sample interfaces (gas, liquid and supercritical fluid chromatographs) have been developed for different purposes but all share the same concept and overall purpose, to introduce the sample into the IMS.

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Investigating the role of an actin-binding protein, palladin, in human fibrotic disease

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Fibrosis is a condition in which the interstitial spaces within organs become filled with excess extracellular matrix fibers, which causes hardening of the organ that can lead to organ failure. Fibrosis can occur in any organ, and it is currently believed that approximately 45% of all disease-related deaths in the United States are caused by some form of fibrosis. Pancreatic fibrosis is caused by binge drinking, and it leads to a progressive, irreversible loss of pancreas function. In the pancreas, in response to alcohol metabolites, pancreatic stellate cells (PSCs) become activated and differentiate into highly proliferative myofibroblasts that display enhanced motility, contractility, and adhesion. These activated PSCs are believed to be the primary cellular mediators of pancreatic fibrosis. Recent studies have shown that the actin-binding protein, palladin, is highly expressed in activated PSCs. In the current study, we tested the hypothesis that palladin plays a key role in the activation of stellate cells to become myofibroblasts. The myofibroblast phenotype arises due to changes in gene expression, which are regulated by transcription factors such as myocardin-related transcription factor (MRTF) and serum response factor. It has been shown previously that palladin can translocate between the cytoplasm and the nucleus, and further, that palladin binds to MRTF and may regulate its transcriptional activity in the nucleus. CDC42 is a small GTPase that plays a critical role in the nuclear translocation of MRTF. Interestingly, our preliminary results suggest that palladin regulates the activity of CDC42 in PSCs: when palladin is knocked down, CDC42 activity decreased dramatically. This led us to hypothesize that palladin plays a key role in regulating the activity of CDC42, and thus that palladin expression is required for the nuclear import of MRTF. We utilized cytoplasmic/nuclear extraction to separate the cytosol and nucleus in the PSCs, and immunoblot analysis and immunofluorescence microscopy to detect MRTF localization in three treatments (normal PSCs, palladin-knockdown PSCs, and CDC42-knockdown PSCs). Our findings show that in normal PSCs, MRTF was detected predominantly in the nucleus. In contrast, MRTF was detected at higher levels in the cytosol of both palladin-knockdown and CDC42-knockdown cells. Together, these results highlight the role of palladin in MRTF localization and suggest that palladin plays an important role in regulating MRTF-mediated gene transcription. Understanding the pathway and roles that these three molecules play in pancreatic fibrosis will aid in our future goals to create drug therapies that are effective in controlling not only the progression of pancreatic fibrosis, but many fibrotic diseases.

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Population Genetics of *Laguncularia racemosa* (white mangrove)

Anthonio Forbes
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Laguncularia racemosa (white mangrove) is one of four mangrove species in the V.I. mangrove community. In the Caribbean, there is little known about the white mangrove's genome due to difficulties in extracting DNA samples from the plant. Past attempts resulted in impure DNA samples due to contamination in the DNA samples before they underwent PCR. With newly developed techniques in DNA extraction and enzyme clean up, we plan to produce pure, viable DNA samples that will be used for analysis of the white mangrove's genetic structure. In order to accomplish these tasks, we must develop an optimal protocol for amplifying *L. racemosa* DNA. Through experimentation with different settings on the thermal cycler and the ingredients required for PCR, a possible combination of the two will yield amplified microsatellite alleles. With this data, we can identify the genetic structure of the *L. racemosa* population and study it in greater detail in terms of genetic diversity and means of dispersal.

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Metagenomic Analysis of the Upper Respiratory Tract of Atlantic Bottlenose Dolphins with High and Low PCB Levels

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Marine mammals are highly susceptible to upper respiratory diseases and respiratory illnesses are the main cause of mortality in wild and captive *Tursiops truncatus*, the bottlenose dolphin. Culture-dependent and independent studies of dolphin upper respiratory tracts (blowholes) have isolated potential pathogens that cause respiratory disease. Polychlorinated biphenyls (PCBs) have been shown to cause decreased immunity and increased susceptibility to infectious diseases in cetaceans. This suppression of immune function may play a role in respiratory diseases in dolphins. In this study, we characterized the metagenomes of the upper respiratory tract of the common bottlenose dolphin in animals from Sapelo Island, GA and Brunswick, GA using next generation sequencing. We hypothesized that the upper respiratory tracts from dolphins that possessed high PCB body burdens would contain a higher proportion of viruses and potential pathogens. A total of 89,466,609 bp were analyzed from seven dolphin samples, each with different levels of PCB exposure ranging from 40 to 769 ppm. The metagenomes were dominated by bacteria, with Bacteroides, Proteobacteria, Actinobacteria, and Firmicutes as the most abundant phyla. A number of bacterial species associated with upper respiratory disease in humans were identified, including *Staphylococcus* spp. and *Streptococcus* spp. The viral sequences were dominated by bacteriophages, and were predominately classified as *Adenoviridae* and *Mycroviridae*. Human adenoviruses, which can cause upper respiratory disease in humans, and Tursiops truncatus papillomavirus (Ttpv), which is linked with genital papillomatosis in bottlenose dolphins, were also detected. In support of our hypothesis, the male and female dolphins with the highest levels of PCBs (769 ppm and 377 ppm, respectively) exhibited the highest percentage of viruses (approximately 10 %). The results of this work indicate that PCB exposure influences the respiratory microbiome of dolphins.

This work was funded by the NIH MARC grant # 5T34GM008422.

Is there a social preference of the rock boring sea urchin, *Echinometra lucunter*?

Tricia Greaux
Dr. Teresa Turner (mentor)
University of the Virgin Islands

The rock boring sea urchin, *Echinometra lucunter*, is a very interesting species that has not been extensively studied and little is known about its social preferences. This intertidal herbivore consumes algae, which harm corals, creating a better environment for the survival of coral reefs. Using transects, a density analysis was conducted in the field to determine the natural occurrence of the *Echinometra lucunter*. The transect data revealed that there is an abundance of juveniles and the high standard deviation means aggregation is present. I observed these sea urchins push each other as a form of intraspecific competition. An experimental study was designed to determine the social preference by using a Y-maze choice experiment conducted at the William P. MacLean Marine Center, St. Thomas campus, University of the Virgin Islands. In the choice experiment, there was a group of juveniles in one arm of the Y-maze with a single adult in the other arm (n=22). This was done to determine if, like other species of sea urchins, the juveniles *Echinometra lucunter* aggregate with other juveniles. The other possibility is that adults protect the juveniles under their spine canopy, therefore the juvenile would travel towards the adult. A chi-squared test revealed there was no strong difference in the choice between both arms (p-value = 0.088). This means the juveniles sea urchins did not respond strongly to the cues of groups of juveniles or single adults. Next steps include testing the response of juveniles in the field or in the presence of predators.

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Elimination of Cruzan Rum's Vinasse Effluent Discharge into the Caribbean Sea

Nathan Gubser
Eric Douglas MSc, PE (mentor)
Cruzan Viril and University of the Virgin Islands

For each gallon of rum manufactured, between ten and twelve gallons of waste product, called vinasse effluent, is generated. Cruzan VIRIL Ltd. operates a rum distillery on St. Croix, U.S.V.I. Until recently, Cruzan VIRIL has been permitted to discharge this undesirable waste product into the Caribbean Sea. However, a change in what will be legally allowable has driven Cruzan Rum to stop this practice, and implement an alternate approach to the processing and disposal of their vinasse effluent stream.

After a review of available alternative technologies, Cruzan Rum has selected an evaporative treatment method, and is currently constructing an evaporative process facility. This facility will concentrate the waste stream into a condensed molasses solids product, which will then be shipped off island to be reused as animal feed additive. Furthermore, the water recovered from this evaporation process will be purified, utilizing reverse osmosis technology and recycled for re-use in the fermentation process.

This development is an engineering solution to an environmental problem. Structural, civil, electrical, chemical, and mechanical engineering design elements are all integral components in the application of this solution. While this experience cannot be considered classical research; through it, immersive exposure to varied engineering applications and plant technology was gained. This was facilitated by the guidance from chemical and mechanical engineers as well as the project's construction manager. The result is a dynamic exposure to and understanding of, multi-disciplinary engineering collaboration and project management, during the construction of this industrial facility.

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Molecular detection of *Rickettsia* sp. in *Rhipicephalus sanguineus* (the brown dog tick) from dogs in the United States Virgin Islands

Akacia Halliday
Dr. Jennilee Robinson (mentor)
University of the Virgin Islands

Rhipicephalus sanguineus is a species of tick common throughout the world. In addition to its commonality, *R. sanguineus* is known to carry bacterial pathogens such *Bartonella* sp., *Anaplasma* sp., and *Rickettsia* sp. (just to name a few). When *R. sanguineus* bites a person or animal it may transmit these pathogens. Spotted fever group rickettsiae are known to cause severe illness in humans. SFG rickettsiae largely affect travelers and people who visit high incidence areas. To evaluate the risk of human or animal infection with vector-borne pathogens in the U.S.V.I., a local veterinary clinic provided us with *R. sanguineus* collected from dogs. We tested the specific hypothesis that SFG rickettsiae are present in *R. sanguineus* in the U.S.V.I. Molecular assays, specifically PCR analyses, using *Rickettsia*-specific primers were employed to test our hypothesis. One set of PCR primers target the citrate synthase (gltA) gene specific to the *Rickettsia* sp. A second set of primers target a fragment of the outer membrane protein A (rompA) gene that is only present in SFG rickettsiae. Total DNA was extracted from 22 individual adult ticks using the Qiagen DNeasy Blood and Tissue Kit, and the PCR assays were employed to screen the DNA extracts for the presence of rickettsial DNA. Preliminary results indicate the presence of *Rickettsia* sp. within 22/22 (100%) of the tick DNA extracts by DNA agarose gel-based visualization of an amplified DNA fragment of the expected gltA fragment size (381 bp). Interestingly, when the PCR products were separated by gel electrophoresis, additional amplified DNA bands were present. These bands were both larger and smaller that the expected 381bp fragment targeted by the gltA PCR primers. However, our rompA PCR results demonstrated an absence of amplified DNA fragments from *R. sanguineus* DNA extracts when PCR reactions were visualized by DNA gel electrophoresis even though controls indicated PCR was working properly, suggesting that that the gltA band may not originate from *Rickettsia* DNA. We plan to further investigate the nature of the ~381 bp fragments generated in the gltA PCR by DNA agarose gel purification and DNA sequencing. Sequences of the gel-purified fragment sand results from PCR screening of additional *R. sanguineus* DNA extracts will be presented.

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The Interplay Between Contingencies of Test Anxiety and Final Course Grades

Tishiro Hanley
Dr. Jennifer Cromley and Dr. Theodore Wills (mentors)
Temple University

In a time when nations are experiencing increased demand in STEM fields, the United States has come to recognize the deficit between position availability and qualified persons to fill those needs. As a result, research into STEM persistence and attrition abounds. Most of this research in STEM participation centers on precursory influencers such as AP class history, GPA, and SAT performance (Thompson & Rust, 2007, Kokkelenberg & Sinha, 2010). In attempt this study examined the relationship between undergraduate college students' test anxiety, and final course grades.

In this study, we used questionnaire data from a larger NSF-funded longitudinal study. Data were collected at two time points (2 weeks and 12 weeks into a 15-week semester) to measure anxiety. Our findings demonstrate the anxiety for students changed. Our results demonstrated anxiety predicted being in the lower grade group students.

Research was supported by National Science Foundation ERP award no. 1036183.

The Interplay Between Contingencies of Self-worth and Test Anxiety in Final Chemistry Grades

Syida Huggins-Richards
Dr. Jennifer Cromley and Dr. Theodore Wills (mentors)
Temple University

Throughout the world, nations have come to recognize that there is a growing demand for qualified people within STEM fields. In the United States, STEM education is a root of concern where it seems that colleges and universities are not producing enough STEM graduates. As a result of this shortage, there is an abundance of ongoing research that examines STEM attrition rates and persistence. For this study we asked (1) What motivational variables influence performance in STEM college programs? (2) How does test anxiety for students with higher and lower contingent self-worth change over one semester and (3) How does test anxiety affect course grades for lower contingent self-worth versus higher contingent self-worth students?

Questionnaire data from a larger NSF funded longitudinal study were used. Data were collected at two weeks and twelve weeks into a fifteen week semester to measure variables such as CSW, implicit self-beliefs, and anxiety. Logistic regression and repeated measures ANOVA were used.

Findings indicate great importance of contingencies of self-worth for STEM majors. This study can serve as a foundation for analyzing and even identifying high-risk students for poor performance and giving universities the opportunity to make corrective actions with those students who are at risks for failing courses and dropping out from STEM majors.

Research was supported by National Science Foundation ERP award no. 1036183.

Mitofusin-2 Protects Renal Epithelial Cells under Stress

Chinaemere Igwebuike
Jonathan M. Gall and Dr. Steven C. Borkan (mentors)
Boston University School of Medicine

The proximal tubule of the kidney is rich in mitochondria, double-membrane organelles that generate ATP and integrate cell signaling events that result in apoptosis. Mitochondria are dynamic and constantly undergo fusion and fission events that alter their morphology and may perhaps, regulate apoptosis and organ function. We hypothesized that Mitofusion-2 (MFN2), a mitochondrial fusion protein, is important in the renal epithelial cell stress response. To test this hypothesis, the MFN2 floxed gene was conditionally knocked out in the murine kidney. Despite marked mitochondrial fragmentation in the kidneys of juvenile MFN2-cKO mice, renal histology, development and organ function did not differ from wild type. In contrast, primary cultures of MFN2-cKO kidney epithelial cells subjected to chemical ischemia were significantly more susceptible to mitochondrial membrane injury, caspase 3 activation and apoptosis than wild type ($P < 0.05$). Bax, a major cause of apoptosis in these cells was equally activated, but far greater mitochondrial Bax accumulation was detected in MFN2 KO cells. We conclude that MFN2 protects renal epithelial cells by preventing Bax accumulation on the mitochondrial membrane.

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An efficient Algorithm for Solving the Berth Assignment Problem

Tyriq Isles and **Kyle Williams**
Dr. Marc Boumedine (mentor)
University of the Virgin Islands

Vessels or ships that arrive at the terminal (port) need to be assigned to berths in order to unload/load containers or passengers effectively. Therefore vessels and ships compete over the available berths and various parameters affect the berth assignment. The berth scheduling problem (BSP) is typically formulated as combinatorial optimization problem. The BSP is an NP-hard problem and heuristics or meta-heuristic algorithms are necessary to produce computationally acceptable solutions given the constraints and available resources. We are given *n* ships and *n* berths; each ship-berth pairing has an associated cost, C_{ij}, corresponding to the cost that the $_i$th ship associates with the $_j$th berth. The goal is to assign exactly one ship to each berth so that the resulting one-to-one correspondence minimizes the total cost. Many algorithms use iterative methods for finding the optimal assignment from the set of *n!* possible assignments. The *Hungarian* or *the Kuhn–Munkres* algorithm, which is one of the best known algorithm for solving the assignment, has been implemented. As expected, the algorithm find the best solution in polynomial time since it complexity is $O(n^3)$. A basic graphical user interface allows the visualization of the solution. This algorithm is currently been adapted to consider additional constraints specific to U.S. Virgin Islands ports.

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Developing an understanding of research design and methods

Danny Lynch, **Tobias Ortega-Knight**, and **Thalia Lake**
Dr. LaVerne Brown (mentor)
Organic Chemistry Lab, University of the Virgin Islands

All research begins with experimental design, and method development. As science majors at UVI and Summer Undergraduate Research Scholars it was our aim to develop a greater understanding of research design and methods.

We followed the scientific method by first formulating a question and developing a hypothesis from this question. A series of experiments was then used to test this hypothesis. The protocols used to perform these experiments were modified when needed. This process of method development was essential to gaining the best results from the experiments. It was determined more research is necessary to further prove or disprove the hypothesis.

A deeper understanding was developed of research design and how to see an experiment through to its completion.

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Yersinia pestis may not be as successful at intracellular growth as previously thought

Blanche Letang
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University of North Carolina-Chapel Hill

Upon infection, macrophages engulf, digest, and transport bacteria to proximal lymph nodes. Some pathogenic bacteria, however, survive and replicate inside macrophages, using them as a tool for dissemination. *Yersinia pestis*, the etiologic agent of plague, is thought to use macrophages for dissemination in the host and has been reported to replicate inside them *in vitro*. To test this hypothesis, we revisited *in vitro* intracellular growth of *Y. pestis* through gentamicin protection assays in which J774 macrophages were infected with two *Y. pestis* strains, CO92 and KIM6. CO92 is a constitutive GFP strain, while KIM6 is a GFP-inducible strain which was used in previous studies reporting that *Y. pestis* grows inside macrophages. Macrophages were infected and first treated with a high concentration of the antibiotic gentamicin for one hour to kill any extracellular bacteria. This served as our initial time point. Cells were then treated with a lower concentration of gentamicin to kill any extracellular bacteria that may have grown at later time points. Infected macrophages were fixed for microscopy or lysed to obtain bacterial counts. We optimized infection procedures to ensure that all macrophages contained bacteria at the desired multiplicity of infection by using sonication to disrupt *Y. pestis* clumps. No bacterial growth was observed at the 24h time point, so we subjected infected macrophages to gentamicin at varying exposure times and concentrations to test whether gentamicin had an effect on our results. Reduced or no exposure to gentamicin resulted in more apparent bacterial growth, which could indicate that extracellular bacteria were not being killed. To test this hypothesis, we used confocal microscopy to visualize the infected cells at our initial time point and found that there were live bacteria outside of the cell, suggesting that the gentamicin treatments did not kill all extracellular bacteria. We tested the susceptibility of bacteria alone by exposing them to different concentrations of gentamicin for different periods of time. CFU counts from this assay suggest that viable bacteria are still present outside of the macrophage after exposure to gentamicin. Infected macrophages were then subjected to this assay. Significant growth was only observed at lower concentrations of gentamicin. Our data suggests that upon infection, most *Y. pestis* bacteria die. However, even at the highest concentrations of gentamicin, we observed that a small fraction of the macrophages contained bacteria that appeared to have replicated. Therefore, *Y. pestis* may replicate and survive inside macrophages, but not as successfully as previously thought. Further investigation, including confocal microscopy, live/dead stain assays, and double stain assays using *Y. pestis* specific antibodies, is needed to confirm our results.

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Studying the Interactions between Coral Reefs and Participants of Marine Conservation Activities

Kyvonn A. Morton and **Antonio Farchette**
Dr. Kostas Alexandridis (mentor)
University of the Virgin Islands

The reefs provide an aquatic habitat that promotes enhanced species biodiversity, protection to our coastal shorelines, and promotion of sustainable marine and natural resource-based livelihoods. Coral reefs are being diminished daily due to anthropogenic and natural threats. In this process the role of information and education of the society regarding coral reef health and conservation is critical to prevent coral loss. In order to determine how people's general knowledge affects how they perceive and interact with coral reefs and how participants interact with each other and the environment during marine conservation activities, we administered a pre-survey to determine their attitudes, beliefs, and behaviors about the coral reefs. The participants undertook a boat trip to three different spatial locations, varying in their characteristics (e.g., difference between inshore and offshore reefs). We interviewed focus groups and used their responses in a content analysis of key perceptions and cognitive characteristics in three key temporal stages: (a) before the trip, (b) during, and (c) after the boat trip. The participant behavior in the water was observed in order to study their interests and the complexity of their interactions, and their encounter patterns with the natural system. The data were used to study the relationship between human interactions and the reefs. The participants realized how their actions affect the coral reefs and they noted the differences between inshore and offshore spatial locations. The study experience enhanced their understanding of how human interactions degrade the reef ecosystem and cause detrimental effects. Our study shows that the participants found the research to be vital, educational and have important impacts to the community perceptions regarding coral reef conservation. Finally, our results present a new approach to studying how social learning from experience can benefit and improve perceptions, beliefs, and attitudes toward environmental conservation.

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Oxygen-methylation of an Aldehyde Precursor for A 14-Membered Macrolide Antibiotic

Marisha M. Perkins[1]
Libing Yu[2] and Dr. Lawrence J. Williams[2] (mentors)
University of the Virgin Islands[1] and Rutgers, The State University of New Jersey[2]

Within the last 60 years, macrolactone glycosides (macrolides), a class of natural products composed of macrocyclic lactones to which one or more deoxysugar residues are attached, have gained recognition as powerful antibiotics. Erythromycin, the first macrolide introduced as an antibiotic for humans, is used clinically to treat certain bacterial infections. Our group has been developing a series of synthetic strategies to prepare derivatives of Erythromycin with modification of the macrolide precursor developed by our group. One synthetic route involves the C4 position as an aldol product, in which the hydroxyl functional group is methylated. Several strategies were developed and tested throughout this study for the oxygen-methylation of the aldol product. However, appropriate o-methylation of the aldol was completed using Meerwein Salt (Me3OBF4) and a proton sponge in DCM. This reaction proved to be efficient based on time, absence of by-products, and 89% yield of the desired methoxy product. In future research, the synthetic route (o-methylation) determined by this study will be used in the synthesis of several of the Erythromycin derivatives proposed by our group.

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The never-ending pairs of (m, n) where $\sigma(m+n) = \sigma(m) + \sigma(n)$

Dwayne C. Richardson
Leonardo José Bardomero & Dr. Douglas Iannucci (mentor)
University of the Virgin Islands

Let $\mathbf{N}$ be the set of natural numbers. We define the arithmetic function $\sigma:\mathbf{N} \rightarrow \mathbf{N}$ by

$\sigma(n) = \sum_{(d|n)} d$, where $\sum_{(d|n)} d$ is the summation of all the *positive divisors d,* of *n.* This operation is known as the *sum - of - divisors* function.

Suppose *m* is another element in $\mathbf{N}$. Given the case $\sigma(mn)$ where (*m, n*) = 1 (which is to say that *m*, and *n* are ***relatively prime***), it is clear that σ is a ***multiplicative*** *function*, i.e.,

$\sigma(mn) = \sigma(m)\sigma(n)$.

We ask ourselves: are there infinitely many solutions (*m, n*) for which

$\sigma(m+n) = \sigma(m) + \sigma(n)$?

(1)

In exploring this aspect of σ, we hope to develop a greater understanding on how numbers work, which may lead to other discoveries within mathematic and scientific disciplines. Some specific cases have been found, where (1) holds for infinitely many pairs (*m, n*). However, the question above still remains unanswered in general.

This is why we seek only for **primitive solutions** (*m, n*). A solution is known as ***primitive,*** if the greatest common unitary divisor of *m, n*, and *m* +*n* is 1.

Upon investigating pairs (*m, n*) that solve (1), it was found that each m, n and m + n would be multiples of primes. Using odd primes *p, q*, and *r*, along with natural numbers a, *b*, and *c*, where $p \nmid a$, $q \nmid b$, and $r \nmid c$, a system of linear equations was developed, as seen here.

$ap + bq = cr$

$\sigma(ap) + \sigma(bq) = \sigma(cr)$

Through applying Linear Algebra and Number Theory, an algorithm was developed which provided some solutions (*m, n*).

This method shows that it is simple, but not trivial to provide several pairs (*m, n*) which satisfy (1). This strengthens the conjecture that there are infinitely many ***primitive*** solutions satisfying

$\sigma(m+n) = \sigma(m) + \sigma(n)$.

This work was supported by NSF HBCU-UP #HRD-0506096.

Characterization of Thermal Residual Stresses in Polycrystalline Alumina

Phil Smith
Benjamin Anglin and Anthony Rollett (mentor)
Carnegie Mellon University

The development of residual stress in polycrystalline Alumina comes as a result of cooling after undergoing thermal expansion. Ceramics are typically used as thermal barriers because of their low thermal conductivity. Some of the applications range from exhaust manifold in vehicles to tiles for the outer surface of a space shuttle. When they are exposed to increasing temperatures, eventually the buildup of residual stress causes micro cracking to occur and the barrier will eventually fail. To observe the behavior of these stresses, synthetic microstructures were first built and using a program called the Thermo Elastic Fast Fourier Transform (FFT) the stresses were simulated. The Thermo Elastic FFT is a C++ program developed and maintained by Prof. Rollett's research group which combines properties of Alumina with the microstructure before simulating the stresses. Residual stress typically occurred near the grain boundary of the microstructure and as the stress value increased, they got closer to the boundary until they eventually occurred on top of the boundary itself. Also, the probability of finding a stress cluster at a short distance from another is high compared to finding grain centers from each other, which tells us that the clusters are more closely packed and that there can have many stress clusters within a single grain.

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Larval settlement of the long-spined sea urchin, *Diadema antillarum,* corroborates seasonality and importance of post-settlement processes in the US Virgin Islands

Jamie Spray
Dr. Teresa Turner (mentor)
University of the Virgin Islands

Rapid phase shifts from coral reefs to algal-dominated reefs have occurred throughout much of the Caribbean following a mass mortality event claiming 95-99% of long-spined sea urchins. Such a strong correlation with this event suggests the herbivorous *Diadema* fulfill an important niche in maintaining coral-dominated reefs. Hence, their reestablishment could enable coral reef recovery. Yet, recovery has been variable and patchy across and within the region. Possible reasons for this could be a difference in larval supply between localities, preferential settlement, and post-settlement processes acting strongly on certain populations. In continuation of a study instituted in Brewer's Bay, St. Thomas, USVI, we are deploying settlement collectors monthly to test the larval supply between two sites with contrasting adult population densities. Data from 2010 show a possibility of a late spring/early summer seasonal peak in settlement. Thus, we predicted a similar peak this 2011 season: May-July. Data show a seasonal peak, with one month's difference between years. The observed annual variation could be due to oceanographic conditions or nutrient flux. Additionally, we predicted that if larval supply accounts for the differences in adult populations, the locality with higher adult density (Runway) would show significantly higher *Diadema* settlement. The results were that Black Point, the site with low adult density, had the higher amount of settlement, whereas Runway had low settlement for the peak month. Because the site with higher adult density had the lower number of settlers, the larval supply hypothesis is falsified. A t-test performed over the entire study period showed no significant difference in the larval supply. Therefore, post-settlement processes must affect juvenile recruitment, and further study on the behavior and nursery requirements of recently-settled juveniles could lend insight into survival mechanisms to ensure healthy adult populations.

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Iowa School Administrators' Perceptions of Home Schoolers

Jamisha S. Stuard
Dr. Stewart Ehly (mentor)
University of Iowa

Home schooling offers an alternative to public and private school attendance while still meeting the legal requirements of compulsory education and attendance laws in the United States. Within the state of Iowa, CPI students are defined as those students whose educational program is managed primarily by their families. This includes students who are schooled in the home, either fully or partially, or in a non-accredited, nonpublic school. These students may be enrolled part-time in campus-based schools or virtual schools or share instruction with other families. Home school assistance programs are operated by the public schools in some communities to support CPI activities. Recent estimates on the number of children in the United States who are being home schooled range from approximately 850,000 to 1.7 million. Home schoolers account for 1.7 to 2.2 percent of students nationwide. *The current study investigated the perceptions of school administrators towards home schooling, considering* (1) roles and responsibilities; (2) programs and activities; and (3) legislation and monitoring. The School District Assessment Survey 2003-05 was emailed to 367 superintendents, which represents the entire population of school superintendents in the state of Iowa. The number of completed surveys was 303, resulting in a return rate of 82.6%. School administrators reported that accountability issues related to home schooling need to be addressed, calling for parity in the assessment of public and home schooled students' academic progress and voicing a concern about the effects of home schooling on No Child Left behind (NCLB) requirements.

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