

*Emerging Caribbean
Scientist Program
Summer Science Research
Symposium 2008*

*Summer Sophomore Research Institute (SSRI)
Summer Undergraduate Research Experience (SURE)
Summer Bridge2Calculus Program.*

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Administration Conference Center 1st Floor

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Welcome

Ms. Tannesia Petersen

Summer Undergraduate Research Experience

Dr. Omar Christian

Summer Bridge2Calculus Program

Dr. Dirk Schlingmann

Summer Sophomore Research Institute

Dr. Michael Thomas

Certificates

Program Coordinators

Closing Remarks

Dr. Marc Boumedine

Sure Participants

Tania Corke

Saskia Corke

Al'Lima deLugo

Andrea Dorsey

Lystra Edwards

Ferlicia Fergusson

Sayvi George

Dennis Powell

Kiara Scatliffe

Johnasha Stuart

SSRI Participants

Shellese Cannonier

Karissa Cave

Neisa Cazaubon

Jamilya Christopher

Dornette Lewis

Agene Rogers

Lynasha Sweeney

Oonijah Thomas

Tameca White

Troi Williams

Bridge2Calculus Participants

Michael Celestine

Kenneth Dawson

Aniqua Dessout

Emilio Edwards

Mahlik Griffin

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Nalinie Ramnaraine

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Regression of U-87 Tumors in Nude Mice after Treatment with Mutant forms of Adenovirus

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Despite the oncolytic efficacy of chemotherapy, radiation therapy, and surgery, these forms of treatment not only kill cancer cells but also destroy living tissues as well. Additionally, chemotherapy and radiation result in lymphedema (fluid retention in arms and legs), memory loss, infertility and secondary cancer in many patients. An ideal cancer therapy therefore should be noninvasive and exhibit minimal toxicity and side effects. Adenovirus (ONYX-015) has been in phase III clinical trials in the United States and is used to treat cancers in China. Although ONYX-015 has been successful in killing cancer cells in tissue cultures, as an individual therapy it is less successful in reducing tumor size. Therefore a virus with the advantages of ONYX-015 with improve killing effects would be beneficial.

Research has shown that the viral early region 4 contains proteins that protect the virus infected cells from dying. Removal of these proteins (E4orf1 and E4orf3) in the context of ONYX-015 may increase virus induced cell death. In this experiment, we compare mutant forms of adenovirus which lacks either E4orf1 or E4orf3 to determine if these mutations may contribute to the reducing the volume of the malignant glioblastoma cell line U-87 (aggressive type of primary brain tumor) in a nude mice model. I hypothesized that treatments with MAT2 (mutant E4orf1) and virus 3112 (mutant E4orf3) would decrease the tumor volume more effectively than ONYX-015. Alternatively, there may be no difference in the tumor sizes after treatment. To test these hypotheses, U-87 cells were implanted in 20 nude mice. The mice were marked for identity purposes and 20% of the tumor volume (width² x ½ length) was the amount of virus administered to each mice. The results established that Virus 3112 had more oncolytic potency than ONYX -015 and MAT2. Although MAT2 was less effective at reducing the tumor size than virus 3112, MAT2 is shown to be more oncolytic than ONYX-015. This data suggests that the mutation of E4orf3 in virus 3112 has increased its oncolytic potency.

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Adenovirus Regulation of The Cell Cycle

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In the United States cancer is the second leading cause of death. Additionally, the therapies used to treat cancer are either highly invasive or toxic to the body. This is because they do not differentiate between normal and cancerous cells. Adenoviruses, however, has been shown in clinical trials to specifically kill cancer cells and are significantly less toxic, making them of interest as cancer treatments. In a recent publication, Cherubini, et.al reported that mutant adenovirus onyx-015 induced endoreduplication in normal cells. However, whether adenovirus induces endoreduplication in cancer cells remains to be determined. In aim 1 of this study, experiments were done to determine if adenovirus induces endoreduplication in cancer cells. Here we report that mutant as well as wild type adenovirus induced endoreduplication in cancer cells.

The adenovirus E4orf1 protein activates the PI3-kinase pathway and members of this pathway have been reported to enhance endoreduplication. Therefore in aim 2 we seek to determine if the PI3-kinase pathways is involved in adenovirus-induced endoreduplication. To test this, various inhibitors of the PI3-kinase pathway were used. In all the samples excluding mock, the drug Rapamycin caused a decrease in endoreduplication with a significant decrease in dl309. The drug LY294002 also caused a decrease in endoreduplication in the samples, though to a lesser degree. This suggests that the P13-kinase pathway is involved in adenovirus induced-endoreduplication.

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Cassava Somatic Embryos from Commercial Cultivars

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Cassava provides food for humans and animals, as well as raw material for the starch industry. Unfortunately, this important crop is not immune to viruses or diseases. Genetic engineering can facilitate us to introduce desirable traits such as resistance against viruses and the modification of cyanide content and protein content of tuberized roots. To genetically engineer cassava, a somatic embryo system needs to be in place for commercial cultivars. Apical shoot meristems and young leaves up to 2 mm were isolated from three cassava cultivars Adira 4 (AD 4), Kasesaert University #50 (KU 50), TMS 60444 (TMS) were transferred to Murashige and Skoog media with 6% sucrose and 2,4-dichlorophenoxy acetic acid (2, 4-D) at 0, 4.5, 9.0 or 13.5 μM to induce somatic embryos. Shoots and leaves were observed twice a week for 20 days. Friable wet callus was first observed on the cut surfaces. Somatic embryos were seen by day 20 arising from the apical meristem. The highest percentage of embryos developed from the apical meristems, 53%, compared to the highest percentage embryos developed from young leaves, 40%, both from the 13.5 μM 2,4-D, indicates that the concentration of 2,4-D positively correlates with the percentage of embryos developed in both meristems and young leaves. The average number of embryos per variety of leaves was higher than the average number of embryos per variety of meristems. Except for the TMS cultivar, which did not develop embryos in the 13.5 μM 2,4-D culture, embryo counts of AD 4, KU 50, and TMS increased as the concentration of 2,4-D increased for each variety. Each variety of cassava responds best to different concentrations of 2,4-D, however meristems should be used to maximize potential of induced somatic embryos. All three of the varieties of cassava developed embryos from both apical shoot meristems and young leaves on the 9 μM 2,4-D, however the 13.5 μM 2,4-D cultivar provides larger numbers of embryos for each variety that develop embryos in that cultivar. This research was funded by the NIH.

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Determining the Most Effective Human Adenovirus That Can Be Used to Kill Cancerous Cells

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Currently, there are three options that can be used to treat the cancer – chemotherapy, radiation therapy, and surgery - that are either toxic or invasive. Therefore, a need for a less invasive and non toxic method to treat cancer is critical. In clinical trials, adenoviruses have been proven to be safe showing minimal toxicity. An adenovirus is a non-enveloped virus that is composed of a nucleocapsid and a double stranded linear DNA genome that is responsible for upper respiratory infections. Additionally, adenoviruses' unique ability to selectively kill cancer cells made it an excellent candidate to treat cancerous cells.

Even though adenoviruses are found to be effective at killing cells in culture, there are aspects that can be improved. In this experiment, we compared eight mutant adenoviruses to determine which would be most effective in killing HeLa cells. Based in the information that was acquired from reading several papers, I hypothesized that dl1520 would have been the most effective adenovirus used to kill HeLa cells compared to the others used in the experiment. Graphs were made to compare the percent of infected cells that were still alive after pre-determined hours. The results illustrated that my hypothesis was false and that dl1005 was most efficient in killing the infected cells. A P-value (.03) was determined, which showed that the percentage viable cells between the dl1520 and dl1005 were significant meaning dl1005 can be a better candidate in treating cancerous cells.

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Cistern Water Quality in the St. Thomas, St. John District

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Water usage in the Virgin Islands is an interesting scientific and an important public health problem. Historically water has been available mostly from catching rainwater and storing that water. The island aquifers are small and provide a small percentage of the water needed. Rainfall averages 40 inches per year and is seasonal. There was a dramatic increase in population starting in the 1960's during which the population increased from 20,000 to the presently over 110,000 resulting in that over 50% of the residences in the USVI use cisterns as their only source of water.

There has not been a study of the presence of environmental pollutants such as polyaromatic hydrocarbons (PAH) or heavy metals in cisterns in the Virgin Islands. Nor has there been a recent study of usage patterns of cistern water.

During this study we have investigated approximately 40 cisterns, collecting liter samples of water. Data on cistern conditions, usage, water source and other relevant historical data have also been investigated. Samples collected will be subject to GC/MS and ICP/MS analysis in the Fall. Information on cisterns and their water analyses will be correlated to establish a baseline to determine if water quality issues exist and to provide a basis for future monitoring of cistern water.

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Developing an AdHu5 vector for a HIV-1 Vaccine

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HIV has developed into a pandemic and the number of infected individuals continues to grow at an alarming rate. Thus the need for a vaccine is desperate. Despite recent antiviral strategies, Human serotype 5 adenovirus has achieved the most success as a potential HIV vaccine candidate. Adenovirus is known for causing illnesses such as diarrhea, pink eye and the common cold. Nevertheless, the benefits of using adenovirus to construct an AdHu5 HIV-1 vaccine vector include the fact that the virus has been proven safe; the virus's ability to naturally induce long lived mucosal and systemic immunity, and ease of manipulation.

The purpose of this research is to determine which AdHu5 mutant virus would be the best candidate for designing a vector to develop an effective HIV-1 vaccine. Cell lines H-1299, U-87, and Hela cells were infected with nine mutant adenoviruses. The nine adenoviruses were analyzed for their ability to produce late viral proteins. The virus that produces the least amount of late viral proteins is expected to elicit the least adverse immune response and hence be the safer candidate. Our results show that in both H1299 and U-87, virus dl223 exhibited the least amount of late viral protein production. In Hela cells, dl1520 produced the least amount of viral proteins. Future research would involve placing the HIV *gag* or *env* gene into one of these vectors and assessing the immune response. Additionally, chimeric viruses made from other Ad subgroups with similar mutation to dl223 or dl1520 may be used in creating an AdHu5 HIV-1 vaccine vector.

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The Effects of Red Mud and Stabilized Red Mud on Seed Germination and Plant Growth

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Red mud is an end-waste product from the process of isolating alumina from bauxite. It has a red-clay color and a mud-like appearance when wet, a pH ranging from 12-14, and poor drainage which severely inhibits plant growth. These factors make growing plants in red mud nearly impossible. However, work was recently done on St. Croix USVI to develop a protocol to stabilize the red mud and reduce the pH. The objective of this research was to compare seed germination and plant growth in red mud and stabilized red mud to determine if the stabilized red mud will sustain and promote plant development. This was done by growing a variety of plants from multiple diverse plant families, Solanaceae, Cucurbitaceae, Caricaceae and Poaceae (tomato, cucumber, papaya and corn, respectively). Seeds were planted in ProMix with 0%, 10%, 25% and 50% red mud and 25%, 50% and 100% stabilized red mud w/v. Seed germination was recorded for 18 days. Plant height, number of leaves and general appearance were also recorded. Deceased plants and water samples were collected from leachate from the pots and pH was determined weekly. Plant growth in the red mud treatments were adversely affected by delayed germination, smaller growth, and a variation in leaf color while the plants in the stabilized red mud treatments did not follow a similar trend except for the 100% treatment which had poor germination. Total plant height and number of leaves gradually decreased as the concentration of red mud increased, whereas the plants grown in the stabilized red mud did not show signs of variation except for the 100% treatment. pH of the red mud treatments ranged from 7-9 and for the stabilized red mud, they ranged from 7-8. These results indicate that the use of stabilized red mud promotes seed germination and normal plant growth over a wide range of concentrations as compared to red mud which caused nutrient deficiency symptoms and early plant death.

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Do the cleaner shrimps *Periclimenes pedersoni* and *Stenopus hispidus* affect the size of flatworm parasites *Neobenedenia mellini* on a Caribbean reef fish?

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Ectoparasites are harmful to reef fish because they damage the fishes' skin and make them more susceptible to disease (Thoney and Hargis 1991). Fish may be cleaned of parasites and other debris by cleaner fishes or shrimps at cleaning stations (Côté 2000). The effectiveness of cleaner fish is well documented, but the role of different cleaner shrimp species is not well studied (Becker and Grutter 2004). Two Caribbean shrimps, Pederson cleaning shrimp, *Periclimenes pedersoni*, and banded coral shrimp, *Stenopus hispidus*, were recently tested for their effect on fish parasite loads (McCammon et al. 2008). Pederson cleaning shrimp reduced parasite loads on fishes but the banded coral shrimp did not (McCammon et al. 2008). Another way cleaners can affect parasite populations is by changing the size of parasites on the host fish. The mean length of parasites from fish that did or did not have access to cleaning shrimp was quantified to test the hypothesis that Pederson cleaner shrimp and banded coral shrimp affect the average size of monogenean parasites on the blue tang fish (*Acanthurus coeruleus*). Blue tang were housed in an aquarium where they were constantly exposed to the infective stages of the parasite; the control treatment contained fish alone, and the experimental treatment included access to cleaner shrimps. After two weeks each fish was recaptured and put in a freshwater bath for five minutes to remove the parasites. Parasites were preserved in ethanol, photographed under a microscope and their lengths measured using NIH Image J. Pederson shrimp significantly reduced the average size of parasites on the blue tang ($p < 0.05$, t-test), suggesting that they prey selectively on larger parasites. Banded coral shrimp did not affect the mean parasite size on the blue tang ($p > 0.05$, t-test), suggesting that not all 'cleaner shrimp' provide a benefit to reef fish infected with monogeneans. The ability of cleaner shrimps to remove parasites, especially larger ones, should have a positive impact on fish health by reducing damage to the host's skin. In addition, reduced parasite size could reduce overall parasite populations on the reef by removing the parasites with the greatest reproductive output.

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Most Young U.S. Virgin Islanders Have Some Understanding of Coral

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It is very important that people possess some knowledge of coral. By understanding what coral is, how useful it is to the environment, and how our actions can affect coral in positive or negative ways, people may be able to alter certain behaviors and help protect the corals. To understand how much Virgin Islanders know, a representative sample survey was conducted in the summer of 2008 by interviewing VI residents, both in person and by web, about their knowledge of corals and the ocean. I wanted to see if males of the age range 18-30 knew more about corals than females of the same age range, or vice versa. Most young Virgin Islanders do have some understanding of coral. For example, 82% females and 74% males could correctly explain what a coral was. And when presented with three photos of ocean organisms, males correctly identified them with an average of 2.28, while females correctly identified the photos with an average of 2.45. The sexes did not differ significantly in knowledge of coral ($p=0.247118$ t-test ($p>0.05$), $p=2.210454$ chi-square test ($p>0.05$)). This suggests that most young Virgin Islanders have some knowledge and understanding of coral. This contrasts with a similar study in coastal Brazil in which only 60% knew what a coral was.

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Feeding behavior of juvenile *Diadema antillarum*, the long-spined black sea urchin

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Diadema antillarum plays an important role in the coral reef community as a keystone grazer. As such they eat algae which compete with coral for space on the reef. Their grazing maintains algal density to a low level allowing corals to flourish and facilitating the recruitment of new corals. However, in the early 1980's there was a great Caribbean wide die-off of the urchin by a still unknown pathogen removing 99% of the population. This created the opportunity for macro-algal cover on reefs to increase. Thus, the return of *Diadema* to reefs is important, but the feeding preference of the juvenile urchin, < 2 cm, is uncertain. Recent availability of the juvenile urchin in Brewers Bay, St. Thomas allowed for an assay in which its feeding preferences were assessed. All urchins and algal samples were taken from Brewers bay, which is adjacent to the University of the Virgin Islands St. Thomas campus. Four trials were conducted. Each trial with 30 juvenile urchins, each fed a single algal thalli for five days. Trials tested: the red alga *Acanthophora spicifera*, the brown alga *Dictyota menstrualis*, the calcified green alga *Halimeda opuntia*, the uncalcified with green alga *Caulerpa macrophysa*. Juveniles had distinct preferences: they ate large amounts of *Acanthophora* and *Caulerpa* but much less of the other species. This information may help managers wishing to return urchins to reefs.

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Generating Large Primes for the RSA Algorithm

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The RSA algorithm or cipher system was invented by Rivest, Shamir, and Adleman in the 1970's. It is a public key cipher system based on modular exponentiation where the keys are pairs consisting of an exponent e and a modulus N that is the product of two large primes p and q . It is an algorithm used to encode and decode messages. It depends on using two large primes which are kept secret and their product is made public. We will show how to find large primes and use them to explain the RSA algorithm and how it works.

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Effects of Oxotremorine on Central Control of a Peripheral Neural Circuit of the Caribbean Spiny Lobster, *Panulirus argus*

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The stomatogastric nervous system (STNS) controls all motor activity of the lobster's foregut and is comprised of four ganglia. The paired commissural ganglia (CoG) are located within connectives of the central nervous system and send axonal projections to two peripheral ganglia: the esophageal ganglion (EoG), and through a single nerve to the most distal stomatogastric ganglion (STG). The STG contains several motor circuits, one of which, the pyloric central pattern generator (CPG) produces rhythmic bursts of action potentials controlling muscles for food sorting. Central nervous system input is necessary for pyloric CPG activity. Cutting all connections from the CoG or EoG to the STG stopped bursting activity. While several neurons in the CoG are known to directly influence the pyloric rhythm, the pharmacology of these influences is not clear. Acetylcholine is an important inhibitory neurotransmitter in the pyloric CPG and acts through muscarinic type receptors. We hypothesized that control by higher motor centers, the CoG, involves neurotransmitters also used in peripheral centers. To test this hypothesis, extracellular and intracellular recordings of pyloric neurons were performed following superfusion of the CoGs with a cholinergic mimic, oxotremorine, at different concentrations ($1 \times 10^{-5} \text{M}$ – $7 \times 10^{-5} \text{M}$), for five minutes followed by a twenty minute wash with lobster saline. Intracellular recordings were taken from pyloric dilator cell bodies. We observed that the pyloric CPG had a wide range of responses to the oxotremorine at different concentrations. If the pyloric CPG had high bursting activity before oxotremorine was placed on the CoG, there was not a major change in the activity; but if the activity was slow, oxotremorine superfusion of the CoG increased the bursting activity. The excitatory effects of oxotremorine on the CoG are similar to previous studies done by Bal et al. Bal and his colleagues placed oxotremorine, on the STG. However, Bal et al. did not observe inhibitory effects by oxotremorine. In contrast, Spitzer et al. observed variable effects of using serotonin treatment on the STG that are similar to our results using oxotremorine on the CoG. We are exploring the possibility that oxotremorine effects converge with serotonin influenced mechanisms in the STG. Thus, we confirm that at least one neurotransmitter common to the STG also influences activity of higher centers known to control the pyloric CPG. Further we hypothesize that oxotremorine and serotonin have converging influences on the pyloric CPG.

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