

#### Name(s)

#### **Rohan Bhushan**

#### **Project Number**

## S1501

#### **Project Title**

## **Gravitactic Effects of Copper Pollution on Glucose Metabolism in Euglena gracilis**

#### **Objectives/Goals**

#### Abstract

Euglena gracilis is a very important part of the marine environment, providing food for many organisms in a similar way phytoplankton does. The organism also has many additional dietary uses and nutritional value for humans. Copper pollution in the Monterey Bay and other local water sources threaten this flagellate in ways that have not been appropriately studied, especially upon the organisms# ability to heterotrophically utilize glucose as a food source. In this project the effect of copper induced gravitactic disturbance on glucose metabolism was observed. I hypothesized that gravitactic agitation would lead to a decrease in glucose metabolism, resulting in reduced biomass and cell size.

#### Methods/Materials

E. gracilis was incubated in two batches CB (Copper Bath) and NC (Neutral Control), in media containing glucose (TMglu), no organic carbon source (TMnoc), glucose and copper (TMglc), and copper (TMcop). CB tests included a primary copper wash. Glucose concentrations were found using a glucose testing kit. A compound microscope was used to record and observe cells in samples. Cell count was found using an automated image analyzer to count the number of cells in a 160x photo, then this number was substituted into a formula I developed. Cell size was found using the same program. Upset Gravitaxis was verified by overlaying two photos of 1 second intervals, then observing the degree in change of orientation off of the preceding path.

#### Results

Glucose metabolism was adversely affected by the addition of copper with an initial decrease in TMglc. TMnoc was found to have a median biomass in comparison to other samples. The results show that copper is impairing Euglena#s ability to metabolize glucose, but once the Euglena is allowed to recover, metabolism is significantly higher than the normal rate. Gravitactic inhibition of photosynthesis was also observed, due to the low cell counts and sizes in TMcop tests.

#### **Conclusions/Discussion**

From the results I could conclude that metabolic rates were negatively impacted by upset gravitaxis caused by copper. In the future, methods for the extraction of paramylon would be developed. Copper pollution will continue to threaten the viability of this organism as a food source for marine organisms, but if there is a need to culture the Euglena for human dietary needs, the Euglena will be able to recover from copper impaired metabolism.

#### **Summary Statement**

I investigated how upset gravitaxis, caused by copper pollution, is affecting glucose metabolic rates in the flagellate Euglena gracilis.

#### **Help Received**

Received advice and help from Dr. James Barry and Dr. Shannon Johnson at the Monterey Bay Aquarium Research Institute and Dr. Shannon Johnson at UCSC.



Name(s)

Simon Cao

**Project Number** 

# S1502

#### **Project Title**

## Investigating the Synergism between Polyphenols and Macrolides: A Method to Combat Antibiotic Resistance?

#### Abstract

**Objectives/Goals** The objective is to compare the individual and combination effects of two common macrolide antibiotics (erythromycin and azithromycin) with two plant polyphenols capable of bacterial membrane damage and permeation (resveratrol and quercetin) against Escherichia coli, in order to test for possible synergism.

#### **Methods/Materials**

A broth microdilution method was used to find the minimum inhibitory concentration capable of inhibiting 50% of E. coli growth (MIC50) for each polyphenol and macrolide alone. By utilizing the microdilution method, the synergy test was done by combining macrolides and polyphenols in 1 to 1 ratios with regards to their respective MIC50 values. Control tests for solvent toxicity, growth, sterility, and chemical turbidity were performed.

#### Results

Alone, erythromycin, azithromycin, resveratrol, and quercetin had MIC50's of 0.77  $\mu$ g/mL, 0.0064  $\mu$ g/mL, 216  $\mu$ g/mL, and 162  $\mu$ g/mL, respectively. In the synergy test, the combinations showed either indifference or antagonism. The resveratrol and erythromycin combination showed no decrease in MIC50, with the inhibitory concentration of erythromycin remaining at 0.77  $\mu$ g/mL. The resveratrol and azithromycin combination showed antagonism, with the MIC50 of azithromycin increasing to 0.0114  $\mu$ g/mL compared to 0.0064  $\mu$ g/mL alone. The quercetin and erythromycin combination showed antagonism, with the MIC50 of erythromycin rising to 1.03  $\mu$ g/mL. The quercetin and azithromycin combination also showed antagonism, with the MIC50 of azithromycin combination showed antagonism, with the MIC50 of azithromycin combination showed antagonism, with the MIC50 of azithromycin combination showed antagonism, with the MIC50 of azithromycin rising to 1.03  $\mu$ g/mL. The quercetin and azithromycin combination also showed antagonism, with the MIC50 of azithromycin rising to 0.0114  $\mu$ g/mL.

#### **Conclusions/Discussion**

Alone, the polyphenols resveratrol and quercetin displayed potent antibacterial activity at high concentrations, showing their potential to act as a novel lead in drug development. However, they did not synergize with macrolide antibiotics. Instead, they had an antagonistic effect in most of the combinations, possibly because both the drugs and polyphenols competed for the same site of antibacterial activity. This unforeseen drug interaction suggested that resveratrol and quercetin, common in dietary supplements, should not be taken together with macrolide antibiotics.

#### **Summary Statement**

The plant polyphenols resveratrol and quercetin were combined with the macrolide antibiotics erythromycin and azithromycin to test for possible synergistic effects.

#### **Help Received**

Research was performed at Universal Biopharma Research Laboratory under the supervision of Dr. Khushoo and Dr. Thusu.



Name(s)

Augustine G. Chemparathy

### Project Number S1503

#### **Project Title**

### Accumulation of the Biodiesel Precursor Triacylglycerol Offsets Oxidative Stress in Chlamydomonas reinhardtii

#### Abstract

**Objectives/Goals** Microalgae accumulate the biodiesel precursor triacylglycerol (TAG) when subjected to nutrient stress, a phenomenon that has become the basis for the field of algal biodiesel. Despite the promise of this technology as a carbon neutral alternative to fossil fuels, low oil yields have constrained adoption. This research seeks to increase oil yield by identifying the process that directly instigates TAG synthesis, which would allow oil accumulation to be induced without starving cells. The putative safety valve function of TAG in relieving electron buildups in the photosynthetic apparatus is evaluated.

#### Methods/Materials

Thin-layer chromatography (TLC) and Gas Chromatography Flame Ionization Detection (GCFID) were used to identify four redox-defective strains, three of which also demonstrated high growth kinetics. Chlorophyll fluorescence showed that photosynthetic electron transport rate and photosystem II yield were enhanced at low light intensities in the high-TAG mutants and provided evidence of excessive electron accumulation in the photosynthetic apparatus when TAG synthesis was halted.

#### Results

Blocking fatty acid synthesis caused the photosynthetic electron carrier plastoquinone to become highly reduced. This result demonstrates for the first time a causative link between reduction pressure and TAG accumulation, and indicates that offset of electron accumulation is a major physiological role of TAG. Four knockout strains that are defective in putative electron carriers where identified and shown to produce up to 30% more TAG than wild-type, as well as higher growth.

#### Conclusions/Discussion

The elucidation of this major physiological role of TAG accumulation in microalgae opens avenues for genetic engineering to enhance biodiesel yields. The four strains identified by this study simultaneously demonstrate high growth and TAG hyper-accumulation, and thus hold significant promise for industrial applications. By specifically activating the pathway that senses abnormal electron accumulation in the photosynthetic apparatus and signals TAG accumulation, biodiesel production can be achieved with no growth constraints.

#### **Summary Statement**

This clean energy project demonstrated that biodiesel synthesis in algae protects the cell from deadly electron buildups, and used this phenomenon to identify new algal lines that produce up to 30% more oil than existing strains.

#### **Help Received**

I used resources and lab equipment at the Carnegie Institution for Science (Department of Plant Biology) at Stanford under the supervision of Dr. Xiaobo Li and Dr. Martin Jonikas.



Name(s)

Rebecca P. Chen

Project Number

# S1504

#### **Project Title**

### Determining Genetic Causes of Streptococcus pyogenes' Susceptibility to Retinoic Acid

#### Abstract

**Objectives/Goals** This project's goal was to find the genes that contribute to Streptococcus Pyogenes' susceptibility to retinoic acid, the active form of Vitamin A.

#### **Methods/Materials**

A genetic screen was used to find the retinoic acid resistant AP M1T1 GAS 5448 mutants created using a Transposon Tn917 library. This was done through screening the transposon library, alternating it in  $5\mu$ g/mL erythromycin antibiotic in Todd-Hewitt Broth (THB) and increasing concentrations of retinoic acid in 80% Roswell Park Memorial Institute media and 20% THB. After a manageable number of mutants were found, their genes disrupted by the transposon insertions were discovered using single-primer polymerase chain reactions, GeneWiz sequencing services, and ncbi.gov's nucleotide BLAST tool.

#### Results

To date, three genes have been identified that contribute to susceptibility to retinoic acid; CitG gene, Csn1 gene and UvrC from the mutants A9 Sequence L-2, B6 Sequence L-2 and B10 Sequence L-2, and D10 Sequence L-2 respectively.

#### **Conclusions/Discussion**

The CitG gene either codes for part of the GntR family of transcriptional regulators, or a catalyst of functions dealing with energy as ATP. The Csn1 gene codes for proteins associated with prokaryotic acquired immunity. The UvrC gene codes for 1 of 3 subunits of an endonuclease that participates in the nucleotide excision repair process. These genes and how they contribute to the bacterium#s susceptibility can be carefully studied further in the future to create novel non-antibiotic treatments of Streptococcus infections, thus reducing society#s dependence on antibiotics and slowing the proliferation of antibiotic-resistant bacteria.

#### **Summary Statement**

A genetic screen is used to discover the genes that cause Streptococcus Pyogenes' susceptibility to retinoic acid.

#### **Help Received**

Used lab equipment at University of California, San Diego under the supervision of Dr. Ericka Anderson; Dr. Jason Cole and Samira Dahesh created the transposon library for me to use.



Name(s)

Yingchen Chen; Yintung Chen

### Project Number S1505

#### **Project Title**

### A Comparative Assessment of the Inherited Resistance of Escherichia coli K-12 against Three Common Disinfectants

#### Abstract

This project compared the ability of bacteria Escherichia coli K-12 to develop inherited resistance against common household disinfectants, triclosan, isopropyl alcohol, and sodium hypochlorite, at different concentrations.

#### **Methods/Materials**

**Objectives/Goals** 

In the experiment, Escherichia coli K-12 were tested under different concentrations of three common disinfectants: triclosan, isopropyl alcohol, and sodium hypochlorite. 10 microliters of each disinfectant at varying concentrations were added via a 6.0 mm filter paper disk to agar plates newly streaked with E. coli. After 24 hours of incubation, the diameters of the zones of inhibition were measured in millimeters. The bacteria that had been exposed to the disinfectants were then transferred to new agar plates to be tested as the next generation. There were a total of three generations and three concentrations for each disinfectant.

#### Results

It was found that the zones of inhibition of triclosan solutions decreased sharply over three generations, while no appreciable decrease was observed for bleach and IPA solutions. For all trials, the percent deviations of the measurements were all less than 6 percent, so the trials were completed with precision.

#### **Conclusions/Discussion**

The hypothesis that triclosan would have effects on the inherited resistance of Escherichia coli K-12 was supported. E. coli K-12 demonstrated inherited resistance against triclosan in 3 generations, but no inherited resistance against IPA and bleach was shown.

#### **Summary Statement**

This project investigated the ability of bacteria Escherichia coli K-12 to develop inherited resistance against triclosan, isopropyl alcohol, and sodium hypochlorite.

#### **Help Received**

Parents helped acquire materials; Equipment borrowed from teacher and friend



Name(s)

**David A. Colton** 

**Project Number** 

# S1506

#### **Project Title**

## The Survival of the Probiotic Lactobacillus acidophilus in a Model Stomach System

Abstract

#### **Objectives/Goals**

This project tested the viability of the probiotic Lactobacillus acidophilus in the acidic environment of the stomach using an in vitro stomach model consisting of artificial gastric juice maintained in an anaerobic environment at 35oC. L.acidophilus was incubated in the model and samples were taken at time points reflective of minimal stomach transit. MRS agar plates were inoculated with the L. acidophilus samples and colonies counted at 2-11 days. As a control L. acidophilus was incubated in deionized water under the same conditions.

#### **Methods/Materials**

MRS Agar (Remel)

Tomato juice, yeast, milk medium and L. acidophilus (Carolina Biological)

Artificial gastric juice (Carolina Biological)

Anaerobic chamber with GasPak (Fisher Sci.)

L. acidophilus capsules (Sprouts)

#### Tums

#### Results

Data from experimental incubations in the stomach model with the probiotic L. acidophilus capsules shown in Figure 10 were analyzed using a 2-tailed unpaired t-test. Data is not shown from the time zero incubation, because there were no colony counts recorded for any of the data groups. Results indicate significantly increased colony counts at 30 and 60 minutes for capsules incubated in deionized water with and without Tums compared to gastric juice with and without Tums as well as gastric juice without pepsin (p<0.05). No significantly higher values were observed with any of the incubations containing gastric juice alone or with either Tums or in the absence of pepsin.

#### Conclusions/Discussion

Conclusions

L. acidophilus delivered as a capsule or liquid culture lost significant viability as measured by colony growth after incubation in an in vitro stomach model. This data supports the original hypothesis for this project. When L. acidophilus was co-formulated with the antacid it was found that there was a no statistically significant improvement in the viability.

This data, along with other probiotic research, hints that the viability of bacteria may not be the real reason that probiotics show significant health benefits.

#### Recommendations Summary Statement

Testing the survival rate of probiotics in a model stomach.

#### Help Received

Supervised in the school's lab.



Name(s)

**Grace M. Dailey** 

#### **Project Number**

## S1507

#### **Project Title**

### The Effect of Increasing Colloidal Silver Concentrations in Combination with Vancomycin against B. cereus and E. coli

#### Abstract

**Objectives/Goals** The antimicrobial properties of silver have been recognized and exercised throughout history. But with the discovery of modern antibiotics in the 1940s, the use of silver for wound treatment, Neisseria gonorrhoeae prevention, etc. declined. However, as antibiotic-resistant bacteria become more pervasive and threatening, silver could reemerge as a significant antimicrobial.

This project is designed to compare the efficacy of Vancomycin when combined with colloidal silver (V+CS) at various concentrations versus Vancomycin alone (V) against Bacillus cereus (Gram+) and E. Coli (Gram-) bacteria.

#### Methods/Materials

Colloidal silver (at 10 ppm) was added via micropipette to blank discs and 30 microgram discs of Vancomycin in various amounts. The V+CS(1) discs along with V(2) discs alone as a comparison, were set on inoculated plates and after 24 hours of incubation, the inhibition zones were measured. A larger inhibition zone indicated a greater efficacy.

#### **Conclusions/Discussion**

Colloidal silver, alone, did not create quantifiable inhibition zones. However, the addition of colloidal silver to Vancomycin resulted in larger mean inhibition zone areas for all concentrations in comparison to Vancomycin alone, with one exception(3). A trend appeared as increased amounts of colloidal silver added to Vancomycin corresponded to increased efficacies. The p-values for the higher V+CS concentrations also indicated statistical significance. However, it is critical to determine the minimum inhibitory concentration (the smallest amount required to inhibit bacterial growth) for added colloidal silver that will increase Vancomycin efficacy, so as to prevent agryria, the blue-grey turning of the skin caused by excessive silver use.

(1) Vancomycin discs administered with colloidal silver

(2) Vancomycin discs containing no colloidal silver. All V+CS discs were compared to V discs.
(3) The exception was V+CS (10-24) for Bacillus cereus. (10-24) indicates that 10 microliters of CS were administered onto the V disc 24 hours in advance prior to placement on the inoculated plate. The majority of the other CS additions were administered onto the V discs immediately prior to placement.

#### **Summary Statement**

My project is designed to see if the addition of colloidal silver to Vancomycin increases its efficacy, for if it does, its addition to existing antibiotics may help to address the growing threat which is antibiotic-resistant bacteria

#### **Help Received**

Mr. Ma helped with statistical analysis; The Branson Science Department provided materials; Neighbor helped edit sections; Kind brother allowed me to download the free-trial of Photoshop onto his computer and then use it to measure inhibition zone areas



Name(s)

Anna D. de la Rosa

**Project Number** 

# S1508

#### **Project Title**

## **Red Hot Fungi Fighters: Effect of Capsaicin Extracted from Kashmiri Chili Peppers on Growth of Aspergillus flavus**

#### Abstract

My objective is to determine the effect of capsaicin extracted from Kashmiri chili peppers on the growth of Aspergillus flavus. I hypothesized that higher capsaicin concentration would result in greater inhibition of fungal growth.

#### Methods/Materials

**Objectives/Goals** 

The effect of capsaicin on growth of A. flavus was determined by using different concentrations of capsaicin.

Acetone was used to extract capsaicin from dried Kashmiri chili peppers. The oily residue was then dissolved in varying amounts of vegetable oil to produce different concentrations of capsaicin solution. Agar plates were swabbed with the different concentrations of capsaicin solution. At the center of each plates was placed a  $1 \times 1 \times 0.2$  cm block containing the A. flavus. The plates were incubated between 24-28 degrees Celsius and the area of growth was measured daily for 5 days.

#### Results

After 5 days, agar plates swabbed with the highest capsaicin solution showed no signs of growth while the agar plates swabbed with the lowest concentration showed minimal growth. The agar plates swabbed only with vegetable oil (control) showed the most growth.

Results of ANOVA test on data show that there was a statistically significant difference between the treatments of various concentrations of capsaicin solutions and control.

#### **Conclusions/Discussion**

My results support my hypothesis that capsaicin extracted from chili peppers can inhibit the growth of A. flavus and is most effective in higher concentrations. This experiment shows that a spice can have other beneficial uses beyond adding flavor to one#s food.

#### **Summary Statement**

The project studied the effectivity of capsaicin extracted from Kashmiri chili peppers in controlling the growth of A. flavus, which is a known plant pest.

#### **Help Received**

Parents assisted in purchasing materials used for the project. Science teacher and advisor provided equipment and guidance on procedures.



**Project Number** 

S1509

Name(s)

Luke Elissiry; Elizabeth Vojvoda

#### **Project Title**

### Using Computer Simulation to Determine the Effect of Fatality Rate on the Spread of Disease

#### **Objectives/Goals**

#### Abstract

The purpose of this project is to create an accurate and useful computer model that portrays the effects of the spread of infectious disease and how different mortality rates allow the disease to affect populations.

#### **Methods/Materials**

A code was created on Netlogo, an agent-based modeling environment, using statistics and reason. The simulation was run using disease variables of SARS (Severe Acute Respiratory Syndrome) and varying mortality rates. Each of the different fatality rates was run in three trials of 1825 ticks.

#### **Results**

The results showed that lower mortality rates caused the SARS virus to infect more people; when the fatality rate was 0%, a median of 7295 million people got infected over 5 years, while when the death rate was 100%, the median was the lowest at 2360 million total infected people. The findings provide that more deaths occurred when the mortality rates were lower; at 100% death rate, there 2680 million deaths, and when the death rate was 5%, 3733.33 million people died.

#### **Conclusions/Discussion**

Because lower death rates allowed the virus to affect more people and survive for a greater duration, viruses can be expected to have evolutionary trends leading to lower mortality rates. While these lower mortality rates may benefit the individual, the results show that lower fatality rates are more harmful to the human race than higher death rates

#### **Summary Statement**

This project is based on using Netlogo to create a simulation of the spread of disease that portrays the effect of varying mortality rates on populations and can be used to predict possible viral evolutionary trends.

#### **Help Received**

Mrs. Groch helped us to focus our project; Used laptops and computers belonging to our parents; Parents helped create poster



**Project Number** 

S1510

Name(s)

Michelle Essien; Everett Kim

### Project Title Efficiency of Viral Phage T4 in Combating E. coli

#### **Objectives/Goals**

The scientific experiment, Viral Phage Efficiency in Combating Escherichia Coli B. will investigate an alternative treatment through the utilization of the viral phage, T4r. The objective of this project is to test the efficiency and pave the way for future studies in bacteriophage therapy and productions of disinfectants and pesticides. We believe that the viral phage T4r will effectively eradicate the Escherichia Coli B, infecting the bacterium exponentially until all of the bacterium have been infected, at which point, the virus will revert into a dormant stage.

Abstract

#### **Methods/Materials**

During the first phase of the experiment, E. Coli was scraped onto the petri dish in a zig-zag formation to produce variations in bacterial formation concentration. The second phase of the experiment tested bacteria cultured in non-uniform concentrations kept at a stable 98.7 degrees Fahrenheit in order to mimic circumstances within the human body.

#### Results

During the first phase of the experiment, E. Coli was scraped onto the petri dish in a zig-zag formation to produce variations in bacterial formation concentration. Viral phages were fairly efficient at eliminating and/or containing bacterial growth higher concentrations of the viral phage at room temperature. The second phase of the experiment tested bacteria cultured in non-uniform concentrations kept at a stable 98.7 degrees Fahrenheit in order to mimic circumstances within the human body. Under these circumstances the viral phage was very effective at killing off bacteria, with higher concentrations of viral phages killing or containing bacterial growth after 12 hours of observation, and all concentrations limiting or ceasing bacterial growth after 24 hours. These

#### **Conclusions/Discussion**

These results demonstrate that the viral phage may be an effective treatment of bacterial infections within warm-blooded organisms. Viral phage treatments may also prove fairly effective in applications such as pesticides, but further testing must be conducted to determine the ability of the viral phages to survive in inconsistent environments.

#### **Summary Statement**

Testingt the efficiency of the viral phage T4r at combating Escherichia Coli B.

#### Help Received

Advisor provided a labratory, some lab supplies, and advice.



Name(s)

Elan E. Filler

**Project Number** 

# S1511

#### **Project Title**

### **Transcriptional Regulators as Drug Targets for Treatment of C. glabrata Infection**

#### **Objectives/Goals**

Abstract

1) Discover transcriptional regulators of the fungus Candida glabrata that govern resistance to antimicrobial peptides and the antifungal drug caspofungin, 2) test the virulence of C. glabrata transcription factor deletion mutants that are sensitive to both antimicrobial peptides and caspofungin in the Galleria mellonella model, 3) identify potential antifungal drugs that inhibit these transcriptional regulators.

#### Methods/Materials

Screen library of 216 C. glabrata transcription factor deletion mutants by plating serial 10-fold dilutions of each mutant and the wild-type strain onto agar containing the antimicrobial peptide protamine or caspofungin. Use bioinformatics to determine the function of transcriptional regulators that were found to govern resistance to protamine and caspofungin. Adapt G. mellonella model of disseminated C. glabrata infection to test virulence of the transcription factor mutants, using survival as the endpoint. Use computer-assisted modelling, docking, and screening to identify potential antifungal drugs.

#### Results

Last year, 91 mutants were screened, identifying 3 transcriptional regulators that formed the SAGA histone acetyltransferase complex. This year, an additional 125 mutants were screened, identifying 6 mutants that were sensitive to both protamine and caspofungin. Bioinformatics showed that 3 transcriptional regulators formed the RPD3L histone deacetylase complex and the COMPASS histone methyltransferase complex. These complexes and the previously discovered SAGA complex govern expression of resistance genes by modifying histones. Virulence studies in G. mellonella showed that only the transcriptional regulator Ada2 of the SAGA complex was required for virulence. Computer screening identified sulfonamides as potential inhibitors of Ada2.

#### **Conclusions/Discussion**

The RPD3L, COMPASS, and SAGA complexes govern resistance by modifying histones, indicating that histone modification is a key mechanism by which C. glabrata resists antimicrobial peptides and caspofungin. Because Ada2 is important for both resistance and virulence, it is a promising drug target. Computer modelling and screening identified sulfonamides as potential Ada2 inhibitors. Because sulfonamides are already used to treat bacterial and parasitic infections in humans, they are promising antifungal drugs.

#### **Summary Statement**

In Candida glabrata, the transcriptional regulator Ada2 of the SAGA histone acetyltransferase complex is required for both resistance and virulence, is a promising drug target, and is likely inhibited by sulfonamide drugs.

#### **Help Received**

Used lab equipment at the Los Angeles Biomedical Research Institute under the supervision of Dr. Edwards.



Name(s)

Anita Garg; Hari Garg

Project Number

# S1512

#### **Project Title**

## A Survey and ANOVA Analysis of Ultra-Low Concentrations of Bacterial Contamination

#### Abstract

**Objectives/Goals** The objective of our project was to devise a method for the detection and analysis of ultra-low concentrations of bacterial contamination in public places, such as bathrooms and elevators, as well as water sources using a particle counter.

#### **Methods/Materials**

Materials: 84 cuvette sample tubes, SIM-FCS software, particle counter, pipettes, SYTO9 bacterial stain, fluorescent beads, cotton swabs.

Procedures: The independent variable was the sample source. The dependent variable was the number of bacteria that the particle counter detected.

There were several major steps that we took to investigate the problem. First, we validated the use of particle counter to measure bacteria by conducting a fluorescent bead serial dilution. Next, we collected samples from various public places such as the mens# and womens# bathrooms, the microwave, a keyboard, an elevator, a water fountain, and samples from several water sites in southern California. There were a total of 28 samples collected, and 3 trials were conducted for each of the samples. Residue on the cotton swabs was transferred to cuvettes filled with 1 mL of water. We measured the number of bacteria detected by the particle counter in each cuvette.

#### Results

We determined the calibration curve of the particle counter using the fluorescent bead data. We calculated an R<sup>2</sup> coefficient of determination of 0.987, which implies a strong correlation between bead concentration and the particle counter hits.

The highest amount of bacteria detected was from the microwave, and the lowest was from the elevator. **Conclusions/Discussion** 

ANOVA analysis of the data verified that the groups we measured were statistically different from each other. The R^2 coefficient of determination was able to validate the use of the particle counter as an accurate method to quantify the number of bacteria in public places.

The particle counter was able to quantify bacteria in public places in ultra low concentrations, from 10 to 105 bacteria per sample. We validated that the particle counter can be utilized for commercial applications, such as detecting pathogens indoors in places such as restaurants, hospitals, and clinical environments.

#### **Summary Statement**

Our project demonstrates that ANOVA and R<sup>2</sup> coefficient of determination is an effective way to analyze and validate bacterial contamination in ultra low concentrations.

#### **Help Received**

Teachers at my school guided me and reviewed my report.



Name(s)

**Quentin A. Gonzalez** 

Project Number S1513

#### **Project Title**

### Can Algae Be Used as an Ink Substitute?

#### **Objectives/Goals**

The purpose of this experiment is to determine if algae, specifically a species of green algae called Spirulina, can be used as an ink substitute and to find what abiotic factors and environmental conditions such as water temperature are best for cultivating algae.

Abstract

#### **Methods/Materials**

Algae, plant fertilizer, 5 gallon container, printer, ink cartridge, fish tank heater, air pump, air pump tubes. **Results** 

During this experiment, I collected some very interesting and conclusive results. From the data I have gathered, I found that it is best to cultivate algae in water that is approximately 80 degrees Fahrenheit, as it had the best growth rate and highest overall weight of the other two temperatures. At the end of the experiment, the Spirulina algae in the 80 degree water had gained and grown an additional 4.33 pounds of algae, greatly overtaking the 50 and 110 degree water. The algae in 50 degree water only added 1.67 pounds and the algae in 110 degree water only added 1.18 pounds, making it the worst performing of the three temperatures. I also observed that the algae in 80 degree water had the best growth rate compared to the other two containers. Surprisingly, the 50 degree water performed better in all aspects than the 110 degree water.

#### **Conclusions/Discussion**

In conclusion, I feel that my science project was successful. Through valid data and testing, I was able to justify and prove that my hypothesis was correct. Algae growth is, in fact, directly affected by the temperature of water that the culture is growing. My hypothesis was also correct in that it is best to cultivate algae in water with a temperature of approximately 80°F, as it provides the greatest potential for algae compared to the 50°F and 110°F water. In summary, optimal algae growth is seen when grown in waters of 80°F, followed by 50°F being the next most efficient, and then 110°F being the most ineffective of the three temperatures. Most importantly, however, I found that, from my experiment, it is possible to use algae as a substitute for common printer ink. Though it did not product the desired result that I had envisioned, I was still able to print the pigments of the algae onto a piece of paper, which in my opinion is a success, considering no one has ever experimented with this technology. The process of experimenting with algae can be improved, and I believe once it has, the potential for this technology is limitless.

#### **Summary Statement**

Use algae to print documents and replace ink sources.

#### **Help Received**

Marine biologist educated me and helped me grow algae.



Name(s)

Sophia M. Hewitt

#### Project Number

## S1514

#### **Project Title**

### **Optimizing Chromosomal Exposure for Fluorescence in situ Hybridization in Tetrahymena thermophila**

#### Abstract

**Objectives/Goals** 

The objective was to test whether a greater drop height onto slides before QFISH would expose more fully the chromosomal DNA.

#### Methods/Materials

Cell samples of Tetrahymena thermophila were grown for 2 weeks in an incubator. They were centrifuged, the supernatant was removed, various chemicals were added, including formaldehyde to fix the cells, and then 80 ul of the cell sample were deposited onto each of 5 different slides, with each sample dropped from different heights (0 cm, 10 cm, 20 cm. 30 cm, and 40 cm). Probes tagged with FITC and coding for T. thermophila telomeres (AACCCCAACCCC...) were added, and the slides were stored at 37 degrees Celsius for two days to give the probes time to hybridize. After this, VectaShielc, which contained DAPI, was added. The slides were observed under a confocal laser scanning microscope. The DAPI staining was detected (although the FITC tagged probes did not). Due to limited time on the microscope, only slides of 0, 30, and 40 cm could be observed. Computer software was used to analyze the results.

#### Results

The DAPI staining hilighted the macronucleus, and diameters from the drop heights of 0, 30, and 40 cm were compared, revealing no substantial difference. There was a lot of autofluorescence on the same channel as the FITC, so there were no results from the telomere probes.

#### **Conclusions/Discussion**

Based on this study, dropping cells from different heights does not seem to affect the diameter of their nuclei. Thus, the heights would not make probes more likely to attach to the DNA, observing results using FISH would not be any easier, and the results would not be improved. However, a small sample size could be skewing the results. Also, when running FISH on T. thermophila, FITC is not a good fluorescent marker. More research is necessary.

QFISH is a good method for measuring accurately the lengths of specific target DNA, lixe telomeres. A simple method for getting better results would be very beneficial in a lot of studies - for example, measuring to see if one had shortened the telomeres, another project I would like to work on. The idea behind this project was that dropping the cells from higher up would expand the nucleus, thus making more room for the probes to attach and brightening overall results. I will attempt to find a working method of improving my results and, using this, I hope to measure telomere lengths.

#### **Summary Statement**

For my project, I tested whether dropping cells of Tetrahymena thermophila from different heights onto slides before QFISH would expand the nucleus and provide more room for the probes to attach to the DNA, leading to more fluorescence.

#### **Help Received**

Research performed at LA Biohackers (public biology lab); Cory Tobin (supervisor, advice, assistance when handling formaldehyde); my parents drove me and got supplies; Beckman Biological Imaging Facility, CalTech (allowed me to use their microscope for 90 minutes without assistance)



Name(s)

Lauren M. Hinkley

**Project Number** 

# S1515

#### **Project Title**

## Effects of Calcium and Vitamin K on Microtubule Depolymerization in Chlamydomonas reinhardtii

#### Abstract

**Objectives/Goals** The objective of this project was to determine if calcium and vitamin K in the forms of menaquinone and phylloquinone had an effect on microtubule depolymerization as expressed through the flagellar motility of Chlamydomonas reinhardtii.

#### Methods/Materials

After culturing Chlamydomonas reinhardtii and preparing solutions of calcium, menaquinone, and phylloquinone in a consistent range of molarities, 1 milliliter samples of the Chlamydomonas were treated with 40 microliters of each of the treatment solutions. Ten images were captured over 20 seconds in order to compare the rate of movement of individual cells in the treatments. Solutions of combined treatments were prepared and tested using the same protocol as in the dose response. Each of these treatments were repeated 4 times to ensure the viability of the results.

#### Results

The only individual solutions which rejected the null hypothesis to suggest statistically significant movement of the cells (as compared to the control with no treatment) was Calcium at 500 micromolar after 30 minutes and 100, 250, and 500 micromolar after 60 minutes, all with p values of below 0.05. A t-test conducted to test for a statistical significance between the 30 and 60 minute analyses failed to reject the null hypothesis with a p value of 0.15. Two t-tests were conducted to tell whether there was a statistical difference between the calcium and the treatment, at both 30 and 60 minutes. The results failed to reject the null hypothesis at 60 minutes (p=0.06), but rejected the null hypothesis at 30 minutes (p=0.03).

#### **Conclusions/Discussion**

Calcium is the only treatment which, individually, has an effect on microtubule depolymerization. The movement of the cells treated with calcium was significantly less, likely due to that fact that calcium increases depolymerization meaning the microtubules shrink and therefore the flagella are shorter and cannot propel the cell as quickly. All of the calcium treatments were significantly different than the control after 60 minutes, but only the 500 micromolar calcium was significant after 30 minutes. This suggests that calcium takes more than 30 minutes to be absorbed by the cells. Menaquinone when combined with calcium sped up the effects of microtubule depolymerization. The results supported my hypothesis because calcium had the most effect on microtubule depolymerization which increased when calcium was combined with menaquinone.

#### **Summary Statement**

This project tests how calcium, menaquinone, and phylloquinone affect microtubule depolymerization as expressed through the flagellar motility of Chlamydomonas reinhardtii.

#### **Help Received**

Professor Merchant, Crysten Blaby, and Sean Gallaher of UCLA suppled Chlamydomonas reinhardtii and provided tips for culturing them.



Name(s)

Ray C. Huang

**Project Number** 

# S1516

#### **Project Title**

### Toward a Strategy for Extending Antibiotic Effectiveness Indefinitely: Introducing Antibacterial Bio-Restriction

#### Abstract

**Objectives/Goals** The objective is to see if antibacterial bio-restriction (a combination of conjugation inhibition, bacterial interference, and antibiotic) will inhibit the proliferation of the model resistant pathogen (E. coli TOP10F' tetR).

#### Methods/Materials

With TOP10F' (tetR donor) as the model resistant pathogen and TOP10 + pVIB (ampR recipient) as the model symbiotic, these two strains were co-cultured at an initial ratio of 1:1000 (F':pVIB). In this media was also 25% MIC of Ampiciliin and non-lytic phage M13 (independent variable). This culture was incubated for 24 hours, and then a 2  $\mu$ L inoculation was added into a fresh media with all the same additional compounds (excluding cells). This process continued over 4 days. Selective plating was used to calculate number of transconjugant, donor, and recipient cells. The differentiating factor between CONTROL and EXPERIMENTAL was the addition of phage M13. In a pilot study, phage M13 showed no significant lytic effect on the model pathogen (TOP10F').

#### Results

In the EXPERIMENTAL, the model symbiotic grew to an astounding  $3.02 \times 10^{11}$  cells/mL while the model pathogen lowered to negligible levels after just 2 days. In the CONTROL, the model symbiotic only grew to  $1.98 \times 10^{11}$  cells/mL, while the model pathogen and transconjugant grew to about 2.2 x  $10^{7}$  cells/mL and were still on the rise.

#### **Conclusions/Discussion**

The greatest danger of antibiotic resistance is the proliferation of resistant pathogens caused by selective pressure. Unlike previous therapies, the new strategy of antibacterial bio-restriction holds the hopes of extending antibiotic effectiveness indefinitely due to the fact that it targets the amount of space/ nutrients available for pathogen growth, a factor that pathogens have limited evolutionary control over despite natural selection. In addition, there are ramifications that may make this approach revitalizable if resistance was ever to occur. However, while only time can tell how "indefinite" this strategy may be, I believe that it's worth a try.

The results suggest that bio-restriction may be feasible and was effective within a controlled environment. Note that the model resistant pathogen wasn't targeted by any direct bactericidal compounds, but rather most likely died due to competition for nutrients.

#### **Summary Statement**

By rethinking antibiotic resistance through the use of a combination of antibacterial methods, extending the effectiveness of antibiotic indefinitely may be within reach.

#### **Help Received**

Universal Biopharma Research Laboratory supplied materials and minor troubleshooting assistance for the experiment.



Name(s)

**Garron W. Ireton** 

Project Number

# S1517

#### **Project Title**

### **Gentamicin Assay: A Study of the Effects of Gene Suppression on E. coli Infectivity Rates**

#### Abstract

**Objectives/Goals** To determine whether exocyst component targeting siRNAs, when introduced to fifty-fifth generation HeLa cells, will cause an effect on InIB-mediated entry levels of Listeria into HeLa cells.

#### **Methods/Materials**

HeLa (cancerous human cervix cells) cells were injected with various siRNAs, changing their genetic makeup. They were then infected with E. coli Yersinia bacteria, the growing plate was sterilized, and the human cells were broken open. The number of bacteria that had survived the sterilization by successfully infiltrating the human cells was counted and compared between the eight different groups of HeLa cells. The number of bacteria present after sterilization was indicative of the effect of the change the siRNA caused on cellular defense.

#### Results

The siRNA seemed to affect the entry rates, especially in the VAMP 3 well samples, decreasing the entry levels from the control siRNA sample#s 5 colony entry to 1.5 in one case. In response to these results, an unpaired t-test utilizing a two-tailed P value was used to compare the VAMP 3 values and the control siRNA#s values. This test resulted in a P-Value of .0376, meaning the VAMP 3 siRNA was significantly different from that of the control. This strongly suggests the siRNA did indeed make a difference in the entry levels of E. coli.

#### **Conclusions/Discussion**

While the data is statistically insignificant due to a lack of time to perform more tests, all tests performed point towards the alternate hypothesis of the siRNA causing a change in infection rates being correct. The fact that the siRNA affected samples had different entry levels when compared to the control strongly indicates that siRNA has a measurable effect on entry levels. Further tests conducted after my project was complete by the lab I had been working at support my findings.

#### **Summary Statement**

A study of the effects of small interfering RNA gene-suppression on the cellular defense of HeLa cells to E. coli Yersinia.

#### **Help Received**

Used lab equipment and learned basic sanitation procedure under the supervision of Dr. Keith Ireton



Name(s)

**Bowen Jiang** 

**Project Number** 

# S1518

#### **Project Title**

## Your Daily Plate of Algae: The Effect of Algal Hydrophobicity on Their Ability to Grow on Solid Media

#### **Objectives/Goals**

Abstract

The main objective of this experiment was to try and determine if cell surface hydrophobicity (CSH) of algae differs among taxa, and also what role hydrophobicity plays in how well algae grow on solid media, in order to possibly begin devising a method to grow certain species on plates which did not grow well in previous experiments.

#### Methods/Materials

The hydrophobicity of algal cultures of 26 species and strains from seven different phyla was tested by using the MATH (Microbial Adherence To Hydrocarbon) assay. This test involved creating an emulsion with water and a nonpolar solvent (decane), and measuring the difference in light absorbance of the culture between the non-mixed and mixed samples. In addition, samples of the same algal cultures were grown on agar plates with f/2 saltwater nutrient media, and the growth of algae on these plates was compared.

#### Results

All of the algae demonstrated wide differences in hydrophobicities, even among similar taxa. Even the same species or strain of algae demonstrated differences in hydrophobicity measurements; older cultures usually had lower hydrophobicities than younger cultures. Some cultures with low hydrophobicities demonstrated excellent growth on solid media; however, other species with higher hydrophobicities showed very good growth as well, and many species with only moderate hydrophobicities grew very little at all.

#### **Conclusions/Discussion**

The data collected in this experiment suggest that while CSH definitely plays a role in determining the growth preference of algae, it is not the only factor, and other cell surface properties likely also influence algal ability to grow on solid media.

#### **Summary Statement**

In this project, I compared the polarity of the cell surfaces of algae with their ability to grow on agar.

#### **Help Received**

All of the experiment was conducted in the lab of Dr. Gordon Wolfe at the Department of Biological Sciences at California State University, Chico.



Name(s)

#### CALIFORNIA STATE SCIENCE FAIR 2015 PROJECT SUMMARY

Joshua	R.	John

Project Number S1519

#### **Project Title**

### What Is the Effect of Dollar Bill Value on the Amount of Microbes Found Present on the Bill?

#### Abstract

The purpose of this project was to investigate any possible correlation between dollar bill denomination and the amount of microbes found on the bill.

#### **Methods/Materials**

**Objectives/Goals** 

Four common bill denominations: \$1, \$5, \$10, \$20, were tested for average microbes found per a bill type. I hypothesized that if the dollar value of a bill is related to the amount of germs found present, then the higher the dollar value of the bills, \$1, \$5, \$10, \$20, the lesser amount of germs are found present. To test this, five bills of each denomination were acquired, and swabbed on both sides, and cultured. The average CFU for each denomination was compared and determined the results.

#### Results

My results show that there is a correlation between bill value and the amount of microbes found present on the bill, but found greatest to least in the order of \$20, \$1, \$5, \$10 respectfully.

#### Conclusions/Discussion

This evidence shows that \$20 bills were the most contaminated, and \$10 bills the least contaminated. This is due to \$20 bills being transferred at a higher rate

#### **Summary Statement**

An Investigation of any possible correlation between dollar bill denomination and the amount of microbes found on the bill.

#### **Help Received**

none



Name(s)

Anthony K. Kang

Project Number

# S1520

#### **Project Title**

### **TraY-cing the Origins of Antibiotic Resistance Spread through F Conjugation**

#### Abstract

**Objectives/Goals** With the increasing problem of antibiotic resistance in bacteria, there is a need for a different approach to combat bacterial infections without the continual abuse of antibiotics. This experiment demonstrates the viability for a novel approach; inhibition of horizontal gene exchange within a bacterial population to slow or halt the spread of antibiotic resistance genes.

#### Methods/Materials

To emulate the effects of TraY inhibition on conjugation rates, a predictive MatLab simulator was written using conditional probabilities to determine the effect of a TraY mutation on the colony populations of F' and F- bacteria. Results were then lab verified using F' DH5alpha E-Coli with an inhibited TraY site to test its effect on tetracycline resistance distribution within the population. Two oligo sequences encoding different TraY gene regions were electroporated into experimental F' groups, which were then integrated into F- populations to conjugate before being transferred into selective tetracycline media. Population densities of surviving F' were finally quantified by spectrophotometry.

#### Results

Simulation results using 0%, 50%, 80%, and 100% mutation in TraY showed that higher mutation percentages in F' resulted in restricted F' population growth. In vitro data similarly displayed significantly increased containment of the antibacterial resistant genes by the F' bacteria when the TraY gene was restricted in both oligo groups compared to the untreated control F'.

#### **Conclusions/Discussion**

Based on ANOVA analysis for simulation and lab experimental results, the simulation and lab data showed matching significant variances between the TraY restricted experimental and non-restricted control groups. This indicates that TraY plays a significant role in conjugation and thus the spread of antibiotic resistance within a bacterial population.

Here, targeting F conjugation is shown to effectively slow the rate antibiotic resistance distribution within a colony. With future research, novel medications targeting such processes within bacterial infections could be developed, allowing for horizontal containment of antibiotic resistance within a smaller population of bacteria.

#### **Summary Statement**

My project addresses the problem of antibiotic resistance in bacterial infections using a novel approach focusing on the genetic transfer of resistant genes between bacteria.

#### **Help Received**

Primarily used the lab space and equipment at my high school. Used the electroporator machine in Dr. Alexandre de Andrade's lab at ThermoFisher Scientific.



Name(s)

Saumya R. Keremane

Project Number

# S1521

#### **Project Title**

## A Rapid Field Detection of Liberibacter Bacteria Using Lateral Flow Technology

#### Abstract

**Objectives/Goals** Citrus huanglongbing (HLB), a bacterial disease spread by the Asian citrus psyllid, has killed 50% of Florida citrus within ten years of introduction. Delayed detection and heavy pesticide use have resulted in serious epidemics and environmental pollution. The goal of this project was to develop an affordable, point of need testing to facilitate instant management decisions for growers and to complement regulatory efforts by helping them focus in high risk areas. Using a tomato model system, and an inexpensive reaction setup, a 30 min dipstick assay was developed using isothermal amplification and lateral flow technology. The results using the home detection system was comparable with lab-based assays. Since new species of Liberibacter have caused problems in laboratory diagnosis leading to epidemics in other countries in the past, a universal real time PCR detection system was developed using bioinformatic analysis of multiple bacterial genomes. Overall goal is to promote rapid early detection to facilitate better disease management and protect the citrus industry in California.

#### Methods/Materials

A \$20 prototype for isothermal amplification, Bst DNA polymerase, primers, primers labeled with biotin and FAM, dipstick assay components (provided by GE Healthcare), anti-FITC antibody conjugated to gold, streptavidin, anti-rabbit antibody, real time PCR machine, healthy and infected tomato, psyllid and bacterial DNAs.

#### Results

An isothermal amplification was developed in the lab and adapted for field detection using a \$20 prototype. A dipstick assay was developed capable of distinguishing healthy and infected plants and insects. Several primers developed in this study detected four species of Liberibacters, but not other closely related bacteria, in qPCR assays.

#### **Conclusions/Discussion**

A simple to use and affordable home detection system based on dipstick assay was developed in this study to help both growers and regulators to avoid delays in the current system of lab-based centralized detection system. It is expected to greatly help growers and home owners to manage their citrus better and for judicial use of pesticide. In 2004, Brazil failed to contain HLB because of emergence of a new species of bacterium. Using bioinformatics analysis of multiple Liberibacters and other closely related bacteria, universal primers capable of selective detection of multiple Liberibacter species was developed here for qPCR.

#### **Summary Statement**

A simple and affordable dipstick assay was developed for instant detection of plant pathogens to help growers protect citrus from the deadly huanglongbing disease and to encourage judicial use of pesticides.

#### **Help Received**

Laboratory facilites provided by Dr. Richard Lee. I appreciate the technical guidance I received from the lab members.



Name(s) **Project Number** Janie Kim S1522 **Project Title Developing a Novel Antimicrobial Using Contact Lens Solution Preservatives** Abstract **Objectives/Goals** This third year project tested a novel combination of antimicrobial contact lens solution preservatives, Benzyl Alcohol+EDTA+Chlorhexidine gluconate (CHD), for effectiveness against the Gram-negative bacteria MRSA and the Gram-positive Pseudomonas aeruginosa. The goal was to create a novel antimicrobial that was both effective and affordable. **Methods/Materials** This project tested the above preservative combination by performing serial dilutions to find the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC). The preservatives' dilutions started at concentrations similar to what they were in commercial contact lens solutions that contain them. CHD was started around 0.003%, Benzyl Alcohol around 0.5%, and EDTA around 0.5%. The combination columns added these together. The serial dilution was performed: the concentration of the preservatives was cut by half each well down. Bacteria were inoculated into the assay plate wells, and the MICs were determined after incubation. Results It was found that the MIC and MBC against both MRSA and Pseudomonas was 0.06769% total amount of preservative (0.03375% Benzyl Alcohol + 0.03375% EDTA + 0.00019% CHD). It was also found that the three preservatives were synergistic against Pseudomonas. **Conclusions/Discussion** It was discovered that Benzyl Alcohol, EDTA, and CHD combined have a synergistic effect against Pseudomonas. Despite no synergy against MRSA, the combination of the three was as effective against it as against Pseudomonas. The novel antimicrobial combination is also cost-effective (\$10 per oz), compared to the expensive but effective CHD (~\$200 per pure oz), or the comparatively ineffective but cheaper EDTA (\$15 per oz) and Benzyl Alcohol (\$4 per oz). This data suggests that using the combination found in this project as the preservative within contact lens solution would result in a solution effective against Gram-negative and Gram-positive bacteria. The preservative combination could also be used in other sanitation settings (e.g. healthcare and hospitals), and would be invaluable as an effective, low-cost antimicrobial in developing countries. **Summary Statement** This project tested the combination of Benzyl alcohol+EDTA+Chlorhexidine gluconate against MRSA and Pseudomonas aeruginosa, and discovered its effectiveness against Gram-negative and Gram-positive bacteria as an affordable antimicrobial. **Help Received** Professor Victor Nizet graciously provided a lab to work in, and Mr. Leo Lin guided me through my

Professor Victor Nizet graciously provided a lab to work in, and Mr. Leo Lin guided me through my experiment. Parents drove me to UCSD to conduct my experiment. Mrs. Elaine Gillum gave me advice on my project.



Name(s)

Kai Kirby; James Schulfer; Kirk Tada

**Project Number** 

# S1523

#### **Project Title**

## **Electric Fungi 2.0: A Practical Approach to the Propagation of Fungi as a Food, Medicine, and Cure for Hunger**

#### Abstract

**Objectives/Goals** The effect of electricity on the growth of fungi was tested. Using various types of fungi and several different voltages of electricity, we analyzed the resulting effects on growth patterns. This is important because of the possibility of growing fungi for food and medicine faster and more efficiently, using electrical shocks.

#### **Methods/Materials**

Using a variable output transformer, we shocked several samples of oyster mushrooms, pioppino mushrooms, and bread mold when they were still spores at voltages of 30 V, 60 V, 90 V, and 120 V, leaving a control group unshocked. We measured their growth each day for a trial period varying from 13 to 40 days depending on the type of fungi, and analyzed their results at the end.

#### Results

We found that, overall, electrical shocks made the fungi grow anywhere from 2-15% better.

#### **Conclusions/Discussion**

The discovery of this phenomenon is very useful for the production of various foods and medicines. Overall, our hypothesis that electrical shocks help the propagation of fungi was correct. There are several reasons for why this is, ranging from electricity activating growth enzymes to the importance of electrical shocks in intramycelium communication, an interesting phenomenon to continue to study.

#### **Summary Statement**

We discovered that by shocking various types of fungi with varying voltages, their growth is positively effected.

#### **Help Received**



Name(s)

Varun R. Mandi

Project Number

# S1524

#### **Project Title**

### Novel HIV & Tuberculosis Combination Therapy: Proliposomal Formulation of Efavirenz & Glutathione for the Dual-Infected

#### Abstract

**Objectives/Goals** With over 37 million people affected by the HIV virus globally and subjected to ineffective treatment, the current problem of low bioavailability of antiretroviral drugs must be addressed. To aid, is the antioxidant glutathione which impairs function of Mycobacterium tuberculosis. The objective is: will a proliposomal formulation of the antiretroviral effavirenz and the antioxidant glutathione confer higher bioavailability thereby aiding HIV & Tuberculosis infected individuals?

#### Methods/Materials

500mg of Efavirenz was used, with 500mg glutathione antioxidant. To stabilize lipid membranes, 125mg cholesterol was used. After finalizing a 1:1:2:0.25 ratio of GSH:EFZ:DMPC lipid: cholesterol, 1 gram of DMPC lipid was required for the formulation. 600mL of ethanol and 400mL nanopure water were required to dissolve constituents of the formulation in; this solution was rotated in a Rotovapor apparatus above 50C water bath to evaporate solvent from the drug-containing liposomes. 212.5mg of dried powder obtained was filled in capsules and dissolution (in vitro) testing conducted with a 6.8pH sodium-phosphate buffer at 37C. The dissolutions of three formulation capsules were conducted in parallel to those of three pure-drug capsules (control). Samples of liquid media of each of the six vessels were taken at 15,30,45, and 60 minutes. These samples were run through HP 1100 HPLC for high pressure liquid chromatography analysis. Malvern Zeta and particle sizing was also conducted to ensure drug encapsulation, and lipid membrane stability.

#### Results

On average, the control capsule released 30.26% of its glutathione amount, and 26.81% of efavirenz content at the end of an hour (Vessels #1-3). In dissolution Vessels #4-6, the formulation released 57.13% and 65.14% of GSH and EFZ drug respectively. Particle size of empty liposomes read 490.8 nm, whereas the formulation particle size read 728.1 nm. 11.4mV average Zeta potential was obtained for formulation membranes, indicating membrane stability and that aggregation/disintegration will not occur.

#### **Conclusions/Discussion**

My hypothesis was affirmed by results found, as a formulation of efavirenz and glutathione conferred higher bioavailability. Glutathione bioavailability of the formulation was 188 percent of the control, and efavirenz absorption 242.97 percent of the control. These results promise lesser use of antiretroviral equals affordability and effective cures for patients.

#### **Summary Statement**

Solving current underlying problem of low drug absorption in HIV patients using a novel method, while providing a Tuberculosis cure to immunocompromised AIDS patients.

#### **Help Received**

Dr. Guru Betageri of Western University of Health Sciences provided me with the lab equipment and funding required to conduct my independent experiment, thus encouraging my unique scientific pursuits.



Name(s)

**Advait Patil** 

**Project Number** 

# S1525

#### **Project Title**

## **CryoSubtilis: A Synthetic Biological Approach to Engineering Cold Resistance in B. subtilis**

#### **Objectives/Goals**

Abstract

B. subtilis is a popular host for the manufacture of enzymes, mainly due to its high competency and status as a GRAS organism. However, there are severe drawbacks to using it as a host for biomaterial production. The cost of maintaining bioreactors and incubators to grow B. subtilis is extremely high, and inadequate disposal of antibiotics used to select for transformed bacteria is one of the leading causes of pathogens gaining resistance to them. The objective of this project was to increase the cold resistance of B. subtilis through expression of CspC and ZeAFP proteins. By making B. subtilis cold resistant, both problems can be solved in one go.

#### Methods/Materials

The genes for the CspC and ZeAFP proteins were inserted into the plasmid vector pHT01 through a series of cloning steps, resulting in two plasmids: pHT01-CspC and pHT01-ZeAFP. Both plasmids were transformed into B. subtilis. Then, the OD (optical density), number of surviving cells, and survival rate was calculated in response to 4 cold shocks (10 minutes on ice for each cold shock).

#### Results

There is a clear difference between the survival rates of the transformed and untransformed bacteria. After the first cold shock, there was a 37.27% difference in the survival rates, and this trend continues. Over time, the difference in the percentages of living cells averages out to 40%. This means that after 4 cold shocks, the bacteria expressing both proteins are 12.06 times more likely to survive than bacteria without the proteins.

#### **Conclusions/Discussion**

The results presented support the hypothesis that the expression of these two proteins will increase the cold resistance of B. subtilis. These proteins can now be used to replace antibiotics as a selecting agent, developing a self-contained means of selection for transformation. They can eliminate the need for incubators by changing the optimal growth temperature of organisms, and can make organisms such as plants more cold resistant.

#### **Summary Statement**

This project aims to increase the cold resistance and tolerance of the bacterium B. subtilis through the expression of CspC protein from P. irgensii and ZeAFP protein from Z. elongatus.

#### **Help Received**

Parents drove me around from place to place; Mr. Lee helped throughout the project; Stephanie Yang of the Wyss Institute helped with questions about experiment design; Dr. Kenny Mok of UC Berkeley helped troubleshoot failed transformations.



Name(s)	Project Number
Titus M. Patton	S1526
	31520
Project Title	
Antimicrobial Properties of Stingray Mucus	
Abstract	
<b>Objectives/Goals</b> Previous research has suggested that wound healing properties in stingrays n	hav be due to an antimicrobial
substance in the animals epidermal mucus. To investigate this, the minimum	inhibitory concentration
(MIC) of the mucus against the various strains was determined and compare Methods/Materials	d.
Fresh epidermal mucus was collected from 16 Cownose stingrays. Tubes of	
with six common bacterial strains (E. coli, P. flourescens, B. subtilis, S. aure luteus). For the MIC assay, varying concentrations of mucus were inserted in	
control 4 tubes containing only the varying concentrations of stingray mucus	
The test tubes were incubated for 24 hours and then optical density was mea	
the MIC. In addition, disk diffusion assays were conducted to determine the against the same six bacteria.	effectiveness of the mucus
Results	
No inhibitory ring was clearly defined around any disc placed on the cultures scene to suggest partial inhibition. The minimum inhibitory concentration was	
than originally expected.	
<b>Conclusions/Discussion</b> This project showed that Cownose ray mucus could still be a possible answe	r to the problem of antibiotic
resistant bacteria. Through this study many methods and procedures were im	
improved, to ensure better results in future research.	
Summary Statement	
This project analyses the effectiveness of stingrays epidermal mucus against determine if it is a possible source to combat the rising problem of antibiotic	
Holp Dessived	
Help Received Used lab at Universal Biomedical Research Laboratory under the supervision	n of Amardeen Khushoo.

PhD.; Brian Tsukimura, PhD advised project; Andrew Strankman advised project



Name(s)

**Ruby A. Rorty** 

**Project Number** 

# S1527

#### **Project Title**

### **Risky Business: Using Physarum polycephalum to Model Comparative** Evaluation

Abstract

**Objectives/Goals** Purpose: I used the single cell, multi-nucleate slime mold Physarum polycephalum to model comparative evaluation risk-taking behavior. Physarum were presented with two food sources of varying concentration placed in the light or dark. Physarum are adverse to light; therefore, this experimental design allowed me to determine whether they would choose low-quality food in the dark over high-quality food in the light (a risky choice). Comparative evaluation is a form of decision-making in which assets are weighed based on their pros and cons.

Investigative Question: Will Physarum polycephalum choose higher quality foods in a brightly lit, risky environment over lower quality foods in a dark, safe one?

Hypothesis: The Physarum will avoid risky environments, even at the cost of foregoing high-quality food.

#### **Methods/Materials**

Materials: Agar, Ground oats, 80 petri dishes, Physarum polycephalum colony, Camera, Black Paper Methods: A Physarum-covered oat flake was placed at the center of a 2% agar dish between two discs of food, each containing a specific percentage concentration of oatmeal, and each 2.5 mm away. The Physarum were offered two concentrations of oatmeal (1% versus 1%, 1% versus 3%, 1% versus 5%, and 1% versus 10%). A control experiment was performed in the dark (n=4 plates/condition). Three risk experiments (n=4/condition) were performed in the light, but with ½ of each plate in the dark. Data were collected at 24, 36 and 48 hours.

#### Results

After 48 hours, the Physarum in trials at 1v1 and 1v3 concentrations showed no consistent preference towards the light or dark choice. Only when food quality increased to 1v5 and 1v10 did all Physarum samples move towards the risky, high-quality, food choice. In the dark control, Physarum chose the high quality choice in every situation but in 1v1 moved equally towards both.

#### **Conclusions/Discussion**

My results demonstrate that a 4% or greater difference in food quality caused Physarum to choose the better food, hazarding the risky environment. These data suggest that a complex neurological make-up is not required for an organism to exhibit comparative evaluation.

#### **Summary Statement**

In this project, I used the slime mold Physarum polycephalum to model decision making behavior.

#### **Help Received**

Agar was poured in Hinck Lab at UCSC.



**Project Number** 

S1528

Name(s)

Akshay K. Srivastava

#### **Project Title**

### The Effects of Temperature on Exoelectrogenic Bacteria with Applications to Microbial Fuel Cell Performance

#### Abstract

The purpose of this experiment was to determine the sensitivity of exoelectrogenic bacteria to temperature, especially with regards to their abilities to facilitate electron deposition on extracellular electrodes at high temperatures.

#### Methods/Materials

**Objectives/Goals** 

In this experiment, a two chamber microbial fuel cell was constructed by immersing one iron electrode into a 200 mL. solution rich in exoelectrogenic bacteria and another identical electrode in a solution of distilled water. The two electrodes were then connected to a multimeter, which measured potential difference, and the two chambers of the fuel cell were connected using a KNO3 salt bridge to allow an exchange of charges between the two chambers. This apparatus was then heated at a constant rate using a hot plate, and periodic temperature and voltage readings were taken.

#### Results

It was found that in the early stages of heat treatment, microbial fuel cells display a strictly increasing set of voltage outputs. However, at some specific temperature, which varied largely from trial to trial, voltage output sharply decreases to zero. After this point, no further voltage was produced by the fuel cell, suggesting that the bacteria themselves had been permanently damaged at this point.

#### Conclusions/Discussion

In this experiment, it was found that exoelectrogenic bacteria display a high sensitivity to temperature, but one that also exhibits a large variance. This suggests that cell death is not the main cause of the sudden loss voltage observed, as bacteria were shown to survive beyond the temperatures of some of the voltage drops, but rather some sort of premature damage to the bacterial structures involved in electron mediation. However, in order to confirm these findings and further understand possible solutions to this problem, further studies on the structures of exoelectrogens involved specifically in electron mediation, such as the nanowires of Geobacter, would have to be conducted.

#### **Summary Statement**

The electron mediation structures of exoelectrogens are sensitive to temperature.

#### **Help Received**

Worked in high school laboratory under supervision of Kathleen Meyer; Had some discourse with Professors Glenn Hicks and David Carter at UCR



**Project Number** 

S1529

Name(s)

Chaitra S. Subbarao

#### **Project Title**

### Molecular Detection of Peronospora effusa during the Latent Period

Abstract

#### **Objectives/Goals**

Downy mildews are a serious disease of many crops in coastal California. In spinach, downy mildew is caused by Peronospora effusa. In California, where nearly 75% of the US fresh spinach is grown, downy mildew disease causes millions of dollars in losses annually. Currently, the disease is controlled by fungicide applications, and some of these fungicides are applied whether or not infection has occurred on spinach because of the explosive nature of these epidemics. Thus, I hypothesized that if downy mildew infection could be detected during the latent period, fungicide applications could be targeted only when the pathogen is present. This in turn may prevent epidemics from developing.

#### Methods/Materials

For specific detection of P. effusa or Peronospora, polymerase chain reaction (PCR) with P. effusa-specific primers were used to detect the pathogen DNA from potential infections on 50 samples weekly for six weeks. Spinach plants were randomly sampled every 3 m from a 36 m plot, consisting of four beds. For the first four weeks, primers to detect generic Peronospora species and for the last two weeks, primers specific for P. effusa were used in the PCR. Following the PCR, gel electrophoresis was performed to determine which of the fifty samples that week carried the pathogen.

#### Results

#Spinach leaf samples collected four weeks after crop emergence were asymptomatic. However, Peronospora spp.-specific PCR detected the presence of the pathogen in three of the 50 samples collected and processed, demonstrating that the pathogen could be detected during the latent period.

#Six of the 50 samples collected five weeks post-emergence were symptomatic and carried the pathogen. Three more samples had chlorotic spots without sporulation also contained P. effusa.

#By six weeks post-emergence, 28 of the 50 samples were symptomatic and strongly positive for P. effusa. Six more samples that provided weak bands were asymptomatic. This was indicative of the utility of the PCR in pathogen detection during latent and infectious periods.

#By week nine, all samples exhibited downy mildew symptoms and carried the pathogen by PCR.

#### **Conclusions/Discussion**

In conclusion, using PCR, downy mildew pathogen DNA was detected on symptomless spinach plants, and application of fungicides on these plants is expected to prevent further downy mildew development. The approach requires additional validation in larger commercial fields.

#### **Summary Statement**

If downy mildew infection can be detected before symptoms appear (during the latent period), fungicide applications could be targeted only when the pathogen is present, which may prevent epidemics from developing.

#### **Help Received**

Dr. Steve Klosterman and Mrs. Amy Anchieta (USDA Agricultural Research Service, Salinas CA) accommodated me in their laboratory and guided me through all phases of my study.



Name(s)

Cecilia C. Wolf

Project Number

# S1530

#### **Project Title**

### The Effect of Amount of Dextrose Sugar on the Carbon Production of Bakers, Brewers, and Champagne Yeast

#### Abstract

To learn how the amount of dextrose sugar given to bakers, brewers, and champagne yeasts affects their carbon production.

#### Methods/Materials

**Objectives/Goals** 

I put the three different yeasts into test tubes along with the desired amount of dextrose and 100 ml of warm water. I measured their carbon output by placing a balloon over the top of the test tube and calculating the volume based on it's diameter after 30 minutes.

Materials: yeast, water, dextrose, round balloons, tape measure, test tubes...

#### Results

I found that the champagne yeast was the most productive when given 1% and 5% dextrose, and that the bakers yeast was the most productive when given 20% dextrose. As I expected, the brewers yeast was the least productive for all amounts of dextrose. All of the yeast were most productive when given 20% dextrose, except for the champagne yeast, which was most productive with 5% dextrose.

#### **Conclusions/Discussion**

My hypothesis was that the bakers yeast would be all around the most productive, and that all of the yeast-except the champagne yeast- would produce the most carbon when given 20% dextrose. The trends indicate that bakers and champagne yeast produced much more carbon than the brewers yeast. I got these results for several reasons. First, the active yeasts (bakers and champagne) were much more productive than the inactive yeasts. Second, the populations of the champagne yeast appeared to exceed the carrying capacity due to exponential growth in the 20% solution. Third, the increase of food source generally increased productivity Also, the type of yeast and what their purpose is.

#### **Summary Statement**

To learn how the amount of dextrose sugar given to bakers, brewers, and champagne yeasts affects their carbon production.

#### **Help Received**

Advised by teacher, mom helped gluing poster board