Biology (and Biology related collaborations) student abstracts 2010
(Use the Ctrl + F keys to search for particular keywords or faculty)

The Effects of Inhibition of Transient Receptor Potential Canonical (TRPC) Channels on Calcium Signals in and Migration of Glioma Cells

Tia Barclay, Pamela Hanson, Harald Sontheimer, and Valerie Bomben

Gliomas are a type of brain tumor derived from glial cells, which outnumber neurons and support neuronal activity. Glioma cells have been extensively investigated to understand mechanisms by which they proliferate and migrate; glioma cells utilize ion channels found on the cell surface to shrink and facilitate easier migration; it is believed that successful glioma cell migration is dependent on a non-selective cation channel known as a transient receptor potential canonical channel (TRPC). It is therefore hypothesized that inhibition of the TRPC channel, via pharmacology or shRNA-mediated suppression of the TRPC gene, will decrease cellular migration. Immunocytochemistry first determined the presence of TRPC1 on the glioma cell surface, and Transwell migration assays with the glycoprotein victronectin were then used to determine if pharmacological inhibition and inducible shRNA constructs decrease migration of glioma cells. Two-dimensional microscopy was also used to track the basal rates of migration of glioma cells with the shRNA knockdown of TRPC1. Finally, spinning disk microscopy was used determine if Ca^{2+} signals in glioma cells are decreased with pharmacological inhibition of the TRPC channels. The proposed research on the TRPC channels in glioma cells will give more insight into more specific chemotherapeutic practices in patients suffering from gliomas.

Increased Tryptophan Uptake as a Means of Phytosphingosine Resistance in *Saccharomyces cerevisiae*

Taylor Binyon and Pamela Hanson

Phytosphingosine (PHS) is a sphingolipid that inhibits growth of *Saccharomyces cerevisae* because it inhibits the uptake of amino acids, particularly tryptophan. For this reason, increased expression and/or function of the tryptophan transporter Tat2p creates phytosphingosine resistance. The deletion of *CSG2,* a gene required for biosynthesis of complex sphingolipids, creates a resistance to phytosphingosine. It was hypothesized that the *csg2Δ* strain deletion creates this resistance due to an increased expression and/or function of the tryptophan transporter Tat2p. Through two experiments this project confirmed the role of *TAT2* in the *csg2Δ* mutant strain's resistance to phytosphingosine. First, PHS resistance and the increased growth in low concentrations of tryptophan were confirmed in the *csg2Δ* strain by comparing the growth of wild type and *csg2Δ* strains on media containing either variable concentrations of PHS or variable concentrations of tryptophan. Next, the project reversed phenotypes in the *csg2Δ* strain by removing the Tat2p tryptophan transporters through the deletion of the *TAT2* gene. By deleting the *TAT2* gene from the *csg2Δ* strain, the wild type phenotypes returned, specifically high PHS sensitivity and little to no growth on low tryptophan. Since the deletion of *TAT2* reversed the *csg2Δ* phenotypes*, TAT2* was likely behind PHS resistance and increased tryptophan uptake. *TAT2*'s role was further confirmed when *tat1Δ* strains faired moderately well in low PHS concentrations and grew readily in low concentrations of tryptophan. This

showed that Tat2p, was the primary transporter responsible for the c*sg2Δ* strain's PHS resistance and growth in low tryptophan concentrations.

Use of *Saccharomyces cerevisiae* as a Model to Determine Whether $[Ru(phen)_2(qdppz)]^{2+}$ Perturbs Topoisomerase Function

Luke Chiou, Laura Stultz, and Pamela Hanson

Interest in metal based cancer drugs has increased since the success of cisplatin as an antitumor drug. $[Ru(phen)_2(qdppz)]^{2+}$ also known as (Ru(qdppz)) shows promise as a anticancer drug due to its binding DNA and topoisomerase I *in vitro*. Topoisomerase (top) is a crucial enzyme used during DNA synthesis to relieve knots or supercoils in the DNA and is usually expressed at high levels in proliferating cells such as tumors. This study focused on the ability of Ru(qdppz) to inhibit or poison top I and II *in vivo* using baker's yeast *Saccharomyces cerevisiae* (*S. cerevisiae*) as a model. Specifically, wild type and top II overexpressing strains were used to test the ability of Ru(qdppz) to perturb top II function, while *pdr1*Δ*pdr3*Δ and *pdr1*Δ*pdr3*Δ*top1*Δ mutants, were used to test for top I perturbation. Minimum Inhibition Concentration (MIC) Assays were performed to determine IC_{50} values of the respective strains in the presence of known top I poison camptothecin, top II poison m-AMSA, and Ru(qdppz). As expected, the top II overexpressing strain was more sensitive to m-AMSA than the control. The average IC_{50} values for Ru(qdppz) are 7.51μM and 7.83μM for WT and top II overexpressing, respectively. Consistent with previous studies, our *pdr1*Δ*pdr3*Δ*topI*Δ mutant was more resistant to camptothecin than the *pdr1*Δ *pdr*3Δ control. The average IC_{50} values for Ru(qdppz) are 3.09μM and 3.29μM for *pdr1*Δ*pdr3*Δ*topI*Δ mutant and *pdr*1Δ *pdr*3Δ, respectively. The IC_{50} values for Ru(qdppz) are not significantly different between the isogenic pairs of strains; thus, Ru(qdppz) does not perturb top I or top II *in vivo*.

Avian Attack Rates on Models of Juvenile and Adult Sizes of Aposematic and Palatable Frogs

Kelly Gronemeyer, Megan Gibbons, and Ralph Saporito

Both warning coloration (i.e., aposematism, indicating unpalatability) and large body size are associated with increased conspicuousness to predators. These findings suggest that large, brightly colored individuals emit a stronger warning signal than small individuals with similar coloration. If body size in aposmematic species is positively related to toxicity, it is advantageous for predators to associate large body size with low palatability, and small individuals should receive more predator attacks than larger individuals. On the other hand, greater conspicuousness in palatable, cryptically colored species is disadvantageous, such that large individuals should receive more predator attacks than smaller individuals. We conducted an experiment at La Selva Biological Station in Costa Rica using different sized models of the aposematic *Oophaga pumilio* (strawberry dart-poison frog) and frogs of the genus *Craugastor* (a common palatable species) to determine if color and body size influenced avian attack rates. Large and small models (representing the coloration patterns of *O. pumilio* or *Craugaster*) were placed throughout secondary forests; the models were then collected and examined for markings that indicated predation attempts. Color did not influence predator

attack rates. However, size did have a significant effect: more brown adult than brown juvenile models were attacked, suggesting that an increase in conspicuousness of cryptic species increases predation rates. Further research should investigate the relationship of toxicity and body size in aposematic species.

Elevational Changes in Community Composition in Montane Longleaf Pine Forests, Oak Mountain State Park, Alabama

Ruth Hanks and Scot Duncan

Longleaf pine (*Pinus palustris*) forests once covered 37 million hectares but have since been reduced to 3% of the original range due to land conversion, logging, and fire suppression. The montane longleaf pine forest is among the least well known of the longleaf communities. Studies throughout the world have shown that elevation plays an important role in community composition. We sought to determine how elevation affects longleaf community composition within two elevational gradients in Oak Mountain State Park, Pelham, Alabama. The longleaf forests of Double Oak Mountain have endured many decades of fire suppression and extensive logging between the 1930s and 1960s. Twenty-six 20x50m plots were established, 13 on the mountain's slopes and 13 on the peaks of the neighboring foothills. Within each plot, the species and diameter at breast height of adults and basal diameter of juveniles of all trees were recorded. Regression analyses related frequency and size data to elevational data taken from GPS and topographic maps. Preliminary results suggest that several species show significant relationships with elevation but directionality varies among species and age groups. It is hoped that these findings will aid in the planning of longleaf preservation and restoration strategies.

The Examination of Constitutive and Induced Alkaloids of *Baptisia tinctoria* and *Baptisia lactea*

Rachal Jones and Peter Van Zandt

Plants present two forms of defenses: constitutive and induced. Constitutive defenses involve either physical or chemical characteristics of the plant that are always present, while induced defenses are usually chemicals produced by the plant as a response to herbivory. Alkaloids are nitrogen containing, organic compounds that can be toxic or distasteful to herbivores and may therefore be defensive. Different species of legumes produce different kinds and levels of constitutive and induced alkaloids. In a preliminary study, I found that herbivorous caterpillars (*Spodoptera dolichos*) were reluctant to eat *Baptisia tinctoria*, but readily consumed *B. lactea*, suggesting that these plants differ in their defensive chemistry. To assess the difference in constitutive and induced defenses in these species, I grew ninety-eight plants in a greenhouse for thirteen weeks before allowing *S. dolichos* to damage the plants at levels of 10%, 20%, 30%, 40%, and 50% of total leaf area. Control plants were undamaged and used to assess the constitutive levels of alkaloids. I then extracted alkaloids using an acid-base protocol and quantified them with gas chromatography. Preliminary results indicate that alkaloids in both species of *Baptisia* increase as levels of damage increase and the *B. tinctoria* contains higher concentrations of alkaloids than *B. lactea*. These results suggest that these alkaloids are defensive and may be responsible for the differences in palatability for these two legumes.

Determination of the Binding Mode of Various Ruthenium Complexes to DNA

Giselle Josof, Laura Stultz, and Clyde Stanton

The two possible binding modes for a chiral ruthenium complex to DNA are electrostatic and intercalative. Previously, $Ru(phen)_3^{2+}$ has been observed for photochemical evidence of intercalation with DNA. Intercalation shields one of the phenanthroline ligands, increasing the emission lifetime while decreasing the quenching constant. This project sought to determine if $Ru(bipy)_3^{2+}$, $Ru(phen)_3^{2+}$, $[Ru(phen)_2(DIP)]^{2+}$, $[Ru(phen)(DIP)_2]^{2+}$, and $[Ru(phen)_2(qdppz)]^{2+}$ intercalate with DNA. An increase in emission lifetime in the presence of DNA and a red shift as well as hypochromism in the UV-vis spectrum determined the interactions of $Ru(phen)_3^{2+}$ with DNA are intercalative. It was found that there was no intercalative binding of $Ru(bipy)_3^{2+}$ to DNA, and possible intercalative binding of $[Ru(phen)_2(DIP)]^{2+}$ and $[Ru(phen)(DIP)_2]^{2+}$ to DNA.

The Contributions of Feed Levels and Population Density to the Incidence of Cannibalism in the Lab-Reared Sea Urchin, *Lytechinus variegatus*

Cady Kimble, Andrew Gannon, and Stephen Watts (Presented Fall 2009)

Demand for sea urchin gonad for consumption as sushi (*uni*) has stimulated the need for an effective method for farming sea urchins. An obstacle to optimizing output in aquaculture is cannibalism. Frequency of cannibalism is affected by population density and feed levels in both field and laboratory studies. Using wild-caught, large (32-37g) and small (12-21g) *L. variegatus* housed in a laboratory aquaria at high (80% coverage) and low (20% coverage) densities under fed (*ad libitum*) and starved feeding regimens for four weeks, we found that in the high density, starved conditions large urchins survivorship was 98.2% (±0.3%SE) survivorship, and small urchins had 81.3% (±2.7%) survivorship, while at low density, starved large urchins survivorship rose to 100% and small urchin survivorship rose to 96.3% (±0.7%). In the high density, fed conditions, large urchins has only an 81.6% (±4.1%) survivorship, and small urchins had 86.3% (±2.6%) survivorship, while at low density, the large urchin survivorship rose to 100% and the small urchin survivorship rose to 98.75% (±0.31%). Survivorship was solely influenced by cannibalism rates. This indicates that cannibalism decreases survivorship significantly for small urchins only when they are held at high density, but in large urchins, only when they are at high density and fed. Thus, optimal density and feeding conditions change as urchins increase in size.

Determining the Role of EPS-I and EPS-II Exopolysaccharides of *Mycoplasma pulmonis* in Biofilm Formation *in vitro*

Shannon Lewis, Jeanette Runquist, and Warren Simmons

Exopolysaccharides are important components of extracellular matrices of biofilms and likely contribute to the pathogenecity of microorganisms. The mycoplasmas are cell wall-less bacteria that cause chronic disease in man and animals. It has been recently found that mycoplasmas form biofilms, which have been shown to protect the mycoplasmas from the lytic effects of complement and the pore-forming molecule gramicidin. The biofilm formed by *Mycoplasma pulmonis* contains two exopolysaccharides (EPS) in its extracellular matrix, EPS-I and EPS-II. As polysaccharides contribute to the structure of the biofilm, mutants of *M. pulmonis* that are deficient in EPS production were studied in order to determine the role of each polysaccharide in biofilm formation in vitro. The amount of biofilm formed by the polysaccharide mutants on glasscover slips were compared to the amount of biofilm formation by wild-type *M. pulmonis* using a Cystal Violet-based assay and a Hoechst-based fluorescence assay**.** The results of these experiments indicated that EPS-II affected biofilm formation on glass.

Runx2 Regulate Endochondral Bone Formation by Controlling Chondrocyte Proliferation and Differentiation

Ronald MacBeth, Pamela Hanson, Haiyan Chen, and Amjad Javed

Mineralized bone formation requires highly synchronized activities of chondrocytes and osteoblasts during intramembranous and endochondral ossification. Runx2 is an indispensable transcription factor for skeletal cell maturation and bone formation. Runx2 null mice are completely devoid of bone tissue. However, the cell type-specific role of Runx2 remains unknown. The objective of this study is to identify Runx2 role specific to chondrocyte cell maturation. Runx2 conditional null mice and transgenic animals expressing Cre-recombinase under the control of the Col2a-promoter were used as model system. In vivo cell labeling, immuno-staining and histological analyses were utilized to compare chondrocyte proliferation, growth and differentiation. Chondrocyte and cartilage tissue-specific Runx2 gene deletion resulted in failed endochondral ossification and lethality at birth. Mutant animals exhibited dwarfism, doomed skull, runted and poorly mineralized long bones. To understand these defects at cellular level, histological analyses were performed. Zones of resting, proliferating, and hypertrophic chondrocytes in femurs from wild type mice indicated normal progression of endochondral-ossification. Homozygous mutant, however, lacked distinct zones and showed only small, rounded cells. To determine if dwarfism in Runx2 null mice was due to altered chondrocyte growth, proliferation and/or differentiation, BrdU labeling was performed. We observed a marked reduction (37-47%) in chondrocyte proliferation in mutant mice. We further confirmed the failed maturation of chondrocyte by assessing the expression of Sox9, an essential protein for chondrogenesis. We observed significantly reduced levels (30-40%) of Sox9 protein in chondrocytes of the Runx2 mutant mice. In conclusion, Runx2 deficiency in cartilage impairs both proliferation and differentiation of chondrocyte.

Fire Suppression and Ecological Succession in Montane Longleaf Pine Forests, Oak Mountain State Park, AL

Michelle Maciejewski and Scot Duncan

Longleaf pine (*Pinus palustris*) ecosystems once dominated the Southeast but now occupy only 3% of their original range. *P. palustris* is a shade-intolerant, poor competitor reliant on fire to suppress fire-intolerant competitors. Found within the uplands of Alabama and northwest Georgia, the montane longleaf is among the rarest of the longleaf community types. There have been no intensive studies published on successional dynamics in these ecosystems. In particular, little is known of how the great range of topographic variation within the montane system affects community dynamics, including responses to fire suppression and logging. We assessed the successional status of the montane longleaf pine forests at Oak Mountain State Park in Pelham, Alabama. The park's forests were logged in the mid-twentieth century and have endured many decades of fire suppression. Twenty-six 20x50m plots were established where longleaf pine still remains: 13 on the southeast-facing slopes of Double Oak Mountain and 13 on the peaks of foothills. Trees over 1.3m tall were identified and measured (diameter at breast height); trees <1.3m tall were identified and measured (basal diameter) in plots or subplots. Population trends of species were analyzed to determine whether they were members of the original longleaf community, invading species establishing soon after the advent of logging and fire suppression, or invading species appearing only recently. Preliminary results suggest the mountain slope community has been invaded less than the foothills community, suggesting topography should be a major consideration in ongoing conservation efforts of montane longleaf forests.

Regulation of Expression and Role of the Pigment Epithelium Derived Factor in Ras-Mediated Pancreatic Oncogenesis

Joanna Maya and Gretchen A. Repasky

With cancer being the second leading cause for deaths worldwide, extensive research in molecular biology is essential for developing more effective treatment methods. A mutation in the *K-Ras* gene has been implicated in over 90% of pancreatic cancer cases. This mutation causes constitutive activation of K-Ras, thereby promoting several tumorigenic properties including cell proliferation and survival. Pigment epithelium-derived factor (PEDF), in contrast, is a secreted protein known to inhibit several properties associated with tumors including angiogenesis and cell proliferation. Additionally, the loss of PEDF expression has been associated with many cases of pancreatic oncogenesis. Our study focused on the nature of PEDF expression in human pancreatic duct-derived cells that either lack or express the activating *K-Ras 12D* mutation. One specific target of this study was to determine the relative levels of PEDF expression in these cells by western analyses on both cell lysates and cell culture medium. Results indicated that PEDF is upregulated both intracellularly under growth and serum-starved conditions and extracellularly under serum-starved conditions in *K-Ras*-transformed cells, suggesting that the overall production of PEDF is upregulated in *K-Ras*-transformed cells. The second target of this study focused on the effect of exogenous PEDF on the viability of these cells. There was an observed decrease in cell viability of only the PEDF-treated *K-Ras*-transformed cells under serum-starved conditions, suggesting PEDF as a specific inhibitor of *K-Ras*-transformed cell viability. Further investigations of the link between Ras and PEDF would be essential for assessing the therapeutic potential for PEDF in cases of pancreatic oncogenesis.

Assessment of Disabilities Caused by Pediatric Multiple Sclerosis by Analysis of the WeeFIM® Instrument and the Expanded Disability Status Scale

Amy Monroe, Jeannette Runquist, and Jayne Ness

Two assessment tests, the WeeFIM® and Kurtzke's Expanded Disability Status Scale (EDSS) evaluate the degree of disability of pediatric patients diagnosed with the demyelinating disease, multiple sclerosis (MS). Demyelination is caused by damage to the myelin, the protective insulation surrounding the nerve fibers in the central nervous system (CNS). The CNS is composed of the brain, spinal cord, and optic nerves. In MS, the body often repeatedly attacks its own myelin, causing possible permanent scarring in multiple areas in the CNS, manifested as various disabling symptoms. Pediatric MS is defined as symptom onset before age 18. The WeeFIM® instrument analyzes the severity of a child's disability based on a seven-level ordinal scale ranging from total independence (level 7) to total assistance (level 1) in three clinical domains: self-care, mobility, and cognition. The EDSS is used to quantify disability in adult MS, by assigning a total score, ranging between 0-9 in the functional systems. There is not a disability assessment tool that is specific to pediatric MS. This study assessed the reliability and validity of the WeeFIM® instrument in comparison to the EDSS, the 'gold standard' in this study, in pediatric patients with MS. The MS patients seen at The University of Alabama at Birmingham's Center for Pediatric Onset of Demyelinating Disease (CPODD) were used for data analysis.

Spectroelectrochemical Characterization of Polymerized Hemoglobins

Serena Murphy and Scott Dorman

In this project, various types of bovine hemoglobin were investigated with spectroelectrochemistry (SEC) and Nernst plot analysis to find the effect of polymerization on the reduction potentials ($E_{red}°$) of bovine hemoglobins. This $E_{red}°$ value signifies the hemoglobin capacity for oxygen binding. The average E° values for native bovine hemoglobin were 139.89±36.71mV and 85.03±19.26mV for wavelengths 406nm and 430nm, respectively. All reduction potentials were measured against Ag/AgCl. HBOC-201, a glutaraldehyde polymerized bovine hemoglobin, was found to have higher $E_{red}°$ values than those reported for native bovine hemoglobin. Average T-state stabilized bovine hemoglobin analyses were found to have lower values than both free bovine hemoglobin and HBOC-201 $E_{red}°$ values. The data reported implies that HBOC-201is less likely to oxidize and more readily able to carry oxygen. This high reduction value of the glutaraldehyde polymerization shows promise for application as a blood substitute.

Correlation of *Cladosporium* and *Alternaria* Spore Concentrations with Meteorological Conditions in Birmingham, Alabama

Ashlyn Rogers and Peter A. Van Zandt (Presented Fall 2009)

Airborne fungal spores are a component of the atmosphere that trigger asthma and hay fever symptoms. These fungal spores are categorized into dry-air and wet-air spora based on certain characteristics. Specifically, dry-air spores are more abundant when temperatures are high and in low humidity. Wet-air spores increase before and after rainfall and in the early morning. This study investigated how meteorological conditions from February to June of 2008 and 2009 affect the fungal spore concentrations of two members of the dry-air spora (*Cladosporium* and *Alternaria)* in Birmingham, Alabama. I counted spores of *Alternaria* and *Cladosporium* because these spores are typically the most abundant and tend to cause the most allergic reactions for those with asthma and respiratory diseases. Past research has found that higher fungal spore counts are associated with higher temperature and wind speed and with lower humidity and precipitation. My results showed that *Cladosporium* was on average fourteen times more abundant than *Alternaria*. Spore counts of both species were lowest in February and March reaching their peak in May and June. Consistent with published studies, I found that increases in average daily temperature were associated with higher *Cladosporium* and *Alternaria* concentrations. Despite my expectation that humidity and rainfall would be negatively related to counts, I found that neither of these factors played a significant role in determining fungal spore concentrations. This project provides information not only about members of the dry-air spora but what weather conditions lead to higher fungal spore concentrations.

Runx2 Differentially Directs Mesenchymal Cell Fate Decision to Adipocyte and Osteoblast Lineage

Jordan Scott, Pamela Hanson, Haiyen Chen, and Amjad Javed

Metabolic diseases, aging, and obesity are reflected by marked changes in the musculoskeletal system including reduced muscle function and bone mass. Decreased osteoblast activity and increased adipocyte activity are commonly noted in these individuals. Both fat and bone synthesizing cells are derived from a common mesenchymal stem cell. Runx2 is essential for the development of bone tissues and osteoblast differentiation. However, the relationship between adipocyte formation and Runx2 molecular signaling remains unknown. The objective in this work was to identify the role of Runx2 in directing mesenchymal cells toward adipocytes and/or osteoblasts by a regulated reconstitution of the Runx2 gene in a Runx2 null cell line that would otherwise be unable to undergo osteoblastic differentiation but preferentially commit to adipocyte lineage. Specifically, we constructed a doxycycline-regulated Runx2 vector which was transformed into two independent mesenchymal cell lines, Runx2 null and C3H10T1/2. Resistant cell lines were selected, FACS sorted, and expanded. Adipocyte formation was induced by treating with rosiglitazone, insulin, and adipogenic media in the presence or absence of doxycycline. Robust adipocyte formation was noted in control Runx2 null cells after 8 days of treatment with adipogenic media. In contrast, induction of Runx2 expression by doxycycline, resulted in strong inhibition of adipocyte formation, suggesting that Runx2 normally inhibits adipocyte formation. Overall, Runx2 deficiency in mesenchymal cells promotes adipogenesis, while reconstitution of Runx2 blocks their ability to undergo adipocyte differentiation.

The *in vivo* Roles of Mac-1 Expression on Macrophages in Regulating the Initiation and Progression of Inflammatory Disease in MRL/MpJ-*Fas*^{lpr} Mice

Pippa Simmons, Megan Gibbons, Daniel Bullard, and Jianming Wu

Systemic lupus erythematosus (SLE) is a polygenic, autoimmune, inflammatory disease characterized by vascular injury and tissue damage, with disease onset of about 16-55 years of age. Why exactly the disease appears is somewhat unclear, and no specific genes associated with lupus have been isolated. The level of expression of Mac-1on leukocytes appears to be important in the development of SLE. However, the specific roles of this adhesion molecule in macrophages during disease onset and progression are unknown. A construct was developed that included cloning Mac-1 into the CD68 macrophage specific promoter. This construct will be used in future experiments for the generation of a transgenic mouse model. The transgenic mice will provide new insights regarding the role of Mac-1 expression on macrophages *in vivo* during progression of this autoimmune disease. These insights will aid in the understanding of SLE and other inflammatory diseases.

The Anticancer Drug KP1019's Damage is not Corrected through the Base Excision Repair Pathway

Amy Strehle and Pamela Hanson

Colorectal cancer is the second most fatal cancer in the western world. The metallopharmaceutical oxaliplatin is a chemotherapy drug widely used for treatment of such cancers. While effective such platinum-centered drugs cause harsh side effects. Therefore, other metallopharmaceuticals have been brought to light including a group known as ruthenium (III) complexes. The ruthenium complex known as KP1019 is a drug that may enter the cell through the tranferrin pathway and has been shown to produce damaging peroxides in mammalian cells. However, oxidative stress does not account for all of the DNA damage caused by KP1019. To show that KP1019 is an effective drug I used yeast as a model to confirm that cell death is actually achieved by KP1019 and it is not just arresting the cells in their cell cycle. By comparing KP1019 sensitivity of a wild type yeast strain and a mutant strain of yeast lacking the BER pathway, I also determined that the base excision repair (BER) pathway does not help cells tolerate the damage caused by KP1019.

The Comparison of Counts from Bacterial Cultures Compared to Human Oral Forsyth Microbial Identification Microarray (HOMIM) in Detecting *Streptococcus mutans* from Oral Samples

Joshua Thompson, Jeannette Runquist, and Noel Childers

Streptococcus mutans, *S. mutans*, is a Gram-positive, facultative anaerobic bacterium commonly found in the human oral cavity. *S. mutans* attaches it's self to the tooth and forms biofilms known as dental plaque on tooth surfaces (Yoshida and Kuramitsu, 2002). This is a key reason why *S. mutans* are thought to be the leading cause of dental caries worldwide and is considered to be the most cariogenic of all of the oral streptococci (Ajdic et al., 2002). Therefore, further understanding the *S. mutans* bacteria could lead to the control or elimination of the leading cause of decay. There are two basic ways to examine the *S. mutans* bacteria. The first is by collecting the sample and isolating the *S. mutans* colonies on an agar plate. Once isolated the individual colonies are counted, in order to determine the amount of bacteria within a sample. The second way is by using the Human Oral Forsyth Microbial Identification Microarray (HOMIM). This is a state of the art service that is able to rapidly determine bacterial profiles (Dewhirst and Paster, 2008). Although the HOMIM is faster way to process the bacteria, it is unknown how accurate the test is in determining the amount of bacteria within a sample. Within this paper we investigated how accurate the

HOMIM was, by quantitatively comparing HOMIM results to the results found by counting the individual colonies at the University of Alabama Birmingham.

Determining the Time Course of the Induced Response in *Baptisia australis* Following Damage by *Spodoptera dolichos* Larvae

Lee Wammack and Peter Van Zandt

Plants have the ability to defend themselves by employing chemical and/or physical defenses. Following herbivore damage, chemical defenses can be increased or newly synthesized. While chemical responses in many plants have been well studied, the time course that these responses follow after induction has not. Many species in the genus *Baptisia* have been found to contain alkaloids that are induced after herbivore damage. I grew 100 blue wild indigo plants (*Baptisia australis*) in a greenhouse for three months and fed them to herbivorous caterpillar larvae (*Spodoptera dolichos)* to initiate the production of alkaloids. I then harvested the plants at 10 different time periods (from 12 hours – 21 days) following damage and assessed the induction of defenses using a bioassay and gas chromatography (GC). These data will provide future experiments with the time at which induced alkaloids are produced at the highest level.

Determination of the Effects of Depth and Rockiness on *Orconectes virilis* Sex Ratio and Body Size in Roebuck Spring

James A. Watters and Megan Gibbons (Presented Fall 2009)

Orconectes virilis, or the northern crayfish, is an invasive non-native species that has been observed in several different habitats including lakes, streams, and rivers. It is the most common crayfish species in Roebuck Spring (Jefferson County, Alabama), and has had negative impacts on the hay crayfish, *Cambarus striatus*, and the endangered watercress darter, *Etheostoma nuchale*. Its impacts were exacerbated when an ecologically crucial dam was removed from the spring on September 22, 2008. As a result, the invasive crayfish density increased and the watercress darter density was drastically reduced, which heightened predation encounters between these species. As part of an ongoing effort to reduce the numbers of *O. virilis* in the Spring, we trapped and removed the invasive crayfish and conducted a study to determine the effects of depth and rockiness on their sex ratio and total body size. We discovered that sites that differed in depth and rockiness influenced total body length, but not sex ratio. As depth increased, total body size increased, and smaller individuals were associated with rockier habitats than larger individuals. Finally, total body size was more influenced by depth in non-rocky environments than in rocky ones. By identifying habitat preferences for different size classes of crayfish, this study should aid in conservation efforts of native crayfish species and the endangered watercress darter.

Maternal Provisioning Trade-off Strategies of *Agalychnis callidryas*

Charles Yeager and Megan E. Gibbons (Presented Fall 2009)

The trade-offs associated with various maternal provisioning strategies in amphibians is well documented in previous studies. Female size is expected to be positively correlated with provisioning of resources to offspring. Strategies for increased maternal provisioning include increasing the size of the clutch (i.e., number of eggs) or increasing individual egg size. Females sometimes produce clutches of varying individual egg size; this strategy is known as bet-hedging. For this study we examined the trade-off provisioning strategy employed by red-eyed tree frogs (*Agalychnis callidryas*). We recorded female mass, individual egg diameter, total yolk volume, and total eggs laid from 31 amplexed pairs of frogs. Female mass had no significant relationship with total yolk output or total number of eggs, but did have a significant positive relationship with egg diameter. A posteriori tests showed a significant difference between the largest and smallest eggs within clutches, and that clutches differed in their average egg size. For females with the lowest egg size variation in their clutches, there was a significant positive relationship between female mass and total yolk output, a marginally significant positive relationship between female mass and total number of eggs, and non significant positive relationship between female mass and egg diameter. Females with greater within-clutch egg size variation used a bet-hedging strategy, and the traditional trade-off of egg size and egg number was not apparent in these clutches. These data suggest that a combination of maternal provisioning strategies are present in this population of *A. callidryas*, which is likely an adaptive response to an unpredictable environment.