Student Abstracts

15th Annual Graduate Student Symposium
Department of Biological Sciences
November 15, 2013

POSTER PRESENTATIONS (10:30-12:00)
Life Science Building Annex Lobby

Biochemistry & Molecular Biology (BMB)

Name: Kelley Nunez
Title: High-throughput detection of insertional mutants in *Chlamydomonas reinhardtii*

The unicellular alga *Chlamydomonas reinhardtii* has been a model organism for studies that involve photosynthesis, flagellar structure, chloroplast biogenesis, and cell-cell recognition (2). The published genome of *C. reinhardtii* has allowed for forward and reverse genetics, enhancing our understanding of this organism's physiology. However, low rates of homologous recombination have forced researchers to use brute force screens to identify specific mutants in insertion libraries (1). Here, we introduce a technique that targets and retrieves regions of the genome that border heterologous insertions. The technique targets a sequence within the insertion and recovers DNA adjacent to the insert, allowing the location of insertion to be identified by DNA sequencing. Initially, the technique was successfully tested on individual isolates carrying characterized insertions. The method was then applied to DNA isolated from a pool of 1,440 uncharacterized insertion mutants. An Illumina HiSeq 2000 was used to sequence the captured DNA. Preliminary evidence suggests this technique may be used to identify those genes potentially disrupted in the insertion library, providing a means of facilitating future analyses of *C. reinhardtii*.

Name: Noelle Bryan
Title: Analysis of Microorganisms in Aerosols Collected at Altitudes Between 1.5 and 36 km

Microorganisms (i.e., bacteria, fungi, and viruses) aerosolized from surfaces and transported in the high atmosphere must tolerate low relative humidity, a high fluence of ultraviolet (UV) radiation, and low temperatures. Concentrations of cells collected at select altitudes in the troposphere range from 10^4 to 10^6 cells m^{-3}, however little work has been done to quantify the number of cells present at higher altitudes (up to 36 km). Recent studies have shown that microorganisms in the troposphere are capable of intercontinental transport, influence precipitation, and metabolize carbon compounds in cloud droplets. Determining the concentration of cells in the atmosphere will be the first step towards understanding if significant environmental contributions are being made at higher altitudes. In addition, the environmental conditions (temperature, pressure, relative humidity (RH), nutrient availability, and radiation levels) at ~30km are similar to Mars surface conditions. The ability to survive multiple environmental stressors in the atmosphere may also prove beneficial to surviving on other planets, thus an atmospheric isolate could serve as a terrestrial analog for investigating microbial life under Martian conditions. During August 2013, we launched several sampling missions to determine how the numbers of total of cells and viable microorganisms vary with altitude. The first payload type was a microbiological sampler with two chambers designed to collect from two separate target altitudes. Each sampling chamber holds forty plastic rods covered with silicon grease. Each rod has a sampling area of 3.5×10^{-5} m^2 and serves as an impact sampler when the chamber doors open at the pre-defined altitudes. The second payload was a rotating platform with two sampling chambers equipped with sampling rods and was integrated with the High Altitude Student Platform (HASP) during the 2013 flight. One of the goals of NASA's Astrobiology Road Map (2008) is to understand the physical limits of life on Earth in order to predict how life might persist elsewhere in

the solar system. The recovery of viable microorganisms from the atmosphere will allow for controlled laboratory simulations to determine the levels of resistance to multiple environmental stressors. A microorganism that is desiccation tolerant, psychrotolerant, and radiation resistant or capable of forming endospores would be well equipped to survive atmospheric transport. In addition, those same traits would prove beneficial for transport through space or survival under Martian surface conditions.

Name: Bilal Jilani
Title: Detection of microbes within Antarctic ice sheets
Abstract not submitted

Name: Narender Kumar
Title: Specific Function and Regulation of SIM Protein Motifs in the Endoreplication of Arabidopsis Trichomes**.**
Abstract not submitted

Name: Tianyun Long
Title: An essential role of DRH-3 in antiviral RNA

In mammals, including humans, RIG-I-like RNA helicases (RLHs) sense invading viruses thereby to initiate antiviral responses mediated by interferon proteins. Intriguingly, RLHs are also conserved in the nematode worm *Caenorhabditis elegans* and are known to play essential role in an RNA-directed antiviral immunity mediated by RNA interference (RNAi), a novel gene silencing mechanism in eukaryotes. DRH-1 is one of the worm RLHs and has been shown to be required for antiviral RNAi but not for RNAi targeting cellular genes. DRH-3 is another worm RLH that exhibits similar domain structure with DRH-1 and, like DRH-1, plays important role in antiviral RNAi. Currently, it remains largely unclear whether DRH-3 differentiates between viruses and cellular genes in mediating RNAi. Here we show that DRH-3 is required for cellular gene silencing triggered by artificial double-stranded RNA or homologous replicating virus. These observations together suggest that DRH-3 contributes to antiviral RNAi through distinct mechanism compared to DRH-1.

Name: Aaron P. Landry
Title: Thiol-mediated reduction of human mitochondrial outer membrane protein mitoNEET [2Fe-2S] clusters

Human mitochondrial outer membrane protein mitoNEET is a novel target of the type II diabetes drug pioglitazone. The C-terminal cytosolic domain of mitoNEET hosts a redox-active [2Fe-2S] cluster via an unusual arrangement of three cysteine and one histidine residues. Here we report that human mitoNEET [2Fe-2S] clusters are fully reduced when expressed in *Escherichia coli* cells. *In vitro* studies show that purified mitoNEET [2Fe-2S] clusters can be partially reduced by monothiols such as reduced glutathione, L-cysteine or N-acetyl-L-cysteine, and fully reduced by dithiol dithiothreitol under anaerobic conditions. *E. coli* thioredoxin reduced by thioredoxin reductase and NADPH can also efficiently reduced the mitoNEET [2Fe-2S] clusters. Addition of hydrogen peroxide reversibly oxidizes the pre-reduced mitoNEET [2Fe-2S] clusters without disruption of the clusters, indicating that mitoNEET may act as a sensor of oxidative signals via the [2Fe-2S] clusters. Additional studies reveal that binding of type II diabetes drug pioglitazone in mitoNEET significantly inhibits the thiol-mediated reduction of the [2Fe-2S] clusters, suggesting that pioglitazone may modulate the function of mitoNEET by blocking the thiol-mediated reduction of the [2Fe-2S] clusters in cells.

Name: Qing Wang
Title: Global Effect of Extra-transcriptional Functions of RNA Polymerase III Complexes

RNA Polymerase III (Pol III) transcribes mainly tRNA genes as well as some limited number of other small RNAs (5S rRNA, 7SL RNA, U6 spliceosomal RNA). Two transcriptional factor complexes, TFIIIC and TFIIIB, are required for Pol III transcription. Recent studies have pointed out other

functions of Pol III transcription system other than transcription, including chromatin boundary, nucleosome positioning, and genome organization activities. Extra-TFIIIC sites (ETC sites), chromosomal locations bound by TFIIIC alone, also have extra-transcriptional functions. So far, Donze lab have found another extra-transcriptional effect of Pol III complexes—block the progression of intergenic transcription by RNA Polymerase II (Korde et al, submitted). Previously published data from high-throughput microarray of coding sequences have revealed differences of genome expression in response to decreased Pol III transcription. Additionally, extensive alteration and diversity of transcripts have been observed across different growth and stress conditions from recent publish. In order to detect the global effect of RNA Pol III extra-transcriptional functions, we are conducting RNA-seq analysis of two yeast strains (wild-type versus Tfc6-mutant). Our preliminary RNA-seq results indicated that several genes with tDNA nearby have various amount of upregulation or downregulation or gene extension due to the defective Pol III complexes. Ongoing analysis should reveal more informative loci and may find couple undiscovered ETC sites and further investigate additional functions of the Pol III complexes.

Name: Dinesh K. Deochand
Title: Dimer interface of *Deinococcus radiodurans* HucR communicates DNA and ligand binding

Deinococcus radiodurans HucR (Hypothetical Uricase Regulator) is a transcriptional regulator belonging to the MarR family of proteins. HucR binds to promoters of *hucr* and *uricase* genes leading to repression of both. In presence of the ligand urate, the repression of both genes is relieved. The crystal structure of HucR reveals that it is a dimer, each monomer with the topology α1-α2-β1-α4-α5-β2-β3-α6-α7. The framework of the dimerization domain is provided by α2/α2′, in which the imidazole rings of His51 and His51′ are stacked. DNA binding affinity assessed by EMSA for HucR and its mutant H51F at pH 8.0 and 5.0 shows that the HucR-H51F mutant is less sensitive to pH. Analysis of tryptophan intrinsic fluorescence spectra shows that HucR binds urate with negative cooperativity whereas HucR-H51F binds with positive cooperativity. Moreover, thermal shift assays of HucR and HucR-H51F demonstrate that HucR is completely unfolded at pH 5.0 and has a T_m ~ 52 °C at pH 8.0 while HucR-H51F is only modestly destabilized at pH 5.0 (T_m ~ 45 °C). Notably, DNA binding stabilizes HucR at pH 5.0 whereas HucR-H51 is destabilized on binding DNA. We suggest that histidine residues at the dimer interface act as a pH sensor and that DNA and ligand binding is communicated through the dimer interface.

Name: Sara Zahraeifard
Title: Investigating the role of histone H2A.Z abundance in modulating responses to Phosphorus- and/or Iron- deficiency in plants
Abstract not submitted

Name: Surabhi Maheshwari
Title: Structure Based Prediction of Protein-Protein Interactions.

Interactions between proteins are critical to numerous biological processes, thus they are often considered as a core of the cellular interactome. Interacting proteins are routinely involved in signal transduction, protein transport and folding. DNA replication and repair, and cell division, just to mention a few examples. A recent statistical analysis has estimated the number of interactions in human proteome to be ~650,000; however, experimentally determined protein-protein interactions (3,565) constitute only a tiny fraction of the putative human interactome. In that regard, computational methods for the prediction of protein-protein interactions are an important area bio-algorithm development. Many sequence-based methods have been developed over the past years; however, due to the general instability of sequence alignments in the "twilight zone" of sequence similarity, homology-inferred interactions may contain a significant fraction of false positives. Structure based methods typically rely on either the availability of evolutionarily and functionally related dimeric template structures in PDB or the accuracy of protein docking algorithms. Encouragingly, the latter

have demostrated recently to be able to identify known interactions from a large set of docking decoys for ~50% of the benchmarking cases, suggesting that current macromolecular docking methods can be used for reconstruction of biological network.

Name: Misagh Naderi
Title: Synthetic Libraries of Drug-Target Complexes for Structure-Based Drug Design
Abstract not submitted

Cellular, Developmental, & Integrative Biology (CDIB)

Name: Amanda Laque
Title: Leptin Receptor neurons co-expressing the neuropeptide galanin, control orexin neurons and mediate sucrose preference

Leptin, an adipose derived hormone that negatively regulates energy balance, has also been shown to regulate reward function via the mesolimbic dopaminergic system. However, the exact mechanism how leptin mediates these effects are largely unknown. Here, we focus on a subset of leptin receptor (LepRb) neurons co-expressing galanin (Gal-LepRb neurons). We show that Gal-LepRb neurons in the lateral hypothalamus (LH) are inhibitory neurons, stimulated by leptin and directly innervate orexin neurons. We developed mice with a deletion of LepRb in galanin neurons (Gal-LepRb^{KO} mice). In line with the hypothesized lack of the inhibitory input from Gal-LepRb neurons, Gal-LepRb^{KO} mice indeed show enhanced activation of orexin neurons. Orexin acts in the VTA and modulates reward behavior, thus, in a two-bottle choice test we evaluated differences in the consumption of palatable food rewards (isocaloric 25%sucrose & 10% intralipid solutions). Interestingly, Gal-LepRb^{KO} mice elicit a robust sucrose preference in contrast to wild-type mice exhibiting no preference. Similarly, Gal-LepRb^{KO} mice showed enhanced motivation to obtain a sucrose-rich treat in an incentive runway paradigm. In line with previous data, we confirm that increased orexin activation positively correlates with reward driven behavior. Our data suggests that leptin acts via Gal-LepRb neurons to inhibit orexin neurons, to decrease the rewarding value of food.

Name: Peng Zhao
Title: Mechanisms involved in the induction of lipocalin-2 expression in vivo and in vitro

Lipocalin-2 is a cytokine highly expressed in mature adipocytes. Its expression and secretion have been shown to be increased in obesity and type 2 diabetes. Our previous studies have revealed that lipocalin-2 expression and secretion can be induced by pro-inflammatory cytokines in cultured 3T3-L1 adipocytes. Now we report that pro-inflammatory cytokines, IFNγ and TNFα, also up-regulate lipocalin-2 expression and increase circulating lipocalin-2 level in vivo. Signaling studies uncover that STAT1 is required for IFNγ-induced lipocalin-2 expression, and NF-κB is essential for the induction effect of TNFα. As our research has shown that activation of ERKs also participates in the modulation of lipocalin-2 expression, We examined how ERKs signaling contributes to this process. Our experiments clearly demonstrate that ERKs activation affects STAT1 and NF-κB subunit p65 serine phosphorylations, which are critical for their maximum.transactivation activities. Study of human lipocalin-2 promoter identified four STAT1 binding sites and one NF-κB binding site.

Name: Bradley Wood
Title: The myomere-myoseptal intersections in a lamprey (*Petromyzon marinus*) and a shark (*Squalus acanthias*)
Abstract not Submited

Name: Vijai Krishnan
Title: The interplay between proton gradients and chloride flux in retinal amacrine cells

Previous research from our lab has shown that nitric oxide (NO) releases Cl^- from an internal store and that the release is dependent upon a transient acidification of the cytosol (Hoffpauir 2006; McMains and Gleason 2011). Our strongest candidates for internal Cl^- stores are endosomal compartments which contain 20-60 mM Cl^- and maintain a low pH via the V-type H^+ pump. Furthermore, coupled antiport of H^+ and Cl^- has been shown for ClC transporters which are known to reside on endosomal membranes. Here we test the hypothesis that the distribution of protons across internal membranes influences the ability of NO to release internal Cl^-. We have previously reported that the weak bases methylamine (MA, 10mM) and chloroquine (CQ, 100μM) reliably reduced the LysoSensor fluorescence signal. This effect is likely due to the buffering action of MA and CQ in acidic compartments such as endosomes where LysoSensor accumulates. To test the consequences of buffering on the NO-dependent release of internal Cl^-, we made whole cell voltage clamp recordings from individual chick amacrine cells in culture. Voltage ramps were delivered in the presence of GABA (20μM) to activate $GABA_A$ and the reversal potential ($GABA_{rev}$) of the current was measured under different conditions. The initial reversal potential of the GABA-gated current was more positive in MA and CQ (internal solution) than under control conditions ($p=0.01$ for both) suggesting that cytsolic Cl^- was elevated when endosomal pH was buffered. After NO, $GABA_{rev}$ moved negative under buffering conditions suggesting that cytosolic Cl^- was taken up into the store or expelled from the cell. To test whether the H^+ gradient across internal membranes was required for the NO-dependent release of internal Cl^-, we used the protonophore carbonyl cyanide 4-(trifluoromethoxy)phenylhydrazone (FCCP, 1μM) to collapse H^+ gradients and bafilomycin (1μM) to inhibit the V-type H^+ pump. In LysoSensor experiments, we consistently observed a reversible reduction in LysoSensor fluorescence with both reagents consistent with H^+ leaking from acidic compartments. In voltage clamp experiments, we held amacrine cells at -70mV under Cl^-free internal and external conditions to isolate the Cl^- store. Pulses of GABA (20μM, 400msec) were applied to test whether cytosolic Cl^- was available to carry an inward current. Under control conditions, no GABA-gated currents were observed until NO was applied. In FCCP external solution, the NO-dependent release of internal Cl^- was not observed. These results suggest that the H^+ gradient across internal membranes is an important component in the machinery that controls the NO-dependent release of internal Cl^-.

Name: Tyler Rodriguez
Title: Localization of Cl^- Transporters in Retinal Amacrine Cells

Retinal amacrine cells are interneurons that mediate signaling between bipolar and ganglion cells in the inner plexiform layer. There are many different types of amacrine cells but the majority are usually inhibitory, using either GABA or glycine as their neurotransmitter. Our lab is particularly interested in GABAergic amacrine cells because we have shown that nitric oxide (NO) can transiently convert GABAergic synapses from inhibitory to excitatory. This is due to an increase in cytosolic $[Cl^-]$ released from an intracellular store that ultimately shifts the $GABA_A$ reversal potential to depolarizing voltages. The mechanisms involved in this NO dependent Cl^- release are still unresolved.

I examined co-localization of $GABA_A$ receptors with the nitric oxide synthetic enzyme (NOS), and two candidate Cl^- transporters that may be involved in the release of Cl^- (CLC5, CLIC4). The objective of this study was to determine if the spatial relationships between these proteins would allow for functional interactions. I used a monoclonal mouse anti-$GABA_AR$ antibody and polyclonal rabbit antibodies against bNOS, CLC5, and CLIC4. Cultured chick retinal amacrine cells (E15 or E16) were fixed in 2% paraformaldehyde and double labeled for $GABA_ARs$ with bNOS, CLC5, or CLIC4. Images were captured with an Olympus IX-70 inverted microscope and edited using Photoshop or ImageJ.

In order to identify other Cl^- transporter candidates, I am currently using RT-PCR technique with gene specific primers designed for amplification of Cl^- transporter transcripts. I am examining

expression of CLIC, Anoctamin, and Bestrophin isoforms in chick retinal amacrine cells. Importantly, these families of Cl^- transporters are thought to be expressed on internal membranes. Protein expression of candidates will be confirmed through antibody labeling.

Future work involves knocking down expression of candidate Cl^- channels by transfecting miRNA constructs. Knockdown efficiency will be determined by qRT-PCR. Whole cell recordings of $GABA_A$ gated Cl- currents will examine if there is loss of function in knockdowns compared to controls. Knockdown studies may prove useful in identifying the Cl^- transporters responsible for the NO induced Cl^- release from the store.

Name: Karen Field
Title: Social Status Influences Different Modulatory Receptors in the Olfactory Bulb of an African Cichlid Fish

Neuromodulators such as peptides and biogenic amines can influence processing of behaviorally relevant sensory information, helping an animal integrate the external environment with its internal physiological state. In teleost fishes, olfaction is crucial for behaviors such as feeding, predator avoidance, kin recognition, and reproduction, and olfactory processing may be influenced by modulators. The first-order processing center in the fish brain, the olfactory bulb, receives information from the olfactory epithelium, then sends it to higher brain centers involved in mediating behavioral decisions. Little is known, however, about which modulators might function in the fish olfactory bulb, and whether the relative sensitivity to modulators might differ between animals motivated towards different tasks. We used the highly social African cichlid fish, Astatotilapia burtoni, to test the hypothesis that mRNA levels of several modulatory receptors in the olfactory bulb differed between dominant and subordinate males. Dominant male A. burtoni are brightly colored, territorial, and reproductively active, while subordinate males are drab colored, non-territorial, and reproductively suppressed. Using quantitative PCR we demonstrate that dominant males have higher mRNA levels of kisspeptin receptor, dopamine receptors, arginine vasotocin receptor, and neuronal nitric oxide synthase in the olfactory bulbs compared to subordinate males. Modulatory receptors were also positively correlated with gonadosomatic index, suggesting a link between olfactory modulation and reproduction. These results suggest that the olfactory bulbs of dominant males are more sensitive to several neuromodulators, which may facilitate detection and fine tune their perception of olfactory signals vital for social assessment during reproductive and territorial interactions.

Systematics, Ecology, & Evolution (SEE)

Name: Clare Brown
Title: The Evolution of Migration in Swallows (Hirundinidae)

Objective: Explore patterns in the gain and loss of seasonal migration and in changes in migratory distance in the evolutionary history of Hirundinidae, a globally distributed family of aerial insectivores comprising approximately 80 recognized species.

Methods: Ancestral character state reconstruction of seasonal migration on a well-resolved multilocus tree (79 swallow taxa) with maximum likelihood estimation implemented in the APE package in R. Migration coded as both a binary and a multistate character; AIC to select the best model.

Results: Migration as binary character: Asymmetrical rates model performed better than equal rates model, ancestral state equivocal, with losses of migration and decreases in migratory distance occurring at a greater rate than gains of migration and increases in migratory distance. Greatest rates were between resident/short-distance and partially migratory state.

Conclusions: Ancestral state of migration in swallows is ambiguous. Seasonal migration in swallows is evolutionarily plastic - multiple gains and losses of migration in the swallow tree. The gain and loss of partial migration is frequent, and loss of migration or decrease in migratory distance is

more likely than the inverse.

Name: Jenessa Kay
Title: Impact of *BP Deepwater Horizon* oil spill on oyster reef commensal communities in Barataria Bay, LA

Oyster reefs provide many ecosystem services, including providing habitat and refuge for many invertebrates and juvenile fish. In turn, these organisms support higher trophic levels, including economically important commercial fisheries. While seasonal fluctuations in the species diversity, richness and abundance of these commensal communities are to be expected, the health of an oyster reef can also impact the organisms living among them. We hypothesized that commensal organisms from oyster reefs impacted by the 2010 *BP Deepwater Horizon* oil spill might be declining. In 2012, we assessed the diversity and relative abundance of commensal organisms at two sites in Barataria Bay, LA that experienced oiling from the spill, and two control sites that received no contamination. At each site, 5 replicate Vexar bags containing oyster cultch were deployed for one month every three months, after which all of the organisms contained in the bags were sorted and identified. Preliminary results show a decrease in species diversity during the summer sampling period at Bay Jimmy, the most heavily oiled study site. These data correspond with a simultaneous decrease in oyster settlement, suggesting that further studies are necessary to determine the cause of these coupled events.

Name: Maria Vozzo
Title: Long term effects of the *Deepwater Horizon* oil spill on oyster recruitment in Barataria Bay, LA

The *Deepwater Horizon* oil spill in April 2010 threatened productive oyster reefs in Barataria Bay, Louisiana. SCAT data revealed that parts of the bay were heavily oiled and while cleanup occurred shortly after the spill, the long term effects of hydrocarbon contamination on the oyster population are relatively unknown. To test for differences in oyster recruitment, two oiled sites, Grand Terre and Bay Jimmy, and two control sites, Grand Isle and Hackberry Bay, were sampled monthly during the 2012 field season. Ten ceramic tiles were deployed at each site and provided both an exposed and refuge surface for settling organisms. Tiles were collected one month later and the number and percent cover of oysters, barnacles, and other sessile organisms were quantified. Preliminary results suggest that hydrocarbon contamination had no effect on oyster recruitment, or barnacle settlement and growth. However, differences in settlement on the exposed and refuge surfaces suggest that predation may have had a greater impact on oyster settlement and survival at the four sites. These data indicate that predation may play a greater role on long term oyster recruitment and survival than hydrocarbon contamination in Barataria Bay.

Name: Mitchell Bogran
Title: Effect of hydrocarbon contamination on oyster reef commensal assemblage colonization rates

Oyster reefs in Barataria Bay, Louisiana provide essential habit for juvenile fish and intertidal invertebrates; after the Deepwater Horizon oil spill in April 2010, the effect to these communities was unknown. Preliminary data revealed the long-term effects of hydrocarbon contamination on these commensal communities diversity and abundance were neither large nor consistent, however, without being able to sample immediately after the spill, the short-term effects of the oil spill on the oyster reef community went unidentified. To study the immediate effects of oil pollution on commensal organisms, eighteen mesh bags filled with oyster cultch were placed in Barataria Bay in June 2013. Half of the bags were filled with shell soaked in oil to provide a hydrocarbon contaminated oyster reef surface. At one, two and four week after deployment, three oil-soaked and control bags were collected, Organisms collected from the bags were identified and the colonization rate of commensal recruitment was analyzed. The results indicated that there was a reduction in the species richness and abundance in the oil treated commensal assemblages compared to the control commensal

assemblages. Results from this study provide further understanding of the impact of hydrocarbon contamination on organisms that take refuge in oyster reefs.

Name: Bridget Rogers
Title: Short term effect of oil exposure on barnacle settlement in Barataria Bay, LA

Barataria Bay, Louisiana is home to productive oyster reefs and fouling communities but was affected by the *Deepwater Horizon* oil spill in April 2010. To monitor the long-term effects of the oil spill on fouling communities, two control sites, Grand Isle and Hackberry Bay, and two oiled sites, Grand Terre and Bay Jimmy, were selected. Results from 2012 indicate that there are few lingering effects on oyster and barnacle recruitment that can be attributed to the oil spill. Since the spill happened three years ago and immediate effects can no longer be measured, a short term study of oiling effects was conducted. In June 2013, ten cement particleboard tiles were placed at each site: five were control tiles and five were soaked in light Louisiana crude oil prior to deployment. Tiles were collected after one month, and barnacle population density and average size were measured. Data showed that there were site-specific differences in barnacle density, and barnacle abundance was reduced on oiled tiles at one site. Reduced barnacle growth was seen at the two high salinity sites. The data also suggest there may be a refuge effect with control tiles but not with oiled tiles. Results from this study contribute to the understanding of how fouling community assemblages are impacted by large-scale pollution events such as hydrocarbon contamination during an oil spill.

ORAL PRESENTATIONS (1:15-5:00)
Life Sciences Building Annex A663

Biochemistry & Molecular Biology (BMB) and Cellular, Developmental, & Integrative Biology (CDIB)

Name: Ashish Gupta
Title: Transcriptional regulator MftR from Burkholderia thailandensis participates in oxidative stress responses

Burkholderia thailandensis encodes the major facilitator transport regulator (MftR), which is a member of the multiple antibiotic resistance Regulator (MarR) protein family. The genomic locus predicts that MftR regulates expression of genes encoding MftR and Major Facilitator Transport Protein (MFTP). MFTP is a drug efflux pump, which belongs to the major facilitator superfamily. It is predicted to extrude phenolic compounds and detergents, which can be harmful to this bacterium. Sequence conservation and biochemical analysis has shown that MftR binds the ligand urate, and that urate attenuates DNA binding. Molecular docking was used to identify the ligand binding site, predicting that W11 and R63 are involved in the ligand binding. These residues were also present in the ligand binding site of a homologous transcription factor encoded by Deinococcus radiodurans. By quantitative RT-PCR it was observed that MftR represses its own expression and that of MFTP, and that this repression is relieved in the presence of urate. MftR also regulates expression of other transcriptional regulators annotated as LysR and SoxR homologs. Since urate is generated under conditions of oxidative stress, andsince SoxR is predicted to function in oxidative stress responses, our data suggest a role of MftR in such events.

Name: Ambuj Kumar Kushwaha
Title*:* *Mycobacterium smegmatis* Ku binds DNA without free ends

Ku is central to the non-homologous end-joining pathway of double strand break repair in all three major domains of life, with eukaryotic homologs being associated with more diversified roles compared to prokaryotic and archaeal homologs. Ku has a conserved central 'ring-shaped' core domain. While prokaryotic homologs lack the N- and C-terminal domains that impart functional diversity to eukaryotic Ku, analyses of Ku from certain prokaryotes such as *Pseudomonas aeruginosa* and *Mycobacterium smegmatis* have revealed the presence of distinct C-terminal extensions that modulate DNA-binding properties. We report here that the lysine-rich C-terminal extension of *M. smegmatis* Ku contacts the core protein domain as evidenced by a decrease in thermal stability and an increase in DNA-binding affinity and intrinsic tryptophan fluorescence upon its deletion. Ku deleted for this C-terminus requires free DNA ends for binding, but translocates to internal DNA sites. In contrast, full-length Ku can directly bind DNA without free ends, suggesting that this property is conferred by its C-terminus. Such binding to internal DNA sites may facilitate recruitment to sites of DNA damage. Our data also suggest that extensions beyond the shared core domain may have independently evolved to expand Ku function.

Name: Zelam Kaluskar
Title: Environmental and Genetic Basis of Phase Variation in the Human Pathogen *Virbio vulnificus*

Vibrio vulnificus is the leading cause of seafood consumption or exposure related deaths in the United States. Disease is caused by strains producing a capsular polysaccharide, loss of which results in an unencapsulated translucent phenotype with diminished or lacking virulence potential. These two phenotypes can produce rugose exopolysaccharide resulting in a dry, wrinkled form, called rugose, that has the ability to form profuse biofilms. Phase variation occurs in this species, which is marked by altered levels of capsule or exopolysaccharide production. In this study, we have attempted to identify environmental factors which cause phase variation in *V. vulnificus.* Specifically, the role of manganese ions in *V. vulnificus* phase variation has been elucidated. We demonstrate

here that, manganese in millimolar quantities induces phase variation of the opaque to translucent or rugose phenotypes. Additionally, we also demonstrated that phase variation observed in manganese involves genetic changes in the Group I CPS operon.

Name: Satya Avva
Title: Characterization of BEAF Insulator Protein and identification of its interaction partners

Like enhancers and promoters, insulators or boundary elements are a specialized class of regulatory DNA sequences. Two of the first insulator elements to be identified are the scs and scs' sequences which bracket two Hsp70 genes at the 87A locus of *Drosophila.* Boundary Element-Associated Factor (BEAF32), a 32 kDa protein, was found to bind to the scs' insulator, which is expressed in two isoforms 32A and 32B. Results from the current studies enabled us to characterize the BEAF proteins. Here we determine what region of BEAF mediates BEAF-BEAF interactions; test the ability of deletion mutants of BEAF to rescue the BEAF knock-out mutation and identify its interaction partners.

Name: Sujeet Kumar
Title: Members of the conserved DedA family are membrane transporters required for drug resistance in *E. coli*

The DedA Membrane protein family members are present in most bacterial genome including *Escherichia coli*. An *E. coli* strain (BC202) lacking partially redundant DedA family genes *yqjA* and *yghB* displays temperature sensitivity and cell division defects. These phenotypes are rescued by overexpression of MdfA, a Na^+-K^+/H^+ antiporter of the major facilitator superfamily (MFS) or by growth in acidic media (pH 6), suggesting roles for YqjA/YghB in maintenance of some aspect of the protonmotive force (PMF), composed of the electrical potential ($\Delta\Psi$) and the ΔpH. We found that BC202 is hypersensitive to several biocides, antibiotics and cationic compounds. Moreover, overexpression of YqjA or yghB corrects the drug susceptibility of BC202 and the function of YghB and YqjA is dependent upon membrane-embedded acidic amino acids, hallmarks of most known proton-dependent antiporters including members of the MFS. Similarly, BB0250, a distantly related DedA family protein of the Lyme disease pathogen *Borrelia burgdorferi* which complements the cell division and temperature sensitivity phenotype of BC202, also corrects the drug sensitivity of this *E. coli* strain. Like its *E. coli* counterparts, functions of BB0250 are dependent upon membrane embedded amino acids. Overexpression of *mdfA* in BC202 restores wild type resistance to substrates of MdfA as well as other drug resistance transporters such as EmrE and AcrAB. These results suggest that YqjA and YghB belong to a new family of membrane transporters and are required for PMF-dependent drug resistance in *E. coli*.

Name: Asawari Korde
Title: RNA polymerase III transcription factor complexes block transcriptional interference from intergenic RNA polymerase II progression in *Saccharomyces cerevisiae.*

In eukaryotes, DNA-dependent RNA polymerases (RNA Pol I, Pol II and Pol III) have allowed a more sophisticated regulation of individual sets of genes. Typically, RNA Pol III transcribes transfer RNA (tRNA), 5S ribosomal RNA and other small non-coding RNA genes. Transcription of tRNA gene (tDNA) requires sequential assembly of transcription factors on the tDNA comprising internal control promoter elements *A-box* and *B-box*. In the process of transcription, first, RNA Pol III transcription factor TFIIIC specifically recognizes and binds at highly conserved *B-box* promoter sequence by bridging *A-box* sequence. Bound TFIIIC recruits TFIIIB-another RNA Pol III transcription factor few base pairs upstream to the transcription start site and stable TFIIIB-tDNA association then allows binding of Pol III enzyme complex at the start site upon which, transcription initiates. Earlier studies have found that, partially or completely bound Pol III transcription complex assembly (Pol III complex) at tDNA exerts significant effects on regulation of neighboring genes or on genome organization. Such effects are found to be independent of tDNA transcription hence, known as 'Extra-transcriptional effects of Pol III complex'.

Here, our results show novel extra-transcriptional effect of Pol III complex bound at *tV(UAC)D* tDNA which lies between two divergently transcribing Pol II genes- *ATG31* and *SES1* in *Saccharomyces cerevisiae*. Pol III complex bound at this tDNA not only functions as an enhancer blocking insulator for *ATG31* gene but also acts as a barrier to intergenic transcription by RNA Pol II. In yeast, pervasive transcription from bidirectional strong promoters causes transcription of intergenic region, mostly by RNA Pol II. Result of such a transcription is a stable unannotated transcript *SUT467* which starts ~300 bp upstream of the tDNA and normally ends at the start of the tDNA. Our Northern and western analyses results have confirmed that mutation of the tDNA allows readthrough of transcription from the upstream *SUT467* start site that prevents normal *ATG31* transcriptional initiation. Also, the extended 5'-UTR inhibits translation of the *ATG31* coding sequence. In yeast, Atg31p is required in autophagy which is a bulk degradation process for the cell survival under cell stress or limiting nutrient conditions. Our results from Pho8Δ60 phosphatase and survival assays confirm lower fitness in strains that predominately produce the extended *ATG31* transcript due to compromised ability of mutants to induce autophagy. From the ChIP analysis, we demonstrated that TFIIIB, as a part of the Pol III complex associated with the *tV(UAC)D* tDNA, serves as a physical impediment to elongation RNA Pol II, initiating at the *SUT467* transcriptional start site and this barrier further protects *ATG31* gene from transcriptional interference effect.

Name: Amanda Achberger
Title: Molecular Analysis of Microbial Communities inhabiting Subglacial Lake Whillans, Antarctica

A complex hydrologic system consisting of lakes, streams, and water saturated sediments exists beneath the Antarctic Ice Sheet. Although subglacial aquatic environments are hypothesized to harbor active microbial communities, direct sampling of these potential ecosystems has been lacking. During January 2013, the Whillans Ice Stream Subglacial Access Research Drilling (WISSARD) Project created a ~800 m borehole to access Subglacial Lake Whillans (SLW) and collected water and sediment samples. The biotic and abiotic particulates larger than 10, 3, and 0.2 µm were fractionated and concentrated *in situ* on 142 mm filters using a custom filtration device that was lowered into the lake. In addition, the upper 40 cm of lake sediments were retrieved and sampled at 2 cm intervals. Nucleic acids were extracted from the samples and phylogenetic analysis based on the V4 region of the 16S rRNA gene was used to characterize the prokaryotic microbial community structure in SLW. The significance of the molecular data for deciphering ecosystem processes in SLW are discussed.

Name: Shawn Doyle
Title: Antarctic Basal Ice as an Analog for Icy Extraterrestrial Habitats

The putative existence of water and ice throughout the solar system (e.g. Mars, Europa, and Enceladus) has raised considerable interest that these extraterrestrial worlds could plausibly harbor cryogenic habitats suitable for microbial life. As such, frozen environments on Earth such as glaciers, permafrost, and sea ice have been identified as ideal systems to investigate the survival and physiology of microorganisms under conditions analogous to those found on extraterrestrial worlds. Measurement of the concentration of gas species in air trapped in sediment-rich basal ice from Taylor Glacier, Antarctica revealed unusually high concentrations of CO2 (60,000 to 325,000 ppmv) occurring simultaneously with decreased O2 concentrations (4 to 18% of total gas volume). The high CO2 and low O2 concentrations occur concurrently with increased microbial cell abundance in the basal ice, suggesting that in situ microbial respiration altered the composition of the entrapped gas. Species of the genus Paenisporosarcina are a numerically abundant member of the microbial assemblage from the ice horizons with elevated CO2 and depleted O2 and were readily culturable from the basal ice samples examined. Metabolic experiments with Paenisporosarcina sp. TG14 revealed its ability to conduct macromolecular synthesis when frozen in basal ice melt-water at -15 °C. These results support the hypothesis that basal ice environments are microbial habitats harboring bacteria with the physiological capacity to remain metabolically active and cycle elements within the cryosphere.

Systematics, Ecology, & Evolution (SEE)

Name: Lori Patrick
Title: Morphological and Phylogenetic Community Structure of North American Desert Bats

Understanding mechanisms governing mammalian community assembly has long interested ecologists. Traditionally, ecomorphological data have been used to infer resource partitioning among taxa within a community. Recent approaches have used a regional species pool phylogeny and community membership data to make inferences about processes shaping local communities, thereby tying ecological to evolutionary processes. Here we integrate morphological community structure data with our previous work on phylogenetic community structure to evaluate effects of spatial and taxonomic scale on bats in the four great deserts of North America. We calculated community structure metrics from a distance matrix of log-transformed morphological measurements of 55 taxa. Community membership data for each desert were compiled from fieldwork, MaNIS specimen records, and published reports. We found that communities were both morphologically and phylogenetically clustered at the largest spatial (all deserts) and taxonomic (all bats) scales. At smaller spatial and taxonomic scales, communities were phylogenetically overdispersed, suggesting an important role for interspecific interactions (such as competition) in structuring communities. However there was an ambiguous pattern for individual deserts based on morphological results, with most communities not significantly different from those randomly assembled at all three taxonomic scales.

Name: Warwick Allen
Title: Latitudinal variation in tritrophic interactions associated with native and exotic genotypes of *Phragmites australis*

Despite theoretical expectations, studies of latitudinal gradients in plant-herbivore interactions have produced mixed results. Contradictory findings may be due to methodological flaws such as ignoring the influence of other trophic levels (e.g. predators, parasitoids, and inquilines), sampling few locations or a narrow latitudinal range, and not accounting for differences between plant species or genotypes. We investigated tritrophic interactions between native and invasive genotypes of common reed (*Phragmites australis*), gall-inducing *Lipara* spp. (Diptera: Chloropidae – *L. pullitarsis, L. rufitarsis* and *L. similis*), and their natural enemies and inquilines at 36 sites along the North American east coast ranging from South Florida (26.6°) to New Brunswick, Canada (48.4°). Contrary to predictions from theory, we found that the proportion of stems infested with *Lipara* spp. galls increased significantly with latitude and was also significantly higher in the native than exotic genotype. The frequency of inquiline fly co-occurrence in galls was also significantly higher in more northern sites but did not differ between genotypes. Parasitism rate of galls was virtually zero, regardless of *Lipara* species, site latitude, or *P. australis* genotype. These results indicate that enemy-release may play a part in the successful invasion of the exotic *P. australis* genotype and its associated species, and that invasion success may vary over a broad spatial scale. Future study directions include a controlled common garden experiment to examine if the gradient has a genetic basis.

Name: Ganesh P. Bhattarai
Title: Test of enemy release hypothesis with native and invasive genotypes of *Phragmites australis*

Enemy release hypothesis suggests that exotic species receive lower enemy pressure in the introduced range than in their native range which may result into a substantial increase in their

population growth rate. The reduced enemy pressure in a novel range compared to co-occurring native species has been considered as one of the primary causes of biological invasion. We examined this hypothesis with *Phragmites australis*. This species has been a member of the wetlands of North America for millennia but an introduced Eurasian genotype has been invading the wetlands of North America in the past century.

We performed a common garden experiment in Louisiana to examine the plant palatability and defenses between native and exotic genotypes of *P. australis*. Plant defense (leaf toughness), chewing damages caused by a generalist herbivore, fall armyworm (*Spodoptera frugiperda*), and herbivore performance (change in weight biomass) were evaluated. Exotic plants produced tougher leaves than natives. Although survivorship rate of larvae did not differ between native and exotic genotypes, feeding damages and larval growth rates were substantially lower on exotic genotype than the native. These results suggest that escape from natural enemies may have contributed to the invasiveness of the Eurasian genotype in North America.

Name: Bill Ludt
Title: Living outside the coral triangle: the evolutionary history and biogeography of the genus *Prionurus*

The surgeonfish family Acanthuridae contains six genera with approximately 80 species. One of these genera, *Prionurus*, has a notable distribution and is poorly studied. While the vast majority of surgeonfishes are tropical, most species within *Prionurus* have anti-tropical distributions, or are restricted to cold-water upwelling areas. Within *Prionurus* there are six species distributed in the Indian and Pacific Oceans and one species, *P. biafraensis*, in the Eastern Atlantic. There have been several previous studies examining the relationships among genera within Acanthuridae, however most of these studies have only included one or two species within the genus *Prionurus*. Here we present the evolutionary relationships within this genus using both mitochondrial and nuclear DNA to discuss this lineages biogeographic and phylogenetic history.

Name: Kalpataru Mukherjee
Title: Multiple *rdh*A Genes are Concurrently Transcribed by *Dehalogenimonas lykanthroporepellens* BL-DC-9T during Dehalorespiration with 1,2-DCA,1,2-DCP and 1,2,3-TCP.
Abstract not submitted

Name: Andrew Flick
Title: How do pathogens influence predators and parasitoids

The interactions between natural enemies can drive host populations to either grow or decline. In biological control, there is often an oversight regarding how these enemies interact, leading to failed control programs. In general, predators are a quick fix that may not be able to control the entire population, while pathogens are slow acting but can affect the entire population. Several studies have shown how the interactions between pathogens and predators can lower the control potential of the enemies. I conducted a meta-analysis of 48 published studies looking at the implications of host pathogens on host predators and parasitoids. I found evidence that pathogens have deleterious effects on life history traits of both predators and parasitoids. For instance, pathogen-infected prey moderately increased the development time of both enemies, decreased longevity of both enemies, and also decreased offspring produced in both enemies. Predators consumed more infected prey, as they were a poor source of nutrients. Parasitoids laid eggs in fewer infected prey, suggesting that the parasitoids would rather not lay eggs at all, than waste them in hosts that are not suitable. These results show a need for understanding how pathogens influence predators and parasitoids before they are applied haphazardly to a crop.