Anthropology

Dietary Reconstruction of Domestic Dogs in Precontact North America (Poster)

Amanda A. Burtt, Indiana University

The archaeology of dog-keeping by indigenous North Americans enriches our understanding of ways people conceptualized their environments in the past. Finding new ways to investigate this topic contributes to broader anthropological knowledge about relationships among humans and the natural world. This poster describes my dissertation research, both method and analysis. My research investigates ways that domestic dogs were maintained and the assumed value of dogs among Native North Americans. I employ Dental Microwear Texture Analysis (DMTA) to examine human-canine connections and dog feeding/provisioning strategies. Analyzing the diets of domestic dogs (Canis familiaris) provides a proxy for human intentions to sustain canine companions. Baseline data from members of the Family Canidae provides comparative microwear textures and a more contextualized insight into the dietary behavior of dogs that lived with humans in geographically and culturally distinct areas of North America.

Load at first bite: comparative analysis of incisor cross-sectional area in anthropoid primates (Poster)

Jacob A. Wood[1,3] and Jeremiah E. Scott[2,3]

[1]University of Indianapolis, [2]Western University of Health Sciences, [3]Southern Illinois University Carbondale

Previous studies have established that frugivorous anthropoids have larger incisor dimensions relative to body mass than folivores. This distinction is often attributed to frugivores using their incisors more intensively during ingestion than folivores, but this assumption has been challenged. In this study, we tested the hypothesis that the ability of incisors to withstand loading varies according to diet. We divided a sample of 87 anthropoid species into three dietary categories: frugivores, folivores, and hard-object feeders (HOFs). Using mesiodistal and buccolingual diameters, we computed incisor cross-sectional areas (CSAs) and size-adjusted them by dividing their square roots by jaw length, a widely used standard in studies of masticatory biomechanics. Comparisons conducted using phylogenetic generalized least squares indicate that HOFs and frugivores tend to have larger CSAs relative to jaw length than folivores, but frugivores and HOFs cannot be distinguished. We also tested for differences between platyrrhines and catarrhines, as catarrhines have been characterized as having larger incisors than platyrrhines, regardless of diet. Such differences were only apparent in folivores: in this group, platyrrhines have significantly smaller incisor CSAs than catarrhines. Our results are consistent with the hypothesis that folivores tend to load their incisors less severely than other primates. We note, however, that the strength of this signal is exaggerated by the unusually small incisors of platyrrhine folivores; in catarrhines, overlap between folivores and other groups

is extensive. Such overlap comports with data on tooth wear and ingestion behavior suggesting that some catarrhine folivores use their incisors as intensively as frugivores.

The detection of nicotine in skeletal remains to assess tobacco use in the prehistoric Midwest

Savannah Leach Newell and Jonathan A. Karty, Indiana University

North American research on tobacco in prehistory has focused on residues in ceramics and pipes, paleoethnobotanical remains, and historical inferences. This project is the first to determine individual tobacco use by testing for nicotine in human skeletal remains from the Midwest. Archaeological rib samples were taken from five Illinois sites spanning from Late Woodland (400-1050 CE) to Mississippian (1050-1400 CE) time periods. Modern samples were added to the study for comparative analysis, including rib samples from donors of Indiana University's Gross Anatomy Lab and a molar from a chronic smoker. Nicotine and cotinine, its primary metabolite, were extracted and detected using liquid chromatography-tandem mass spectrometry. Based on archaeological evidence, higher tobacco levels were expected from Mississippian than Late Woodland samples. The highest levels of nicotine and cotinine, respectively, were present in modern remains with a mean of 21.37 ng/g (3.12 ng/g); Mississippian nicotine levels had a mean of 6.07 ng/g (1.37ng/g) and Late Woodland individuals had a mean of 0.97 ng/g (1.32 ng/g). Recovery for nicotine was particularly low and inconsistent across triplicates for each individual. Relative standard deviations ranged from 47.24% to of 163.17%. Cotinine was more consistently recovered with relative standard deviations ranging from 1.19% to 59.37%. The methods presented here will be further refined to increase recovery and replication with the ultimate goal of broadening the evidence base for studying the ceremonial use of tobacco in prehistoric North America.

"What's Wrong with Forensic Anthropology These Days? How We've Forgotten the Science and Can't Do the Math." (Hot Topic)

Stephen Nawrocki, University of Indianapolis

The forensic sciences play an important role in the medicolegal system by providing independent analysis of physical evidence. The fundamental assumption is that these specialists are so well-trained in their scientific specialty that the resulting expert opinions can be relied upon to convict or free those accused of wrongdoing. Unfortunately, forensic anthropology continues to flounder in a traditional social sciences mindset: few of its practitioners receive adequate training in the scientific method, experimental design, or statistical analysis, and like most members of our modern non-agrarian society, they have little real understanding of the natural world. As a result, we continue to endure ambiguity and disagreement over basic issues involving human skeletal variability, and thus a mature forensic anthropology continues to elude us even after ~50 years of development. This presentation outlines some of the most dire problems in the field as reflected in the recent anthropological literature.

Botany

The Fort in the Doctor's House: Using Tree-Ring Growth Patterns to Discover Historic Fort Wayne

Darrin L. Rubino, Hanover College and Christopher Baas, Ball State University

Tree rings are an effective way to date the construction of historic buildings. Local Fort Wayne, Indiana lore says that the Dr. Merchant Huxford House contains timbers from Fort Wayne, the US military outpost constructed in 1794. The wilderness fort played roles in Native American and British conflicts and was the genesis for Indiana's second largest city. Using standard tree-ring methods, we sought to confirm or dispel accounts that the house contains recycled timbers from the historic fort. Through the use of crossdating, a method of comparing tree-ring growth patterns in timber of unknown age to those of a known age, each ring was assigned a calendar date. We successfully crossdated 34 samples from 28 timbers representing 6 timber types (white oak, red oak, American beech, tulip poplar, hard maple, and sycamore). White oak was the most commonly encountered timber type (28 samples from 22 timbers) in the house. We constructed a white oak chronology (a series of accurately dated and measured tree rings) of 2,291 rings spanning from 1546 to 1850. We established an 1852 construction date for the Huxford House and identified timbers contemporary to the third and final version of Fort Wayne. Using historic primary sources, we hypothesize the identity of the building we believe was recycled into the doctor's new house. The data collected from this project was a valuable addition to our tree ring database because we were able to establish chronologies in northeastern Indiana, an understudied geographical area in the state.

Creating a regional mycoflora – an Indiana case study

Stephen Russell and M. Catherine Aime, Purdue University

Attaining a reasonable understanding of the biodiversity of macrofungi for any given region has mostly eluded researchers working in North America. Despite over 200 years of mycological progress, few regions have up-to-date surveys of the macrofungi that occur in their area; this is especially true for surveys that are broadly supported by genetic data. As a part of the North American Mycoflora Project, a full-scale, state-wide survey of macrofungi is currently underway in Indiana. This includes three primary components – extensive field collecting, broad environmental sequencing, and high-level engagement of citizen scientists. Here we report preliminary progress towards a mycoflora of Indiana involving over 4,000 newly vouchered and sequenced specimens deposited in the Purdue Kriebel Fungarium since 2015. These sequenced specimens will provide the "faces" for large-scale environmental sampling efforts being conducted across the state. Methodologies for successful citizen scientist engagements are also discussed. Together, these three components combined will significantly enhance our understanding of regional diversity and phenology. It will also form a core dataset to be used in future ecological research.

Soybean: time-lapse examination of sleep movement and response to cold temperature

Jacob Hamilton, Stephen Randall, Indiana University Purdue University Indianapolis

Soybean, *Glycine max,* is known to be a cold intolerant species. The crop has major agriculture importance supplying food to a vast amount of livestock which provides food for millions of people. Plants will move in response to certain stimuli to optimize exposure to resources. For example: the closing of the petals of a flower at dusk. This study characterizes the influence of light intensity and cold temperature on the sleep movements of soybean. Ultimately a relationship between the physical changes of soybean, in response to cold, is sought to be related to physiological pathways within the plant. Soybeans from cultivar Williams 82 were planted and acclimated to a temperature of 22 ºC and 16-hour days (16: 8; light: dark) for twelve days. After this period, the plants were exposed to a cold treatment of 4 ºC. Time lapse videos were created to document physical responses. Leaf angles were defined as the angle between the stem of the plant and the unifoliate leaf being measured. Through averaging the sum of 6 individual soybean unifoliate leaves, 5 distinct characteristics were observed consistently in response to cold, dark, and light: 1) leaf angles decreased in response to dark; 2) leaf angles increased in response to light; 3) leaves appeared to oscillate in the dark as if “searching for optimal positioning”; 4) anticipation of the dark light by decreasing leaf angle prior to exposure to dark; and 5) cold treatment causes a continued reduction of sleep movement. Currently, these five characteristics are being observed over a spectrum of light intensities ~ as low as 20 µmol of photons / m^2 * s and as high as 400 µmol of photons / m^2 * s.

Soil transfer from a mesic prairie to conserve a native seed bank and control non-native plants (Poster)

Tessa M. Aby, Adam R. Warrix, and Jordan M. Marshall, Indiana University-Purdue University Fort Wayne

Non-native species colonize and can become dominant in managed ecosystems. Such species often invade disturbed areas, which are a common result of restoration or other management activities. In dredging open water bodies at Eagle Marsh Nature Preserve, artificial mounds were constructed with the soil removed. This movement of soil created a disturbance allowing the recruitment and colonization of non-native species. The purpose of this study was to quantify the effectiveness of transferring a native seed bank from a restored mesic prairie to the artificial mounds as a method of controlling non-native plant species, which was tested with greenhouse and field experimentation. Soil samples containing the seed bank were moved to a greenhouse to quantify plant emergence following different physical soil treatments. Prior to the field experiment, a plant survey of the mounds was conducted in July 2014. In October 2014, 144 m^3 of soil was transferred from the donor site to the recipient mounds. During the growing season immediately after the move (July 2015), native species diversity increased slightly while the diversity of invasive species decreased dramatically at the recipient location. After allowing establishment of the community (July 2017), both native and invasive species diversity increased dramatically – matching greenhouse values from the donor site before the transfer. Across all surveys, native species outnumber non-native species 2:1. While the soil transfer greatly decreased the number of non-native species immediately after transfer, after an

additional 2-years there were more non-native species than before the transfer. Fortunately, a similar increase in native species did occur. The primary goal of this management was to move an existing native seed bank. Some benefits may arise from moving soil locally to transfer an established seed bank. However, such management may be facilitating colonization of non-native species.

Evaluation of Second generation transgenic soybean plants for resistance against soybean aphids (Poster)

Shailesh Raj Acharya and Vamsi Nalam, Indiana University Purdue University Fort Wayne

The soybean aphid continues to be an important pest in major soybean growing regions around US. The current consensus management recommendations that have been developed over more than a decade of research include cultural, genetic, economic and chemical controls. Although, these strategies reduce the threat, they do not completely eliminate soybean aphid as a pest. As a complement to current management practices we propose utilizing a biotechnological approach to enhance plant immunity. The genetic manipulation of components of the plants own defense signaling pathways offers an attractive strategy for boosting plant defenses. Regulatory genes that control the expression of multiple defenses are excellent targets for developing broad spectrum of pathogens. This approach has several important advantages including a minimal impact on crop yield and economically important plant, reduced impact of the environment and on natural enemies of the aphid due to reduced insecticide sprays. A defense regulatory gene PHYTOALEXIN DEFICINT4 (PAD4), is an excellent candidate for over-expression soybean plants due to its important role in plant defenses against a broad-spectrum of pathogens. Previously, we have developed transgenic soybean plants that over-express the native PAD4 gene under the control of a constitutive promoter. Evaluation of T0 and T1 plants from 5 transgenic events indicates that the PAD4 gene is indeed present and expressed in the transgenic plants. Soybean infestation assays indicate that the transgenic plants display enhanced resistance to soybean aphids. Data from the screening of second-generation transgenic plants for response to the soybean aphid indicate that the transgenic plants continue to show enhanced resistance to soybean aphids. Our research shows a central role for PAD4 in plant defense against soybean aphids and the genetic manipulation of regulatory genes in crop plants can produce broad spectrum of resistance associated with a lack of detrimental effects on crop yield and plant traits.

Cell Biology

G4R1/RHAU (DHX36) is phosphorylated upon entry into mitosis (Poster)

Adam Richardson, Philip Smaldino, Ball State University

G-quadruplexes (G4) are guanine-rich DNA or RNA secondary structures composed of two or more guanine-tetrads bonded by Hoogsteen hydrogen interactions and further stabilized by monovalent cations. These self-associating structures have been observed throughout the human genome, particularly in gene promoters, telomeres, DNA replication origins, untranslated regions (UTRs), and pre-mRNA. These structures influence genomic stability and cellular processes, such as DNA replication, telomere maintenance, transcription, translation, and pre-mRNA processing. The human enzyme G4-Resolvase1 (G4R1), also known as RHAU or DHX36, is responsible for the majority of G-quadruplex resolvase activity in human cells. G4R1 is also involved in the regulation of many genes involved in cell cycle regulation, and knocking out G4R1 led to an enrichment of G-quadruplexes at their promoters, blocking transcription. Overexpression of G4R1 has been found in breast cancer cells and also associated with transcription factors involved in cell proliferation and cancer. A quantitative phosphoproteomic study found that G4R1 is phosphorylated at amino acid residues S136 and S161 during mitosis. Also, the last three amino acids, Y1006, Y1007, and S1008, at the C-terminus are phosphosites, which might be involved in cellular localization and inhibition of enzymatic activity. However, the details of this modification during different stages of mitosis are unknown. Phosphorylation affinity gel electrophoresis followed by western blot analysis was done to assess the phosphorylation status of G4R1 during different mitotic stages. Initial results suggesting that G4R1 is phosphorylated upon entry into mitosis and dephosphorylated upon exiting mitosis will be presented and future directions will be discussed, including approaches to investigate cellular localization of G4R1 and the effects of phosphorylation on specific residues.

Rescue of Zoledronate-Induced Apoptosis in Human Oral Keratinocytes (Poster)

Jesse Maguire, Preston Rippe, Ethan Blake, Nur Bashira Binti Shaharuddin, Wen Lin Chai, and Dan Jones, Indiana Wesleyan University, University of Malaysia

Zoledronate (ZOL) is a nitrogen-containing bisphosphonate, commonly used as an osteoclast inhibitor in diseases such as bone cancer and osteoporosis. It also binds to hydroxyapatite, decreasing calcium release, and thereby increases bone density. ZOL has a side effect in cancer patients—osteonecrosis localized to the actively remodeling bone and to the soft tissue of the jaw. ZOL blocks completion of the mevalonate pathway, leading to cell death. Geranylgeraniol (GGOH) is a naturally occurring intermediate of this pathway, acting downstream of the ZOL block to potentially rescue cells. Other potential rescue agents include 100 microM citrate (naturally occurring salivary calcium chelator), 10 microM melatonin (antioxidant), and 2 milliM Etidronate (non nitrogen-containing bisphosphonate thought to block cell uptake of nitrogen-containing bisphosphonates). The purpose of this study was to compare the four agents in terms of rescue from ZOL-induced apoptosis (programmed cell death) in oral

epithelium. Apoptosis was measured by two methods: 1) determination of caspase 3 (apoptosis executioner) levels in treated cells and 2) determination of the number of treated cells displaying phosphatidyl serine (a cell marker for apoptosis) on their surfaces through the flow cytometry-based Nexin assay. These experiments revealed that normal human oral keratinocytes show a mild but significant increase (1.8-fold) in apoptosis when treated with 50 microM ZOL for 72 hours. However, the immortalized OKF6/TERT2 oral keratinocytes showed no significant increase in apoptosis with ZOL treatment, and no rescue when paired with 10 microM GGOH or any of the other agents.

Investigation of FSHR-1 as a Potential Anaphase Promoting Complex Substrate in *C. elegans* Neurons (Poster)

David Emch, Kyle Cherry, Amy Godfrey, and Jennifer R. Kowalski, Butler University

Healthy nervous system function requires a balance of excitatory and inhibitory (E:I) neuronal signaling. This E:I balance relies upon the ubiquitin system, a process that regulates many proteins within cells through enzymatic tagging with ubiquitin polypeptides to affect their activity and abundance. Proper function of ubiquitin enzymes like the Anaphase Promoting Complex (APC), a ubiquitin ligase, and its substrates is needed for regulation of E:I balance. E:I imbalances occur in many neurological diseases, so a better understanding of the APC and its substrates may lead to improved treatments for diseases like Parkinson's and epilepsy. Our previous data showed the APC acts in inhibitory *C. elegans* GABA neurons to promote GABA neurotransmitter release, but APC substrates relevant for this effect are unknown. Follicle Stimulating Hormone Receptor 1 (FSHR-1) is a possible APC substrate. FSHR-1 possesses recognition sequences for APC interaction, and both *fshr-1* and *fshr-1;apc* loss of function mutants exhibit decreased muscle contraction, indicating FSHR-1 acts after the APC to control signaling. I am testing the hypothesis that FSHR-1 is an APC substrate by generating worms expressing fluorescently tagged FSHR-1 and quantitatively measuring changes in neuronal FSHR-1::GFP concentrations when the APC is non-functional. Current work is also focused on determining the cell types in which FSHR-1 is endogenously expressed. If FSHR-1 is a target of the APC enzyme, I expect higher FSHR-1::GFP concentrations in APC loss of function worms and for FSHR-1 to be expressed in neuron classes in which the APC is expressed.

Effects of Endoplasmic Reticulum Stress on Hrd1-Mediated Protein Quality Control

Bryce Buchanan, Laura Scanameo, Eric M. Rubenstein, Ball State University

Protein quality control is critical for cellular health. The Endoplasmic Reticulum (ER)-Associated Degradation (ERAD) protein quality control system degrades aberrant proteins at the ER membrane. The ubiquitin ligase Hrd1 mediates the degradation of three different classes of ERAD substrates: ERAD-L (lumen), -M (membrane), and -T (translocon). These substrates differ in the position of aberrations relative to the ER membrane. Proteins with aberrations within a membrane-spanning segment are ERAD-M substrates. Those with aberrations in a luminal portion are ERAD-L substrates. Finally, proteins that aberrantly engage the translocon are ERAD-T substrates. ER stress occurs when misfolded proteins accumulate in the ER. Multiple human diseases, including Parkinson's disease and diabetes, are characterized by elevated

levels of ER stress. The molecular consequences of ER stress in disease states and the extent to which ER stress contributes to pathology is not clear. In this study, we systematically evaluate the effect of ER stress on ERAD in the model organism *Saccharomyces cerevisiae* by comparing the abundance and rate of degradation of a panel of model substrate proteins in stress and non-stress conditions. We also determine the impact of overexpression of a luminal chaperone, Kar2, on ERAD-T during acute ER stress. Our data suggest that ERAD-T and -L are impaired by ER stress, while ERAD-M is unaffected. Further, while previous studies demonstrated that Kar2 overexpression rescues ERAD-L during acute ER stress, increased expression of Kar2 does not rescue ERAD-T during ER stress. This is consistent with a novel mechanism of impairment of ERAD-T by ER stress. A better understanding of the effects of ER stress on cellular physiology may facilitate the discovery of therapeutic targets for diseases characterized by increased levels of ER stress.

Localization of the SUMO conjugating enzyme UBC-9 in *C. elegans* neurons (Poster)

Elly B. Mawi and Jennifer R. Kowalski , Butler University

The nervous system relies on tight regulation of proteins to maintain a balance of excitatory to inhibitory signaling for proper function. SUMO (small ubiquitin-like modifier) polypeptides are attached to cellular proteins by the UBC-9 conjugating enzyme, thereby regulating such functions as neuronal signaling. The molecular mechanisms by which SUMOylation affects neuronal signaling balance remain unknown, yet multiple neuronal proteins are SUMOylated, including several implicated in neurodegenerative diseases. I imaged transgenic worms expressing fluorescently tagged UBC-9 to investigate where UBC-9 is expressed in neurons and whether this enzyme colocalizes with the *Caenorhabditis elegans* SUMO polypeptide (SMO-1) at neuromuscular synapses. My hypothesis is that UBC-9 acts in presynaptic motor neurons to control signaling via its SUMO-conjugating activity. I constructed a UBC-9 strain that fluoresces green in inhibitory motor neurons and crossed it to a strain in which inhibitory synapses fluoresce red. Imaging results indicate that green UBC-9 overlaps with red synaptic vesicle proteins, suggesting UBC-9 can localize to neuromuscular synapses. Additional imaging of strains in which SMO-1 fluoresces green and synapses fluoresce red are underway. UBC-9 and SMO-1 strains will eventually be crossed and imaged to see if the two proteins also colocalize. Future studies will investigate whether the UBC-9 catalytic site is required for UBC-9's effects on neuromuscular signaling and identify UBC-9 substrates. Given the similarities between *C. elegans* and mammalian nervous systems, understanding how SUMO enzymes control the balance of normal neuronal signaling may provide important information related to human neurobiology and contribute to research on diseases of imbalanced signaling.

A pharmacological model of TRPA1-mediated nociception in the zebrafish for therapeutic discovery (Poster)

Emre Coskun, Logan Ganzen, MeeJung Ko, and Yuk Fai Leung, Purdue University

According to a new analysis of National Health Interview Survey data, NIH estimates that 25.3 million adults are suffering from chronic pain as of August 2012. Along with the increasing prevalence of chronic pain, the use of narcotic painkillers to relieve it has also been increasing. These drugs are often associated with side effects, overdose, and dependence. In addition, the current opioid crisis in Indiana costs approximately $1 billion to the state each year according to the IUPUI School of Public Health. The Transient Receptor Potential Channel, subfamily A1 (TRPA1) is a channel involved in chronic neuropathic pain transmission (nociception) in humans, and it is also expressed in zebrafish. Agonizing TRPA1 channels via pharmacological treatment in zebrafish larvae results in a locomotor swimming response. The goal of this project is to utilize this nociceptive-like swimming behavior to develop a behavioral assay that models chronic neuropathic pain in humans and use it to identify novel non-narcotic treatment options. We have identified a TRPA1 agonist (ASP7663) that results in a sustained locomotor response when zebrafish larvae at 5 days post-fertilization (dpf) are exposed to the chemical in their water. We interpret this sustained nociceptive-like behavior as modeling a chronic pain condition in human patients. To determine if this nociceptive-like behavior can be blocked by antagonizing TRPA1, we pre-treated 5 dpf zebrafish larvae with the TRPA1 antagonists HC30031 and TCS5861528 before challenging them with the ASP7663 agonist. We found that these antagonists will block the agonist-mediated swimming response and prevent the nociceptive-like behavior in the zebrafish. Utilizing this developed behavioral assay, we will screen drug libraries to identify compounds that can block the nociceptive-like swimming behavior induced by agonizing TRPA1 in zebrafish. We expect that this work will provide both novel therapeutics for chronic pain treatment, and mechanistic insight into TRPA1-mediated pain transmission.

A Potentially General Role for a Conserved Protease in Translocon-Associated Quality Control (Poster)

Avery Kirschbaum, Eric Rubenstein, Ball State University

Translocation into the endoplasmic reticulum (ER) is essential for many proteins, as the ER provides an environment suitable for protein folding and modification. Proteins are transported into the ER through a channel called the translocon. Occasionally, proteins moving through the translocon aberrantly or persistently occupy the channel. Although functional ER translocation has been intensely studied, quality control mechanisms have not been fully elucidated. In humans, the protein apolipoprotein B (apoB), the major protein component of low-density lipoproteins (i.e. bad cholesterol), can become stalled in the translocon if its lipid binding partners are not present in the ER. In yeast, the ubiquitin ligase Hrd1 ubiquitylates the model translocon-clogging protein *Deg1*-Sec62, thereby targeting the protein for degradation. It has previously been shown that a conserved protease cleaves a different artificial model protein that aberrantly engages in the translocon. However, the extent of this protease's involvement in quality control at the translocon is not clear. In this study, we investigate the effects of this protease on abundance of the translocon-clogging protein *Deg1*-Sec62 and growth

of *Saccharomyces cerevisiae.* Analysis of protein abundance suggests that this protease cooperates with Hrd1 to target the model translocon clogging protein *Deg1*-Sec62 for degradation. Yeast strains lacking both *HRD1* and the gene encoding the protease exhibit a synthetic phenotype, growing more slowly than wild type or single knockout strains. A better understanding of the mechanisms that clear aberrantly clogged translocons could potentially lead to new treatment options for high cholesterol.

Characterization of learning, memory, and seizures in complement C3 knockout mice (Poster)

Zoe D. Carlson-Stadler, Nicole D. Schartz, and Amy L. Brewster

The classical complement pathway is part of the immune complement system. Activation of this pathway is initiated by the complement C1q and leads to downstream activation of the complement C3. C3 is cleaved into C3a and C3b to regulate inflammatory and phagocytic responses, respectively. We recently found increased activation of the C3 in human and experimental epilepsy. In order to investigate the role of C3 in epilepsy we obtained constitutive C3 knockout (KO) mice. The objective of this study was to determine the baseline learning and memory, locomotion, anxiety, and seizure threshold of C3KO compared to wild type (WT) mice. We tested hippocampal-dependent learning and memory using the Barnes Maze (BM) and the Novel Object Recognition (NOR) test, locomotion and anxiety with the Open Field (OF), and determined seizure threshold to two different doses of the chemoconvulsant pilocarpine (325 and 350 mg/kg). We found that WT and C3KO mice displayed similar learning in the BM assessed as decreased time to reach a hidden box in an open arena during 4 days of training. Similarly, C3KO and WT mice were able to differentiate and preferentially explore a novel object compared to a familiar one in the NOR test. OF showed that WT and C3KO mice displayed similar locomotion (distance traveled and speed), and anxiety behaviors (freezing and % in inner area of the OF). Lastly, following administration of pilocarpine, C3KO and WT mice developed prolonged behavioral seizures consistently according to the Racine seizure scale (1-6). Both pilocarpine doses (325 and 350 mg/kg) promoted the development of stage 4.5-6 seizures in all genotypes. Taken together these data suggest that C3KO and WT mice have similar basal learning behaviors and seizure thresholds. Future studies will test whether C3KO mice are protected against seizure-induced injury and the subsequent development of epilepsy.

Cold Regulation of the Dehydrins in Diploid Strawberry

Zach Deitch, Stephen Randall, Indiana University Purdue University Indianapolis, Nelson Osuagwu, Rob Wilson, Inland Norway University, Isam Fattash, American University of Madaba, Muath Alsheikh, Graminor Breeding Ltd. and Norwegian University of Life Sciences

Strawberry (*Fragaria* spp*.)* is an agricultural crop grown in temperate regions that has high variability in its susceptibility to freezing injury. In order to breed cultivars for frost and freezing tolerance, identification of molecular markers associated with low temperature tolerance is advantageous. Previous work identified seven putative dehydrins in *Fragaria* and demonstrated that cold tolerance positively correlated with dehydrin protein expression levels. In an effort to understand the cold-regulated expression of dehydrins as a function of cold exposure time, two dehydrins were purified and positively identified by mass spectroscopy and determined to be COR47-like (SKn) and XERO2-like (YnSKn). Utilizing RT-qPCR and western blots, the levels of RNA transcripts and subsequent proteins corresponding to the two dehydrins were examined in strongly cold tolerant (ALTA) and lesser cold tolerant (FDP817, NCGR1363) *Fragaria* cultivars. The COR47-like (SKn) and XERO2-like (YnSKn) dehydrins both had higher transcript accumulation and protein levels in the more cold tolerant line in comparison to the other two less cold tolerant lines. Discrepancies in transcript and resulting protein with COR47 in Alta were observed at different timepoints where protein preceded an increase in RNA, but this trend was not seen with the XERO2 data. Investigation into this non-congruence is necessary to identify at what level COR47 is being regulated. Multiple isoforms of the gene exist for COR47 and could possibly account for the missing transcript counts. Primers were designed for both isoforms and RT-qPCR was performed to examine the transcripts of COR47 more closely. Overall it is concluded that dehydrin transcripts are potential biomarkers for identifying low temperature tolerance in diploid strawberry. The specificity of individual dehydrin contribution to cold tolerance is still being investigated.

Comparison of Soybean and Arabidopsis using the RD29A Cold-Regulated Promoter

Adib Behrouzi, Jennifer Robison, Stephen Randall, Indiana University Purdue University Indianapolis

The inability of certain agriculturally important plants to tolerate extreme environmental conditions is a concern for the maintenance and improvement of food production. Previous work shows that the *Arabidopsis thaliana* promoter RD29A is responsive to a variety of abiotic stresses, and the RD29A gene is involved in cold-stress adaptation. The RD29A reporter construct (AtRD29A::GUS/GFP) was constructed and transgenically introduced into *Arabidopsis thaliana* (At) and *Glycine max* (Gm). Homozygous AtRD29A::GUS/GFP transgenic lines (four At and three Gm) have been identified. Examination of Arabidopsis seedlings showed GUS activity in trichomes, roots, and leaf tips. Tissue and organ expression during cold treatment of RD29A::GUS/GFP in a cold-tolerant plant (At) was compared to the cold-intolerant soybean plant (Gm). Lines of At and Gm were examined quantitatively at different time points in an assay using the substrate 4-methyl-umbelliferyl-β-D-glucuronide (MUG) with extracts of control (22°C) plants or plants treated for 2 days in the cold (4°C). The GUS assay showed a strong increase in cold-driven RD29A expression of GUS activity in both At and Gm. These data

support the hypothesis that RD29A is cold-regulated in both Arabidopsis and soybean. This promoter will be a useful tool to help understand how cold tolerance occurs in cold-tolerant plants and may help determine cold-responses in intolerant plants such as soybean.

Expression of IRBIT along the gastrointestinal tract (Poster)

Marie Dix*, Payton Klosa*, Natasa Petreska, Amanda Bazaldua, Chase Jones, Emily Hughes and Patrice G Bouyer. * equally contributed author. Valparaiso University.

Over the past decade, IRBIT (inositol-1,4,5-trisphosphate (IP_3) receptors binding protein released with IP_3) has been shown to be a powerful regulator of fluid and electrolyte transport in the pancreatic duct and other epithelial cells. However, the regulation and expression of IRBIT is not well characterized. The laboratory demonstrated that inflammatory cytokines (*e.g.,* interleukin 13, tumor necrosis factor alpha) decrease IRBIT expression in T84 and Caco2 cells. It is believed that IRBIT is ubiquitously expressed in cells. In this study, the laboratory tested the expression of IRBIT along the gastrointestinal tract in human biopsies, rat and mouse intestinal epithelial cells. From human histological sections, we found that IRBIT is expressed in the parietal cells of the stomach antrum and in the intestinal epithelial cells of the duodenum. On the other hand, faint to no expression was found in the human colon. Next, we tested IRBIT protein expression using Western blotting. IRBIT was expressed in mouse duodenum epithelial cells. In addition, from a duplicate experiment, we found that IRBIT is expressed in the duodenum, jejunum, ileum, proximal and distal colon rat intestinal epithelial cells. In rats, IRBIT expression was higher in ileum compared to the rest of the other segments. In conclusion, we report that IRBIT is present in the gastrointestinal tract but its expression varies along the different segments of the intestine and it is different between human and rat especially in the colon.

Forkhead protein-regulated genes mediating cardiac progenitor cell specification and division

Andrew J. Kump, Manoj Panta, Ye Chen, Xujing Wang, Shaad M. Ahmad.
Indiana State University; National Heart, Lung and Blood Institute, NIH.

While at least eight Forkhead (Fkh/Fox) transcription factors (TFs) are required for proper cardiac development in mammals and mutations in four Fkh genes have been linked to human congenital heart defects, relatively little is known about the molecular mechanisms or the downstream target genes by which these Fkh TF-mediated cardiogenic functions are brought about. We previously showed that the *Drosophila* Fkh gene *jumeau* (*jumu*) mediates both cardiac progenitor cell specification by regulating the expression of Heartless and Frizzled, the receptors of the FGF and Wnt signaling pathways respectively, and cardiac progenitor cell division by regulating the activity of the Polo kinase. However, the significant enrichment of Fkh TF binding sites in the enhancers of cardiac genes suggested that *jumu* might be utilizing additional downstream target genes to regulate these two cardiogenic processes. Using RNA-sequencing to compare genome-wide transcriptional expression profiles of flow cytometry-purified mesodermal cells from wild-type and *jumu* loss-of-function embryos, we detected 1,272 putative *jumu* targets, i.e. genes exhibiting significant differential expression in *jumu* mutants

compared to wild-type. Our ongoing phenotypic analysis of a prioritized subset of these downstream targets with amorphic and hypomorphic mutations shows that *jumu* does indeed transcriptionally activate at least eight additional genes mediating both asymmetric and symmetric cardiac progenitor cell divisions—*Incenp*, *barr*, *neb*, *Cks30A*, *SMC2*, *scra*, and *glu*—and yet another gene, *tum*, required for the cytokinesis of cardiac progenitor cells. Intriguingly, *Cks30A*, one of the *jumu*-regulated genes mediating cardiac progenitor cell division, also appears to play a role in cardiac progenitor specification. We are using more detailed phenotypic analysis, genetic interaction assays, epistasis tests, and rescue assays between these *jumu*targets themselves and also between the *jumu* targets and genes previously known to be involved in cardiac progenitor specification and cell division to determine their roles and positions in these cardiogenic pathways.

Targeting the host cell rather than the bacteria to limit infection

Susan A. McDowell, Ball State University

By hijacking host cell machinery, invading bacteria establish an intracellular niche that provides a barrier from first-line antimicrobials. Bacteria that re-emerge after antimicrobial treatment is complete and enter adjacent host cells initiate a cycle of recurrent infection. The goal of our research group is to identify host-targeted inhibitors that limit bacterial invasion to break the cycle of recurrent infection. We discovered the host regulatory protein CDC42 is essential to invasion by *Staphylococcus aureus* and by *Streptococcus pyogenes*, two invasive bacterial pathogens associated with elevated antibiotic failure rates and recurrent infection. We found the statin drug simvastatin limits function of RHO-GTPase family members, including CDC42, and decreases host cell invasion by both pathogens. We are exploring repurposing of simvastatin and targeted CDC42 inhibition by the first-in-class small molecule inhibitor ML141 as novel approaches for recurrent staphylococcal and streptococcal infection.

Identification of a gene signature associated with elevated bone formation rate in aging mice

Krista Jackson, Aaron Hudnall, **Jonathan W. Lowery**, Marian University College of Osteopathic Medicine

Osteoporosis results from bone resorption exceeding bone formation and places individuals at enhanced risk for fracture, disability, and death. By 2020, an estimated 1.3 million senior adults in Indiana will either have osteoporosis or be at high risk for developing it. We reported that deletion of the *Bmpr2* gene in skeletal progenitor cells of mice causes substantially elevated bone mass in young adulthood. Current work indicates the age-related decline in bone mass of *Bmpr2* mutant mice is reduced approximately three-fold compared to control mice; quantification of serum bone turnover markers indicates this is caused by a sustained increase in bone formation rate to at least 35 weeks of age. Here, we determine the gene signature associated with elevated bone formation rate using genome-wide transcriptome profiling in bones of 35-week-old control and *Bmpr2* mutant mice. Stringent criteria comparing the expression data to eight well-accepted housekeeping genes reveals that, out of 24980 exon-containing transcripts detected in both genotypes, 120 genes were up-regulated and 131 were

down-regulated at least two-fold compared to controls. An additional 237 genes were detected in only one genotype. We refined this putative signature by performing transcriptome profiling in these animals at 55 weeks of age when bone formation rate is no longer elevated. Of those genes altered at 35 weeks of age, 178 (71%) were either no longer up-regulated or down-regulated in *Bmpr2* mutant mice by 55 weeks of age. Bioinformatic analyses on this refined gene set indicates elevated bone formation rate in *Bmpr2* mutant mice correlates with enrichment for genes containing binding sites for transcription factors associated with skeletal homeostasis. Further, several genes corresponding with osteoblast differentiation and activity are up-regulated in *Bmpr2* mutant mice. Collectively, our findings provide insight into the mechanisms regulating age-related bone loss and highlight potential targets for therapeutic modulation of bone mass.

Inhibition of breast cancer cell proliferation by dietary flavonoids fisetin, hesperetin & luteolin (Poster)

Hayley Sabol, Tawan Beaumont, Mariah Castañon, and Kimberly M. Baker,
University of Indianapolis

Epidemiological studies suggest that a diet rich in plant flavonoids may prevent cancer. Fisetin, luteolin, and hesperetin, dietary flavonoids found in a variety of fruits and vegetables, have been shown to exhibit anticancer activity *in vitro*. In this study, we evaluated the antiproliferative effects of fisetin, luteolin, and hesperetin, individually and in combination, using MCF-7 breast cancer cells. We found that fisetin, luteolin, and hesperetin each inhibited MCF-7 cell proliferation in a dose dependent manner. Furthermore, we found that co-administration of lower doses of fisetin and luteolin or hesperetin and luteolin led to a greater inhibition of cell proliferation than either agent alone at the same dose and indicate an additive effect. These results suggest that combinatorial treatments using fisetin, luteolin, and hesperetin may be an effective chemotherapeutic strategy against breast cancer.

Investigating the Potential Roles of G4R1 in the neurodegenerative disease, ALS (Poster)

Antonio Chambers and Phil Smaldino, Ball State University

Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disease that results in muscle and respiratory failure. The most common mutation in hereditary ALS occurs in the *C9orf72* gene. In this mutation, a guanine (G)-rich segment of DNA (GGGGCC) is repeated hundreds to thousands of times; compared to healthy individuals who have less than 30 of these repeats. The expansion of this repeat forms an over-abundance of G-quadruplexes (G4) that contribute to ALS pathology. G4s are structures that form from G-rich segments of DNA and RNA. G4s play several roles in the cell, including functions in regulation of replication, transcription, translation, telomere maintenance, and cell development. Cellular enzymes have evolved to regulate these G4 structures. The enzyme that accounts for the majority of all G4 helicase activity is G4 Resolvase 1 (G4R1) (aliases: RHAU, DHX36). Because G4 abundance contributes to ALS pathology, we expect G4R1, as the major G4 helicase, to be dysregulated. We hypothesize that G4R1 expression is upregulated in ALS patient cells compared to healthy cells. We are testing this hypothesis using western blot analysis of ALS patient cell lysates.

Furthermore, we hypothesize that overall G4 helicase activity is dysregulated in ALS patient cells. I am currently quantifying the overall G4 DNA helicase activity in cell lysates. This work will likely lead to a better understanding of the molecular biology of this disease and contribute to rational design of future therapeutics and treatment.

Investigation of FSHR-1 and its downstream pathway components in neuromuscular signaling (Poster)

David Ryskamp, Sarah Olofsson, Allyson Munneke, Alexandra Jennings, Julie Kolnik, Jennifer Kowalski, Department of Biological Sciences, Butler University

G protein-coupled receptors (GPCRs) regulate diverse processes in cells ranging from heart cells to neurons. FSHR-1 is a conserved GPCR that controls reproductive processes in metazoans, but is also expressed in nervous systems, such as that of *Caenorhabditis elegans,* Behavioral data indicate that FSHR-1 regulates signaling at the *C. elegans* neuromuscular junction (NMJ) where a balance of acetylcholine (ACh) and GABA signaling controls muscle contraction. Loss-of-function (lf) *C. elegans* mutants in *fshr-1* exhibit reduced muscle contraction, which is rescued by re-expression of *fshr-1* in either neuron type. However, the pathways activated by FSHR-1 at the NMJ are unknown. In the *C. elegans* germline, FSHR-1 lies upstream of GSA-1 and ACY-1; PKA functions downstream of GSA-1 and ACY-1 to control neuromuscular signaling. We hypothesized that in *C. elegans* GABA and ACh neurons, FSHR-1 promotes muscle contraction using the GSA-1/ACY-1/PKA pathway to regulate synaptic vesicle release. We used paralysis induced by aldicarb, an acetylcholinesterase inhibitor, to measure neuromuscular signaling in worms with mutations in suspected FSHR-1 pathway components. Worms with gain-of-function (gf) mutations in *gsa-1*, *acy-1*, or *pka* paralyzed faster on aldicarb than wild type controls. Worms with *fshr-1(lf)* and *gsa-1(gf)* or *acy-1(gf)* had paralysis rates comparable to *acy-1(gf)* or *gsa-1(gf)* single mutants, suggesting PKA, ACY-1, and GSA-1 influence neuromuscular signaling downstream of FSHR-1. Ongoing experiments are testing whether the transcription factor CREB or the active zone protein UNC-10 RIM link FSHR-1 to synaptic vesicle release further downstream. Future investigations will test proposed G protein pathway components in a cell type-specific manner.

Investigation of FSHR1 in regulation of UNC-10 RIM synaptic localization in C. *elegans* (Poster)

Morgan Buckley and Jennifer R. Kowalski, Butler University

Regulation of neuronal signaling (synaptic transmission) is essential for nervous system function. Release of neurotransmitters from presynaptic vesicles, which are necessary for neuronal signaling, are controlled by presynaptic regulatory proteins. G protein-coupled receptors (GPCRs) are a class of synaptic membrane proteins with diverse functions across eukaryotes and are targets of many drugs. In the nervous system, GPCRs are receptors for neurotransmitters or neuropeptides, which regulate neurotransmitter release. Follicle Stimulating hormone receptor (FSHR1) is a conserved GPCR that regulates reproductive processes; FSHR1 is also present in the nervous system, where it regulates neuromuscular signaling in the roundworm *Caenorhabditis elegans*. Inhibition of *fshr1* expression leads to

reduced muscle contraction and synaptic vesicles accumulation at presynapses in worms, likely reducing neurotransmitter release; however, the specific pathway activated by FSHR1 to affect synaptic transmission is unknown. UNC 10 (RIM in mammals) is a candidate effector of FSHR1 that regulates synaptic vesicle release at the *C. elegans* neuromuscular junction. To determine whether FSHR1 regulates UNC-10 synaptic localization, the dorsal nerve cords of wild type and *fshr-1*loss of function mutant worms, in which UNC-10 is tagged with green fluorescent protein, were imaged on a fluorescence microscope and the level of UNC-10 abundance measured. Changes in synaptic levels of UNC-10 in *fshr-1* mutants would indicate that FSHR-1 impacts UNC-10 at neuromuscular synapses, potentially explaining FSHR-1 effects on synaptic transmission. Given the high conservation of gene and nervous system organization with humans, determining the roles of FSHR-1 and UNC-10 may aid understanding of human nervous system disorders.

Investigation of new Notch target genes in stem cell maintenance (Poster)

Joseph Ballard, Hunter Jones, Erika Sorensen-Kamakian, Wabash College

The *C. elegans* roundworm is a powerful genetic model used to study genes important in human biology and disease because ~38% of worm genes have human orthologs and most components in known signaling pathways are conserved (1). *C. elegans* exists primarily as a self-fertilizing hermaphrodite; therefore, the germline (the lineage of cells responsible for producing gametes) generates both oocytes and sperm. The germline maintains a population of germline stem cells (GSCs) in order to create a continuous supply of gametes that can give rise to a new animal (3). Stem cells are undifferentiated, have the capacity to self-renew, and are totipotent (capable of giving rise to all cell types). In *C. elegans*, the conserved Notch signaling pathway functions to maintain GSCs in a totipotent state and is the only known pathway known to do so (3). While much research has been done to discover the mechanisms of the Notch signaling pathway, a large question remains unanswered; which Notch target genes are used to control self-renewal, totipotency and differentiation? Specifically, we want to determine if *lst-4*, *ccar-1*, *mcm-6*, and *csr-1* are important for GSC maintenance using *in situ* hybridization and RNAi gene depletion.

Investigation of SYD-2 as a neuronal substrate of the anaphase-promoting complex in *C. elegans* (Poster)

Taylor Prechtel, Jennifer Kowalski, Butler University

Neurons communicate at specialized cell junctions called synapses by releasing chemical neurotransmitters (NTs) that bind to postsynaptic neurons, causing either inhibitory or excitatory responses. Proteins in pre- and post-synaptic neurons work to regulate synaptic transmission. One regulator of synaptic signaling is ubiquitin (Ub), a polypeptide that causes protein degradation. The ubiquitin-proteasome system (UPS) degrades most proteins, and a failure to properly degrade proteins occurs in many neurodegenerative diseases. The Kowalski lab showed the anaphase-promoting complex (APC), an enzyme that adds Ub to proteins, regulates inhibitory synaptic signaling at neuromuscular synapses in *C. elegans* roundworms. Potential APC substrates at this synapse include SYD-2, a synaptic scaffolding protein, and we

found synaptic SYD-2 levels increase in APC loss of function mutants. However, it is unclear if the APC or proteasome directly control SYD-2 ubiquitination. First, if SYD-2 is a substrate of the APC at the *C. elegans* NMJ, then Ub-SYD-2 levels should fall and overall SYD-2 abundance increase in APC mutants compared to wildtype worms. Thus far, a Ub-SYD-2 band has been detected, but additional tests are needed to determine if the band is specific and how it compares to Ub-SYD-2 levels in wildtype animals. Second, if the proteasome regulates SYD-2, then total SYD-2 and Ub-SYD-2 levels should increase with proteasome inhibition. Preliminary results have shown no apparent difference in SYD-2 levels at the inhibitor concentrations or exposure times tested. Results of additional trouble-shooting will be presented. These studies are important in determining if SYD-2 is a potential substrate for the APC.

Investigation of *oxi-1* function in neuromuscular signaling under oxidative stress (Poster)

Barry Wei and Jennifer R. Kowalski, Butler University

Oxidative stress is a condition in which the balance between free radicals and antioxidants is disrupted within an organism. Prolonged oxidative stress can lead to neurodegeneration. The gene *oxi-1* encodes a protein homologous to the UBE3B ubiquitin protein ligase, which is involved in protein degradation important for reducing oxidative stress effects. *UBE3B* mutations are linked to a human intellectual disability syndrome. Preliminary data focusing on the role of *oxi-1* in neuromuscular signaling using *Caenorhabditis elegans* roundworms as a model indicated that *oxi-1* loss of function mutants have reduced synaptic vesicles in their motor neurons; however, it is unknown if *oxi-1* helps to regulate neuromuscular signaling under oxidative stress and what cell type requires its expression. Here, I investigated the hypothesis that *oxi-1* acts in motor neurons to control signaling for muscle contraction under normal or oxidative stress conditions. Thrashing assay results showed that *oxi-1* mutants exhibited decreased motility by 10.3% compared to wild type worms (p=0.00869), indicating that the gene is required for neuromuscular signaling. Since *oxi-1* is known to be upregulated in the presence of increased reactive oxygen species (ROS), current experiments are testing the impact of increased ROS on *oxi-1*'s activity in the neuromuscular junction by exposing worms to oxidative stress for two days using paraquat followed by thrashing assay tests of neuromuscular activity. Furthermore, to examine the cell types in which *oxi-1* expression is sufficient for normal muscle contraction, *oxi-1* mutants will be injected in the gonads with functional *oxi-1* genes. Offspring of the injected mutants will have a functional *oxi-1* gene expressed exclusively in specific neuronal cells and will be subjected to a thrashing assay to identify whether neuronal restoration of *oxi-1* restores wild-type behavior. Potential findings of the gene's involvement in neuromuscular regulation may have applications in future studies of neurodegenerative diseases influenced by oxidative stress.

Mapping NKCC1 in the endocytic pathway during PKC activation in Mardin Darby Canine Kidney cells (Poster)

Amanda Bazaldua*, Emily Hughes*, Chase Jones, Natasa Petreska, Payton Klosa, Marie Dix, Nicholas Barron and Patrice G Bouyer. *Equally contributed authors. Valparaiso University.

Gut clearance (*i.e.,* fluid secretion) represents an important defense mechanism of the gut. Fluid secretion not only flushes luminal bacteria and toxins, it also helps expanding the mucus layer, another defense mechanism. In the colon, transepithelial chloride fluid secretion drives fluid secretion. The basolateral Na-K-2Cl cotransporter 1 (NKCC1) is the main mechanism for loading cells with chloride for its secretion by apical chloride channels (*e.g.,* cystic fibrosis transmembrane regulator). The lab has previously shown that protein kinase C (PKC) activation causes internalization of NKCC1, thus blunting chloride secretion. On the other hand, the fate of NKCC1 in the internalization pathway remains unknown. To determine whether NKCC1 is recycled or degraded, we used Mardin Darby Canine Kidney (MDCK) cells that stably express eGFP-NKCC1 and then mapped NKCC1 in the endocytic pathway with endosomal markers. For immunocytochemistry, MDCK cells were cultured on coverslips in a six well plate until confluence. Cells were exposed to either phorbol 12-myristate 13-acetate (PMA), an activator of PKC, or DMSO (vehicle). Cells were fixed with 1% paraformaldehyde, incubated with specific primary antibody against endosomal markers and mounted for immunofluorescence. Images were acquired with an Olympus compound microscope equipped for fluorescence and processed using ImageJ. In these experiments, we did not find colocalization of NKCC1 with Rab5, a marker of the early endosome. We found occasional colocalization of NKCC1 and Rab11, a marker of vesicles recycling to the plasma membrane. Finally, we did not find colocalization of NKCC1 with either LAMP1 or P20S markers of the lysosome and the proteasome respectively. Our results suggest that some NKCC1, after internalization recycles to the membrane in MDCK cells. Further investigation will be needed to determine the fate on NKCC1 in the endocytic pathway.

MicroRNA depletion results in Notch loss of function phenotypes and loss of stem cell totipotency (Poster)

Hunter Jones, Joseph Ballard, Ben Geier, **Erika Sorensen-Kamakian**, Wabash College

Notch signaling regulates stem cells and differentiation during normal animal development and when dysregulated can lead to cancer. In the model organism *C. elegans,* Notch signaling functions to maintain germline stem cells (GSCs) in a totipotent state (capable of differentiating into all cell types) by promoting the expression of target genes that function in GSC maintenance. Recently, microRNAs (miRNAs) *mir-61* and *mir-250,* termed collectively *mir61-250,* were identified as Notch target genes in GSCs. miRNAs are non-coding RNAs that act post-transcriptionally to limit the expression of other genes. To ask if *mir61-250* affects GSC maintenance, we performed assays using a CRISPR-Cas9 generated mutant, which lacks the *mir61-250* promoter. Unfortunately, no GSC defect was observed. This lack of defect is consistent with previous reports where many single-gene miRNA deletions do not produce strong mutant phenotypes unless placed in a sensitized background. Progressing forward, our lab has placed *mir61-250* mutant animals in sensitized backgrounds and assayed them for

defects in animal development, GSCs, and fertility. We find that *mir61-250* mutants exhibit developmental deformities in their egg laying apparatus, have fewer GSCs, whereas embryonic development appears normal. In addition, we find that loss of these miRNAs promotes reprograming from a germ cell fate to a somatic fate (*i.e.* neurons).

Natural killers are made, not born: Cancer immunotherapies with genetically-engineered immune cells (Hot Topic)

Sandro Matosevic, Jiao Wang, and Andrea Chambers, Purdue University

Despite extensive research into personalized therapies and a deeper understanding of its molecular mechanisms, cancer remains a major cause of mortality. Solid tumors have been particularly resistant to many traditional anti-cancer therapies. Immunotherapy—specifically, the engineering of immune cells to enhance their anti-cancer function—has emerged as a potentially transformative approach for the treatment of tumors, particularly those refractive to other treatments. Recently, scientists have begun pursuing approaches aimed at enhancing T or natural killer (NK) cells' anti-tumor activity through genetic engineering. Immunotherapies designed to trigger killing of tumor cells with immune cells engineered to bear sophisticated genetic machinery targeting specific cancer cells with the use of chimeric antigen receptors (CARs) have shown remarkable clinical results. In this hot topic, we will discuss the foundational principles of engineering immune cells by equipping them with genetic machinery aimed at enhancing their anti-tumor effector function, and how cutting-edge nanomedicine approaches can help these advances toward safer clinical therapies. With specific examples from our lab's own work, we will illustrate how engineering NK cells can enhance the targeting of solid tumors and the profound translational relevance for the treatment of complex cancers. Additionally, we will discuss next-generation immunotherapeutic cell reprogramming approaches that can evade cancer mutations and control the harsh tumor microenvironment that is created around metastatic tumors and which fuels their growth.

Notch activates expression of pericardial genes using distinct permissive and instructive mechanisms

Manoj Panta, Andrew J. Kump, John M. Dalloul, Kristopher Schwab, Shaad M. Ahmad. Indiana State University

The development of a complex organ involves the specification and differentiation of diverse cell types constituting that organ. The *Drosophila* heart is comprised of two major cell types: contractile cardial cells (CCs) that constitute an inner tube and pericardial cells (PCs) that form a sheath surrounding the CCs. We showed previously that binding sites of Suppressor of Hairless [Su(H)], an integral transcription factor in the Notch signaling pathway, were enriched in the enhancers of genes specifically expressed in the PCs. Furthermore, by using *cis*- and *trans*- assays with enhancer-reporter constructs for a PC-specific gene, *Holes in muscle*(*Him*), we demonstrated that Notch signaling activates *Him* expression in PCs in a permissive manner: in the absence of Notch signaling, Su(H) forms a repressor complex with co-repressors and binds to the *Him* enhancer, repressing its transcription; upon alleviation of this repression by Notch signaling, *Him* transcription is activated. Using relevant enhancer-

reporter constructs, we now show that Notch signaling activates the expression of *Zn finger homeodomain 1* (*Zfh1*), another PC-specific gene, in a distinctly different, instructive manner: mere alleviation of repression by preventing the binding of the Su(H) repressor complex to the *Zfh1*enhancer is not sufficient to activate transcription in PCs. Our results suggest that, in the case of *Zfh1*, upon Notch signaling, the Notch intracellular domain binds with Su(H) to change the Su(H) complex bound on the *Zfh1* enhancer from a repressor to an activator complex, and that this activator complex is necessary for bringing about *Zfh1* transcription. Collectively, these data show how the same feature, enrichment of Su(H) binding sites in the enhancers of PC-specific genes, can be utilized by two distinct mechanisms, one permissive, the other instructive, to contribute to the same overall goal: the specification and differentiation of pericardial cell types by activation of the pericardial gene program.

Potential Roles for ER-Phagy and a Golgi-Localized Ubiquitin Ligase in Translocon Quality Control (Poster)

Danielle L. Overton, Samantha M. Turk, Christopher J. Indovina, Sarah M. Engle, and Eric M. Rubenstein, Ball State University

Hrd1 is a ubiquitin ligase that functions in the degradation of proteins associated with the endoplasmic reticulum (ER) translocon. One such translocon-associated protein is the major protein component of low-density lipoproteins (“bad cholesterol”). Other proteins that function with Hrd1 in this process have yet to be identified. A recent genome-wide screen in *Saccharomyces cerevisiae* identified Atg39, Atg40, and Dsc3 as candidate proteins that cooperate with Hrd1 in promoting degradation of translocon-associated proteins. Atg39 and Atg40 are involved in the process of ER-phagy (autophagy of ER segments), while Dsc3 is a subunit of the Golgi-localized DSC ubiquitin ligase complex. We hypothesize that ER-phagy and the DSC complex contribute to the degradation of translocon-associated proteins. We performed yeast growth assays that indirectly report on protein degradation to test the roles for the Atg39, Atg40, and Dsc3 proteins in the degradation of translocon-associated proteins. Yeast strains lacking the *ATG39*, *ATG40*, and *DSC3* genes exhibited similar growth phenotypes to *HRD1*-knockout yeast, consistent with a role for these proteins in degradation of translocon-associated proteins. Because ER-phagy and Golgi-associated protein degradation are conserved across eukaryotes, these data suggest that the analog human genes may represent therapeutic targets for diseases linked to translocon-associated proteins, such as elevated cholesterol.

Rescue of Zoledronate-Induced Necrosis in Human Oral Keratinocytes (Poster)

Ethan Blake, Morgan Smith, Jesse Maguire, Preston Rippe, Nur Bashira Binti Shaharuddin, Wen Lin Chai, and Dan Jones, Indiana Wesleyan, University of Malaysia

Zoledronate (ZOL) is a nitrogen-containing bisphosphonate used in the treatment of bone resorptive diseases, including osteoporosis and cancer metastasis to bone. ZOL is linked to bisphosphonate-related osteonecrosis of the jaw (BRONJ), a disease in which oral lesions occur, particularly after oral surgery. Geranylgeraniol (GGOH) rescues some cell types negatively affected by ZOL, likely by providing an alternate route in the mevalonate pathway, a

pathway blocked by ZOL and necessary for cell viability. This study aimed to examine ZOL-induced human oral keratinocyte necrosis, as well as the rescue effect of 10 microM GGOH on ZOL-induced necrosis. In addition, we investigated possible ZOL-induced necrosis rescue effects from 10 microM melatonin (known reactive oxygen species scavenger), 2 milliM Etidronate (non-nitrogen bisphosphonate thought to block nitrogen-containing bisphosphonate entry into cells), and 100 microM citrate (endogenous calcium chelator in saliva). Two human oral keratinocyte lines were used: normal human oral keratinocytes (NHOKs) and immortalized human oral keratinocytes (OKF6/TERT2s). Necrosis measurements via lactate dehydrogenase (LDH) release assays indicated a mild but significant increase in NHOK necrosis due to 72 hour treatment with 50 microM ZOL (6.4%), along with a significant but modest rescue by 10 microM GGOH (6.7% decrease to untreated levels). OKF6/TERT2 cells demonstrated a greater increase in necrosis (13.3%, $p<0.05$) but no rescue in the presence of 10 microM GGOH. Data describing comparative rescues from melatonin, citrate, and Etidronate in OKF6/TERT2 cells suggests that each of these agents do not rescue these cells from ZOL-induced necrotic effects at the doses tested.

Rescue of Zolendronate-Induced Senescence in Human Oral Keratinocytes (Poster)

Preston Rippe, Jesse Maguire, Ethan Blake, Nur Bashira Binti Shaharuddin, Wen Lin Chai, and Dan Jones, Indiana Wesleyan University and University of Malaysia

Bisphosphonates are used to treat osteoporosis, Paget's disease, cancer that metastasizes to bone, and other bone resorption diseases. An adverse side effect occurs within roughly 12% of bisphosphonate-treated oncology patients: bisphosphonate-related osteonecrosis of the jaw (BRONJ). Zolendronate (ZOL) is a nitrogen-based bisphosphonate that functions by blocking farnesyl pyrophosphate synthase in the mevalonate pathway. This pathway plays an important part in synthesizing sterol isoprenoids (cholesterol), promoting prenylation of CAAX proteins, and maintaining osteoclast function. Geranylgeraniol (GGOH) is a mevalonate pathway intermediate that is a potential rescue agent for the ZOL-induced death effects of BRONJ. In the oral mucosa, ZOL can cause excessive senescence characterized as the decline of DNA replication, resulting in lower cell density and delayed wound healing. In this study, we compared 10 microM GGOH to other agents in terms of their relative ability to rescue from 72 hour, 50 microM ZOL-induced keratinocyte senescence. Overall, ZOL induced senescence in normal human oral keratinocytes (NHOKs, 2.8-fold, $p<0.05$) and immortalized OKF6/TERT2 keratinocytes (5.6-fold, $p<0.05$). Significant senescence recovery occurred in both keratinocyte lines in response to 10 microM GGOH and an additional booster of 10 microM GGOH 24 hours after initial treatment. In OKF6/TERT2 cells, exposure to 2 milliM Etidronate, 100 microM citrate, and 10 microM melatonin each rescued ZOL-induced senescence to a level comparable to that of GGOH.

The Effects of Combined Alcohol and Nicotine in a Two-bottle Choice Paradigm in *C57BL/6J* Mice (Poster)

Shriya Vinjimoor, Eliza Martin, & Jennifer Berry, Butler University

Alcohol and nicotine are both very commonly abused and are also widely abused in combination with one another. Smoking tobacco cigarettes has been shown to increase the number of alcoholic drinks per day and increase the chances of alcohol dependence. As both substances are known to individually elevate dopamine levels in the mesocorticolimbic reward system and also activate the hypothalamic-pituitary-adrenal (HPA) axis to release stress hormones known as glucocorticoids, including corticosterone in rodents. Using a two-bottle choice paradigm, we investigated the effects of the acquisition, maintenance, and withdrawal from chronic, combined alcohol (5-20% v/v) and nicotine (10-40 µg/ml) in male and female C57BL/6J mice. Withdrawal behavior and plasma corticosterone levels were measured approximately 18 hours after alcohol and nicotine were both removed. Mice exposed to alcohol, nicotine, or the combination of alcohol and nicotine exhibited slightly elevated anxiety-like behavior as evidenced by increased number of marbles buried during the withdrawal period compared to water controls in the marble burying task. All mice tested including those exposed to alcohol and/or nicotine had elevated levels of the rodent stress hormone corticosterone during the withdrawal period compared to basal corticosterone levels taken before nicotine or alcohol exposure. Given the large number of individuals who are co-dependent on both alcohol and nicotine, future studies should continue to examine the effects of combined alcohol and nicotine.

The role of Forkhead domain transcription factors in mediating proper positioning of cardiac cells (Poster)

Harjas Singh, Manoj Panta, Andrew J. Kump, Caithlin M. Helle, Sierra A. Crowe, Ye Chen, Xujing Wang, Neal Jeffries, Shaad M. Ahmad, Indiana State University; National Heart, Lung and Blood Institute, NIH.

The development of a complex organ requires the specification of appropriate numbers of each of its constituent cell types as well as the correct positioning of these cell types within the organ. Our previous work on *Drosophila* embryonic heart development had shown that the Forkhead (Fkh/Fox) domain transcription factors Checkpoint suppressor homologue (CHES-1-like) and Jumeau (Jumu determine the correct number of different cardiac cell types by regulating the division of cardiac progenitor cells through a Polo-dependent pathway. Here we show that *CHES-1-like* and *jumu* are also required for the correct positioning of these cardiac cell types: null mutations in either gene result in the misalignment and incorrect location of both cardial and pericardial cells within individual hemisegments. We show that these positioning defects in Fkh mutants have two causes: (1) steric effects due to contralateral hemisegments possessing different cardiac cell type numbers as a consequence of defective cell divisions, and (2) errors in a cell division-independent Fkh-mediated pathway. In order to identify and analyze downstream targets utilized by *CHES-1-like* and *jumu* to bring about correct cardiac cell positioning through the cell division-independent pathway, we compared genome-wide transcription expression profiles of purified mesodermal cells from wild-type embryos and

embryos lacking functional copies of *CHES-1-like*, *jumu*, or both Fkh genes. We detected 2,131 genes exhibiting significant differential expression in single or double Fkh mutants compared to wild-type. Our preliminary phenotypic analysis of a prioritized subset of these downstream targets suggests that the Fkh transcription factors bring about the correct positioning of cardiac cell types by restricting the expression of *G protein gamma 1* (*Gγ1*): *CHES-1-like* and *jumu* function in a mutually redundant manner to repress *Gγ1* expression levels, while ectopic overexpression of *Gγ1* in the mesoderm phenocopies cardiac cell positioning defects observed in *CHES-1-like* and *jumu* loss-of-function mutants.

A brief history of CRISPR and its potential roles in feeding the world (Hot Topic)

Doug Bernstein, Ball State University

*Clustered regularly interspaced short palindromic repeat (*CRISPR) mediated genome editing continues to take molecular biology world by storm. These technologies have dramatically enhanced the scientific community's ability to manipulate the genomes of model organisms and has facilitated significant progress in fields for which classical genetic techniques were impractical. In this hot topic we will discuss some of the foundational discoveries made by scientists in the United States and around the world that have paved the way for these advances. Furthermore, we will discuss the required components of a CRISPR genome editing system and the role these components play in genome editing. In addition, we will examine how CRISPR is being used in plant agriculture and how CRISPR is/could be leveraged to increase food production and increase farm efficiency. Finally, we will discuss how CRISPR mediated genome editing has the potential to impact natural product and drug production.

Chemistry

Deep Eutectic Solvents for Chiral Discrimination

Todd Hopkins, Butler University

Controlling the interaction between chiral molecules (chiral discrimination) is one of the most important goals in chemistry. Deep eutectic solvents (DES) are mixtures of high melting point compounds that have a lower melting point together. The properties of DES can be controlled by the choice of the components of the mixture. This presentation will describe efforts to determine the chiral discrimination of chiral DES created by combining ammonium, or phosphonium chloride salts with amino acids, including glutamic acid, proline, and arginine. A racemic mixture of chiral luminescent lanthanide complexes was dissolved in the DES to measure the impact of the solvent on this mixture using the circularly polarized luminescence (CPL). Results from CPL studies show that the preference of the handedness of dissolved lanthanide complexes is controlled by the handedness of the amino acid. The impact of phosphonium chloride vs. ammonium chloride and temperature dependence of chiral discrimination will be discussed.

Optimizing the adsorption of Brooker's merocyanine by Zeolite L (Poster)

Thomas Dabertin, Shyann Mattis, Laura Engerer, Benjamin Henning, Kelsey Weber, Jennifer Holt, Valparaiso University

Host-guest chemistry is the study of the unique relationships between a host material and the guest molecule inserted into that host. Zeolites have unique structural designs that allow for the formation of intricate channels that can be used as molecular traps to hold and organize molecules. When a host zeolite constrains a guest molecule, such as Brooker's merocyanine, new properties can be achieved that are not observed in the individual substances. Zeolite L was synthesized via a hydrothermal process, and then Brooker's merocyanine was adsorbed from solution by the synthesized Zeolite L microcrystals. In order to better understand how to maximize the amount of molecules absorbed by the zeolite, different variables were studied during the dye loading process. Some of the variables tested included: pH, light exposure, heating and stirring. Zeolite L was also added to neutral, basic, and acidic dye solutions of Brooker's merocyanine. Each sample was analyzed using UV-Vis spectroscopy to quantitatively determine the amount of dye adsorbed by the zeolite. It was found that changing light exposure, heating, and stirring could be used to increase the amount of dye molecules absorbed per gram of zeolite, and the maximum absorption was 5x10^19 dye molecules absorbed per gram of zeolite. However, the pH of the dye solution did not significantly affect the dye adsorption by Zeolite L. This optimized host-guest system can now be used to investigate the possibility of new properties, including nonlinear optical behavior.

Using 3D Printing and Arduino Technology to form a 'Reaction Chamber' (Poster)

Carrie Caldwell and James Mendez, IUPUC

3D printing opens a gateway to new learning possibilities. Recently, I have began to design a type of 'reaction chamber' that recognizes different elements and compounds that it is exposed to. The desired results would be a pedestal (containing an LED screen on the front) and a circular platform for the reaction site. By dropping in a series of individually 3D printed elements that represent different molecules onto the platform, for example two hydrogens and an oxygen on the platform it would display that together they formed water. Another example would be when a student adds two oxygen molecules and one carbon molecule and the LED screen on the front would display that together they created carbon dioxide (CO2). To get there, a base able to house the Arduino computer system is connected to sensors (either light or magnetic). Whenever a molecule is within close proximity to, say, a magnetic sensor the welcome screen on the LED monitor would change to sulfur, indicating the presence of a sulfur molecule.

We plan on utilizing this technology in college classes as well as younger students to provide a tangible way of learning how reactions form and what certain molecules can form when put under the 'right' conditions. It will create a hands on way to get students involved with the learning process, and allow introductory Chemistry courses to see different structures and molecules being formed. The hope for this technology is that students are provided with an easier method to learning basic chemistry knowledge that will help them on their path through their field of study.

Synthesis and Characterization of Some Alkoxy-Substituted Meldrum's Acid Adducts (Poster)

Austin Steppey[1], Micah Gerakinis[1], Jacob DeBoest[1], Ryan Buzalski[1], Nathan C. Tice[2] and Chad A. Snyder[1], [1]Grace College [2] University of Findlay, Findlay, OH

Metal cyclopenta[*c*]thienyl complexes are of significant interest in both catalysis and materials chemistry (i.e. polymerization). These relatives of the low-band-gap polymer polybenzo[2,3-*c*]thiophene show great promise due to their unique electronic properties as environmentally stable, conductive polymers and as energy-efficient, light-emitting diodes (LEDs). Recently, cyclopenta[*c*]thiophenes have received further attention as studies have shown that these molecules have potential as photodynamic anticancer agents particularly effective against leukemia cells. The current method of synthesizing these compounds is lengthy, costly, and has a very low yield. Our group has developed an alternative synthetic route fusing making use of traditional organic chemistry reactions (S_N2, PCC oxidation, LAH reduction, Dieckmann condensation, etc), proven to consistently work in the laboratory. Prior to this target compound's completion this research highlights the ring-opening step of the 5-7-6 oxpeine product pgenerated by the C,O-dialklyation of 3,4-bis(chloromethyl)-2,5-dimethylthiophene and Meldrum's acid. A family of novel alkoxy products were produced from these reaction conditions.

Adsorption of Pharmaceutical Contaminants at the Air-Aqueous and the Lipid Monolayer Interfaces (Poster)

Giada Dalla Pozza, Tyler Williams, and Mahamud Subir, Ball State University

Numerous pharmaceutical compounds have been detected in the aquatic environment. Selected examples include amlodipine (AMP) – a calcium blocker used for treating high blood pressure, and carbamazepine (CBZ) – an anticonvulsant used in the treatment of epilepsy. These organic molecules can interact with various natural surfaces and living matter. However, limited information is available with respect to their interaction with cell membranes and their affinity toward the neat air-water interface. To this end, we have explored the interaction of AMP and CBZ with a model dipalmitoylphosphatidylcholine (DPPC) monolayer and their binding affinity for the neat air-aqueous interface. Drug-monolayer interaction studies were carried out using the Langmuir trough technique. Pressure vs. area isotherms were collected for different aqueous concentrations of AMP and CBZ. Comparison of these isotherms with that of the neat (no pharmaceutical) DPPC reveals that there is a change in the structure of the DPPC monolayer. This suggests a significant interaction between the DPPC monolayer and the drug molecules at the micro-molar concentration range. Calculation of the elasticity modulus of the DPPC monolayer in the presence and absence of AMP and CBZ shows that these molecules have a destabilizing effect on the monomolecular film. Furthermore, our studies using surface tensiometry show that AMP has an affinity for the neat air-aqueous interface whereas CBZ does not. The affinity for the air-water interface appears to be pH-independent for both AMP and CBZ. Together, these experiments show the potential effects of pharmaceutical pollutants on aquatic organisms as well as the plausible pathways by which these emerging contaminants are transported in the aquatic environment.

Effects of Conjugation on Dual Amide Hydrogen-Bonding in CCl_4: Theory and Experiment

Shannon Lieb and Paula LeBlanc, Butler University

The doubly hydrogen-bonded dimer of pyrrolidinone (γ-lactam) is used as a benchmark for the effect of conjugation on the strength and extent of hydrogen-bonding of the compounds Oxindole and Isoxindole. The experimental portion of this project consists of collecting the FTIR spectrum of γ-lactam, Oxindole and Isoxindole in CCl4. The concentrations and temperatures were varied to determine the thermodynamic properties of ΔHd, ΔSd and Kd. In addition, the spectroscopic parameters of the difference of the monomer and dimer N–H stretching frequencies (Δν) and the ratio of the molar absorptivities of the dimer and monomer (εd/εm) are experimentally determined. The values of ΔHd, ΔSd, Δν and εd/εm are computed using the Gaussian03 program implementation of MP2 and B3LYP methods with 6-31G** and 6-31+G** basis sets. The experimental values are best described by B3LYP/6-31G**with the incorporation of the dielectric effect of CCl4 using the Polarized Continuum Method. The experimental and computational results support the results of increased and decreased hydrogen-bond strength relative to γ-lactam due to conjugation effects on the polarization of the monomers in forming dimers

Interaction of Emerging Pollutants with Colloidal Natural Organic Matter (Poster)

Keith T. Murray, Tyler Williams, and Mahamud Subir, Ball State University

Populations of urban centers are living longer and healthier. Accordingly the use of pharmaceuticals and personal care products (PPCPs) are on the rise. PPCPs have been detected in the environment; however, their impact on the aquatic environment remains unknown. Thus, the need to understand the fate and transport of PPCPs is of significant research interest. One possible pathway by which PPCPs can be transported in the environment is through their interaction with the surfaces of colloidal natural organic matter (NOM). The focus of this research has been to study the adsorption of amlodipine (AMP) and carbamazepine (CBZ), two PPCPs commonly detected in the environment, to model NOM surfaces. Magnetic particles (MPs) containing either $–NH_2$ or –COOH functional group have been used to mimic the NOM surface. Adsorption study using UV-Vis spectroscopy revealed that AMP adsorbed to both $–NH_2$ and –COOH functionalized MPs with large binding energies of –40.2 (+/–0.4) kJ/mol and –41.5 (+/–0.5) kJ/mol, respectively. CBZ also binds to these particles but with lesser degree of affinity. To determine the colloidal stability, we have further explored the impact of PPCP adsorption on the zeta-potential and size of the MPs using dynamic light scattering. We determined that the surface of both MPs are negatively charged (zeta potentials are –50.9 mV for $–NH_2$ and –54.3 mV for –COOH MPs at pH 7), which became neutral as the AMP and CBZ adsorption occurred. The size of the MPs remained constant with increasing PPCP concentration, indicating aggregation does not occur and that MPs remain relatively stable upon PPCP binding. Along with these results, adsorption studies involving humic acid, a major component of NOM, will be presented to highlight the importance of NOM-PPCP interaction for the transport and fate of these emerging contaminants in the aquatic environment.

Investigating the Interaction of Aryl Tetrazolones with Anions

Erika Nord and Sundeep Rayat*, Ball State University

Anions play an important role in biology, industry, and environment. Compounds that have the capability of binding anions may find applications in the treatment of various diseases and disorders, in removal of harmful anions from drinking water and much more. Tetrazolones possess an N-H moiety that offers the potential to interact with negatively charged species through hydrogen bonding, however, the anion-binding ability of tetrazolones remains largely unexplored. In this study, a variety of substituted aryl tetrazolones were synthesized by reacting the isocyanates with trimethylsilyl azide under reflux for 24 hours. The interaction of synthesized tetrazolones with anions such as acetate, hydrogen sulfate, bromide, nitrate and chloride was investigated in acetonitrile by UV and NMR spectroscopy. Significant UV changes were observed upon addition of the acetate anion to the solution of tetrazolone in MeCN. Our studies showed that tetrazolones underwent an acid-base reaction with the acetate anion owing to its higher basicity. A downfield shift in the NMR spectrum was observed for the acetate protons in the presence of tetrazolones, indicative of proton transfer. To gain further support for deprotonation, UV titration of tetrazolones with triethylamine was carried out which showed similar spectroscopic changes as observed in case of the acetate addition. Further, the UV and

NMR spectroscopic titrations indicated that tetrazolones interact weakly with Cl-, Br-, NO3-, and HSO4, most likely through hydrogen bonding. Job's plots were employed to determine stoichiometry of interaction of these anions with tetrazolones.

Adsorption and Self-assembly of Benzoic Acids at Au(111) Electrode/Electrolyte Interfaces (Poster)

Cody Leasor and Zhihai Li, Ball State University

The self-assembly of supramolecular structures is based on noncovalent interactions such as hydrogen-bond, van der Waals force, and metal ion-ligand coordination,[1] etc. Both the molecule-molecule interaction (intermolecular) and molecule-substrate (often metal electrode) interaction play a crucial role in determining the structural motif of supramolecular architectures formed. Self-assembly provides unique routes to create highly organized molecular nanostructures at well-defined surfaces.[2] In the present work we employed in-situ scanning tunneling microscopy (STM) in combination with electrochemical techniques to explore the adsorption and self-assembly of benzoic acids (BA) at Au(111) electrode/electrolyte interfaces. Depending on the applied electric field, benzoic acids form distinctly different, highly ordered adlayers on Au(111). At low potential protonated BA molecules are adsorbed at interfaces with a disordered manner. At higher potentials, the increase of the electrode charge triggers the interfacial deprotonation of BA molecules, forming long-range ordered patterns. The role of intermolecular hydrogen bonding, concentration of molecular solution, and electrochemical potential on the formation of surface nano-architectures will be discussed.

An improved Method to Access 1,4-diaryl tetrazolones

Benjamin Goka and Sundeep Rayat*, Ball State University

Tetrazoles and their derivatives are widely employed for agricultural, medicinal and industrial applications. Previously, we reported the synthesis of a series of 1,4-diaryl tetrazolones through a highly efficient copper catalyzed N-arylation of 1-phenyl-1H-tetrazol-5(4H)-ones with aryl boronic acids in dichloromethane. However, this approach suffered from some drawbacks such as low to moderate yields and long reaction times. In this presentation, we will discuss our attempts to overcome these limitations. Our recent work shows that 1,4-diaryl tetrazolones can be obtained in higher yields and in shorter times by decreasing the amount of solvent, and by using triaryl boroxines instead of boronic acids. The 1-phenyl-5*H*-tetrazole-5(4*H*)-one for the reaction was produced by reacting phenyl isocyanate with trimethylsilyl azide via 1,3-dipolar cycloaddition, and the triaryl boroxines were synthesized from aryl boronic acids using Dean Stark apparatus.

Chiral Discrimination by Amino Acid based Deep Eutectic Solvents (Poster)

Cameron Wright and Todd Hopkins, Butler University

Deep eutectic solvents (DES) are composed of a hydrogen bond donor (HBD) and a halide salt that when combined give a melting point lower than either component. The properties of DES can be controlled by the choice of HBD and salt, including chirality. Chiral DES's can be used as a solvent for chiral discrimination. Amino Acids such as Glutamic acid will be used as the chiral HBD combined with tetrabutylammonium chloride as the salt to create the DES. The chiral discrimination of the DES is determined by measuring the impact of the solvent on a racemic mixture of luminescent europium tris(dipicolinate) ($Eu(dpa)_3^{3-}$). The chiral discrimination results in a non-racemic mixture, and is measured using circularly polarized luminescence (CPL). The sign and magnitude of the CPL is directly related to the chiral discrimination of the DES. The relationship between DES structure and chiral discrimination will be described in this presentation.

Geometry, electronic structure and physico-chemical properties of the carcinogen NNK diazonium ion (Poster)

Maverick L. Grayer, Christos Deligkaris, University of Southern Indiana

The compound 4-methylnitrosamino-1-3-pyridyl-1-butanone, abbreviated NNK, is a carcinogen found in tobacco products. Although NNK does not directly form DNA adducts, it is metabolized by cytochrome P450 into a variety of intermediates that can methylate and pyridyloxobutylate DNA. The NNK diazonium ion is a carcinogenic intermediate that is produced when α-hydroxylation occurs at the methyl carbon of NNK, creating α-hydroxymethyl NNK. α-hydroxymethyl NNK can spontaneously produce formaldehyde and pyridyloxobutyldiazohydroxide, the latter of which can form the diazonium ion. The ion is of particular interest, as it can either directly react with nucleophiles, or it can become a cyclic oxonium ion or α, β-unsaturated ketone, both of which react with nucleophiles. This study involves the analysis of the NNK diazonium ion's ground state geometry and electronic structure, along with physico-chemical properties such as atomic partial charges, molecular dipole moment, and electrostatic molecular suface using the B3LYP hybrid generalized gradient approximation density functional and the 6-311+G(2d,p) basis set. We will present the results and discuss the implications the data has on the diazonium ion's physical binding and damage to DNA.

Infrared Spectroscopic Investigations of the Interactions of *Lactams* with Nanoparticles

Joe Kirsch, Butler University

Infrared spectroscopy was used to investigate the interactions between a series of lactams (*beta-lactam*, *gamma-lactam*, *delta-lactam*, and *epsilon-lactam*) and the nanoparticles C_{60} and C_{70} in a toluene solvent environment. Changes in the shape and position of the infrared carbonyl absorption in the lactams were used to identify interactions and complex formation between the lactams and the nanoparticles. The challenges of the study were that the interactions between the lactams and the nanoparticles are governed by an equilibrium process

and that the carbonyl absorption of the lactam interacting and not interacting with the nanoparticles may be located near the same wavenumber. Addition of lactam to the nanoparticles drives the equilibrium toward complex formation; however, it also increases the amount of non-interacting lactam potentially obscuring the carbonyl absorption of the complex. Spectra were collected at different concentrations of the lactam in a saturated toluene solution of nanoparticles in order to determine an optimal concentration level to allow complex formation and also observe the infrared absorption of the complex. Infrared spectra indicated significant interactions for the *gamma-lactam* with C_{60} and *delta-lactam* with both C_{60} and C_{70}. Marginal or no changes in the infrared spectra were observed when *beta-lactam* and *epsilon-lactam* were treated with either C_{60} or C_{70} nanoparticles.

Microplastics and Water Quality of Waterways in Northwest Indiana

Kathryn Rowberg, Brandon Stieve, and Joshua Miranda

Microplastics are small pieces of plastic, <5 mm, that can form from the breakdown of larger pieces of plastic, or synthetic textiles. Much research has focussed on the oceans and beaches, but little work has been done in Northwest Indiana. The water quality observables: dissolved oxygen, pH, conductivity, surface water temperature, and turbidity are important to determining the quality of life for aquatic organisms. This study aims to quantify the concentration of microplastics, and evaluate a water quality “snapshot” in the waterways of Northwest Indiana. It was found that most of the waterways in Northwest Indiana have relatively few microplastics, Lake Michigan on the other hand has substantially higher concentration of microplastics. The observed water quality also shows healthy waterways.

Optimized routes to *7-carboxymethyl-pterin*, a useful building block for medicinal chemists (Poster)

Kassidy Grumbles, Zachary Bennett, Jeffrey Pruet, Valparaiso University

Pterins are a class of nitrogen-heterocycle with wide application in the field of medicinal chemistry. The importance of pterin derivatives stems from discovery of numerous biologically relevant pterins, such as folate and bioterin, which play key roles in cell division, nucleic acid/amino acid formation and degredation, neurotransmitter biosynthesis, and nitric oxide production. Due to this biological relevance, several pterin-based inhibitors have been developed for targets such as ricin, leishmaniases, nitric oxide synthase, and dihydrofolate reductase. Likely the most wide known medically relevant pterin is methotrexate, used to treat cancer and rheumatoid arthritis among other uses. In this regard pterins can be viewed as a privileged scaffold, as the discovery of new pterin analogs gives rise to a vast array of potential drug candidates. *7-carboxymethyl-pterin*(7-CMP) has previously been shown as a useful scaffold for the rapid generation of structurally diverse pterin amides. We are currently exploring multiple routes towards 7-CMP to assess the most efficient method of generating this useful scaffold. We intend to use 7-CMP as a building block in our search for selective inhibitors of fungal methionine synthase.

Pond System Chemical and Biological Monitoring within the African Journey Biome (Poster)

William Lorenz, **Brileigh Malott**, Christopher Moseti, Nathan C. Tice, Joe Smith, and Chad A. Snyder

The Ft. Wayne Children's Zoo (FWCZ) contains over 1,000 animals living in four self-contained biomes occupying a footprint of over 40 acres. Of these biomes, the *African Journey*, contains a water system of six artificial, connected ponds that is home to 11 species of bird and mammal inhabitants. The Snyder research group and the FWCZ have partnered to ensure the zoo's pond system health by performing regularly chemical and biological testing of its ponds. Analysis have included pH, surface water temperature, dissolved oxygen, and algae species distribution and concentration (measured in cells/mL). Additionally, the following ions associated with algae growth were measured. Those included calcium, chloride, phosphate, nitrate, and nitrite. The research reported herein displays the pond water analyses collected over two seasons beginning in 2016 until now.

Research in General Chemistry Laboratories: Pharmaceutical and Dye Degradation Using an Oxy-Catalyst (Poster)

Ruth E. Nalliah, Huntington University

The design of some types of research experiments is compatible with investigations which can be performed in undergraduate teaching laboratories with beginning students. We report preliminary results obtained from students in general chemistry laboratories investigating the degradation of mixtures of the pharmaceuticals acetaminophen, caffeine, and quinine, and mixtures of the FD&C food dyes Blue 1, Red 40, and Yellow 5, using a proprietary oxy-catalyst from Hydrogen Link, Inc. of Canada, and comparing the effects of adding hydrogen peroxide or sodium percarbonate. Degradation was measured by the disappearance of the absorbance peaks of the compounds over time periods of 24 and 48 hours, in order to allow for the disappearance of interfering absorbance from undegraded hydrogen peroxide. All substances were found to degrade with the catalyst and added hydrogen peroxide in both acidic and neutral solutions, and the catalyst itself was found to have some ability to degrade the caffeine and quinine in purified water, perhaps due to low levels of oxidants. The food dyes were not found to degrade significantly in 24 hours using hydrogen peroxide without catalyst under these concentrations, while Blue 1 and Red 40 were found to degrade with added sodium percarbonate alone. In the presence of the catalyst, caffeine and quinine degrade more slowly with the sodium percarbonate than with the hydrogen peroxide, perhaps because sodium percarbonate makes the solution basic, or because of the limited release of hydrogen peroxide from the solid due to poor solubility. We determine that it may be useful to study the effects of sodium percarbonate with added acid as well as catalyst. Students report that the experiment raised their awareness about the emerging problem of pharmaceutical pollution; and students tend to be highly motivated to obtain results that contribute toward useful solutions.

Synthesis and Characterization of Dichloride and Meldrum's Acid Adduct (Poster)

John K. Marhefka, Danila Kourkoulin, Santiago Maya, Chad Snyder, Nathan Tice

Heterocycles comprise an important class of organic compounds and offer a wide range of applications that include electronic materials, hydrodesulfurization (HDS) modeling, medicine, and catalysis. Our research focuses on the synthesis of thiophene derivatives intended to eventually serve as semiconductor substrates. Our research presents a proposed synthetic route toward thiapentalenes and highlights the synthesis and characterization of the first third of this novel route. Characterization will include IR, ^{1}H and ^{13}C NMR spectroscopy as well as X-ray crystallography for 3,4-bis(chloromethyl)-2,5-dimethylthiophene and its oxepine product.

Ecology

Effect of site and seasonality on vocal behavior of mixed-species flocks (Poster)

Cara Christensen, Anupriya Karippadath, Jeffrey Lucas, Purdue University

In this study, we examined variations in behavior and vocalizations of mixed-species flocks consisting of Carolina chickadees, Tufted titmice, White-breasted nuthatches and Downy woodpeckers across three forest sites in Indiana. Calls of a known predator of these species (Eastern screech owl) were played near naturally occurring mixed flocks, and the response of the flocks was recorded. Preliminary data suggested that all three populations respond to a predation threat and there is no difference in latency of flock approach between sites. Additionally, we found differences across sites in latency and approach order at the species level. Furthermore, we analyzed call and song rate for Carolina chickadees and Tufted titmice before, during, and after the predator playback. Significant differences were found between both season and site for call rates for both species.

The effect of cutting and burning on spider abundance, composition, and diversity in North Carolina.

E. R. Stern and M. A. Milne, University of Indianapolis: J. Campbell, University of Florida; C. Vigueira, and P. Vigueira, High Point University; K. Greenberg, US Forest Service Southern Research Station.

The biodiversity of invertebrates in forests may be impacted by both artificial and natural habitat disturbances such as fire, tree falls, timber harvesting, and prescribed burning. To simulate these types of disturbances, we conducted mechanized felling and/or prescribed burning to selected sites in the Green River Game Lands, NC and then allowed these habitats to naturally reform over a fifteen-year period (2001-2016). The change in spider diversity and abundance were measured in response to four different treatment scenarios within these forested plots: an unmodified control, repeated prescribed burns, mechanical thinnings, and a combination of mechanical thinnings followed by repeated prescribed burns. We collected and identified spiders from these plots between 2014 and 2016. We found that spider abundance was significantly greater in the mechanically felled and burned plots when compared to control plots. There were also significantly different taxonomic compositions (both at the genus and species level) among treatment types. However, sites did not differ in guild composition or taxonomic composition at the family level. Moreover, there was no significant difference in Shannon diversity at the family level among treatments. During the spider identification process, we also discovered multiple new distribution records for NC and some potentially undescribed species. These data demonstrate the significant impact that environmental manipulation – through forest management and prescribed burning – can have on invertebrate abundance and composition.

Examining differences in anuran communities between man-made and natural wetlands in central Indiana (Poster)

Marin Bray, Spencer Siddons, and Catherine Searle, Purdue University

Human-created wetlands are becoming increasingly common across much of North America and have the potential to support native plant and wildlife communities. However, these wetlands are often established for rainwater retention or river impoundments which may create unsuitable habitat for some species. Patterns in habitat quality and ability to support wildlife between man-made and natural wetlands remain unclear. Therefore, we aimed to examine differences in the community of wetland-dependent anurans between man-made and natural wetlands, and identify what habitat characteristics are affecting anuran communities. We hypothesized that because man-made wetlands are often established for water retention in urbanized areas, these habitats are less suitable for anuran communities than natural wetlands. We predicted that natural sites would have higher anuran diversity, density, and species richness than man-made sites. Our methods consisted of multiple visual encounter surveys of anuran communities at nine localities in Tippecanoe County (IN) from May through August 2017. Sites were chosen based on known presence of at least one anuran species, and the known origin of the wetland (man-made or natural). Habitat characteristics, water parameters, and surrounding land use practices were also analyzed to examine their influences on anuran communities. Our results revealed non-significant patterns that anuran diversity (measured as Shannon-Wiener Diversity Index) and anuran density were higher at natural sites than man-made sites, while species richness showed no pattern. Man-made wetlands had significantly higher average salinity, conductivity, and total dissolved solids than natural wetlands. Our findings suggest that man-made wetlands are more prone to poor water quality than natural wetlands, which may be the underlying cause of lower anuran diversity and density patterns we observed. This study can help guide strategic management efforts in anuran conservation to improve habitat quality of existing man-made wetlands in the ever-increasing urbanized landscape.

Influence of two-stage ditch segments on fish assemblages within channelized streams (Poster)

Jennifer Troy, **Gabe Curtis, Abhijeet Bisht**, Robert B. Gillespie, Indiana University- Purdue University Fort Wayne

Although recent research has shown that two-stage ditches are successful in ameliorating loads of suspended solids and nutrients to receiving streams, few studies have documented the impacts of the two-stage morphology on the communities of aquatic organisms. We predicted that fish populations that inhabit two-stage segments and/or downstream reaches of two-stage segments have higher diversity, richness, and abundance than those in channelized segments of headwater streams. Our study area comprised segments of Creel Ditch and West Branch, located in the Fish Creek catchment of the St. Joseph River Watershed in northeast Indiana. We collected data on fish assemblages, water chemistry, physicochemical parameters, and instream habitat quality from 15 segments during 2015-2017. Among these sites were 50-meter stream segments comprising Upstream Channelized (2), Upstream Unchannelized (4), Two-

Stage (5), and Downstream Channelized (4) habitat. Diversity of fish assemblages was greater in West Branch than in Creel Ditch. However, there was no apparent correlation between abundance, richness, or diversity and ditch characteristics. A greater richness and diversity of fish communities in West Branch suggests a relationship between diversity and quality of habitat, but perhaps at a greater landscape scale than was captured by these segment assessments. Species diversity was slightly greater at two-stage old segments than channelized segments, but were similar to that of downstream channelized segments in Creel Ditch and West Branch. The short distance between segments and mobility of fishes makes it difficult to assess the relationship between fish community metrics and stream characteristics.

Indy Wildlife Watch: Habitat Indicators of Urban Wildlife in Indianapolis (Poster)

Alexi Zaniker, Butler University

Urbanization creates unique opportunities and challenges to wildlife living within urban and periurban areas. For example, fragmented and degraded habitats can be potentially threatening to some animal species while creating more anthropogenic or edge food sources for other species. In order to mitigate the negative impacts of urbanization on resident wildlife, it is necessary to understand the types of habitats urban wildlife is utilizing and how these locations vary in species diversity. Based upon previous research, we hypothesize that vegetation structure will indicate preferences of medium-to-large urban wildlife toward certain habitat types. The Indy Wildlife Watch project at Butler University has been sampling urban wildlife along two transects spanning downtown Indianapolis northward into periurban areas for nearly two years. Each transect contains a total of 24 motion-sensitive cameras deployed at a variety of habitat types (parks, remnant forests, cemeteries, etc.) to monitor the presence of wildlife. For this study, we sampled vegetation at 17 camera traps that were selected to represent a variety of habitat types found in periurban environments. To assess vegetation structure at each site, a nested sampling design was used to assess canopy cover, density, and variability of each stratum (overstory, understory, shrub, herbaceous, etc.). Vegetation structural parameters will be compared among sites to determine significant differences using ANOVA and principal component analyses. Images of wildlife collected via motion-triggered cameras during July and October 2017 (roughly 27,000 photos) will be tagged to species and will be used to determine how wildlife species presence/absence and diversity vary with vegetation structure. This knowledge will be beneficial to urban green space planning and development by determining how land should be managed to maximize wildlife diversity.

Influence of two-stage ditch segments on macroinvertebrate assemblages within channelized streams (Poster)

Deanne E. Jensen, Hannah C. Hunting, and Robert B. Gillespie, Indiana University-Purdue University Fort Wayne

Two-stage ditch design has been shown to reduce nutrient and suspended solid loads in receiving streams thus improving water quality and ecological function. However, there are few analyses of the effects this channel morphology has on the resident aquatic biota. We predicted that macroinvertebrate communities within the two-stage segments and/or downstream of two-stage segments would have greater diversity, richness, and abundance compared to those in channelized segments of headwater streams. Our study area comprised segments of Creel Ditch and West Branch, located in the Fish Creek catchment of the St. Joseph River Watershed in northeast Indiana. Data on macroinvertebrate assemblages, water chemistry, physiochemical parameters, and instream habitat quality were collected from 15 segments during 2015-2017. 50-meter stream segments comprising Upstream Channelized (2), Upstream Unchannelized (4), Two-Stage (5), and Downstream Channelized (4) habitat were included in the sites. Segments within West Branch had greater quality of macroinvertebrate communities than those of Creel Ditch segments. QHEI scores suggest higher quality instream and riparian habitat in West Branch likely resulted in greater diversity of macroinvertebrate communities. The highest macroinvertebrate scores were recorded within two-stage old segments of both Creel Ditch and West Branch. Furthermore, upstream and downstream reaches of both Creel Ditch and West Branch, where riparian habitat is predominately forested, showed less macroinvertebrate diversity than that of two-stage old segments. This suggests that two-stage morphology resulted in higher quality instream habitat for macroinvertebrates than that of channelized ditches, even in segments with higher quality riparian habitat.

SNP identification in *Fraxinus* linking genetic characteristics to tolerance of emerald ash borer

Cecelia E. Smith, Vamsi J. Nalam, and Jordan M. Marshall, Indiana University- Purdue University Fort Wayne

Ash populations in North America have been decimated by the introduced pest emerald ash borer (*Agrilus planipennis*), having both negative impacts on forest ecology and economic consequences. While the majority of trees succumb to attack by emerald ash borer, some trees have been found to be tolerant to infestation. These surviving, tolerant trees remain as part of the forest canopy. In this study, restriction site-associated DNA (RAD) sequencing was used in order to sequence 47 individual ash trees, 28 classified as tolerant and 19 classified as susceptible based on crown dieback, overall vigor, and signs of emerald ash borer attack. A reference genome was constructed and single nucleotide polymorphisms (SNPs) were called using SAMtools. After several filtering criteria were implemented, over 17,000 SNPs were generated. SNP diversity was visualized using principle component analysis (PCA) and some clustering occurred related to geography; however, five tolerant trees clustered separately from all other samples despite geographic diversity. Identification of the outlier SNP loci within this group could lead to the discovery of variation in functional genes that contribute to the ability of

these trees to tolerate infestation. Our goal is to further link these genetic characteristics to identifiable phenotypic differences between tolerant and susceptible trees in order to better identify tolerant ash at sites prior to emerald ash borer infestation.

Suspended solids, turbidity, instream habitat, and fishes of headwater streams in NE Indiana (Poster)

Jennifer L. Troy, Robert B. Gillespie, Indiana University-Purdue University Fort Wayne

Extensive agriculture in the Midwest United States has led to headwater streams being used as drainage ditches. These streams are subject to contaminants from fields, increased sedimentation, and degradation of natural habitat. Human induced alterations to the aquatic ecosystem have been shown to negatively impact the integrity of the aquatic stream community. Previous research has shown that instream habitat alteration better explained variation in fish-community metrics than water chemistry. However, these studies did not include suspended solids (TSS), which are considered a major freshwater contaminant. Our research attempts to better understand the impacts of total suspended solids to aquatic communities in an effort to provide important information to guide conservation efforts. Our study area included three 125-meter sites among three channelized headwater streams of the Cedar Creek watershed; seven, 100-meter sites among two headwater streams of the Fish Creek watershed; and one 125-meter site in a headwater stream of the St. Joseph River in Osseo, MI. Weekly water samples were collected and analyzed for total suspended solids, turbidity, and agrochemicals. Instream habitat, riparian habitat, and fish surveys were characterized. Preliminary data suggest that total suspended solids, turbidity and fish assemblage metrics are not negatively correlated (R^2=0.00-0.11). Discharge, however, was negatively correlated with species richness, diversity, and abundance (R^2=0.23-0.32), suggesting that hydrology may be a stronger influence on fish assemblages than TSS. Additional data will be collected during summer of 2018 to increase sample size in systems whose water chemistry and instream habitat parameters vary with agricultural activity and weather.

The Taylor University Arboretum: forest community sampling and structure (Poster)

Levi Sherman and Robert T. Reber, Department of Environmental Science, Public Health, and Sustainable Development, Taylor University

In 2006, Taylor University Arboretum consists of a diverse range of plant communities including upland forests, floodplain forests, early successional forests, old fields, and agricultural row crops. The principal focus of this study is the determination of forest community types. In 2010, one hundred sixty, 200m^2 circular plots were sampled in the forested sections to assess the forest community structure. All trees (dbh> 2cm) were identified and basal area of each was recorded. Resampling occurred in the summer of 2017. Coverage, density, frequency, relative cover, relative density, relative frequency and relative importance values were calculated for each species sampled. These metrics were used to compare the data collected in 2010 to the 2017 data. Similar trends exist between study years. Three distinct community types were recognized: an upland-dry forest community, a mixed hardwood community, and a floodplain/early successional community. In the upland-dry forest community, *Carya*

ovata and *Ostrya virginiana* dominated the relative densities; *Carya ovata* and *Quercus alba* attained the greatest relative coverages. In the mixed hardwood community, *Aesculus glabra* and *Ostrya virginiana* dominated the relative densities; *Carya cordiformis* and *Quercus alba* had the greatest relative coverages. In the early successional /floodplain community, *Aesculus glabra* and *Asimina triloba* dominated the relative densities; *Juglans nigra* and *Quercus rubra* attained the greatest relative coverages. Across the entire arboretum, *Juglans nigra*, *Carya ovata*, and *Quercus alba* exhibited even-aged distributions. *Acer saccharum*, *Ostrya virginiana*, and *Aesculus glabra* displayed uneven-aged distributions. The stand shows the results of past disturbances with an even-aged overstory and an uneven-aged understory. Current loss of *Fraxinus americana* due to the Emerald ash borer has occurred. Only small diameters of *Fraxinus americana* remain, and gaps in the canopy have developed. Overall, the forest community is currently undergoing a successional change to more shade tolerant species.

What is the relative influence of bed sediment chemistry on aquatic macroinvertebrate metrics? (Poster)

Tyler C. Shuman, Robert B. Gillespie, Indiana University-Purdue University Fort Wayne, and Peter C. Smiley Jr., USDA-ARS Soil Drainage Research Unit, Columbus, OH

Channelized headwater streams in the Midwest are prone to erosion and often receive runoff with herbicides and excess nutrients. These degraded headwater streams possess communities of macroinvertebrates that are exposed to these stressors. We hypothesized that macroinvertebrate assemblages would be less diverse at sites with low variability in grain size distribution of bed sediments than those at sites with high particle-size diversity. We analyzed grain size distribution of channelized watershed headwater streams within Cedar Creek, Indiana and a reference site in Hillsdale County, Michigan. Composite sediment samples were collected at each site and particle-size distribution and percent organic matter were analyzed. Physiochemical data consisting of temperature, pH, and conductivity and dissolved oxygen were recorded. Macroinvertebrate assemblages were sampled using Hester-Dendy plates and leaf packs. Preliminary results show that channelized headwater streams within Cedar Creek, have benthic sediments consisting mostly of silt and sand, whereas, those at the reference site, East Branch St. Joe (EBSJ), included silt, sand and cobble. Preliminary results from the macroinvertebrate sampling show that species richness is lower in the channelized headwater streams (mean= 7 families) than in the reference stream (mean= 15 families). Because sediment chemistry (e.g. agriculture chemical contamination) may influence macroinvertebrate assemblages, we will analyze sediment concentrations of selected pesticides and nutrients to determine their potential impact and relationship with particle-size distribution on macroinvertebrates of channelized headwater streams.

Resource competition and its effect on disease dynamics in freshwater zooplankton populations (Poster)

Mackenzie Chapman and Catherine Searle, Purdue University

Interspecific competition can reduce population densities of competing species. However, the effects of competition on infectious disease, particularly when only one species can become infected with a given parasite, is less understood. We performed two experiments to study the effects of resource competition between two species of zooplankton and its effects on disease dynamics. First, to quantify potential resource competition, we estimated feeding rates in both species to quantify how much an individual of each species may consume in a given time. *Simocephalus sp.* and *Daphnia dentifera* feeding rates were studied in this experiment with 15 replicates of each species. Each replicate was an individual in a falcon tube with a known amount of food. After two days of grazing, we measured absorbance values using a fluorometer and body size. This experiment was conducted once using young individuals (juveniles) and once using older individuals (adults). We did not find any significant difference in feeding rate between the two species, but older (and larger) animals consumed more food. Second, to quantify the effect of competition on disease susceptibility, we set up populations of *D.* dentifera with two treatments: with and without *Simocephalus sp.* present. Within each treatment, half of the beakers were exposed to *Metschnikowia bicuspidata*, a fungal parasite. Population densities and rates of infection were quantified every two weeks. Population densities of *D.dentifera* were lower in the presence of *Simocephalus sp.* indicating that competition between these two species does occur. Additionally, the presence of *Simocephalus sp.* reduced densities of infected *D.dentifera*, but not infection prevalence. There was a trend of higher infection prevalence when *Simocephalus* are absent in a population. These studies demonstrate that intraspecific resource competition can influence both population densities and patterns of disease in this system.

An ecological analysis of benthic foraminifera in the Venice Lagoon, Italy (Poster)

Caz Bejger and Jennifer Latimer, Indiana State University

Benthic foraminifera are eukaryotic, unicellular protists characterized by their shells (tests). They live in abundance at both the sediment-water interface and within seafloor sediments. Benthic foraminifera are sensitive indicators of environmental change, and the presence of contaminants can result in aberrant morphologies and altered geochemistry of foraminiferal tests. The Venice Lagoon has been influenced by anthropogenic contaminants for thousands of years. As a result, a gradient in contaminant concentrations, especially metals, has developed within the Lagoon. The purpose of this project was to evaluate the impact of these contaminants on the ecological distribution and microhabitat preferences of benthic foraminifera in the Venice Lagoon using core samples collected between June 2005 and November 2006. Overall, the Lagoon is an area of low benthic foraminiferal diversity, with only eight species represented across all sites. Relatively high densities of benthic foraminifera were observed at the most contaminated sites, suggesting that abundant food sources outweigh the impact of exposure to contaminants. Significant differences in vertical distribution and microhabitat preferences of benthic foraminifera were observed between sites. This study also

included the evaluation of core samples collected from two artificial subtidal banks constructed in the Lagoon in November 2005 using dredged sediment. The purpose of the banks was to evaluate potential impacts on the ecology of benthic foraminifera resulting from sediment disturbance and potential remobilization of contaminants. By May 2007, the percentages of benthic foraminifera with aberrant test morphologies were similar to those at the nearest sample site, but they remained lower than the percentages of aberrant morphologies at the site closest to the sediment source. One species, *Haynesina germanica*, exhibited the highest percentages of aberrant morphologies deeper within the sediment, suggesting that this species may be exposed to greater concentrations of contaminants deeper within the sediment than at the surface.

Autumn Habitat Selection of Eastern Red-backed Salamanders (*Plethodon cinereus*) in Delaware Co, IN (Poster)

Zachary Laughlin and Kamal Islam, Ball State University

The Eastern Red-backed Salamander (*Plethodon cinereus*), a temperate woodland salamander, is a common species found in the Midwestern United States. This species is commonly used as an indicator species of forest health because it is sensitive to changes in its environment. Few data have been collected on the preferred habitat characteristics of this species. Based on previous research, the Eastern Red-backed Salamander is typically present in heavily wooded and moist environments. We are interested in determining presence or absence of this species using habitat characteristics that may impact them. We present preliminary results using an adaptive random cluster sampling technique from the autumn field season of 2017 in Delaware County, Indiana. By identifying the habitat characteristics of the Eastern Red-backed Salamander, natural resource personnel will be better informed, able to more effectively assess forest health, and plan even more effective forest management strategies for this species.

Beetle community resilience to harvest

Jeffrey D. Holland, Purdue University

Timber harvest can alter the biodiversity and sustainability of forest ecosystems. Resilience is one measure of ecosystem response to disturbance that can act as an indicator of the sustainability of the management regime. In this study, the resilience of the longicorn beetle community (Coleoptera: Cerambycidae) to timber harvest was assessed to examine differences in the trajectory back to pre-disturbance communities under different management regimes. I hypothesized that different silvicultural regimes result in different levels of resiliency in the beetle community. I used data from the first decade of the Hardwood Ecosystem Experiment in south-central Indiana, USA., to test the following predictions: (1) communities would be different following harvests than the communities pre-harvest and different from unharvested controls, (2) both treatments, even-aged and uneven-aged harvested landscapes, would show resilience with a community trajectory back to pre-harvest communities, (3) the more concentrated disturbance of even-aged management would lead to a lower community resilience than the more diffuse disturbance of uneven-aged management, and (4) the main change in the communities would be a an increase in the relative abundance of beetle species that develop

within freshly killed wood. We found that beetle communities did change, but that the magnitude of the change was the same for both treatments but along different species-trajectories. Communities subject to either treatment did show resilience in a return to pre-harvest communities and unexpectedly at the same rate. The changes in species most responsible for postharvest shifts in communities were as predicted with species with larvae that develop in living wood increasing and those that develop in decaying wood decreasing.

Do Cerulean Warblers (*Setophaga cerulea*) exhibit geographic song variation?

Garrett J. MacDonald and Kamal Islam, Ball State University.

The Cerulean Warbler (*Setophaga cerulea*) is one of the fastest declining North American wood-warblers (family Parulidae); however, little is known about how consistent or variable its songs are across its breeding range. Geographic song variation may be informative about patterns of population connectivity and dispersal, and since song functions in courtship and territoriality, it has the potential to promote speciation by acting as a pre-mating isolating mechanism. We examined geographic song variation in the Cerulean Warbler by measuring 9 acoustic and temporal variables from the songs of 246 individual male warblers from 9 Bird Conservation Regions (BCR) spanning the breeding range. All measurements were made using Raven Pro 1.5. Audio recordings were obtained from public collections and colleagues and were supplemented by recordings we personally made with a parabola. Variables corresponded to measures of frequency, duration, amplitude, and song composition. We conducted parallel analyses on the entire song and the 3 distinct sections of each song using principal component analysis (PCA), MANOVAs and individual ANOVAs on each of the principal components, and linear discriminant analysis. We performed all analyses in Program R. We found a few differences between populations in some of the acoustic variables, but these may have been driven by low sample sizes in some of the BCR regions. Song structure was similar across the breeding range in a number of characteristics, including composition, duration, frequency, and bandwidth. We failed to find evidence for geographically structured singing (e.g., dialects). This is the first study to assess patterns of geographic song variation across the species' entire breeding range. Additionally, this research may support the likelihood of gene flow between breeding populations, which may have important implications for the conservation of the declining Cerulean Warbler.

Examining the fungal pathogen, *Batrachochytrium dendrobatidis*, in anurans during winter in Indiana

Spencer Siddons, Marin Bray, and Catherine Searle, Purdue University

The aquatic pathogenic fungus, *Batrachochytrium dendrobatidis* (Bd), infects the skin of amphibians and is a leading factor in recent global amphibian declines. Bd infections in North America often fluctuate across seasons, but no studies have examined infection in winter in northern temperate climates. Investigating Bd during winter can provide unique insight into which species are sustaining infections and how Bd infection impacts overwintering amphibian hosts. Some species, such as the American bullfrog (*Lithobates catesbeianus*), overwinter in ice-covered ponds, commonly have high Bd infection rates, and may retain Bd infection through

winter. We hypothesized that the colder temperatures during winter would reduce Bd reproduction and survival, thereby decreasing the pathogen's ability to infect amphibian hosts. Therefore, we predicted that Bd infection prevalence and infection loads would be lower in winter than in early summer in amphibian hosts. From August 2016 – November 2017, we collected skin swabs of anurans across eight localities in Tippecanoe County (IN) to examine Bd infection prevalence and load. We sampled eight of the 17 anuran species native to Indiana. The majority of samples (78%) were collected from species that overwinter in water (i.e. American bullfrogs, *L. catesbeianus*, and green frogs, *L. clamitans*), and 22% of swabs were collected prior to or during their overwintering period. Our findings can facilitate management of anuran species that harbor infections through winter as these species may be more susceptible to disease symptoms, which can decrease their overwintering survival. By examining infection during winter, our findings will help describe a more complete understanding of seasonal Bd dynamics in northern temperate climates. This study provides vital information on the status of a potentially deadly pathogen in amphibian communities of Indiana, which can guide conservation efforts to prevent Bd outbreaks and preserve amphibian communities across this region.

Factors Influencing Mammalian Activity in an Urban Campus Environment (Poster)

Jacob Reeves, Julia Angstmann, Butler University

With growing urban populations across the globe, college campuses have the potential to provide a wide array of resources and habitats to animals living in increasingly fragmented urban environments. Omitting city parks and nature preserves, college campuses provide one of the only large swaths of land that is owned and managed by the same entity over long periods of time. This stable ownership coupled with an investigation of how various campus spaces are utilized by mammals can help land managers maximize these properties for both humans and animals. The present study surveyed the presence/absence of mammals using motion-triggered cameras at eight different locations on Butler University's campus. Using presence/absence data collected during 2017, correlations of animal activity with spatial location, habitat type, shrub layer density, and various geospatial parameters were explored to determine which habitat variables may drive species activity and diversity. Each camera location represented a spatially and structurally variable campus habitat type (park-like spaces, highland woods, riparian areas, etc.). Shrub density data was determined using the line-intercept method and was selected as a key vegetation parameter because of the high level of invasion by honeysuckle species. Geospatial parameters were quantified using ArcMap and include distance to central campus, distance to water, site isolation, and fractal dimension. Mammalian diversity was the lowest near the campus interior and highest in areas further from the campus center. This will establish spatial and vegetation parameters that may be utilized to inform habitat management on urban college campuses.

How fungal endosymbiont within *Ipomoea tricolor* affects *Meloidogyne incognita* gall formation

Lekeah Durden, Indiana University, Dong Wang, Inner Mongolia Agricultural University, China
Daniel Panaccione, West Virginia University, and Keith Clay, Indiana University

Some species of morning glory (Convolvulaceae) form symbioses with seed-transmitted fungal endosymbionts in the genus *Periglandula*. These fungi produce ergot alkaloids within the plant, which may contribute to plant-fungal defensive mutualisms. Allocation of seed-borne ergot alkaloids to various tissues of several *Ipomoea* species has been demonstrated, including differential allocation of ergot alkaloids to the roots of *I. tricolor*. The Southern root-knot nematode (*Meloidogyne incognita*), colonizes young root tissues of broad range of host species, causing systemic damage to the plant and significant economic losses. The aim of this study was to determine if infection of *I. tricolor* by the fungal endosymbiont affects root-knot nematode gall formation and host plant fitness. In particular, we hypothesized that *I. tricolor* plants infected by the *Periglandula* endosymbiont (E+) would have fewer nematode-induced galls on the root system compared to non-symbiotic plants (E-). E+ or E- status of plant lines was confirmed by grinding individual seeds and testing methanol extracts for ergot alkaloids. To test the effects of *Periglandula* on nematode colonization, E+ and E- *I. tricolor* seeds were germinated in petri dishes and grown for one week. Seedlings were then potted in soil infested with high densities of *M. incognita* nematodes (N+) or no nematodes (N-). All plants were then grown in the greenhouse for four weeks before harvesting when nematode colonization was visualized microscopically and total gall number and plant biomass were quantified. We found that the presence of the endosymbiont-produced ergot alkaloids in roots significantly decreased nematode gall formation by 50%, independent of root biomass, as well as significant differences in total plant biomass among treatments. These results demonstrate the *Periglandula* endosymbiont's potential defensive role against belowground biotic stressors, such as plant parasitic nematodes. These results also inform potential agricultural applications for pest management by decreasing use of synthetic nematicides.

Prolonged Effects of Severe Drought on Oak Species in Eastern North America (Poster)

Alayna Blum and David LeBlanc, Ball State University

This study evaluated the duration (years) of Oak radial growth decline after severe droughts by comparing the post drought ring width to the average ring width in the five-year period before the drought in 153 sites across eastern North America. The duration of post-drought growth decline was categorized into immediate recovery (0-3 years), protracted decline (5-10 years), or improbable recovery (over 10 years). The percentage of trees at each site in each decline class was compared among the four geographic quadrants of the region to evaluate differences across eastern North America. The percentage of trees exhibiting decline for more than 10 years was significantly lower in the north-east quadrant than the south-east or south-west. These results suggest that Oaks in the southern United States are more likely to be weakened or injured by a drought and have increased susceptibility to other stresses, such as disease or insect attack.

Roads Impact Massasauga (*Sistrurus catenatus*) Movement Behavior

Monica Matthews, Jillian Josimovich and Bruce A. Kingsbury, Indiana University-Purdue University

Research has shown not only that snakes in addition to many other taxa can be negatively impacted by roads, but that snakes also perceive roads as dangerous, implying that roads affect snake behavior. However, how roads affect Massasauga (*Sistrurus catenatus*) movement behavior in particular has been little-studied and with the Massasauga's recent federal listing of threatened, it is pertinent that we gain a better understanding of Massasauga road ecology to better inform conservation and management practices. Through radio-telemetry monitoring, we have been able to observe the movements of 36 Massasaugas in Northern Michigan to allow us to determine more definitively how roads impact Massasauga active season movements. With a sample size of 18 snakes that met the threshold of radio-tracking events per season, we used the behavioral modeling program, Netlogo, to establish expected Massasauga road crossings and RStudio analysis software to compare expected versus observed road crossings. We then evaluated how qualities of the roads, including road substrate, road type, road usage and age of the road, impact frequency of road crossings. The significant difference specifically between observed and expected road crossings of females suggests that at least female Massasaugas are apprehensive to cross roads. Nonetheless, a handful of males within this population are willing and capable of crossing regularly traversed roads successfully numerous times through their lifetime. Of the four qualities of the roads, the analyses indicated that age of the road had the greatest impact on road crossing frequency of any given stretch of road within our site. Canopy cover also appeared to play a role in road-crossing behavior, though this relationship requires further examination. The relationship between age of roads and frequency of road crossings suggests that habitual road-crossing snakes either will alter that behavior, or be removed from the population through road mortality.

Roosting habitat use by Cerulean Warblers (*Setophaga cerulea*)

Clayton D. Delancey and Kamal Islam, Ball State University

The Cerulean Warbler (*Setophaga cerulea*), a Neotropical migratory songbird, is listed as state-endangered in Indiana, and a species of concern across its range. This species is declining faster than any other species of wood-warbler in North America. Since 2007, we have been monitoring Cerulean Warbler breeding populations in Yellowwood and Morgan-Monroe state forests as part of a 100-year project called the Hardwood Ecosystem Experiment. This long-term study aims to determine the effects of different forest management techniques on plant and animal communities. One of the least studied aspects of avian biology in North America is roosting ecology. Only one other study has examined habitat use of a roosting Passerine, the Wood Thrush (*Hylocichla mustelina*), in North America. We were interested in examining habitats used by adult male Cerulean Warblers as roosting cover, while females were nesting. In 2017, data were collected over multiple nights for ten individuals. Data were analyzed in program R using generalized linear models with random effects. Analyses showed that the adult males selected areas with higher canopy cover, fewer shrubs, less white oak, steeper slopes, more grape, and less basal area. Territory mapping of individuals showed that many individuals

chose to roost outside of their respective territories more often than inside their territories. Information gathered from this study can potentially be used to suggest forest management prescriptions that provide Cerulean Warblers with all of their required habitat needs during the breeding season.

The effect of wildflower strips on spider abundance and diversity in Florida

BaoThu Dinh and Marc Milne, University of Indianapolis; Joshua Campbell and Jamie Ellis, University of Florida

Wildflower strips have recently been used near agricultural fields to increase pollinator biodiversity and to improve habitat health. Spiders are often good indicators of pollinator biodiversity to their predatory relationship with insects. To investigate the effectiveness of these wildflower strips at increasing biodiversity, we examined the abundance and diversity of spiders in agricultural habitats with and without wildflower strips in Florida. In 2015 and 2016 and at eight sites throughout Northern and Central Florida, we set up wildflower (treatment) plots and fallow (control) plots. We collected spiders with bowl traps, pitfalls traps, and random sweep net samples during the growing season in Florida (April to November). The data - from two years study - indicates a significant difference in the abundance and maturity of spiders between control and treatment plots. Moreover, there was a significant difference between control and treatment plots in the taxonomic family, genus, species, and guild composition. However, there was no significant difference between the treatment and control plots in the sex ratio or Shannon diversity (at the family level) of the spiders. Furthermore, within these data, we discovered new spider distribution records and multiple potentially undescribed spider species. These data show the importance of wildflower strips at increasing pollinator abundance near agricultural plots.

The impact of infectious disease on toxic effects in a freshwater zooplankton model (Poster)

Juliana Ilmain and Catherine Searle, Purdue University

Contaminants in a freshwater environment can have harmful effects on wildlife. To determine a contaminant’s harmfulness, the lethal dose is often tested by exposing healthy animals to various concentrations of contaminant over a short period of time. However, toxic effects may differ in communities where disease is present; diseased animals may be more susceptible to the toxic effects of contaminants. This may suggest that current studies are providing an underestimation of toxicity, resulting in a more severe ecological and environmental impact than anticipated if disease were absent. To understand if there is an interaction between infection and toxicity, we exposed *Daphnia dentifera* to varying levels of salinity in combination with pathogen exposure in laboratory experiments. Half of the treatments were exposed to the fungal parasite *Metschnikowia bicuspidata*, and each individual was exposed to one of three salinity levels. Mortality was tracked over 120 hours, and survival was compared across treatments using a Cox proportional hazards model. Infectious disease and high salinity both significantly increased mortality, but the effects were not interactive. This project will be expanded to test

other contaminants such as insecticides to determine if disease significantly increases the lethal impacts of contaminated environments.

Translocation Approaches for the Imperiled Massasauga Rattlesnake

Bruce Kingsbury, Jillian Josimovich, and Monica Matthews, Purdue University Fort Wayne; Michael Ravesi, Michigan Department of Military and Veterans Affairs; Sasha Tetzlaff, University of Illinois Urbana-Champaign; Brett DeGregorio, U.S. Army Corps of Engineers

Purposefully moving (translocating) wildlife is a frequently employed if poorly studied practice. Here we report on activities underway to refine such approaches for snakes. Snakes may be moved because they are deemed a nuisance, a hazard, or as part of an effort to augment or repatriate other populations. A challenge has been that displaced animals appear lost and may exhibit behaviors such as long-range movements as they look for familiar territory. A strategy that may reduce such behaviors is “soft release”, temporarily holding animals on site, rather than a hard release - simply relocating them, with the hope that the snakes will acclimate and more likely accept their new location. Here, after some review of previous translocation efforts with snakes, we provide an update on our ongoing study investigating the utility of soft versus hard release for translocating the federally threatened Massasauga (*Sistrurus catenatus*). This constitutes some of the earliest research of the pros and cons of soft-release for translocating snakes, and to our knowledge is the first to report on how effective this technique is when translocating wild-caught snakes. Since 2013, we have radio-tracked over 50 soft-released (i.e. held in enclosures for approximately two weeks prior to release), hard-released (i.e. released immediately after translocation), and control massasaugas (i.e. released at capture location). We are comparing measures of survival and behavior to evaluate the “success” of each translocation effort. Preliminary analyses suggest that soft release may not help translocation efforts for this species, and that translocation in general is challenging. This work is continuing until 2019, and we will develop models exploring how factors like distance translocated, sex, and size may influence translocation success. This research will help inform conservation and management efforts to conserve the Massasauga as well as other imperiled herpetofauna.

Bat summer habitat use before and after timber harvest on two Indiana State Forests

Scott Haulton, Indiana Department of Natural Resources, Division of Forestry

The Hardwood Ecosystem Experiment (HEE) is the largest, most comprehensive study of forest management impacts on the flora and fauna of Indiana’s State Forests. Bat summer habitat use has been an important field of study within the HEE project since its inception in 2006. One aspect of this study included annual acoustic monitoring of bat activity in HEE research areas at Morgan-Monroe and Yellowwood State Forests. Two years prior to experimental harvesting, acoustic bat monitoring stations were established in three separate, replicated treatments; two treatments were due to be harvested using even-age and uneven-age silvicultural methods, while the third treatment would remain unharvested mature forest as a control. Even-age treatments consisted of 10-acre clearcuts and 3-stage shelterwoods within a matrix of unharvested mature forest; each uneven-age treatment included several patch cuts (1-5 acres)

within a matrix harvested using single-tree selection. Ultrasonic bat detectors (Anabat II/ZCAIM) were deployed annually at acoustic monitoring stations during the two summers prior to harvesting and three summers immediately post-harvest. Bat calls were identified to species based on call characteristics using the automated classification software package BCID (version 2.7). Multiple-season occupancy models were used in program Presence to estimate site occupancy (i.e. “use”) and detection probabilities. Post-harvest, total bat activity was greater in both harvested treatments than unharvested controls. Total bat use of even-age harvest treatments increased after harvesting. During the post-harvest period, most species were detected more often in harvested treatments, relative to controls, and no species was found to use unharvested controls more often than harvested treatments. No species used harvested treatments less following harvest.

Habitat preference of Migrating Northern Saw-whet Owls in Delaware and Henry counties, Indiana: preliminary results.

Kaitlin Gavenda, Kamal Islam, Clayton Delancey, Ball State University

This study aims to test if Northern Saw-whet Owls (*Aegolius acadicus*) exhibit a preference between two potential habitat types in Indiana, an old growth deciduous forest and a Christmas tree farm, during migration. Previous studies have documented the use of old growth deciduous forest. Christmas tree farms contain the coniferous trees preferred by these owls; however, it is an artificial environment that may not contain the necessary cover that Northern Saw-whet Owls favor. This study will examine if there are any differences in capture rates between these two habitat types based on season, and if males and females have different preferences in the type of habitats that they select. Two mist-netting stations have been established to monitor fall and spring migration periods: one at Ginn Woods (Ball State University property) in Delaware County, and the other at Whitetail Tree Farm in Henry County. Each station uses six mist-nets: a line of four nets, with one net on either side of the middle to form a cross. A recording of a Northern Saw-whet Owl call is played at the center of the cross to increase owl capture rates. During the 2017 fall migration period, we caught 10 owls at Ginn Woods, and 8 owls at Whitetail Tree farm, only one of which was a definitive male. Of these owls, 9 were hatch year, 5 were second year, and 4 were after second year. This project will continue during the 2018 spring migration period.

Engineering

Implementation and Testing Hydraulic Machinery Employing Lexan Designs (Poster)

Keith Pate, Joseph Marx, Farid Breidi, University of Southern Indiana

This work presents the build and test of hydraulic machinery utilizing uniquely layered Lexan designs. A miniature hydraulic excavator arm was designed and built to demonstrate the advantages of using multiple layers of Lexan, which allows observation of the mechanical components and mechanics of the arm. The excavator arm is mounted to a tool chest that also acts as the fluid reservoir for the hydraulic system. The hydraulic system used to control the excavator consists of four main components including an electrical water pump, mechanical valves, hydraulic cylinders, and flow control valves. The hydraulic fluid used in this system is water due to its availability and environmental benefits compared to other fluids typically used in hydraulic systems. An electric water pump is used to transfer water from the reservoir to mechanical valves. The mechanical valves are controlled using two individual joysticks. Each joystick controls two mechanical valves and is positioned for movement in two axis, x and y, and transfer fluid to hydraulic actuators that rotate the arm. Push-to-connect fittings are used on each of the valves and cylinders to make the hydraulic circuit easy to assemble. Flow control valve fittings are used on the hydraulic cylinders which utilize a metering out system that permits each fitting to regulate the flow through the cylinders, restricting the speed of the excavator arm. This mechanical system illustrates how hydraulic equipment transforms fluid energy into mechanical energy.

Design and Construction of Mechanical Components Utilizing Layered Materials

Joseph Marx, Keith Pate, Farid Breidi, University of Southern Indiana

The objective of this work is to design and construct mechanical parts using layered materials, which are typically manufactured in sheets. Sheet materials can be cut into intricate shapes using simple hand tools and can be assembled/disassembled for ease of portability. This makes layered material designs good for demonstration purposes and proof of concepts, where strength is not a main concern. One main advantage of such an approach is the freedom in sizing the parts, which is currently limited in additive manufacturing processes typically used in proof of concept designs. Using this layered material approach, a hydraulic miniature excavator arm was designed and constructed using a sheet of Lexan. One of the main purposes of this demonstrator is to create a visual of how hydraulics can transfer fluid power into mechanical power. Some of the mechanical components of interest on the arm include hydraulic cylinders, which are actuated to rotate several components on the arm making the arm mimic the movement of typical industry excavator arm. This transparent layered design provides students with a visual tool to see how fluid power technology is used in today's industry and can be used in multiple laboratory experiments including courses such as fluid power, statics, and dynamics.

Experimental Analysis of an Air Foil in Engineering Education

Michael Ivanyo, **Alexander Wray**, Nuri Zeytinoglu, Connor Feeney, Purdue University Northwest

Applying and assessing data from the usage of wind tunnels to the study of Fluid Mechanics has led to understanding the technical aspects of lab equipment necessary for research. To further understand the usage of new technology, this study will be geared on understanding the data acquisition and software used in utilizing the *Subsonic Flotek 1440 Wind Tunnel*, with the focus on utilizing the data acquisition to obtain key elements of the *NACA 2415* airfoil design. Additional focus will be on measuring the changes in *lift and drag forces*, as well as mapping *pressure distributions* and acquiring sets of *Reynold's numbers* whilst changing the *angle of attack* in the experiment. Experimental results will then be compiled and assessed, and compare to against the theoretical data. This research will also include the generation of a *NACA 2415* airfoil using *solid modelling*and *CAD programs*, after which the model was printed via *3D* printing with *PLA Plastic*.

Analyzing a Small-scale, Constructed Wetland for Stormwater Treatment (Poster)

Juliann Apple, Pascal Schlee and Michelle Marincel Payne, Rose-Hulman Institute of Technology

Stormwater treatment by means of constructed wetlands has the ability to effectively remove pollutants such total suspended solids, nitrite, nitrate and ammonia. Utilizing two small, lab-scale constructed wetlands, one free water flow system and one subsurface flow system, our research team analyzed the levels of these pollutants at different locations in the wetland. Our team of two Civil Engineering undergraduate students tested a variety of different water samples including tap water, storm water, and a high nitrate solution. A consistent decrease in nitrate and nitrite was observed throughout the systems. While there was not an overall decrease in ammonia, decreases were observed between individual basins. This research provides valuable data in determining the potential implementation of constructed wetlands as a lower cost, environmentally friendlier alternative to reduce combined sewer overflows, or treatment via traditional wastewater treatment plants.

Production of Long Chain Dicarboxylic Acids from Distillers Corn Oil Using *Candida viswanathii* (Poster)

Jennifer A. Mobley, Nicholas A. Palmer, Katharine S. Ryan, Irene M. B. Reizman, Rose-Hulman Institute of Technology

Dicarboxylic acids are specialty chemicals that are used in the production of lubricants, fragrances, and biodegradable polymers such as polyesters and polyamides. Long chain dicarboxylic acids (LCDCAs) are commonly made through ozonolysis of oleochemicals and petrochemicals, which results in low yields and smaller chain byproducts. Distillers corn oil (DCO), a co-product in dry mill ethanol production, can be used as a renewable feedstock to manufacture these LCDCAs via fermentation with *Candida viswanathii* (ATCC 20962), which results in a higher yield of LCDCAs with fewer byproducts. In this study, we examine the

technical and economic feasibility of adding a bio-based LCDCA production process to an existing dry mill plant that produces approximately 50 MM gallons of ethanol per year.
The DCO produced from dry mill ethanol plants contains triglycerides and long chain free fatty acids, which can be converted to methyl or ethyl esters with a transesterification reaction. The proposed production process involves conversion of the methyl esters to LCDCAs by *C. viswanathii*, followed by a separation train for purification of C16 and C18 chain length diacids with centrifugation, flash tank, and distillation steps. A preliminary cost analysis and process flow diagram have been developed, and potential scale up of these technologies will be investigated with a capital cost assessment.
To support process simulations, laboratory experiments were conducted on C. viswanthii to determine the optimal conditions for growth and biotransformation. Growth curves were analyzed for the yield of biomass on glucose, xylose, and glycerol. This yeast strain was found to grow well on xylose, which can be derived from corn cobs, and glycerol, which is a byproduct of the transesterification reaction, indicating a potential for cost savings for carbon substrate. Studies are currently in progress to determine the best conditions for LCDCA production from methyl ester standards and DCO.

Exploration of the recent transition to roundabouts from intersections in West Lafayette, IN

Terry R. West and Alivia Shively, Purdue University

Roundabouts have been utilized since the 1960s by Great Britain, but just in recent years, the city of West Lafayette, Indiana has chosen to implement several roundabouts in place of intersections within the city's limits. Why now? What makes roundabouts so unique and preferred over intersections? West Lafayette's assistant city engineer, Michael Susong, explained the philosophy behind the recent construction of roundabouts was to lessen the severity of accidents. The city of Carmel, Indiana and other communities within the area who had implemented roundabouts had seen the number of injury accidents reduced by about 80% and the number of accidents overall by about 40%. However, roundabouts cannot be installed at every intersection due to specific requirements. When deciding if a roundabout is an appropriate solution, cities need to consider the amount of space available, the concentration of pedestrians at that location, and the traffic volumes. Once implemented, roundabouts contribute to the community by potentially reducing costs, increasing the traffic capacity, cutting down vehicle emissions and fuel use, improving the aesthetics within the area, and greatly reducing serious injuries and accidents. After conducting a detailed evaluation, the city selectively chose several locations in which they believe will benefit the city's future.

Environmental Science

Paleoclimate History and Seismic Survey of Pretty Lake, Indiana (Poster)

Hillary Johnson, Emily Starbuck, Broxton Bird, and Jeffery R. Stone
Institutions: Indiana State University and Indiana University-Purdue University Indianapolis

Pretty Lake is located in northern eastern Indiana; it originally formed around 16,000 years ago as a kettle lake that resulted from the Laurentide glaciation. Since its formation, Pretty Lake has filled with a diatom-rich sediment record and has been shown to be isotopically responsive to synoptic upper atmospheric variation caused by the Pacific-North American (PNA) teleconnective pattern. Here we present information on the fossil diatom history and lake seismic basin analyses developed to explore for evidence of past hydroclimate fluctuations and potential changes in lake stratification. Fossil diatoms are especially significant because they are sensitive to multiple environmental variations and can be used to help distinguish large-scale changes in lake level, water chemistry, and nutrient flux throughout the lake's history. The upper 2000 years of the sediment record is varved, providing certainty that sediments were undisturbed by substantial bioturbation. To support the diatom record, we also created high-resolution bathymetric map using seismic data from a chirp sub-bottom profiler. The bathymetric map was used to create a three-dimensional model of the lake basin surface. Bathymetric information will be used to support the interpretation of fossil diatom record by allowing for the creation of a model of the available diatom habitat under differing lake-level scenarios.

Bioavailabity of Lead in Indiana Soils (Poster)

Caroline Meunier, Jennifer C. Latimer, Joshua Pigg, Indiana State University

Urban soils frequently have a high lead burden from the past use of leaded paint and fuel, as well as industrial emissions. Once lead is in soils, it has low mobility and often stays near the surface in undisturbed areas. Because soil and soil dust can have high lead concentrations, communities are often concerned about childhood exposure to lead. Young children, in particular, have hand to mouth behaviors that increase their potential exposures if their surroundings include environmental sources of lead. The purpose of this research was to further evaluate lead bioavailability following digestions of contaminated soils using a simulated gastric solution. After extraction, all samples were analyzed using ICP-OES. Previous studies in Terre Haute and Indianapolis have documented areas with elevated soil lead concentrations. Additional analyses of these soil samples revealed that a high percentage of the total lead was extractable using a simulated gastric solution after reaction for 2 hours, which was meant to mimic the digestive process. For some samples, as much as 100% of the lead was found to be bioavailable; however, typical ranges were between 45-75% bioavailable. To further understand bioavailability, we are increasing the pH of the samples after the 2-hour extraction to simulate the movement of material from the stomach into the intestines where most absorption occurs. These analyses are ongoing, but the results will help us to better understand and quantify lead bioavailability.

Spatial econometric modeling for determining relationships between crop yield and landscape features

Ashley Kissick, James Camberato, and Robert Nielsen, Purdue University

Sound agronomic recommendations are crucial for today's agronomists as they strive for improved yields, profits, and sustainability. Determining the spatial relationships between yield and landscape variation including soil properties and terrain attributes may improve management decisions, particularly with regards to proper nitrogen application for minimizing both costs to farmers and environmental impacts. Here we investigate relationships between landscape features, soil texture, soil properties and corn yield as part of a preliminary study to model corn yield with variations in landscape attributes, soil properties, and weather. We used yield monitor data collected from 2010 – 2015 at a 12 ha field at the Davis Purdue Agricultural Center in Randolph County, IN, USA. We obtained 25 digital elevation-based models of terrain attributes that describe morphometric and hydrologic characteristics of the field. We also sampled the field to obtain soil texture and soil property estimates. For each year we used the random forest method to select the variables that were most important for predicting corn yield across the field and used these in spatial error models. The variables with the most significant relationship with corn yield each year were soil texture and topographic wetness index. These results demonstrate that models for predicting corn yield in Indiana need to include landscape features for increased model performance. This analysis met one objective of a larger investigation that will incorporate soil properties, terrain attributes, and weather patterns into models of corn yield across Indiana landscapes.

A Multi-Proxy Analysis of Lake Sediments to Assess Impacts of Coal-Fired Power Plants in Indiana (Poster)

Erika Smith, Jeffery R. Stone, and Jennifer C. Latimer, Indiana State University

Indiana is known for its coal mining industry and massive coal-fired power plants. Some of the power plants in southern Indiana have historically been ranked among the largest contributors of atmospheric pollution. Our research explores how pollutants from the coal-fired plants may impact water quality and aquatic ecosystems in several lakes near two major power plants using a multi-proxy approach to evaluate sediment cores. These proxies include diatom indicators of water quality, quantification of carbonaceous spheroidal particles (CSPs), and geochemical analyses of heavy metals associated with coal combustion. Diatoms are a type of brown algae that are sensitive to the introduction of nutrients and pollutants in lake systems. For this reason, fossil diatoms are useful biotic indicators of changes in water quality through a lake's history. SCPs form as a result of the incomplete combustion of coal and are rapidly deposited and preserved in local lake sediments. SCPs can be used to track changes in atmospheric pollution produced by nearby power plants. Heavy metals commonly associated with coal combustion include Hg, Pb, and Cd among others. We will use consensus-based sediment quality guidelines to evaluate the risk to benthic organisms and the impact of the metals on the local ecosystem. The down-core records will be compared to power plant generation history to determine if higher levels of power generation directly relate to the observed trends. This research will contribute to our understanding of how industries such as coal-fired power plants

are impacting our local ecosystems. Findings may also demonstrate how effective regulation and improved smokestack scrubber technology are at filtering out harmful atmospheric pollutants.

Analyzing the water quality of the Wabash River using diatom species assemblages and geochemistry

E. O'Neil, J. Stone, J. Latimer, Indiana State University

The Wabash River is a large meandering stream that stretches for over 471 miles from Ohio through Indiana, where it ultimately joins the Ohio River. The majority of Wabash River tributaries drain agricultural landscapes. Ramifications of this agricultural drainage are such that the Wabash River Valley comprises some of the highest nutrient yields in the US, which contribute to excessive algal blooms and nutrient exports into the Gulf of Mexico. This study uses diatom assemblages and geochemistry to analyze the water quality of the Wabash River. Sampling sites were selected to focus on the agricultural influxes from the Terre Haute area and Otter Creek tributary into the Wabash River. Over the past 15 months we collected weekly live diatom samples, water chemistry data, and water samples for nutrients and phosphorous analyses from the Wabash River to create a high-resolution dataset. Nutrient data, specifically nitrate and ammonium are collected using a YSI. Water samples were analyzed for phosphrous using the molybdate blue technique. Analyzing specific geochemical parameters provide insights into nutrient load preferences of specific diatom species. These trends will ultimately be used to give a macorscopic picture of the Wabash River's impact on hypoxia in the Gulf of Mexico.

Comparative water quality assessment of Lake Sno-Tip, a small Midwestern impoundment (Poster)

Adrienne C. Funderburg and F. Collin Hobbs, Huntington University.

Lake Sno-Tip is a 2 hectare impoundment located on Huntington University’s campus in Huntington, IN. The lake has a reputation among the student body for being polluted and of poor quality, but no recent studies have been performed to confirm or deny that perception. I measured secchi depth, dissolved oxygen, temperature, and total phosphorus once a week for five weeks in October and November 2017. The data were compared to a Sno-Tip water quality assessment from 1977-78, a study performed on Prairie Creek Reservoir in east-central Indiana in 2006-07, and IDEM Water Quality Targets. Average total phosphorus was half as high in 2017 (0.043 ppm) as it was in 1977-78 (0.082 ppm), and the 2017 range fell within the 1977-78 range. Average secchi depth also fell between previous measurements, but average dissolved oxygen was lower in 2017 (7.31 ppm, surface) despite inclusion of summer months in the 1977-78 sampling period (10.15 ppm, surface). Compared to IDEM Water Quality Targets and Prairie Creek Reservoir, Lake Sno-Tip is of average and acceptable water quality for a lake of its type, though phosphorus levels are close to the EPA recommended maximum.

Cs-137 Fallout Across Indiana's 92 Counties (Poster)

Rick Whitman and Carson Wright, Ball State University, Natural Resources and Evironmental Management

The atomic bomb detonations, especially those at megaton yield, tested by the United States, Soviet Union, China and France in the Northern Hemisphere dispersed fallout throughout the Hemisphere, with highest concentrations centering on 40 degrees North latitude. Most testing of soil for fallout since then consisted of a single sample in an area, or on sloping ground as part of soil erosion studies. Cesium-137 with a long half-life (30.17 y) and with a unique gamma signature (663 kev) appears more than others in the literature and its half-life makes it the ideal non-natural isotope for this study. The current study measured for Cesium-137 on locally level and mostly private property in all 92 Indiana Counties during the summer of 2017. Forested and Grassland areas unplowed since 1940 revealed distinctly greater retention in forested areas as compared to meadows with a correlation to clay and precipitation levels. Sampling took place from 2 to 12 cm depth, oven dried, ground and sieved at Ball State University to <2 mm sizeand then placed in Marinelli beakers and read on a nitrogen cooled and calibrated Germanium-Lithium detector at Purdue University for an hour each. The results disclosed patterns of retention and invite additional work at sub-county and across different terrain.

Fossil Diatom Record from a Proglacial Lake in Indiana (Poster)

Jordyn Loveall, Jeffery Stone, Henry Loope, Indiana State University

A set of sediment samples were collected from a 17-meter long core near Brush Creek, southeast of Columbus, Indiana. These samples were dated using radiocarbon techniques to between ~25,000 and ~24,000 years before present and they are thought to be the remnants of part of an ancient proglacial lake that extended from the front of the Laurentide Ice Sheet. The sediments had previously been identified as lake sediments because of the presence of ostracodes. Here we present further evidence that the samples represent a lake environment through analysis of fossil diatoms recovered from the sediments. Diatoms are highly sensitive environmental indicators in lakes and because the skeletons are composed of silica that does not decompose, they are often preserved in lake sediments. Our preliminary observations show that multiple diatom species are present in the sediment samples including genera such as *Staurosirella*, *Mastogloia,* and *Amphora*. These groups typically represent shallow to intermediate water depths, but at least a few of the samples have plankton present, indicating open water. Further quantitative analysis of the fossil diatom assemblages in these samples should provide more insight into the lake environment and history of changes in lake level and water chemistry throughout the sediment record.

Heavy Metal Analysis in Sediments Recovered from the Wabashiki Wetlands and Izaak Walton Lake (Poster)

Matthew Barley and Jennifer Latimer

Although water quality has improved significantly over the past several decades in the Wabash River, little is known about how efficiently heavy metals are being stored in sediments in the surrounding wetland areas. The Wabashiki Fish and Wildlife Area (WFWA), located in west-central Indiana, is currently undergoing restoration to mitigate flood damage and to develop outdoor recreational areas. During spring floods, the WFWA is inundated, and given the past history of pollution along the Wabash River, it is possible that bottom sediments in the WFWA are storing contaminants such as heavy metals. In contrast, Izaak Walton Lake, which is due north of the WFWA, has been used for several decades for boating and does not have a direct connection with the Wabash River. The purpose of this project is to evaluate the geochemistry of sediments collected from both WFWA and at Izaak Walton Lake to determine if these areas are acting as sinks for heavy metals. Sediment quality guidelines will be used to evaluate ecological risks posed by metals such as lead and mercury. Samples from the WFWA were recovered using a Livingstone coring device in permanently inundated areas, while cores from the Izaak Walton Lake were recovered using a gravity coring device. As a result, the Izaak Walton cores are significantly longer and represent an older record. Once collected, cores were extruded and samples were freeze-dried. A heavy metal analysis is ongoing via inductively coupled plasma-optical emission spectrometry.

Hook, Line and Sinking Metals (Poster)

Kathryn Mudica, Indiana State University

Anthropogenic activities such as mining and industry have caused an accumulation of trace heavy metals. Bioremediated areas often become a sink for the toxic contaminants. These metals enter the food web affecting plant and animal life as well as humans. Biomagnification of heavy metals in lake environments can have detrimental effects on humans that rely on fish as sources of protein in their daily diet. Learning disabilities, nervous system damage, decreased muscle and bone growth, and kidney damage in humans are linked to low exposures of metals. Current testing methods do not closely match consumption habits so therefore do not give a correct assessment of the amount of exposure of heavy metals on humans. Fish are an important nutrition and economic source. . The global fishing industry generates well over $100 billion annually and employs more than 200 million people. Analytical methods that more closely match consumption habits in necessary to properly analyze metal contaminants. Safe fish consumption is a global concern.

Phytoremediation of Soil Contaminated by Produced Water (Poster)

Lucas H. Clay and John Pichtel, Ball State University

In recent years hydraulic fracturing has been increasingly used in oil and natural gas production. The process involves injection of specialized fluids into a target geologic formation to initiate hydrocarbon recovery. So-called produced water (PW) is recovered along with the hydrocarbons. Disposal of PW is a concern, as it may contain hundreds of different chemicals, many known to be harmful to human health and the environment. In the greenhouse, constructed wetlands were tested for their ability to treat synthetic PW. Test plants included sedge (*Cyperaceae*) and cattail (*Typha latifolia*). Growth media included peat or Glynwood soil. Over the course of 13 weeks, six wetlands were contaminated with increasing PW concentrations. During this period, the plants grown with PW maintained robust growth and showed no signs of toxicity. Leachate, soil, and plant tissue (shoots and roots) were tested for concentrations of Na, K, Pb, Cr, Cu, Mg, Fe, Li, Ni, and Cd. Overall trends show metal concentrations in the leachate to be decreasing. It is expected that several of these contaminants have been removed from soil and taken up into plant tissue. Current work involves determination of possible metal sequestration in various plant parts. If these plants are capable of withstanding metal toxicity, they may be feasible for remediation of contaminated field sites.

Response of Tidal Wetlands to Rising Sea Level in Three Estuaries along the US Southeast Coast

Elena Solohin and Christopher B. Craft, Indiana University

Tidal wetlands are increasingly threatened by global climate change and anthropogenic activities. Future rates of sea level rise (SLR) and altered sediment supply due to land use change may have an impact on future tidal wetland ability to keep pace with rising seas.
We used a combination of soil and plant measurements, and remote sensing to understand how tidal marsh health and resiliency has changed over time along three Southeast estuaries: Cape Fear (NC), Edisto (SC), and Altamaha (GA). Soil accretion and mineral deposition were measured using radiometric dating to determine marsh response to current SLR. Preliminary results revealed that accretion rates for cores collected in GA, NC and SC saltmarshes are on the order of 2-4 mm yr^{-1}, indicating that marshes are keeping pace with current SLR (2-3.2 mm yr^{-1}). However, remotely sensed time series analysis of tidal marshes' biomass and vegetation height reflect regional patterns of vulnerability to SLR, with greatest decline in the health of NC marshes. This is supported by RTK measurements of marsh elevation, which suggest that NC marshes have lower elevation in tidal frame, making them more susceptible to SLR than GA and SC marshes.
Understanding the response of marshes to changes in land use and human disturbances will inform coastal adaptive management in the face of climate change and SLR.

Ohio River Levee Performance Evaluation under Flood Conditions

John A. Mundell and Forrest Kunkel, Mundell & Associates

Many of the Nation's levees were built more than 50 years ago with the intent of protecting crops from flooding. Over the last several decades, as housing and commercial development on land protected by levees has taken place, the risk to public health and safety and property damage associated with potential levee failure has also dramatically risen. This is notable along many segments of the Ohio River, where the levee systems constructed since the 1940s by the U.S. Army Corps of Engineers (USACE) have long since exceeded their design lives, and as changes to the River's hydrologic dynamics has been modified both by development and the construction of dams. This presentation will focus on a case study of the evaluation of one levee system along the Ohio River and lessons learned from its performance since its construction in 1940. Current efforts being made to upgrade the levee system, rehabilitate and evaluate the existing pressure relief well system, and assess potential failure modes based on the occurrence of sudden, rapid and uncontrollable events will be discussed. Within the case history, past levee design methods utilized by the USACE will be reviewed as well as new methods for levee risk assessments. Finally, the value of levee operation and maintenance activities including the collection of actual levee performance data during flood conditions will be highlighted to demonstrate the challenges faced by levee sponsors and communities in maintaining USACE certifications and FEMA accreditations within limited budgets.

Chlorinated herbicide leaching from utility poles and potential impact on private water wells: first documented case from Indiana.

Rachel Walker and John Mundell, Mundell & Associates.

The chlorinated herbicide pentachlorophenol (PCP) has been shown to migrate from wood utility poles into the environment. Once released, PCP readily adsorbs to acidic soils, but in neutral or alkaline soils, PCP is more mobile and dissolves into groundwater over time. It was generally assumed that a limited extent of groundwater could be impacted as a result of PCP leaching from utility poles. However, PCP contamination of water wells near wood utility poles has been documented. In the state of Vermont, cases of PCP groundwater contamination from wood utility poles were identified in 2009. One involved the contamination of a shallow dug water well by a pole installed upgradient approximately 50 feet away.

Recently, a case of utility pole related contamination of private water wells was identified in southern Indiana. Two wells were found to have experienced PCP contamination linked to newly installed PCP-treated wood utility poles that had leached into shallow groundwater. Both impacted wells were shallow dug water wells. Well #1 was located 16 feet from a pole, while Well #2 was 52 feet from the nearest pole. Well #1 dropped to non-detect after the first quarter of monitoring, while Well #2 experienced very high initial PCP levels followed by an overall decreasing trend that did not reach non-detect until the ninth quarter of monitoring. This well also experienced occasional spikes in PCP concentration over eight quarters that appeared to be related, in part, to seasonal rainfall events. Another factor contributing to contamination of

this well was construction-related, with sediment infiltration of the concrete casing allowing PCP containing soils to enter the well bore.

These cases of PCP contamination illustrate a potential threat to private water wells that was previously unrecognized in the Midwest, as well as further indication that shallow dug wells are chronically prone to contamination.

Efficiency of combined chlorination and silica filtration for antibiotics removal from wastewater (Poster)

Kayla Burch, Bangshuai Han, Natural Resources and Environmental Management, Ball State University

The occurrence of antibiotics in freshwater ecosystems is a growing concern among industrialized nations. Antibiotics place selective pressure on resistant bacteria in the environment, leading to the emergence of multi-drug resistant bacteria. Wastewater treatment effluent is a significant contributor of antibiotics release to the environment. Conventional secondary wastewater treatment technologies, which are primarily biological in nature, cannot completely eliminate antibiotics; therefore, tertiary processes are critical in their removal. Chlorination, the most widely implemented tertiary treatment method for water disinfection, has demonstrated high removal efficiency for three classes of common antibiotics (macrolides, sulfonamides, and tetracyclines). Combining chlorination with another tertiary method, silica filtration, may result in synergistic effects in the removal of these classes of antibiotics. Few studies have determined the removal efficiency of antibiotics for a combined chlorination and silica filtration method. This study examine the potential of combined chlorination and silica filtration for antibiotic removal from wastewater. Samples are collected from the influent and effluent of the Muncie Wastewater Treatment Plant. Solid phase extraction is conducted to purify the samples and liquid chromatography-mass spectrometry is utilized to determine the presence and concentrations of antibiotics in the samples. The removal efficiency of the chlorination/silica filtration method for each antibiotic class will be analyzed. The results will increase our knowledge base regarding the effectiveness of antibiotic removal from wastewater and may direct future wastewater treatment design.

Earth Science

***Euglena mutabilis*—Stromatolite builder in an acid mine drainage environment** (Poster)

Lucas Monroe and Sandra Brake, Indiana State University; Benjamin Magnin, DePauw University

Euglena mutabilis, a photosynthetic, acidophilic protozoan, forms benthic biofilm layers under the harsh conditions of acid mine drainage (AMD). In the AMD environment at the Green Valley coal mine site near West Terre Haute, Indiana,*E. mutabilis* contributes to the formation of Fe-rich biolaminated structures (stromatolites). This study examines the ability of *E. mutabilis* to use secreted mucilage to bind chemical sediment that form continuously in the AMD system. Simulated AMD and powdered Al hydroxide was used to make a slurry to simulate chemical sedimentation. The slurry mixture was applied to a glass slide containing pipetted *Euglena* cells and examined microscopically. Microvideography was used to document cell response to chemical sedimentation. The results indicate that *E. mutabilis* is very productive worker. Cells were capable of binding and transporting bundled slurry particles to a growing accumulated mass of particles that was, in turn, reworked by other *Euglena* cells. Time-lapse microphotographs show enlarging areas swept clean of particles via the back and forth sweeping locomotion of *E. mutabilis* cells. Particles encountered by the cells during sweeping locomotion were transported down the sides of the cell and bound in a bundle that was dragged behind the cell to a centralized mass of particles. This study will also use ImageJ software to quantify spatial clearing of simulated chemical sediments by *E. mutabilis* cells. We propose that the microbial activity of sweeping, gathering, binding, and transport of chemical sediments is a mechanism used to construct thinly laminated layers as stromatolites in the AMD environment. This study has important implications for understanding microbial activity responsible for the formation of similar stromatolitic structures on early Earth and for the exploration of similar features on such extraterrestrial bodies as Mars.

Detailed Soil and Terrain Maps for the Conterminous United States

Darrell G. Schulze, Purdue University, and The Isee Network

We have developed approaches for visualizing soils and landscapes data at scale, and for delivering these highly detailed maps via the SoilExplorer.net website and the Soil Explorer app (for iPad now, iPhone and Android versions coming soon). This presentation will highlight a number of new maps recently added for the conterminous United States. Soil Explorer uses a hillshade base map derived from the best available elevation data from the US Geological Survey's 3D Elevation Program (3DEP) (https://nationalmap.gov/3DEP/) and other sources such as Indiana's state-wide lidar data (http://IndianaMap.org). The hillshade is calculated to ~5 meter resolution (~1:18,000) and is exceptionally detailed where 1/9 arc second (~3.5 meter) 3DEP or other lidar data is available (~25% of the conterminous US), and less detailed where only 1/3 arc second (~10 meter) data is available. The U.S. Soil Survey Geographic Database (SSURGO), available as a gridded version at 10 meter resolution, provides the data for a

growing number of soil property maps. Maps currently available for the conterminous US include Soil Orders and Natural Soil Drainage Classes. A Dominant Soil Parent Materials map currently includes 7 states, with 10 more to be added soon. Soil Explorer also displays aerial imagery and a topographic map from the USGS. These maps can be used for a variety of purposes including soil and environmental science education, habitat assessment, and geomorphology studies.

Mapping Soil Spatial Variability at the Purdue Agronomy Center for Research and Education (ACRE)

Shams R. Rahmani and Darrell G Schulze, Purdue University

Soil variability is an important factor in field-based plant phenotyping research because a given genotype is likely to express a slightly different phenotype on different soils in the same field. Climate, organisms, relief (topography), parent material, and time are the soil-forming factors that cause soil variation across soil landscapes. Within individual farm fields, however, most of these factors are constant and relief is usually the major factor determining soil variability. Topographic differences influence plant growth and yield through water, nutrient, and sediment movement from higher spots to lower spots in the field. Additional variability is introduced by tile drainage, and by ice-wedge polygons that formed on parts of ACRE at the end of the Wisconsin Ice Age about 15,000 to 18,000 years ago. We are using aerial photography and computer modeling to capture soil variability and develop high-resolution soil maps for ACRE. Aerial photography taken at different times shows different features and is used to capture soil variability due to long-term differences in plant growth in the original pre-settlement vegetation, tile lines, and ice-wedge polygons. Terrain attributes calculated from high resolution lidar data is used for quantifying surface topographic variation.

Patterns and Changes in Atmospheric Mercury Transport to Indiana Landscapes

Martin R. Risch, U.S. Geological Survey and Donna M. Kenski, Lake Michigan Air Directors Consortium

Mercury (Hg) is a persistent environmental contaminant that accumulates and concentrates in aquatic and terrestrial food webs to present health risks to humans and wildlife. Most of the Hg in these ecosystems comes from the atmosphere and Hg in the atmosphere predominantly originates from human activities. In the past 16 years, regulatory actions and economic factors have affected the transport and transfer of atmospheric Hg to landscapes in Indiana and surrounding states where a third of the stationary sources of anthropogenic Hg emissions in the continental USA were located. The transport and transfer of atmospheric Hg to landscapes was measured by analysis of precipitation samples to quantify wet deposition and by analysis of forest litterfall samples to estimate dry deposition. An extensive monitoring record for wet and dry deposition of Hg was investigated, which revealed spatial patterns and temporal changes in Indiana and surrounding states during this time. Statistical and graphical analysis suggested that local and regional, rather than exclusively continental or global Hg emissions sources were likely contributing to the observed spatial patterns of wet Hg deposition. Some locations had recurring, episodically high Hg deposition. Statistically significant decreases of Hg

concentrations in precipitation were identified during the 16-year study. These temporal changes coincided with reported reductions in Hg emissions in the USA required by national rules and related to economic responses to the rules. This study illustrates the value of long-term environmental monitoring for understanding the status and trends in statewide conditions framed with a regional and national context.

Renewal of Archival Legacy Soil Data: A Case Study of Busia Area, Kenya

Joshua O. Minai, Darrell G. Schulze, Purdue University

Much older soils information, collectively known as 'legacy soil data' lies idle in libraries or in the personal collections of retired soil scientists. The probability of this legacy data being lost or destroyed is very high. We demonstrate the stepwise process of bringing legacy soils data 'back to life' using the Reconnaissance Soil Survey of the Busia Area in western Kenya as an example. The first step, *data archeology*, involves locating and cataloging legacy soil data from key institutions, which often requires numerous site visits and the assistance of individuals familiar with the desired data. The second step, *data rescue*, involves converting paper copies of data into a digital format by scanning the maps, narrative descriptions, and tables, and storing the information in a database. The third step, *data renewal*, consists of bringing the data to modern standards by taking advantage of technological and conceptual advances in geo-information technology. In our example, the resulting digital soil map of the Busia area is a significant upgrade from the fragile old paper map. Careful interpretation of the agronomic information available within the legacy soil survey allowed us to produce ten land quality maps showing the ability of land to perform specific agronomic functions, and nineteen different crop suitability maps that were not available originally. We have made some of these maps available in the Soil Explorer app and SoilExplorer.net website. These rescued maps and their associated tabular and narrative data, while useful themselves, also provide crucial inputs for generating more detailed soil maps using digital soil mapping techniques that were unavailable when the original mapping was conducted.

Secrets within the varves: A Late Holocene climate reconstruction of Herd Lake (Poster)

Bethany L. Kile, Jeffery R. Stone, Jennifer C. Latimer, Indiana State University, Mark Shapley, Bruce Finney, Idaho State University

Herd Lake located in Idaho is a eutrophic lake with high productivity and high sediment burial rates. These conditions have resulted in thick, nearly continuous seasonal laminations (varves) since the formation of the lake approximately 2,500 years ago. A short (53-cm) surface core was collected and sub-sampled by individual lamellae, resulting in a record extending back to 1923CE. Fossil diatom assemblages, major/minor element concentrations, and detailed phosphorus geochemistry were analyzed, and results were compared to instrumental climate data of the region. We found that diatom productivity, represented by changes in valves per gram (VPG), were closely linked to the regional effective moisture. The fossil diatom assemblages throughout the record were dominated by alternating blooms of *Stephanodiscus* c.f. *yellowstonensis* and *Stephanodiscus parvus*. *Stephanodiscus* c.f. *yellowstonensis* appears to be associated with the mixing and nutrient

cycling of seasonal succession, evident by their dominance in the light (spring) laminae of varves and correlating changes in the burial of oxide phosphorus. A longer (10.99-meter) sediment core was collected in 2013, which extends the record back to 623CE. This core was also sub-sampled by individual lamellae and evaluated using the same approach as the short core. The correlations between the short core and the instrumental climate record are being used to reconstruct regional climate conditions throughout the Late Holocene using this longer record.

Anthropogenic influence (?) on Earth-tides in the Inglefield Sandstone, Southwestern Indiana (Poster)

Payton L. Lykins, Paul K. Doss, and Eric Greenwood: The University of Southern Indiana

Water-levels from a deep-shallow piezometer nest in the Inglefield Sandstone portray a dynamic groundwater system. Ground water-levels at the 33.5 and 18.3 m depths fluctuate significantly, and include high frequency and high amplitude barometric effects and low-amplitude periodicity from Earth-tide stresses. Barometric influences were removed from water-level data using the Kansas Geological Survey-Barometric Response Function software (KGS-BRF). Fast Fourier Transform (FFT) analysis of tidal periodicities in corrected water levels suggests that the solar stressor signal was stronger than the lunar stressor signal, yet the moon is the dominant tidal force on Earth. We hypothesize that the higher-amplitude solar stress periodicity may result from an anthropogenic influence operating on a solar cycle, such as the diurnal loading of campus by people and vehicles. FFT analyses of 2016 hourly water-level data for the deep (WMW) and shallow piezometers (EMW), and hourly data from winter 2015, fall 2016, and winter 2017 identified periodicities of water-level change at 12.4 hrs and 12.0 hrs for WMW, 12.5 hrs and 12.0 hrs for EMW, 12.0 hrs for winter 2015 and fall 2016, and 12.2 hrs for winter 2017. Periodicities of winter and fall water-level data correlate with a solar stress. Lunar stressors were observed for both wells in the 2016 year dataset. Solar signal amplitude remained unchanged between winter 2017, fall 2016, and winter 2015 datasets. Our hypothesis suggested we would observe a smaller amplitude solar signal in the winter data because no daily aquifer loading was occurring on campus. However, our results provide no conclusive evidence to suggest that daily aquifer loading is the source of the stronger solar stress signal.

Body Size and Locomotor Performance of Non-Avian Theropod Dinosaurs: Evidence from Fossil Trackways

James O. Farlow and Dan Coroian, Purdue University Fort Wayne

Non-avian theropod dinosaurs spanned an enormous size range, including the biggest bipedal animals that ever lived. Interpretations of how they moved, and how their locomotor performance varied with size, have been based on functional morphology of their skeletons, and on comparisons with modern animals. Fossil trackways (sequences of footprints made by the same animal) provide an independent source of information. We collected data from the literature and our own observations for more than 1800 trackways attributed to theropods, determining the mean footprint length (a proxy for body size) and stride length (distance between successive footfalls of the same foot). Most data points in the stride: footprint length

relationship fall along a "main sequence" that presumably describes the performance of walking animals. The relationship is curvilinear, with bigger dinosaurs taking relatively shorter steps than smaller dinosaurs. Some points plot above the main sequence, and probably reflect animals moving at speeds faster than a walk. We compared the performance of theropods of different body size (footprint length) using three different measures: 1) the ratio of stride length to footprint length (all trackways); 2) the ratio of observed stride length to expected stride length from the stride: footprint length relationship for the main sequence (potential runners); 3) estimated trackmaker speed (all trackways). The stride/footprint length ratio may increase from the smallest to slightly larger theropods, but then progressively declines as the animals get bigger, whether one looks at main sequence trackways or those attributed to running dinosaurs. For potential running dinosaurs, the observed/expected stride length and the estimated speed both peak at footprint lengths of 20-40 cm, but then decline among the biggest trackmakers. The biggest theropods either did not run rapidly, or did not do so very often.

Comparison of Soil Classes in the Public Land Survey System to Current Soil and Terrain Maps (Poster)

Erica R. Wyss and Darrell G. Schulze, Purdue University

The Public Land Survey System (PLSS) was developed under Thomas Jefferson with the creation of the Land Ordinance of 1785. Under the PLSS, states were divided into 6 x 6 square-mile townships, and the townships were further divided into 36 one-square mile sections. Surveyors were required to note a variety of natural features, including “the quality of the soil.” The purpose of our study was to compare the land classification in the PLSS with current soil maps to gain insights as to how concepts of soils have changed over the past 200 years. Our study area is Township 23 North, Range 5 West, 2nd Principle Meridian, which was surveyed in 1822. The area is bisected by the Wabash River and includes part of Purdue University, West Lafayette campus. The original field notes and plat were downloaded from the Tippecanoe County Surveyors Office website. We georeferenced the plat map from the original survey to the modern-day PLSS grid available from IndianaMap.org using ArcGIS Pro version 2.0. We read the narrative descriptions given in the original survey notes, extracted the land classifications and added them to our GIS dataset. The land was characterized as first rate, second rate, or third rate, and then also by the any notable geographic characteristics such as whether the land was under prairie or forest, and whether it was flat, rolling, or broken. We then compared the PLSS soil rating to modern soil maps available on the SoilExplorer.net website.

Developing a Comprehensive Natural Hazards Map for the Indiana-Illinois-Kentucky Tristate Area (Poster)

Hannah R. Walker, Paul K. Doss, James M. Durbin: Department of Geology and Physics, University of Southern Indiana

Natural hazards include geological and meteorological events that threaten human health, life, economic security, and property. Tools such as Geographic Information System (GIS) have been used to create natural hazards maps to improve prediction and mitigation techniques and lessen risk to communities in the United States. The intent of this study is to create a comprehensive map of natural hazards occurrence in the Indiana, Illinois, Kentucky tristate area from 1997 to 2017. The tristate area includes 36 counties from Indiana, Illinois, and Kentucky. These states seek to share stable and secure business, work force, and financial resources and will benefit from a comprehensive natural hazards map of the area. Natural hazards to be included in this study are droughts, earthquakes, floods, winter storms, severe storms, and tornadoes. Droughts defined by Standard Precipitation Index values of -1.0 or less will be included, as will all earthquakes and tornadoes that occurred in this region during the time frame. Winter and severe storms designated as major disaster declarations by FEMA will be included. It is unclear at this point how flood events will be designated, but they may be defined by flood inundation areas and/or FEMA declarations. The development of a map that locates the occurrence and distribution of natural hazards from the previous 20 years can be used by local and regional governments and emergency responders to improve economic development and prediction and mitigation responses to natural hazards. The findings of this study will be an important educational tool and can be useful for proactive land-use planning. Further efforts might include mapping the occurrence of human-caused natural hazards such as mine subsidence or fires.

Petrified wood from the Inglefield Sandstone (Upper Pennsylvanian) in Vanderburgh County, Indiana (Poster)

William Elliott and Scott Beard, University of Southern Indiana

The Patoka Formation (Upper Pennsylvanian) overlies the West Franklin Limestone of the Shelburn Formation and is capped by the Carthage Limestone of the Bond Formation in the western part of Vanderburgh County, Indiana. The lowermost part of the Patoka is subdivided into the Ditney Coal and overlying Inglefield Sandstone Member. The Inglefield Sandstone consists of tan to gray, thin to thick bedded, cross-stratified, fine- to medium-grained sublithic to quartz arenite, interpreted to have been deposited in an incised paelovalley during a lowstand systems tract. Ten samples of petrified wood were collected from the Inglefield Sandstone west of Evansville near the campus of the University of Southern Indiana. Tangential and transverse thin sections were prepared from silicified and hematite replaced specimens. Transverse sections exhibit uniseriate rays consisting of two to twelve cells. Most cells are circular in shape, while others are angular or distorted. This distortion is most likely caused by compaction. Tangential sections show narrow rays in contrast to broad tracheids. Growth rings were not observed in any of the studied specimens. Four specimens are identified as *Dadoxylon*, four specimens are unidentified *Cordaites*, and two specimen may be coniferous. Episodic

regression of continental seas due to periodic Gondwana glaciation in the Late Pennsylvanian resulted in repeated exposure of depositional systems across the Illinois Basin. The association of Cordaitalean and coniferous petrified wood from the Inglefield Sandstone is consistent with dry lowland vegetation that accumulated during a lowstand systems tract. The lack of growth rings in the petrified wood specimens suggest a lack of seasonality. Because of the decreased preservation potential of lowstand deposits, this study provides additional insights into the xerophytic floras of the Late Pennsylvanian (Kasimovian) of North America.

Predicting Soils Types and Properties with Limited Data in the Uasin Gishu Plateau, western Kenya

Mercy W. Ngunjiri, Joshua O. Minai, Darrell G. Schulze, Purdue University, Zamir Libohova, USDA Natural Resources Conservation Service, Cornelius Serrem, University of Eldoret

Digital soil mapping approaches can be used to create new soil maps or enhance existing maps, particularly in areas where only very general soil maps are available. In this study, we utilized a knowledge-based inference soil mapping approach to develop a first generation digital soil map for part of the Uasin Gishu Plateau in western Kenya. Knowledge-based inference soil mapping integrates environmental covariates with existing soils information obtained from historic soil surveys and from expert knowledge. This approach works efficiently with limited data, which is often the case in emerging economies like Kenya, while at the same time rescuing legacy data and expert knowledge that is at risk of being lost. The following environmental covariates derived from the Shuttle Radar Topographic Mission 30 m digital elevation model were selected for establishing and quantifying soil-landscape relationships: slope gradient, multiresolution valley bottom flatness index, multiresolution ridgetop flatness index, topographic position index elevation and profile curvature. These covariates were used along with existing soil information and expert knowledge to produce new raster-based maps of soil class, effective soil depth, and soil moisture storage capacity. The soil class maps predicted using clustering analysis and fuzzy logic methods showed good agreement with field observations based on the overall accuracy values. The fuzzy logic map performed slightly better (Kappa Coefficient = 0.68) than the map based on clustering analysis (Kappa Coefficient = 0.59). The soil effective depth map accuracy was better for the fuzzy logic map (R^2 = 0.56; RMSE = 11; ME = 1.1) compared to the existing soil map (R^2 = 0.34; RMSE = 27; ME = 8). The results of this study generated more detailed and improved predictions of soil classes and properties at 30 m grid resolution that will be useful for soil, crop and land use management decisions in the future.

Soil fertility, weed biomass accumulation, and corn yield response to legume cover crops (Poster)

Carson Wright and J.L. Ghezzi

Over applicaiton of pesticides is an environmental problem that's led to herbicide resistant weeds and environmental degradation. Cover crops are a popular conservation technique. Legume cover crops can offset the application of nitrogen fertilizer. When interseeded into standing row crops, certain cover crops can help reduce weed biomass accumulation and have the potential to reduce anthropogenic inputs of fertilizers and herbicides. A study site in northeastern Delaware County was utilized in a cover crop study. Study objectives included a) utilizing a winter triticale and hairy vetch cover crop mix on weed biomass accumulation and available soil nutrients; and b) Spring planted sweet corn interseeded with red clover at varying times was utilized with the objective to examine red clovers' effect on available nutrients and weed biomass accumulation as well as sweet corn yield. Mehlich 3 extraction for soil analysis included potassium, calcium, sodium, and magnesium by Mehlich 3 extraction while phosphorus was analyzed using the Strong Bray method. Organic matter was determined using the loss on ignition method. Soil nitrate and ammonium was determined using the KCl extraction method with a cadmium reduction column. Weed biomass was collected in quadrats at regular intervals using a randomized block design. Statistical variance was analyzed using ANOVA. Preliminary results indicate suppression of weed biomass, indicating reduced need for future herbicides.

I need my space! Holes and Minerals

Nelson R. Shaffer, Nannovations

Minerals make much of the natural world. Most minerals are small requiring microscopes, electron microscopes, microprobes, and such for proper study. The public knows minerals mainly by large, beautiful crystals. Large perfect crystals need space to grow well. Availability of natural holes such as vugs, solution cavities, mirolitic cavities, geodes, lava tubes, even caves provide appropriate open spaces. We shall look at holes, their kinds, origins, and implications to mineral growth and some examples of fine minerals formed within holes.

Abating Indianapolis' Combined Sewer Overflow System – An Overview of the DigIndy Tunnel System (Hot Topic)

Michael Miller, Citizens Energy Group

Citizens Energy Group is implementing a Long Term Control Plan (LTCP) to comply with a federal consent decree requiring combined sewer overflow (CSO) abatement. The DigIndy Tunnel System is the backbone of Citizens' $2B LTCP, and includes six primary components. They are; the Deep Rock Tunnel Connector Pump Station, the Deep Rock Tunnel Connector, the Eagle Creek Deep Tunnel, the White River Tunnel, the Lower Pogues Run Tunnel, the Fall Creek Tunnel and the Pleasant Run Tunnel. Each tunnel is constructed in the bedrock nearly 250 feet below ground. After more than five years of construction, two of the six key projects are nearly complete. The remainder of the system is in various phases of design and construction. Once the system is fully constructed, it will prevent up to six billion gallons of combined sewage

from entering Indianapolis' waterways annually. The intent of this presentation is to provide an in depth look into the DigIndy Tunnel System and share the benefits to our environment that the program will provide.

Microbiology & Molecular Biology

Reduction in the Early Stage of Logarithmic Growth in *Salmonella* Exposed to EDTA (Poster)

Theresa Emeli, Rajdeep Bomjan, and Daoguo Zhou

The rise in antibiotic resistant bacteria has placed pressure on the need to understand the modes of resistance as well as alternatives to combat bacteria who have evolved resistance towards commonly-used antimicrobials. Amongst the rise of antibiotic resistant bacteria, most bacteria that are gaining resistance are gram-negative bacteria, such as *E. coli,* which are of concern when treating patients in a healthcare setting. This has led to research on new antimicrobials and drug targets within bacteria which are less susceptible to resistance by bacteria. Ethylenediaminetetraacetic acid, EDTA, has been of interest due to its effect on the beginning stages of logarithmic growth in Salmonella bacteria. The purpose of this project is to understand the effects of EDTA on the growth and pathogenesis of Salmonella bacteria and understand its effect on gene expression within Salmonella bacteria. By understanding its effect on *Salmonella*, this information can be applied to other types bacteria and help combat the issue of antibiotic resistance.

Human Umbilical Vein Endothelial Cell Viability is Maintained during Long-term Exposure to ML141 (Poster)

Melissa D. Tinsley, Nathan C. Hahn, Rhashaan L. Canty, Robert E. Sammelson and Susan A. McDowell, Ball State University

Staphylococcus aureus is a leading cause of bloodstream infection, skin and soft-tissue infection, pneumonia, and infective endocarditis worldwide. *S. aureus* can reside within host cells and evade antimicrobial agents and immune defenses. CDC42, a host membrane protein, mediates *S. aureus* invasion of host cells. When CDC42 is mutated, there is decreased internalization of bacteria. Therefore, the lab examined whether ML141, an allosteric inhibitor of CDC42, would decrease invasion. We found that short term ML141 exposure decreases intracellular infection by 90%. Host cell viability is maintained during short term exposure to ML141. For the current study, we have examined whether host cell viability is maintained during long term exposure to ML141. CellTiter 96 Aqueous cell proliferation assay indicated decreased absorbance in samples treated with ML141. To confirm whether this decrease was caused by a loss of host cell adhesion or host cell viability, a propidium iodide flow cytometry assay was run. We found ML141 decreases host cell adhesion, but host cell viability is maintained. Long-term exposure to ML141 could be used to treat *S. aureus* infections by preventing the bacteria from invading host cells.

The development of CRISPR mediated genetic engineering in *Candida viswanathii* (Poster)

Elena North, Douglas Bernstein, Irene Reizman. Ball State University, Rose-Hulman Institute of Technology

Candida viswanathii is a fungal species in the *Candida* clade. It is closely related to *Candida tropicalis*, a prominent human fungal pathogen. Using the *Candida albicans* CRISPR system as a template, we have begun to develop CRISPR mediated genetic engineering in *C. viswanathii.* We have identified guide sequences in the *C. visawanathii ADE2* gene and cloned these sequences into a vector that also expresses Cas9. After generating these constructs, we cotransformed these vectors along with repair template that will introduce stop codons when incorporated into the *C. viswanathii ADE2* locus. We were able to obtain dozens of transformants and we are now testing if these transformants have incorporated the repair template.

Toward a model system to investigate fungal endophytic suppression of human pathogens in spinach (Poster)

Justin S. Golday, Lindsay Gielda, Evan Curl, Chris Weliczko, and Scott T. Bates, Purdue University Northwest

Symbiotic microbes are known to benefit both human and plant hosts by influencing metabolic processes, immune defenses, and microbial colonization. Fungal endophytes (endo = ‘within’, phyte = ‘plant’) are known to benefit plants in many ways, from stimulating plant growth to providing protection against phythopathogens, thus promoting plant health overall. Recently we isolated pleosporalean fungal endophytes in the genus *Stemphylium* from commercially available spinach plants that showed *in vitro* antimicrobial activity against pathogenic bacterial strains, including *E. coli* O157:H7. Uncharacterized secondary metabolites produced by our fungal endophytes, including *Stemphylium* PNW-2016-02, were subsequently shown to be responsible for this antimicrobial activity. Because *E. coli* O157:H7 is also known to grow endophytically within spinach, thus potentially increasing consumer risk to disease, we have focused our research efforts on developing a model system for examining *in plantae* supression of *E. coli* O157:H7 by our *Stemphylium* isolates. This spinach model system also holds the potential for examing whole phytobiome microbial interactions with the aim of understanding mechanisms that lead to pathogen supression as well as produce plant health. Toward this effort, and in preparation of future *in plantae* supression experiments,we have developed an efficient method using 96-well plates to inoculate germinating spinach plant seeds with our *Stemphylium* endophytes. We have also developed a molecular assay for confirming successful fungal endophyte inoculation in spinach plants that uses PCR with specific primers for pleosporalean fungi, amplifying the nuclear ribosomal internal transcribed spacer (ITS) region, combined with a *Stemphylium*-specific restriction enzyme digestion (*AluI* and *SnaBI*) on the ITS amplicons. Initial inoculation runs with our 96-well plate system showed spinach seed inoculation with our *Stemphylium* isolate (PNW-2016-02) persisted in the spinach plant tissue over two weeks, at which point we also observed a statistically significant enhancement of spinach plant growth.

Altering lipid droplet homeostasis affects *Coxiella burnetii* intracellular growth

Minal Mulye, Marian University College of Osteopathic Medicine, Brianne Zapata, California State University, and Stacey D. Gilk, Indiana University School of Medicine.

Coxiella burnetii is an obligate intracellular bacterial pathogen and a causative agent of culture-negative endocarditis. While *Coxiella* initially infects lung macrophages, it may disseminate and cause endocarditis up to 20 years after initial infection, which can be fatal if untreated. Even for those receiving treatment, the mortality rate is 19%. Hence, designing better treatments is crucial. To this end, it is important to determine how *Coxiella*infection leads to endocarditis. The occurrence of *Coxiella* endocarditis several years after the initial infection suggests that the bacterium survives long term by subverting host responses normally used to clear bacterial infection. To determine the host processes *Coxiella* manipulates, we performed gene expression analysis of *Coxiella*-infected macrophages which revealed differential regulation of lipid storage related genes. Further we observed that in host cells *Coxiella* induces accumulation of lipid storage organelles called lipid droplets (LDs). Interestingly, *Coxiella* has been found in LD-containing foamy macrophages in the cardiac valves of endocarditis patients. Further, this LD accumulation was dependent on the *Coxiella* Type 4B Secretion System (T4BSS), a major virulence factor that manipulates host cellular processes by secreting bacterial proteins into the host cell cytoplasm. To determine the importance of LDs during *Coxiella* infection, we assessed the effect of manipulating LD homeostasis on *Coxiella* intracellular growth. While blocking LD formation in macrophages increased *Coxiella* growth, preventing LD breakdown almost completely blocked bacterial growth suggesting LD breakdown is essential for *Coxiella*. Together these data suggest that maintenance of LD homeostasis, possibly via the *Coxiella* T4BSS, is critical for bacterial growth. Since LDs are known precursors of immune system suppressors, ongoing studies are identifying the contribution of LDs and the immune system in *Coxiella* long-term persistence in host cells.

The second messenger c-di-AMP is involved in regulating diverse cellular activities and virulence in *S. pyogenes*.

Tazin Fahmi[1], Gary Port[2] & Kyu Hong Cho

[1] Indiana State University [2]Washington University School of Medicine, St. Louis, MO

c-di-AMP has been shown to play very important role as a second messenger molecules in the regulation of physiologies and virulence in many Gram-positive bacteria including *Staphylococcus aureus, Listeria monocytogenes, Mycobacterium* spp., *Clostridium* spp. and *Streptococcus* spp. The appropriate level of intracellular c-di-AMP is necessary to carry out their functions. Diadenylate cyclase (DacA) and phosphodiesterse (PDE) enzymes regulate the level of intracellular c-di-AMP and any mutation in these two enzymes altered the downstream gene regulation processes. Our study emphasizing the roles of c-di-AMP in the cell physiologies and infections of the human pathogen *Streptococcus pyogenes*, which causes a wide variety of mild to life threatening severe diseases. *S. pyogenes* mutant strains have been created by deleting *dacA* and *PDE*

genes to observe the changes in the regulation of physiologies and virulence activities. We observed the growth of DacA,, Pde2, and GdpP, null mutant strains in different salt concentration, pH, and oxygen stressed environment. We also observe changes in biofilm formation ability, antibiotic sensitivity of all the strains. DacA null mutant exhibit higher sensitivity to salt, pH and oxygen stress environment in compare to the other mutant strains. All the mutant strains produced less biofilm except the Pde2 null mutant that produced 1.6 times high than the wild type. DacA and Pde2 null strains exhibit negative SpeB activity that is an important virulence factor of *S. pyogenes*. In conclusion, disturbance of c-di-AMP level by deleting genes involved in its biogenesis changed diverse cellular activities and expression of the virulence factor SpeB.

Identification of a critical interdomain loop structure of mouse hepatitis virus protease nsp5 (Poster)

Mansi C. Pandya, Sean M. Callahan, Megan E. Franke, Emily F. Hasik, Benjamin C. Nick, and Christopher C. Stobart, Butler University

Coronaviruses are enveloped, positive-sense single-strand RNA (+ssRNA) viruses that infect a wide range of animals including humans. Human coronaviruses are associated with diseases ranging in severity from the common cold to severe acute respiratory syndrome (SARS). Despite a significant clinical need, there remain no vaccines and limited therapeutic options for coronavirus infections. Immediately upon entry, the coronavirus genome is translated into a long polyprotein comprised of 16 nonstructural protein (nsp) domains. Nonstructural protein 5 (nsp5) is a viral protease that is required for proteolytically processing the viral polyprotein at 11 distinct sites and is essential for virus replication. Due to its importance, nsp5 remains a major target for inhibitor design. Nsp5 consists of 3 distinct structural domains with a long loop connecting domains 2 and 3. Very little is known of the function of this loop region. Site-directed mutagenesis was performed on the nsp5 interdomain loop of the coronavirus mouse hepatitis virus (MHV). We show that two amino acid residues within a horseshoe region of the loop, when mutated, result in lethality and an additional two amino acids at the base of this region when mutated to alanine permit recovery of temperature-sensitive MHV mutants. These studies provide new insight into nsp5 function and identify a new critical region of the nsp5 protease, which may be targeted for future coronavirus inhibitor design.

LAMP assay: shedding light on a technique to detect *Mycoplasma ovis* (Poster)

Kathy Johnson, Pierre Deshuillers, Joseph Camp, Jr., and Joanne B. Messick, Purdue University

Mycoplasma ovis, the causative agent of hemoplasmosis in sheep, has a worldwide distribution. Acute infection is characterized by hemolytic anemia in sheep that can be life threatening, whereas chronic infection leads to loss of body weight and ill thrift. It is believed the latter animals are the main source for spreading and maintaining infections within a herd. Thus, there is a need for a rapid, inexpensive assay to facilitate accurate detection of *M. ovis*-infected sheep and gain a better understanding of its transmission in the herd. The objectives of the present study were to develop, optimize and validate a loop-mediated isothermal amplification

(LAMP) assay targeting the dnaK gene of *M. ovis*, and to compare the performance of the assay to cPCR (conventional PCR) using 145 field samples from naturally-infected sheep previously tested by qPCR (quantitative PCR). To accomplish this, three sets of primers specifically designed to amplify the dnaK gene of *M. ovis* under isothermal conditions were used. Upon optimization of the LAMP assay reaction conditions, *M. ovis* was consistently detected in 45 minutes using a reaction temperature of 64°C. A metal ion indicator, hydroxynaphthol blue (HNB), was added prior to the reaction and allowed for easier visual detection of LAMP positive samples as indicated by a color change from violet to sky blue. The limits of detection of the LAMP and cPCR assays were 10^2 and 10^3 plasmids per reaction, respectively. A kappa coefficient of 0.754 (95% CI: 0.642 – 0.865) was obtained, indicating substantial agreement between cPCR and the LAMP assay. To the best of our knowledge, this is the first report of a LAMP assay for the detection of *M. ovis*. In conclusion, the LAMP assay provides a fast, reliable method to detect *M. ovis* using minimal equipment.

Susceptibility of respiratory syncytial virus (RSV) strains to antimicrobial peptide inactivation (Poster)

Caitlin E. Haas, Katelyn R. Castiglia, Karina Latsko, Nathan A. Junod, and Christopher C. Stobart, Butler University

Antimicrobial peptides are small proteins that play a critical role in the innate immune response. Previous studies have established an important role of antimicrobial peptides in providing defense and limiting growth of bacterial, fungal, and viral pathogens in the respiratory tract. Respiratory syncytial virus (RSV) is an upper and lower respiratory pathogen which is associated with severe disease in young infants and the elderly. Efforts to develop a vaccine for RSV have been hindered by physical and genetic instabilities, poor immunogenicity, and a legacy of vaccine-enhanced disease. Therapeutic options after onset of symptoms are limited for RSV and the host events associated with RSV clearance remain unclear. Recently, several studies have shown that a class of antimicrobial peptides called defensins and a separate antimicrobial peptide, cathelicidin (LL-37), were able to inactivate the RSV laboratory strain A2. However, these studies were limited to this laboratory strain and appreciable differences in the structure and replication among RSV strains are known to exist. In this study, we evaluated the efficacy of human defensins and LL-37 on inactivation of a panel of recombinant RSV strains that express the attachment and fusion proteins of laboratory and clinical strains of RSV. We used a combination of inactivation assays and replication assays to more clearly define the susceptibility of RSV strains to antimicrobial peptide inactivation. These studies illustrate a clear role of antimicrobial peptides in limiting RSV infection and may provide new therapeutic targets to treat active RSV infections.

The second messenger c-di-AMP regulates SpeB biogenesis and virulence in *Streptococcus pyogenes* (Poster)

Sabrina Faozia[a], Gary Port[b] and Kyu Hong Cho[a]

a Indiana State University b Washington University School of Medicine, St. Louis, MO

Cyclic di-adenosine monophosphate (c-di-AMP), an important secondary messenger molecule, is produced by human bacterial pathogen *Streptococcus pyogenes* where it is synthesized by a diadenylate cyclase DacA and degraded by phosphodiesterases (PDEs) GdpP and DacA. In this study, biogenesis of the virulence factor SpeB was investigated in the gene deletion mutant strains, ΔDacA and ΔPde2. SpeB is a secreted cysteine protease that has a major role in *S. pyogenes* pathogenesis. SpeB not only damages host proteins directly but also disturbs the host immune system by liberating streptococcal cell surface virulence factors such as M protein, protein F and C5a peptidase. Protease detection plate assay and western blot were performed to detect the production of SpeB. Quantitative PCR (reverse transcriptase real time PCR) was also done to determine the relative SpeB transcript amount in the mutants. It was demonstrated that both ΔDacA and ΔPde2 mutant strains did not show any protease activity in the protease detection plate assay and no SpeB is detected in the Western blot assay, either. Both mutants expressed insignificant amount of SpeB transcript in quantitative PCR, indicating that c-di AMP influences SpeB biogenesis in ΔDacA and ΔPde2 at the transcriptional level. Though ΔDacA and ΔPde2 are involved in the opposite reaction in c-di-AMP biogenesis, their phenotype regarding SpeB production was almost identical. Herein, another major virulence determinant, M protein of *S. pyogenes* was also investigated by quantitative PCR analysis. Both ΔDacA and ΔPde2 showed significant amount of the transcript for M protein, indicating that c-di-AMP does not regulate M protein biogenesis at least at the transcriptional level. We also examined the virulence of the mutants with a murine subcutaneous infection model where both mutant strains showed no lesion in the mouse tissue. Taken together, c-di-AMP plays an important role in *S. pyogenes* pathogenesis at least regulating SpeB biogenesis.

Deletion of the Gabapentin Receptor and VSCC Subunit a_2d_1 Impairs Bone Formation and Strength (Poster)

Madison Kelly, Karan Sharma, Christian Wright, Xin Yi, Aaron Gegg, Taylor Gorell, Megan Noonan, Alexander G. Robling, Julia Hum, Mary C. Farach-Carson, William R. Thompson, Marian University; Indiana University; University of Texas Health Science Center

Voltage sensitive calcium channels (VSCCs) influence bone structure and function. While the pore-forming (a_1) subunit of VSCCs enable Ca^{2+} influx, auxiliary subunits influence biophysical properties of the pore. $a_2d_1$binds the a_1 pore, and has a large exofacial domain. Gabapentin (GBP), an anti-epileptic and neuropathic pain drug, binds a_2d_1, impairing channel activity. GBP relieves epilepsy and neuropathic pain, yet it has detrimental skeletal effects, impairing bone formation. The function of a_2d_1 in neurons has been well-studied; however, the mechanisms through which a_2d_1 regulates bone formation *in vivo* are unknown. We examined the skeletal consequences of deleting *Cacna2d1*, the gene encoding a_2d_1. DXA analyses from 6 to 18 weeks old, showed reduced whole-body bone mineral content (BMC) in male ($p<0.0001$) and

female (p=0.0003) *Cacna2d1*$^{-/-}$ mice compared to WT controls. Whole-body bone mineral density (BMD) was also significantly reduced in both sexes (male:p<0.0001, female:p=0.014). Reductions in femoral and vertebral BMD of *Cacna2d1*$^{-/-}$ mice were significant in both sexes (p<0.001). µCT analysis of femurs and L5 vertebrae of *Cacna2d1*$^{-/-}$ mice showed ~39% reduction in trabecular bone volume (BV/TV) in male and female *Cacna2d1*$^{-/-}$mice compared to controls. Trabecular BMC was reduced in male (47.7%, p<0.05) and female (40.9%, p<0.05) KO mice. A 37.1% reduction in trabecular connectivity and a 13.43% decrease in thickness were seen in male *Cacna2d1*$^{-/-}$ mice (p<0.05). Male and female *Cacna2d1*$^{-/-}$ mice had reduced cortical BMC of 14.1% (p<0.01) and 10.4% (p<0.05) respectively. Notably, the polar moment of inertia was reduced by 33.53% in males and 19.18% in females (p<0.01). As such, a_2d_1 KO mice have impaired bone formation, structure, and strength revealing a previously unidentified role of a_2d_1 in regulation of bone content, density, and strength. Further understanding of the extracellular sensing mechanisms and intracellular signaling pathways regulated by a_2d_1 may reveal innovative approaches to combat bone pathologies.

Determining Genetic Requirements for Translocon-Associated Protein Degradation (Poster)

Cade J. Orchard (Delta High School; Ball State University), Christopher J. Indovina, Sarah M. Engle, and Eric M. Rubenstein. Ball State University

High concentrations of low-density lipoproteins (LDL) are associated with cardiovascular disease and other cholesterol-related pathologies. Statins are used to treat afflicted patients by lowering LDL levels but have been shown to be incompletely effective. Because of this, there is a demonstrated need for new methods to lower bad cholesterol levels. LDLs are synthesized at the endoplasmic reticulum (ER) membrane in hepatocytes (liver cells). During synthesis, the protein component of an LDL particle enters a translocon, a pore within the ER membrane, and stalls until lipids inside the ER bind to the protein. Under certain circumstances, an enzyme called Hrd1 targets for destruction proteins that have stalled in the ER translocon, such as the protein component of LDL particles. How Hrd1 recognizes these proteins is unknown. A genome-wide screen conducted by a former laboratory member revealed several candidate genes with potential roles in degrading translocon-associated proteins in the model organism, *Saccharomyces cerevisiae* (budding yeast). We hypothesized that a specific group of these genes functions with Hrd1 in a protein degradation pathway. Using small-scale growth assays that indirectly report on abundance of a Hrd1 substrate, we identified genes that are integral to this process. Our results suggest that a gene encoding the protein chaperone, Ydj1, and a gene encoding an exomannosidase of the ER, Mnl1, is required. Because protein degradation processes are highly similar in yeast and humans, genes found to be involved in this pathway may represent potential therapeutic targets for elevated cholesterol and could lead to more effective treatments.

Enzymatic regulation of G-quadruplex structures in *C9orf72* ALS (Poster)

Peter Beerbower and Philip Smaldino, Ball State University

Higher-order nucleic acid structures called G-quadruplexes (G4 structures) can form in regions of both DNA and RNA that contain high numbers of localized guanine (G). There are ~750,000 possible G4-forming sequences in the human genome. A G-rich hexanucleotide repeat expansion in the first intron of *C9orf72* (*C9*) has been found to be the primary cause of inherited amyloid lateral sclerosis (ALS), a neurodegenerative disease causing motor neuron degeneration. The accumulation of excessive G-quadruplex DNA and RNA derived from the repeat is a defining characteristic of C9 ALS. We propose that the human enzyme G4 Resolvase 1 (G4R1), the enzyme responsible for the majority of G-quadruplex resolving activity in HeLa cells, is likely involved in C9 ALS due to the high prevalence of G4 structures. Preliminary data suggests that G4R1 protein levels are upregulated in C9 ALS patient cells. We hypothesize that G4R1 functions by unwinding G-quadruplexes forming in the DNA, which, if left unwound, inhibit C9 gene transcription. Furthermore, we hypothesize that by upregulating C9 transcription, G4R1 increases the levels of G-rich *C9orf72* transcripts. To determine this, I have begun quantifying the levels of *C9orf72* transcripts and toxic RNA foci derived from *C9orf72* transcripts relative to G4R1 levels.

Identification of Functions of Pseudouridine Synthase 7 in *Candida albicans* (Poster)

Ethan Pickerill and Doug Bernstein, Ball State University

Candida albicans is the most prevalent human fungal pathogen. Likewise *Candida spp.* are the fourth most commonly isolated nosocomial pathogens, and these infections range in severity from oral or vaginal candidiasis to severe systemic infections While antifungal drugs exist, courses of treatment are lengthy and antibiotic resistant isolates are prevalent, highlighting the need for new therapeutic targets. In order to identify new drug targets we must better understand what differentiates fungi from humans at the molecular level. This research investigates the effects of CRISPR mediated deletion of Pseudouridine synthase 7 in *C. albicans*. We find *pus7/pus7* has similar initial fluconazole resistance to wild type, but *pus7/pus7* exhibits greater trailing growth. Additionally *pus7/pus7* has defects in filamentation, grows slower than wild type *C. albicans*, and has decreased virulence in the *Galleria mellonella* infection model. Comparison of *pus7/pus7* and wild type transcription via RNA sequencing has identified numerous changes in expression. Notably, RNA from *pus7/pus7* grown at 37^{o}C has increased unprocessed RNA indicating defective rRNA processing, and changes in oligopeptide transporter expression and other cell surface protein expression levels have been observed as well. In the future we will identify *C. albicans* Pus7 RNA substrates via primer extension analysis, and will further evaluate *pus7/pus7* virulence by assessing cell surface hydrophobicity.

Manipulation of drug lipophilicity as a strategy to combat vancomycin-resistant *Enterococci* (Poster)

Kevin Hammond, Adam Lesniak, Victoria Yagodinski, and Colleen L. Doçi, Marian University Indianapolis

Increasing bacterial resistance in clinical settings has recently prompted urgent investigation into finding antibiotic compounds that inhibit novel pathways in bacteria. Among these resistant bacteria, vancomycin-resistant enterococcus (VRE) is one of the most prominent, having developed resistance to the commonly-used vancomycin. The discovery of novel antibiotics has recently slowed while antibiotic resistance has accelerated, creating a healthcare crisis for treating bacterial infections. Rifampicin has the potential to be one such novel compound. Rifampicin can effectively in treat resistant enterococcal infections, particularly in combination with other antibiotics. Rifampicin acts by inhibiting bacterial RNA polymerase, meaning it must cross the cell membrane to be effective. This suggests that strategies that help effectively target this drug may improve its efficacy. While not feasible for many compounds, increasing the lipophilicity of some drugs may improve their membrane permeability. This research aims to increase the sensitivity of antibiotic-resistant bacteria to rifampicin through drug lipophilicity modulation. We have shown the feasibility of this approach by demonstrating a sensitivity increase in *E. coli* to tunicamycin. However, tunicamycin does not show significant resistance patterns in bacteria, and *E. coli* is sensitive to most antibiotics. Thus, a study validating these preliminary results in a relevant model such as VRE is necessary to determine if lipophilicity modification is a viable method of increasing antibiotic efficacy. To test this hypothesis, we first optimized a standard turbidity assay to quantitatively measure its correlation with overall culture viability. We tested the effect of varying concentrations of vancomycin-inoculated cultures, rifampicin-inoculated cell cultures, and liposome-encapsulated rifampicin-inoculated cultures on VRE viability. We hypothesized that the average turbidity value of VRE cultures inoculated with liposome-encapsulated rifampicin would be significantly lower than that of VRE cultures inoculated with vancomycin, indicating a more effective antibiotic treatment.

ML141 protects against *Staphylococcus aureus* invasion and sustains macrophage immune response (Poster)

Nathan C. Hahn, Courtney D. Klug, Robert E. Sammelson, Heather A. Bruns, Susan A. McDowell, Ball State University

Persistent infections by *Staphylococcus aureus* are a serious global threat despite availability of antimicrobial treatments. *S. aureus* can persist despite treatment by invading deep tissues exposed during invasive procedures. In past work, we found that ML141, a novel small molecule inhibitor for host cell CDC42, limits *S. aureus* invasion of host cells. Though ML141 limits host cell invasion, Rho-GTPases such as CDC42 serve a variety of functions within the host cell, including regulation of proinflammatory cytokines. Currently the effect of ML141 on the regulation of proinflammatory cytokine and other elements of the immune response is unknown. In this current work, we demonstrate that ML141 inhibits *S. aureus* invasion without disrupting immune response in macrophage. Our findings demonstrate the potential of ML141 to provide a novel therapeutic for decreasing intracellular *S. aureus* without disrupting immune response.

PCR-based differentiation of *Culex pipiens* pipiens and *Culex pipiens* quinquefasciatus (Poster)

Monique T. Huynh, Glené Mynhardt, and Pamela R. Pretorius, Hanover College

Culex pipiens pipiens and *Culex pipiens* quinquefasciatus are two mosquito forms of interest because they are both potential West Nile virus vectors. Differentiation between *C. pipiens* pipiens and *C. pipiens* quinquefasciatus is difficult due to both forms being morphologically identical. Using molecular techniques, the cytochrome oxidase I (*COI*, mitochondrial gene) and angiotensin-converting enzyme 2 (*ACE2*, nuclear gene) genes can be targeted to differentiate between the two forms of *C. pipiens* species. The goal was to establish a protocol to isolate and differentiate between previously identified genetic variations in *COI* and *ACE2 in C. pipiens* pipiens and *C. pipiens* quinquefasciatus. Both forms of *C. pipiens* were collected using a CDC Gravid Trap Model 1712 placed near the Hanover Water Treatment Plant. Of the 100 specimen collected, 79 were all identified down to the *Culex* genus using morphology. Molecular identification of the two forms is ongoing.

Phenotypic consequences of *Candida albicans* PUS4 overexpression

Caleb Embree, Rebecca Kurtz Paulette, and Douglas Bernstein, Ball State University

In *S. cerevisiae*, Pus4 modifies uridine to pseudouridine in cytoplasmic and mitochondrial tRNAs at residue 55 and also modifies some mRNAs. C. albicans Pus4 is thought to have analogous functions but these have not been investigated. We aim to verify the function of Pus4 in *C. albicans* and demonstrate how overexpression leads to differences in drug resistance and virulence.

Restriction digest screening of *Candida albicans* transformants (Poster)

Douglas Bernstein, **Ben Evans**, Olivia Smith, Ball State University

Introducing point mutations is an important tool in assessing gene functionality. CRISPR mediated genome mutation has allowed for a more efficient transfer of the desired mutation into a wide range of model organisms. Traditionally standard genomic sequencing is used to scan for successful mutations, however, when working with model organisms such as *Candida albicans,* transformation efficiency is highly variable, making sequencing costly and time consuming. Restriction screening is an efficient tool to identify transformants using PCR and restriction enzyme digest. Using CRISPR we introduced a restriction site coding for desired point mutations into *UME6*, a known regulator of *C. albicans* filamentation. Repair templates coding for different restriction sites were not equally successful in inserting at the cleavage site created by Cas9. Restriction screening was used as fast and cost-efficient way to identify for successful mutations. Mutated isolates were compared with wildtype *Candia* and found to have clear defects in filamentation. Our data suggests restriction digest screening is an efficient method of screening for point mutations.

Rett Syndrome- A Beacon of Hope for Autism (Hot Topic)

Daniel Tarquino, Rett Syndrome Clinic, Emory University Hospital

Abstract coming soon.

The effect of *PUS5* deletion on mitochondrial protein expression in *C. albicans* and *S. cerevisiae*

Allyson R. Morris and Douglas A. Bernstein, Ball State University

While RNA is made of only 4 bases, these bases can be modified in over 100 distinct ways. These modifications play critical roles in modulating RNA function. Pseudouridine is the most common modified nucleoside, and is found in all kingdoms of life. However, the role of pseudouridylation in translation and RNA function is not well understood. Pseudouridylation is found at dozens of sites in Eukaryotic cytoplasmic rRNA and cytoplasmic ribosomes translate thousands of proteins. As such, it is challenging to study the effects individual sites of pseudouridylation have on translation. In contrast, in fungi, mitochondrial ribosomes translate only eight genes encoded by the mitochondrial genome and mitochondrial rRNA contains only one highly conserved pseudouridine, which is made by the pseudouridine synthase Pus5. I find *Saccharomyces cerevisiae pus5Δ* deletion is more sensitive to drugs that inhibit oxidative phosphorylation such as oligomycin, suggesting they have a defect in mitochondrial function. I hypothesize Pus5 mediated pseudouridylation is important for mitochondrial protein expression. I will use mass spectrometry to determine if mitochondrial rRNA pseudouridylation is required for wild type mitochondrial gene translation. Furthermore, from this mass spectrometry data, I will determine if *C. albicans* uses an alternative mitochondrial genetic code. Investigation of this highly conserved RNA modification will lead to a better understanding of how defects in pseudouridylation lead to human disease and could lead to the identification of novel antifungal drug targets.

Phylogenetic Diversity of Rhizosphere Bacteria of Amur Honeysuckle (*Lonicera maackii*) at the Nina Mason Pulliam EcoLab as Determined by 16S rRNA Gene Sequence Analysis

Azeem Ahmad, Marian University

The Nina Mason Pulliam Ecolab (NMPE) is a 55-acre ecological research laboratory located on the campus of Marian University in Indianapolis. Once a very diverse prairie land with many animal and plant species, Ecolab has suffered many environmental stresses and only a few healthy remnants of the original prairie remain. The prairie restoration efforts at the Ecolab have been ongoing for many decades but major hindrance is posed by non-native plant species which may outcompete and crowd native species reducing biodiversity. There is substantial concern that the incidence and severity of these non-native or invasive plant species has increased in the last few decades as the result of environmental changes at the local and global scale. Indiana has more than 2,000 species of vascular plants, approximately 25 percent of these are non-native to Indiana. As a component of prairie ecosystem, bacteria play a unique role in regulating plant growth, mineralizing nutritive elements, increasing and maintaining soil fertility, converting energy forms and cycling materials. The established invasive plants can alter

soil biogeochemistry and may affect these rhizosphere bacterial communities for shifts in cycling of essential nutrients advantageous for invasive plant composition, diversity, and succession within a community. We plan to investigate compositional relationships of bacterial communities in the rhizosphere of invasive plant, Amur honeysuckle (*Lonicera maackii*) at NMPE. Through 16S ribosomal RNA gene sequencing and RFLP (restriction fragment length polymorphism), we will characterize total bacterial types and their relative abundance. The role and importance of these bacteria in establishment of Amur honeysuckle is still unknown and available data is not conclusive. An improved understanding of the rhizosphere community could allow better understanding of these important interactions. Preliminary data from our laboratory suggests that specific bacterial phyla inhabit honeysuckle rhizosphere that may be implicated in the establishment of soil conditions conducive for Amur honeysuckle growth and establishments. Our research on the function and diversity of bacteria at Ecolab will promote sustainable utilization of microorganism in prairie restoration efforts on a larger scale.

Optimization of PCR methods for identifying HF183 in water (Poster)

Christain Chauret and **Oluwatimilehin Soyoola**

Bactereroides fragilis is an anaerobic bacterium that is normally found in human intestine tracks. *Bactereroides fragilis* causes endogenous infections that can be found in all parts of the body such as chest, soft tissue, and the central nervous system. *Bactereroides fragilis* has the most antibiotic resistance mechanisms and highest antibiotic resistance rates among all anaerobic pathogens. The host specific marker HF 183 marker from *Bacteroides fragilis* was studied as a potential indicator of fecal pollution in water. In this study, the goal was to optimize two Real-Time PCR methods to detect the DNA sequence of interest. One method utilized SYBR- Green, whereas the other method utilized a TaqMan® asssay. In our experiments using the TaqMan® assay, several dilutions of untreated municipal wastewater (such as 1/100, 1/500, 1/10,000 and 1/5,000) were tested to assess the sensitivity of the method. Detection of the HF 183 DNA marker was possible even with highly diluted wastewater samples. It was concluded that the TaqMan® assay was more sensitive than the SYBR-Green assay; it gave the best result because of its ability to amplify and detect DNA from more diluted wastewater samples. In the future, we plan to test this assay with various samples from natural waters as well as from treated and untreated wastewater. This will give us the opportunity to evaluate the sensitivity of **TaqMan® assay.**

The End of the Antibiotic Era: Is Resistance Inevitable? (Hot Topic)

Jenny Fisher, Indiana University Northwest

Antibiotic resistance has become a global health crisis. “Superbugs” are occurring more frequently, and no new classes of antibiotics have been developed in over 25 years. Some scientists are even predicting an end to the era of antibiotic treatment. Where do these superbugs come from, and how do they become resistant to such a wide array of antibiotics? Industrial animal farms and health care facilities are the two major sources of antibiotics and resistant organisms to runoff and wastewater. Genetic and cultural evidence of resistant bacteria is often observed in water resources downstream from animal feeding operations and

wastewater treatment facilities, indicating that these organisms are surviving in the environment. How did we get here, and can we do anything to fix the problem? This lecture will explore how humans have contributed to widespread antibiotic resistance, the different mechanisms by which bacteria acquire resistance, and the current state of the ever-growing global “resistome.”

Examination of the persistence of secondarily transferred DNA on regularly used knives

Erica Cantor, **Jessica Miller**, Kayla Mueller and Krista Latham, University of Indianapolis
Cynthia Cale and Gay Bush, Strand Analytical Laboratories

This study tests for the persistence of transfer DNA over seven days on objects that have both important daily functions and potential evidentiary importance. Participants were assigned a sterile knife and instructed to handle it twice a day for two days, simulating daily use. The knives were stored in evidence bags between handling events to prevent cross-contamination. On the third day, participants shook hands with another person before handling their knives, which were then stabbed several times into styrofoam cups. Before each handling event, participants were instructed to come in contact with the entire knife handle, which was divided into three sections using a permanent marker. The first area was swabbed an hour after the stabbing simulation, the second area after one day, and the third area after one week. The knives were stored in evidence bags between sampling events. The DNA was purified from the swabs, quantified and amplified for DNA profiling. The specific combination of analytical and stochastic threshold used for interpretation was dependent upon the quantity and quality of the DNA samples. The samples that were collected after one day were lost during an equipment malfunction, so results are based on comparisons of DNA quantity and profile quality between one hour and one week samples. In the samples collected after one hour, 70% showed mixed profiles, with DNA from both the handler and their partner present. In the samples collected after one week, 90% showed mixed profiles with DNA from both the handler and their partner present. Additionally extraneous alleles that could not be contributed to either person were detected in over half of the total samples. This study builds upon previous research and demonstrates the persistence of transfer DNA profiles that were obtained from evidentiary items using next generation DNA kit technologies.

Utilizing human toenail clippings for use in genetic studies and forensic identification

Jonah W.P. Stone, Haley E. Rock, Rachel M. Kreher, and Krista E. Latham, University of Indianapolis; Cynthia Cale and Gay Bush, Strand Analytical Laboratories

A variety of biological sources, such as blood and bone, have commonly been utilized for genetic investigations. However, the methods for procuring these samples can be both time consuming and invasive to the human subject. Clipping human toenails is a less invasive method of obtaining genetic material and has the potential to be utilized in large-scale population studies as well as in forensic identifications. This study aims to test the ability to generate informative DNA profiles from toenail material. Participants were instructed to provide toenail clippings, which could have originated from any toe, to the analysts. The DNA extraction process started by immersing 15mg of toenail clippings in 400µl of Qiagen Buffer ATL and 10µl proteinase K solution. The toenails were incubated in the solution for 5.5 hours with the

expectation that the tissue would dissolve. However, the toenails did not dissolve in solution, so an additional 20µl of proteinase K was added and the samples incubated for an additional 6 hours. Again, the toenails did not dissolve so the liquid surrounding the tissue was removed and extracted with a Qiagen DNA mini kit as per manufacturer's instructions. The DNA was then quantified and amplified for DNA profiling. The specific combination of analytical and stochastic thresholds used for interpretation was dependent upon the quantity and quality of the DNA samples. The amount of DNA obtained from each sample ranged from 7.28ng to 228.48ng. Each sample produced a single source DNA profile that was consistent with the contributor. The present study suggests that human toenail clippings can be a reliable source of genetic material for DNA profiling and forensic identification. When compared to blood collection, clipping toenails is a less invasive process for collecting DNA, and a less destructive method than collecting DNA from skeletonized remains.

Effects of biofertilizer and amino acids on nitrogen and the microbial community in soil (Poster)

Carlena Crawford, Jordan M. Marshall, and Tanya Soule, Indiana University-Purdue University, Fort Wayne

Rhizosphere microorganisms are able to change soil properties in various ways, including available nitrogen, pH, and organic carbon contents. Certain microorganisms in the soil are known to fix nitrogen and many microbes can also process amino acids to yield usable nitrogen sources for plants. Biofertilizers are microbial broths applied to soil which contain microorganisms with many useful attributes, such as the ones mentioned above, to aid in plant health and growth. This study examined the effect of a biofertilizer (Environoc 401) and an amino acid fertilizer (Micronized N) both in conjunction and alone against untreated controls on greenhouse-grown tomato plants and soil health. Plant biomass, chlorophyll content, and percent nitrogen was measured, as well as the soil pH and soil nitrogen content. Furthermore, the abundance of *Bacillus amyloliquefaciens* and *Pseudomonas taiwanensis* in the soil were quantified temporally throughout the duration of the 12-week experiment and the activity of the soil microbial community on common carbon substrates was measured. No significant differences were found among treatment groups concerning plant health except that treated plants yielded less percent nitrogen in leaf tissue than untreated controls. Many differences were also found among treatment groups when analyzing carbon substrates used by soil microbes, including l-arginine, l-asparagine, tween 40, l-serine, a-D-lactose, D,L-a-glycerol phosphate, and putrescine ($p \leq 0.05$). Overall the abundance of *B. amyloliquefaciens* was generally constant throughout the study, although there was some variation between treatment groups, while *P. taiwanensis* concentrations mostly increased over the course of the experiment. This study is the first to compare changes in plant health and the soil microbial community following the use of a biofertilizer and amino acid fertilizer in a controlled greenhouse setting. Future studies will examine ways to improve overall plant and soil health using different combinations of treatments on a variety of plants.

Studies of Growth Re-initiation Following HipA-induced Growth Arrest in *E. coli* (Poster)

Sanya Hatimi and **Hisako Masuda**, Indiana University Kokomo

Bacteria regulate their growth by utilizing a number of Toxin-Antitoxin (TA) systems. Different toxins inhibit a variety of essential cellular processes, such as translation, transcription and cell division. In normally growing cells, antitoxins form tight complexes with toxins, thus regulating the toxin's deleterious effects. Under stress conditions, antitoxins are preferentially degraded, releasing free toxins to block essential cellular functions. Once the stress is removed, cells resume their growth by once more forming TA complexes.
HipA is a toxin of an *E. coli* HipBA TA system, which has been proposed to inhibit the translational elongation factor, EF-Tu. Ectopic overexpression of HipA immediately halts cellular growth. HipA is known as a persistence factor, as its expression produces multidrug tolerant cells. These persisters are different from resistant mutants in that they are phenotypic variants of the wildtype. HipA-induced persisters are shown to resume their growth unlike other TA-toxin induced growth arrests. We will discuss the variations in the patterns of restarting growth following toxin overexpression after repeated induction of plasmid-encoded HipA, seen in our high throughput measurements.

Visual Motor Response of a Transgenic Retinitis Pigmentosa Zebrafish Model

Logan Ganzen, Purdue University; Chi Pui Pang, Chinese University of Hong Kong; Mingzhi Zhang, Shantou University; Motokazu Tsujikawa, Osaka University Graduate School of Medicine, Yuk Fai Leung, Purdue University

Purpose: Retinitis Pigmentosa (RP) affects approximately 1 in 4000 individuals globally, and there are currently no effective treatment options available. To identify new drugs, we optimized a visual-behaviour assay, termed visual-motor response (VMR), around a transgenic zebrafish carrying a truncated human rhodopsin transgene (Tg(rho:Hsa.RH1_Q344X)). The Q344X larvae experiences significant rod degeneration by 7 days post-fertilization (dpf).

Methods: To assess the vision of the Q344X zebrafish, the VMR assay was run under a dim-light condition based on recorded rod b-waves in larval fish and the minimum cone activation threshold in mice. Specifically, Q344X and control larvae at 7dpf were placed into a 96-well plate and acclimated to a dim-light source (1.802e-05 μW/cm2 at 500nm) for 1 hour. The VMR was tracked and quantified during light offset. The total distance travelled was averaged and analyzed at one-second post-stimulus. Retinas were dissected from Q344X and control larvae and whole-mounted to validate the rod degeneration in the Q344X model.

Results: We found that the Q344X larvae displayed an attenuated VMR (0.121 ± 0.041cm) to the dim-light offset as compared to the control larvae (0.2751 ± 0.038cm). Analysis of whole-mounted retinae indicated significant rod degeneration at 7dpf compared with controls (Control: 87 rods/retina, Q344X: 9.3 rods/retina, Welch's Two-Sample T-test p-value=1.4e4,). As the only apparent difference between the two groups of larvae is significant rod degeneration, it can be concluded that the behavioral phenotype was a result of the degeneration.

Conclusions: These results suggest that the attenuated Q344X VMR is a result of the rod degeneration. This behavioral phenotype can be utilized to screen chemical libraries to identify compounds that ameliorate the rod degeneration. Compounds that prevent degeneration are expected to result in a significant increase in VMR in response to the dim-light stimulus.

Physics & Astronomy

Charge Transport through a Double-Ring Molecule with Multiple Outputs

Eric Hedin, Robin Klause, and Yong Joe, Ball State University

Electron transport through a double-ring quantum dot structure, in the form of a naphthalene molecule, is modeled using a tight-binding approximation to the Schrodinger Equation. Transmission properties are analyzed in terms of their dependence upon a choice of two input connection sites and four output connection sites. Interference effects between the propagating electron waves in the two rings produce markedly different transmission features at different outputs. In addition, under certain conditions, the output at one site is shown to depend sensitively upon the level of coupling to the output lead at another site. This effect produces a conduction sensor at the nanoscale level. An external magnetic field applied perpendicularly to the plane of the rings is shown to modify the outputs in a specific manner through the Aharonov-Bohm effect.

Harnessing Spin Crossover Phenomena

Aaron Mosey and Ruihua Cheng, Indiana University-Purdue University Indianapolis

Functional molecules with lockable spin states provide a promising platform for material design in the post-Moore's era. The spin crossover complex [Fe{$H_2(pz)_2$}$_2$ (bipy)] (pz=pyrazol-1-yl, bipy=2,2'-bipyridine) which exhibits two distinct spin states, and thermodynamically stable behavior at room temperatures, may be switched between the S=0 low spin and S=2 high spin state by an external stimulation. [Fe{$H_2(pz)_2$}$_2$ (bipy)] has also been shown to couple to local electric fields and ferroelectric substrates. This opens the door to many novel spintronic applications. We examine thin films of [Fe{$H_2(pz)_2$}$_2$ (bipy)] which were fabricated using high vacuum thermal evaporation. The samples are characterized by atomic force microscopy and scanning tunneling microscopy. Conductivity as a function of temperature is investigated to identify distinct high and low spin state switching. Future works involving spintronic devices are outlined.

Magnetic Switching in a Double-Ring Molecule with Multiple Outputs

Robin Klause, Eric Hedin, and Yong Joe, Ball State University

A double ring structure formed of ten quantum dots (QDs) is modeled using a tight-binding computational technique, in order to calculate the electron transport through the device. The model incorporates two alternative sites for connections to input leads, and four output leads connected to different QD sites. System parameters can be varied to show the calculated transmission through any combination of the inputs and multiple outputs. An external magnetic flux through the rings is shown to modify the outputs as a result of the Aharonov-Bohm effect. Certain levels of magnetic flux create sharp spikes in the transmission curves at selected electron energy values. In addition, a change in the magnetic field causes large variations in

transmission probability at the output sites, producing a magnetic switching mechanism.
This work is partially supported by Indiana Academy of Science.

Measuring the Resistance of the YBCO Superconductor (Poster)

Oscar Matter, Jacob Millspaw, and Mark Masters, Indiana University-Purdue University Fort Wayne

Yttrium barium copper oxide (YBCO) is a commonly studied type-II superconductive material. Measuring its resistance can depend on how the superconductor was created or the technique used to measure its resistance. This project explores various techniques to measure the resistance of a YBCO samples.

Multi-Band Ensemble Phometry of the Eclipsing Binary Star NSVS 4161544 (Poster)

Tyler J. Redfern and Robert C. Berrington, Ball State University

We report new multi-band ensemble aperture photometry for the Northern Sky Variability Survey (NSVS) eclipsing star candidate NSVS 4161544. All multi-band images were taken by the Ball State University Observatory 0.5-meter telescope in the Johnson B and V, and Cousins R band passes. All images were reduced using the ccdred image reduction package in the Image Reduction Analysis Facility (IRAF) software suite. Ensemble aperture photometry was performed with the AstroImageJ (AIJ) software package. Measured light curves are presented. The measured light curves are analyzed by the physics of eclipsing binaries (PHOEBE) software package, and best fit orbital parameters and stellar models are reported.

Non-magnetic multilayered thin films with net spin for hypothetical long-range forces study (Poster)

Saeed Yazdani, Joseph Soruco, Aaron Mosey, and Ruihua Cheng, Indiana University Purdue University Indianapolis

In this investigation, we try to develop engineered magnetic materials with certain magnetic ordering which possesses a net spin alignment without a net magnetization. We designed and fabricated magnetic multilayer thin films on silicon substrate. Ferromagnetic thin films in our samples are rare earth $Nd_2Fe_{14}B$ and transition metal Fe separated by a thin film of non-magnetic material (Ti). The magnetization of the ferromagnetic layers which are separated by spacer layers, are coupled by electrons in spacer layer. To have a multilayer structure with nonzero spin and without magnetization, a transition metal ferromagnetic layer and a rare earth ferromagnetic layer with antiferromagnetic alignment between them is needed. The crystalline properties of the multilayer structure is characterized using X-Ray diffractometer (XRD) and the magnetic properties are studied using magnetometry measurements.

Substructure in Galaxy Cluster Abell 154 (Poster)

Wes Tobin, Indiana University East; and Robert Berrington, Ball State University

Galaxy clusters, the largest gravitationally bound structures in the universe, grow through hierarchical merging of smaller clusters to form large clusters. Cluster interactions constrain and quantify current knowledge in dark matter and dark energy, which provides a basis for the large-scale structure of the universe. In this way, actively merging clusters appear as multiple relaxed systems superimposed in position and velocity space. The structure of one galaxy cluster, Abell 154, is presented, including 176 new radial velocity measurements. The cluster exhibits a clearly defined foreground group in addition to the main group of galaxies, which is confirmed by X-ray data from the literature. The main group of galaxies also presents with a velocity distribution that is not fully consistent with a relaxed state. Statistical tests and gravitational binding tests are used to identify and analyze subclustering as well as to estimate mass and gravitational binding.

The Neutron Star Merger GW170817 (Hot Topic)

Patrick M Motl, Indiana University Kokomo

On August 17, 2017 gravitational waves were observed through the combination of the LIGO and VIRGO gravitational wave observatories for only the second time. Previous gravitational wave signals were seen only by LIGO. Shortly afterwards, a brief pulse of gamma rays – a gamma ray burst - was seen by the Fermi and INTEGRAL observatories. These coincident detections were quickly analyzed, and their importance became manifest. Unlike previous gravitational wave transients, this was a merger involving neutron stars not just black holes; with matter to radiate electromagnetic signals a hunt began by astronomers from around the world to locate the source on the sky. Within hours, a transient source was found in the relatively nearby galaxy NGC 4993 and astronomical observatories from around the world and several facilities in orbit focused in on the post-merger remnant. GW170817 has helped to answer many questions including firmly connecting neutron star mergers with gamma ray bursts, the origin of kilonovae from these mergers and the role of such mergers in the rapid neutron capture process (r process) that fills in the periodic table of elements. As is often the case in science, this discovery has raised at least as many questions as it has answered, and I will conclude by highlighting some of these outstanding questions.

The Schönberg – Chandrasekhar Instability in Stars with a Core – Envelope Structure

Patrick M Motl, Indiana University Kokomo

We will discuss an instability in stellar structures that have a bipolytropic structure where the core has a polytropic index of 5 and the envelope has a polytropic index of 1 that was explored originally by Eggleton et al. In the original work of Schönberg and Chandrasekhar, they found that a star with an isothermal core and an n = 3/2 envelope has a limiting fraction of the mass of the star that can be supported in its core. For the analogous situation of a (5,1) core - envelope structure, we explore whether the instability operates on a dynamical time scale.

3D Printed Instruments as an Exploration of Acoustics (Poster)

Justin Yoder, Panayioti Panayi, and Mark Masters, Indiana University Purdue University Fort Wayne

We explore using 3-D printing as a manufacturing process for clarinet reeds and trumpet mouth pieces. For the reeds we control stiffness and density with design and compare the printed reeds with natural reeds. For the mouth pieces we explore the impact of shape and density on sound. We examine the sound produced using Fourier Transforms as well as the performer's experience.

Charge transport through tilted double-stranded DNA molecules (Poster)

Elijah Halliwell, Eric Hedin, and Yong Joe, Ball State University

We study quantum mechanical electron transport along the long axis of the DNA molecule in an advanced two-dimensional tight-binding model, considering hopping integrals for the next nearest-neighbors and implementing a strain-dependent DNA helix conformation in conjunction with the theories of Slater-Koster and linear elasticity. The transport properties of a 30 base-pair ds-DNA molecule tilted with respect to the inter-contact electric field direction with a mechanical strain are investigated. Specifically, we present contour plots of single electron transmission spectra and nonlinear current-voltage (I-V) characteristics as functions of tilted angles and source-drain voltage for both selected electron energy and percentage strains. The observed negative differential resistance in the I-V curve is characterized by a peak-to-valley ratio (PVR). We show that a high value of PVR is achieved as either percentage strains or tilted angles increase. This higher value of PVR for an I-V curve implies a greater ability for the realization of potential applications such as logic devices and low-power memory.
This work is funded by Indiana Academy of Science.

Plant Systematics & Biodiversity

Results of the 2017 Red-Tail Land Conservancy Biodiversity Survey in East-Central Indiana

Donald G. Ruch, Ball State University

On June 10th – 11th, 2017, a biodiversity survey (also known as a bioblitz) was in east-central Indiana on two properties owned or maintained by the Red-Tail Land Conservancy. The two sites were the White River Woods (WRW), a 117 acre nature preserve located along the White River in Delaware County, and McVey Memorial Forest (MMF), 249 acre forest located along the Mississinewa River in Randolph County. Over 70 scientists and volunteers working on 19 different taxonomic teams have reported 1079 taxa to date. Taxonomic teams included ants, aquatic macroinvertebrates, bats, bees, beetles, birds, butterflies, odonates (dragonflies & damselflies), fish, freshwater mussels, herpetofauna, moths, mushrooms, bryophytes, singing insects, small mammals, snail-killing flies, spiders, and vascular plants. Many Delaware and Randolph County records have been reported. The results for each group will be presented. As an example, from the vascular plant team 476 plant taxa were reported (405 from MMF and 289 from WRW; 218 taxa occurred at both sites, 71 only at WRW, and 187 only at MMR).

How to Use the Consortium of Midwest Herbaria Data Portal in Research and Teaching (Workshop)

Paul E. Rothrock and **Eric B. Knox**, Indiana University-Bloomington

The Consortium of Midwest Herbaria (CMH) data portal currently holds over 181,000 Indiana vascular plant specimen records, including 76,990 specimens from the Indiana University Herbarium and 43,301 records from the Butler University Herbarium. This workshop will introduce participants to the features of the CMH data portal, with tips on how to search and download botanical information for a range of disciplines from plant systematics to community ecology. The CMH data portal also has online plant identification tools that can be used for teaching a broad range of audiences, from primary school through advanced college courses. The taxonomic structure of the data portal includes most of the old names previously used for Indiana plant species, so you do not need to be a professional botanist to use the site. The portal provides an up-to-date inventory of Indiana's flora, along with the species descriptions from Gleason & Cronquist's (1991) *Manual of Vascular Plants* and the species observations from Deam's (1940) *Flora of Indiana*. The CMH site can host floristic inventories from individual conservation areas, and the National Park Service has already posted a species list for Indiana Dunes National Lakeshore. Note: Participants should bring a fully charged laptop with wifi capability in order to access the CMH data portal.

Post-glacial colonization of the Midwest by *Lobelia siphilitica*

Eric B. Knox, Indiana University-Bloomington, Andrea L. Case, Kent State University, Hannah Appiah-Madson, Northeastern University, Christina M. Caruso, University of Guelph

Plastid genomes are typically inherited uniparentally (usually maternally) and do not recombine, which makes plastid DNA-based phylogenetic estimates essentially matrilineal gene genealogies. DNA sequencing of *Lobelia siphilitica* collected near Yellowwood State Forest (Brown County, IN) revealed an unusual situation in which seed collected from a single plant (which are minimally maternal half-siblings) grew into plants that possessed two markedly different plastid haplotypes that were equally divergent from the inferred ancestral DNA sequence. Genome sequencing demonstrates that the western variety *ludoviciana* is sister to the eastern var. *siphilitica*, and a sample of the named hybrid *L. ´ speciosa* has var. *siphilitica* as the maternal parent and *L. cardinalis* as the paternal parent. The most rapidly evolving region is in and around a foreign gene of unknown function (ORF262) inserted between *trnQ* and *rps16*, and 580 plants from 86 Midwestern populations have been genotyped for this region. A 49-bp minisatellite repeat array upstream of ORF262 is hypervariable and 121 haplotypes have been discovered. Phylogenetic analysis demonstrates that multiple lineages survived the glacial maxima, and biogeographic analysis indicates that most terminal variation is associated with post-glacial colonization. Long-distance seed dispersal has been common during the past 10,000 years, but individual populations tend to be inbred with one or a few haplotypes that can be quite different from nearby populations.

Efforts to Conserve the Indiana Genotype of Heart-leaved Plantain (*Plantago cordata*) in Nature

Michael A. Homoya, Indiana DNR Division of Nature Preserves

Heart-leaved plantain (*Plantago cordata*) is a critically endangered vascular plant in Indiana and throughout much of its natural range. With the exception of Missouri, where its conservation status is considered secure, it is either rare, threatened, endangered, or extirpated in all of the states and the one Canadian province within its range. It was thought to be extirpated in Indiana until the 1989 rediscovery of a small population in Whitley County. That population's numbers fluctuated in subsequent years until in 2013 only one plant remained. In June 2016, approximately 50 seeds were collected from that individual and grown into 32 seedlings in a nursery. In September 2016, 21 of those seedlings were introduced into a geographically appropriate habitat considered suitable to support the species' growth requirements. In May 2017 all were accounted for and growing vigorously, with 11 of them flowering and fruiting. The population will be monitored for survival and success of recruitment in upcoming years.

Filling in the Gaps: The Discovery of *Rhexia mariana* L. var. *mariana* in Northwest Indiana

Scott A. Namestnik, Orbis Environmental Consulting

Rhexia mariana var. *mariana* (Maryland meadow beauty) is an herbaceous perennial species in the family Melastomataceae that occurs in dry to moist sandy soils primarily in the southeastern United States, stretching inland from the Gulf of Mexico and Atlantic coastal plains. Populations bordering Lake Michigan in southwest Michigan represent coastal plain disjunct extensions of the geographical range of this species. Within Indiana, *Rhexia mariana* var. *mariana* had previously been documented from the southern third of the state (scattered within the Southern Bottomlands Natural Region, the Driftless Section of the Southwestern Lowlands Natural Region, the Plainville Sand Section of the Southwestern Lowlands Natural Region, and the Bluegrass Natural Region), where it represents the northern extent of its core range. The author recently documented a thriving population of the State Threatened and S1 *Rhexia mariana* var. *mariana* in Porter County, Indiana, within the Lake Michigan Border Section of the Northwestern Morainal Natural Region, approximately 200 miles from the nearest known Indiana populations and approximately 80 miles from the nearest known disjunct Michigan populations. This new record represents the third county (and the first in Indiana) where *Rhexia mariana* var. *mariana* occurs within proximity to Lake Michigan as a coastal plain disjunct.

Science Education

Science for Everyone: Informal Science Education in Our Communities (Panel Discussion)

Melanie Fox, Science and Innovation, Indiana State Museum and founder of Central Indiana Science Outreach (CINSO); **Karen Jepson-Innes**, WonderLab Museum of Science, Health and Technology; **Mitch Luman**, Evansville Museum

Community organizations and educational institutions are increasingly engaging people of all ages—from preschoolers to retirees—outside traditional educational environments to share the excitement of scientific inquiry and discovery. What types of science programs are most effective? How does informal engagement complement traditional educational environments? How can professional scientists and students get involved to share their own science, or contribute expertise to content development? This workshop features a panel discussion with three museum professionals from across Indiana who will share information about some of their organization's science engagement and outreach activities. Come ready to share and discuss ideas with colleagues from across the state!

Development of a Novel Laboratory Approach for the Colorimetric Detection of Vitamin C (Poster)

Paula Angarita, **Devan Gabbard**, Kegan Main, Katherine Timmermann, Kristy J. Wilson, Karla B. Kinkade, and Colleen L. Doçi, Marian University Indianapolis

The use and social discourse regarding dietary supplements is increasingly prevalent in American society. Current regulatory laws do not closely monitor nor distinguish in the different formulations of many of these supplements, and social media platforms frequently contain anecdotal or incomplete information regarding their mechanism of action and appropriate application. Using Vitamin C as an example, we have developed a simple laboratory approach for science instructors to use in the instruction of core biology and chemistry techniques. Since Vitamin C is something students recognize from wider culture, they are more readily engaged in learning to perform an assay to determine its concentration in a solution. Further, Vitamin C is available in a diverse array of natural and synthetic formulations, which provides a platform discussions touching on metabolism, supplement solubility, and bioavailability. In this laboratory experiment, we have adapted a chelation-based transition metal catalysis reaction to facilitate a colorimetric quantification of Vitamin C concentration. Students generate a standard curve and calculate the Vitamin C content from other compounds, which could include food sources, vitamin supplements, or alternative medicines. The laboratory exercise has been designed with flexibility that will allow the instructor to optimize the experiment for his or her classroom, including different conceptual introductions and unknown compounds. Pre- and post-lab assessments indicate that students increase their understanding of spectrophotometry, construction and application of standard curves, calculation of unknowns, as well as more conceptual issues such as the relationship between solubility and bioavailability. This laboratory exercise not only presents a novel colorimetric detection method, but couples this methodology

with an innovative classroom approach that facilitates greater classroom engagement and learning.

Becoming a Professional Scientist: How to Get Started! (Workshop)

Jessi Ghezzi

Students will learn how to introduce themselves as a candidate within a one-minute time frame, what exactly constitutes professional dress in various environments, interview styles and common questions, small steps they can take at conferences to get noticed, small steps they can take at the start of a new position to get noticed in the right way, one-on-one resume and cover letter help and the importance of professional mentorship, as well as networking tips for introverts. By the end of the workshops students will have an arsenal of tools to work towards becoming a successful, professional scientist. It is never too soon to start moving up the ladder, let us help you jump-start your career while you are still in college! Students will get advice and one-on-one tips/edits on the different resume/CV styles for the wide-array of job opportunities from each sector (academia, industry and consulting).

Teaching Dendrochronology to Preservation Students: Linking Historic Sources to Scientific Inquiry

Christopher Baas, Ball State University; Darrin Rubino, Hanover College; and Jonathan Spodek, Ball State University

The preservation and interpretation of historic structures and landscapes is dependent on the accurate identification of significant historic dates. Historians use fieldwork and primary sources to document sites (i.e. photographs, newspapers, legal documents, maps and atlases, journals and diaries). However, rarely do primary documents explicitly identify precise construction dates for historic sites, leaving historians to infer dates based on available primary evidence. Dendroarchaeology is the use of tree rings to assign construction dates to historic structures. The results of tree-ring analysis, paired with primary resources, provides managers of historic sites the most accurate information for making preservation and interpretive decisions. Dendrochronology is typically taught in the sciences (botany, ecology, etc.). Due to its specialization it can only be taught in limited detail to historic preservation students. We recently assembled teaching materials for the instruction of dendrochronological methods in establishing the construction date for a historic log house, and tested them on students currently enrolled in a historic preservation program. Students were provided instruction in dendrochronological methods as applied to the dating of historic structures. In the ensuing lab they were provided the products of field work (timber samples, photographs, measured drawings, etc.). Students were asked to date timber samples by crossdating tree-ring samples (crossdating is a method of assigning calendar dates to samples of unknown age by utilizing series of accurately dated tree rings). Students were also required to provide results in the form of a construction date. Finally, students were asked to reflect on the relationship of primary sources to the dendrochronological results. This presentation will report our process, the effectiveness of the exercise, and how teaching a combination of methodologies from both the sciences and humanities instructs students in the best means for preserving and interpreting historic resources.

Biology Indianapolis Outreach: A new undergraduate service learning course at Butler University

Erin Gerecke and Philip Villani, Butler University

Service learning in the community can be an important element of an undergraduate education. In STEM fields, connecting students with opportunities outside the classroom to apply their knowledge and reflect on their own participation in science can help them consider career opportunities and learn about broader issues in science and society as a whole. We present an overview of our planned offering of a new service learning course in the Department of Biological Sciences at Butler University. As part of the University's Indianapolis Community Requirement curriculum, our new course, Biology Indianapolis Outreach (BIO), will allow students to apply their study of biology to various projects in partnership with community agencies in Indianapolis. The main focus of the course will be developing and presenting STEM educational activities for children in partnership with the Indiana State Museum and Celebrate Science Indiana. Other projects involving environmental engagement will also be included.

A Simple Intervention to Increase Inclusive Pedagogy in a Biology Laboratory

Jennifer Robison, Patrick Gentry, Nicolas Berbari, and Anusha Rao, Indiana University Purdue University Indianapolis

Inclusive pedagogy incorporates dynamic practices, integration of multicultural content, and varied assessment to promote student success. In this intervention, we aimed to increase integration of multicultural content in a 300-level genetics laboratory course of 20 students. This intervention would be simple to adapt into any curricula in any discipline. The assignment instructed students to create a special edition of a mock magazine featuring a diverse set of experts in the field. Students were randomly assigned a geneticist from a pool of modern geneticists spanning a wide range of cultural backgrounds and identities. Next, they wrote a 500 word biography, read their peer’s biographies, and, in groups, categorized these biographies into a Table of Contents. Out of five groups, two focused only on research themes of the geneticists, while three categorized based upon both research themes and multicultural content (i.e. geography, sex, advocacy, etc.) in their Table of Contents. Additionally, a pre-test was administered on the first day of class where students reported the names of all the geneticists and their contribution of the field to the best of their knowledge and repeated as a post-test on the final exam. Scientists reported in the pre-test were 92% male, 8% female and in the post-test the distribution shifted to 76% male, 24% female. Scientists identified in the pre-test were 100% non-minority while the distribution was 76% non-minority and 24% minority in the post-test. These data suggest that this simple assignment can increase student’s awareness of the diverse and multicultural nature of the scientist’s populations. An additional benefit is that it provides role-models for under-represented groups within their field which has been suggested to improve student retention in STEM; however, longer term studies would be required to reliably measure the effectiveness of this intervention and its impact on student learning and retention.

Improving Bone Health Knowledge in Selected Age 9-12 Indiana Students (Poster)

Jaylin Miller and Dan Jones Indiana Wesleyan University

Studies by Winzenberg *et al.* regarding bone mass and osteoporosis indicated that a 10% increase in bone mass during childhood potentially delays the onset of osteoporosis by 13 years. Further, Golden *et al*. reported that between the ages of 9 and 12, peak bone mass lags behind peak height by 6-12 months, making children susceptible to bone fractures. Taking this data into consideration, our laboratory investigated bone health knowledge in students at Fairfield Community of Elkhart county and Lakeview Christian of Grant county. The purpose of the study was to see if an educational presentation would help students understand how to build healthier bones. After institutional review board approval and informed consent from the students and their guardians, our laboratory tested 273 students ages 9-12 in 4^{th}, 5^{th}, and 6^{th} grades. We gave each student a 12-question survey. The first questions identified variables in the students. The second tier of questions probed their knowledge of foods containing calcium and vitamin D, two important nutrients for healthy bones. Then students were asked if they could define osteoporosis. Lastly, students were asked if they knew why exercise was important. The 12-question survey results established their baseline understanding of bone health. Students had an awareness that consuming calcium and vitamin D was important (95.3%), but did not correctly identify which foods contain these nutrients (only 14.7%). Only 7% knew the definition of osteoporosis. The students were aware of the importance of exercise (97.4%) and participated in daily exercise (96.3%). The second part of the research involved a live presentation that gave students information about appropriate nutrition and exercise for good bone health. For the last part of the research, the same survey will be given to the students and results will be analyzed to assess the effectiveness of our educational efforts.

Making Spatial Decisions using Geographic Information Systems (GIS) and the Geo Inquiry Process

Josephine Shireen Desouza, Ball State University

Through web-based hands-on Geospatial technology, middle and high students can participate in making decisions making about real world data, which is a key component to finding solutions to societal issues. The project presented, will show how GIS skills are developed using ArcGIS online (AGO) which employs analytical tools requiring students and teachers to use problem solving and critical thinking skills. GIS is the science of where? Geographic inquiry and STEM pedagogy of scientific inquiry are very similar in process except for the spatial component of objects, events and phenomenon. This presentation advances and supports the IAS conference themes in that it discusses how we can examine societal issues through the lens of geospatial technologies, specifically GIS and ArcGIS online to educate science teachers about designing hypothetical scenarios using innovative technologies. The hands-on experience to learning science or technology can create an interest that could potentially lead to more students ultimately pursuing STEM fields as careers. Additionally, environmental projects are advantageous to society when students, in conjunction with their teachers, learn how to gather and interpret environmental data. The awareness of the meaning of the data analyzed and the

evaluation of whether there are consequences enables youth to play the role of citizen scientists.

A new approach for student debates on controversial topics in agriculture (Poster)

J.L. Ghezzi, Ball State University

It is essential that university-level students be adept at discussing controversial issues from an educated and professional standpoint. Classroom debates serve as a valuable resource for educators to instill such traits in students; concerns exist, however, that debates only reinforce students' existing positions. A new pedagogical method for employing debates in a STEM/Agricultural classroom was investigated with the objective of preparing students to communicate controversial topics and expand content knowledge, and to determine whether opinions are affected after the assignment. On the first day of class 90 students were administered written surveys (with IRB board exemption) with a Likert scale to respond to statements addressing GMOs, organic food production versus conventional, food safety perceptions, humane treatment of animals in CAFOs, meat consumption, and other controversial topics in modern agriculture. Students were then assigned to a debate team that held the opposite opinion as indicated by the survey. Following the debates, students completed the same survey and response differences were analyzed. Students reported that debating the opposite of their opinion was interesting and broadened their perspectives on the issue. A significant number ($p < \alpha = 0.05$) of students changed their initial opinion in the final survey. Overall, there was a significant shift in consumer confidence in food safety and how their food was grown or raised ($p < \alpha = 0.05$). This pedagogical approach to preparing students to communicate demonstrates that students pre-conceived notions on controversial topics were challenged while expanding student's knowledge base.

Charles' Law and Internal Energy (Poster)

Zachary B. Stichter, Caleb E. Yoder, and Chad A. Snyder, Grace College

A simple demonstration and an indirect approximation of the internal energy of a gas were developed. The volume of a flask was measured, and Charles' Law was used to determine the change in temperature of the air within the flask as it was heated. The change in temperature was used to approximate the change in the internal energy of the gas. Students will be instructed that the change in internal energy was responsible for the demonstrated change in volume, and that the temperature change was responsible for changing the internal energy of the gas. The goal of the project is to create a lab exercise where students are tasked with calculating the expected change in internal energy for the gas, assuming ideality and perfect heat transfer.

Establishing Practices Integrating Commuter Students – Year 1 (Poster)

Mindy Capaldi, Michael Watters, **Kristi Bugajski**, Karl Schmitt, Jon Schoer and Bonnie Dahlke Goebbert, Valparaiso University

The EPIC grant focuses on the integration of STM (Science, Technology, & Mathematics) commuter students into the Valparaiso University (VU) campus community through shared undergraduate research experiences, as well as skill-building and social activities. Goals of the EPIC grant include improving recruiting and retention of STM students, increasing the number of STM graduates prepared for research careers, and increasing the number of research intensive majors within STM departments. Students are recruited with scholarships averaging $5,300 per year.
During year 0 the EPIC program developed the infrastructure and tools to recruit, select, enroll, and support the first cohort of students in fall 2017. We report the results of our recruitment initiatives on the poster. In year 1 we refined these practices to include earlier and more varied contact with high schools. Recruitment of the second cohort is underway.
Assessment procedures were initiated through the establishment of pre-EPIC baselines measuring retention, academic, and demographic data for STM students at VU. Data were collected through institutional research and multiple student surveys. This data includes GPAs, STM student retention, graduation rates, and the impact of undergraduate research. Some data is broken down demographically according to gender, race, ethnicity, and commuter status. Preliminary analysis revealed several notable results, such as a 44% retention rate for STM students remaining in their original major and 25% of VU STM students from the 2012 freshman cohort not graduating at all. Additionally, VU commuters are less likely to be retained in a STM major and are less likely to graduate. This group of students is the primary focus of the EPIC program; we predict that our model will improve their retention numbers.

FRESH – Freshman Research Engagement in Science: Early Results (Poster)

Michael Watters, Patrice Bouyer, and Robert Clark, Valparaiso University

We describe here some early results of the FRESH (Freshman Research Engagement in the Sciences) program. A program with the goal to expose freshman to an ongoing research project during the academic year to promote student growth and improve retention in the STEM disciplines. Freshmen worked with a faculty mentor and were also chaperoned by a more senior student researcher in order that they learn lab techniques and the capacity to work independently. Participants were fully engaged in a research project (performing experiments, analyzing and discussing results), not a classic classroom projects, but discovery based projects. By bringing students into the research lab at this early stage, our aim was to improve retention by allowing science students to actually act as scientists, providing an enhanced experience over the usual freshman survey course content. Of the 13 students in two cohorts who joined the program as freshmen, 12 are still in their major and have co-authored over 20 different papers and conference presentations to date. Based on these initial successes, we have modified our approach, tracking qualified applicants who we were unable to fund to serve as our control group in order to study the impact of the FRESH approach on student success.

How to Effectively Teach Biological Writing to College Freshmen and Sophomores

Marc Milne and Leah Milne: University of Indianapolis

Teaching writing to college students is notoriously difficult. In college biology classrooms, writing is largely focused on introducing, describing, and explaining experiments in the form of lab reports or manuscripts. Oftentimes, these assignments are provided early and collected late in the semester with no interaction in between. Using this method, biology college students fail to receive proper training in writing and often produce work that is, as one colleague put it, "so bad that it makes me question my career path." Through an interdepartmental collaborative approach, we introduced a unique method to teach manuscript writing that slowed down the learning process, set achievable writing goals, and provided ample feedback to improve writing over the semester. Compared to previous semesters, writing quality drastically improved and students responded overwhelmingly positive on end-of-course evaluations about the writing process.

Increasing Life Science Interest for Elementary Education Majors (Poster)

Richard C. Roberts and Nathan S. Bosch, Grace College

In teaching life science to Elementary Education students at Grace College, we have found that many of them are hesitant to engage in the study of the life sciences (an attitude similar to math phobia). For these students, their perception of the life sciences is typically that the topic is too hard to understand. We believe that it is key for these students to gain an appreciation for the life sciences if they are to effectively instill a love for this topic in their future students and support STEM educational initiatives. One of the required courses in Elementary Education programs is a basic life science course; at Grace this course is titled “Bioscience Survey” and includes both lecture and laboratory components. In the past, this has been a dreaded course among Elementary Education majors. We have made several changes to the course to combat this attitude with the goal of instilling an excitement for the life sciences in these students. These include two specific assignments: one in which students work in groups to develop a life science lesson, then videotape themselves presenting the lesson for critique by their classmates. The other is finding or developing a life science experiment appropriate for elementary students which directly connects to state educational standards, and presenting their experiment to their classmates for potential future use in their own classrooms (thus providing the students with different ideas about life science experiments). The laboratory portion of the course has been modified as well, providing not only hands-on practice following the scientific method with an independent research project, and also offering examples of experiments that can easily and inexpensively be done in their future elementary classroom. Student feedback has demonstrated that these changes have indeed increased appreciation for the life sciences within the Elementary Education major.

Replacing Old Myths About Secondary and College Teaching with New Opportunities (Workshop)

Chris Edwards, Fishers High School

Educational reform is needed to address a teacher shortage and to better prepare secondary (grades 7-12) students for college and career readiness. For science teachers and and professors, educational reform must first begin with a conversation about what the roles of secondary teachers and college professors are. Only then can the myths about secondary and college teaching be dispelled. What are those myths?

- The job of the secondary teacher is merely to teach out of curricular materials created by textbook-and-workbook companies.
- High school teachers do not need to have a high level of content-area mastery.
- High school teachers and college professors who take risks in the classroom are at risk of being punished by administrators.
- A classroom curriculum falls outside the realm of "knowledge production" as traditionally defined.

For the last three years, a new summer institute program that brings high school science teachers and university professors together has sought to erase these myths and redefine the work of the secondary teacher and professor. This workshop will provide an opportunity for educators to discuss the role of the teachers and professor and to engage with each other on the topics of scientific content and classroom practice. Specifically, the format and results of a new summer institute program which has brought secondary teacher and college professors together for the last three summers will be presented and analyzed.

Science + Art = Symbiotic Learning (Hot Topic)

Roger P. Hangarter, Indiana University

Historically, science and art worked hand-in-hand but over time became separate disciplines, which lead to a general decline in understanding and appreciation of science (and art) among large portions of the public. However, scientists and artists have always been motivated by a passionate curiosity, an appreciation of the subjects they explore, and for creating/discovering new things. By recognizing these commonalities, scientists and artists are collaborating more again in efforts to bring a greater appreciation of both science and art to a broader spectrum of the public. In this presentation we will look at examples of science/art and discuss their effectiveness for engaging public audiences.

Using Immediate Feedback Quizzes and Box.com as Learning Tools in the Active Learning Classroom

Ryan Jeske, Ball State University

I will discuss my continued development of an active learning format for my organic chemistry class at Ball State University. In recent semesters I have started to collect images of student drawings during class in real time that are uploaded to a cloud folder on Box.com. These images are displayed to the class and used to discuss examples of common student errors and excellent answers and drawing skills. In addition, as a team building exercise, I have implemented team quizzes using an answer-until-correct multiple choice answer blank. These answer forms are available online, and provide the students with an answer sheet with scratch off blanks, much like a lottery ticket. These team quizzes have helped encourage, and sometimes force, the team members to work closely with each other. I will discuss the pros and cons of each of these techniques, and share students' thoughts on their experience with the Box.com folder and team quizzes.

Stand out from the Crowd: Communicating Your Science in an Online World (Hot Topic)

Bill Sullivan, Indiana University School of Medicine

The times are a-changing. The traditional peer-review model of scientific publishing is undergoing rapid evolution, and several novel platforms are emerging to report your research findings. Open access and post-review journals, in conjunction with a bevy of social media channels, are revolutionizing the way research is communicated to your colleagues and the world. In addition to promoting your research, social media offers a diverse assortment of tools that can boost lab efficiency, strike up new collaborations, and raise funds for projects. In this presentation, we will describe the changing landscape of scientific publishing and provide tips on effectively using the various forms of social media tools to enhance your research career without falling prey to their potential pitfalls. Using real-world examples, we will discuss tips on communicating your science to diverse audiences, whether they be fellow scientists, grant reviewers, the press, or the public.

Indiana Soil Microbes Producing Anti-Microbial Factors to *Staphylococcus aureus*
(Poster)

John Parroquin, Amy Cuevas**,** Johnathan Potter**,** Iredell Sanders and George Twaddle
Ivy Tech Community College of Indiana – South Bend/Elkhart

Microbes have evolved antimicrobial compounds in their competition for resources in the soil. Among the class of antibiotics, some hold promise as the next generation drugs to fight present antibiotic-resistant human pathogens like MRSA. Following a modified version of Yale University's "Small World" antibiotic discovery initiative, undergraduate students surveyed 11 soil types for putative antibiotic-producing microbes. Candidates were selected on soil-conditioned tryptic soy agar and screened for the ability to produce zones of inhibition when grown in the presence of *Staphylococcus aureus* (surrogate of MRSA), and *Pseudomonas putida*(surrogate of *Pseudomonas aeruginosa*) and *Escherichia coli* C. Fifteen soil bacteria were isolated from over 1000 tested which constitutively produce factor(s) which inhibited the growth of *S. aureus* in

broth culture or that produced growth inhibitory factors after induction following exposure to *S. aureus* conditioned media. Two of seven characterized bacteria isolates characterized so far produced growth inhibitory factors for *Pseudomonas*while two isolates produced factor(s) that surprisingly increased the growth of *Pseudomonas* relative to the controls. Six bacteria produced growth inhibitory factor(s) for *E. coli* while a single bacteria produced a growth inhibitory for *E. coli* only after induction. Three of seven characterized bacteria were found to constitutively produced bactericidal factors to S. aureus with the remaining producing bacteriostatic activity. Further characterization of the factors produced by these bacteria could yield commercially viable products for medical use.

Zoology & Entomology

Effect of Supplementation with □-Lipoic Acid on Nuclear Reduced Glutathione Levels in Rat Kidney

Marianna Zamlauski-Tucker and Bingwei Ye, Ball State University

Dietary supplementation with the antioxidant alpha lipoic acid has been found to increase reduced glutathione (GSH) levels in mitochondria from kidney and heart tissue in old rats. GSH is the major antioxidant inside cells and provides protection against damage by free radicals produced as a consequence of oxidative metabolism. The purpose of this study was to investigate the effect of dietary supplementation with alpha lipoic acid on GSH levels in the nucleus of rat kidney cells. Old female Lewis rats (i.e., 22 months of age; n = 4) received alpha lipoic acid (100 mg/Kg of body wt) via i.p. injection for one week. Age-matched Control rats (i.e., n =4) did not receive any supplementation. The kidneys were harvested from anesthetized rats and the cortex and medulla were separated and homogenized. The nuclear fractions were isolated using differential centrifugation. The GSH levels were measured by a spectrophotometric assay and compared using as a Student's T Test. There was a significant increase in nuclear GSH levels in both the rat kidney cortex and medulla with supplementation. This finding suggests dietary supplementation with alpha lipoic acid may also be beneficial to the cell nucleus by increasing the GSH levels and thus, providing protection against free radical damage.

Fluctuating temperatures impact embryonic growth and hatchling size in *Chelydra serpentine* (Poster)

Megan R. Moss and Michael S. Finkler, Indiana University Kokomo

Eggs of many reptile species are exposed to diurnal fluctuations in temperature that could influence incubation duration, embryonic development, and hatchling phenotype in ways not predicted by incubation at a constant temperature of the same mean. Moreover, as the thermal sensitivity of embryonic growth and development appears to decrease over the course of incubation, thermal fluctuation during early stages of development may have greater impact on overall incubation duration and hatchling phenotype than would fluctuation later in development. We incubated snapping turtle eggs under different combinations of constant temperature (25°C) and fluctuating temperature (25±2 °C). Exposure to fluctuating temperatures accelerated embryonic growth and development rates, based on both the mass and development stages of Day 21 and Day 42 embryos. Hatchlings from eggs exposed to fluctuating temperature over the entirely of incubation ("Fluctuating") hatched earlier and had longer carapace and plastron lengths than did those exposed to constant temperatures over the entirety of incubation ("Constant") or those exposed to fluctuating temperature only during the last ~40% of incubation ("3rd Trimester"). Eggs exposed to fluctuating temperature during the first ~30% of incubation ("1st Trimester") hatched sooner than did those in both the 3rd Trimester and Constant treatments. There was no significant difference in hatchling dry mass among any of the

incubation treatments, but dry yolk sac mass was significantly smaller in the Fluctuating and 2nd Trimester (eggs exposed to fluctuating temperature during the middle 30% of incubation) treatments, than in the Constant treatment. Carcass nonpolar lipid contents did not differ significantly among treatment groups. These findings indicate that diurnal fluctuations in temperature accelerate growth and influence hatchling morphology. Moreover, fluctuating temperatures during early incubation appear to have a more pronounced effect on incubation duration than similar fluctuations during late incubation.

Histology Germinal Epithelia of Spermatogenesis in Alligator Snapping Turtle, *Macrochelys temminckii* (Poster)

[1]**Alanna Mayberry**, [2]Stanley Trauth, [1]Kevin Gribbins,
[1] University of Indianapolis [2]Arkansas State University, Jonesboro, AR

We investigated the cytology of the seminiferous epithelia of the Alligator Snapping Turtles (*Macrochelys temminckii*). Spermatogenic and regressed testes were assessed from 2 large individuals collected in Arkansas in May and September of 1993. Specifically, we focused on the cellular phases of germ cell development and maturation. The germ cell morphology and developmental strategy within the germinal epithelium of *M. temminckii* appear similar to that of other genera of turtles previously studied. Interestingly mitotic, meiotic, and spermiogeneic cells are nearly identical to that of other studied turtles. There are also 6 recognizable steps to spermiogenesis, which is slightly different that the 7 steps of most turtles. Although this study only uses two individuals (because of its endangered status), *M. temminckii* appears to start spermatogenesis in the spring and the climax of spermiogenesis occurs in the fall similar to that of other temperate turtles studied to date. Peculiar to both turtles in this study were the regular appearance of very large germ cells in the basal compartment of the germinal epithelium. Based on previous research and our histological analysis, these enlarged spermatogonia exhibit hypertrophic characteristics typical of cells undergoing apoptosis.

Prairie Quest Farm: Working land conservation and restoration (Poster)

Mary C. Damm, Indiana University Bloomington and Prairie Quest Farm

Prairie Quest Farm is a 120-acre farm in Northeast Iowa along the bluffs of the upper Mississippi River. The majority of the farm is planted in perennial cool-season grasses and forbs for rotationally grazed pastures and hay fields. Over the past two decades, the farm has been managed for the production of grass-fed beef and dairy cattle as well as habitat for breeding grassland birds. Grassland bird populations (Bobolink, Eastern Meadowlark) have declined by 75% in the past 50 years due to habitat loss, originally from plowing the prairie and more recently from the conversion of diversified farms with long crop rotations and livestock to intensively managed farms of corn and soybeans. I recently purchased Prairie Quest Farm with the goal of continuing the established management of the farm. With the advice of my farm mentor and renter, I enrolled the majority of the farm in the NRCS Conservation Stewardship Program (CSP) to achieve my dual management goals of producing food and providing wildlife habitat. The CSP was first introduced in the 2002 Farm Bill to financially reward farmers for conservation practices on working farms. In 2015 the CSP offered 119 practices under the

broad categories of improving soil and water quality, conserving energy, and planting diverse habitat for wildlife and insects. Within the CSP practices are provisions for maintaining and restoring farmland for plant species diversity and ecosystem function. I will describe how I selected 9 CSP practices to maintain the high quality pasture-grassland and restore farm borders on Prairie Quest Farm.

Ultrastructure of Spermiogenesis within the Testis of the Earless lizard, *Holbrookia elegans* (Poster)

[1]**Cailin Davis**, [1]Alanna Mayberry, [2]Justin Rheubert, [1]Kevin Gribbins,
[1] University of Indianapolis [2]University of Findlay, Findlay, OH

Variation in sperm maturation has been utilized for phylogenetic studies as well as ecological and toxicological research. This study purpose is to supplement our sparse knowledge of spermiogenesis within the Squamata. This study provides the first examination of the cellular events during spermiogenesis within the testis of *Holbrookia elegans*. Three male specimens were collected from southeastern AZ in the month of June, 2015. Testicular tissue was processed for transmission electron microscopy, sectioned, and evaluated to determine the major morphological events of spermiogenesis. Spermiogenesis can be divided into three phases as sperm mature; acrosome development, nuclear elongation, and flagellar development. Most of the ontogenic features of spermiogenesis in *H. elegans* appear to be highly conserved and similar to what has been observed in other lizards studied to date. Two key characteristics that may be unique to *H. elegans* are a flatter basal perforatorial plate and chromatin extensions into the clear nuclear shoulders of the during fossa development in the caudal flagellum. This lizard does share several characters (filamentous chromatin condensation, nuclear lacunae, well-developed manchette, shallow nuclear acrosomal indentation) with several sceloporine lizards, which are found in the same family, Phyronsomatidae. Though these data offer valuable insight to the already accumulating information on spermiogenesis in phrynosomatid lizards, caution must be maintained on its robustness until more genera are studied in this lineage of saurians.

A Test of Local Enhancement in Fathead Minnows (*Pimephales promelas)* (Poster)

Ciara J. Mergler, Theresa E. Wrynn, Joseph R. Titus, and Brian G. Gall, Hanover College

Local enhancement is a form of learning in which individuals are attracted to areas where conspecifics have previously been located. We tested the responses of Fathead Minnows (*Pimephales promelas*) in circular arenas in two separate experiments to test whether these fish exhibit this form of learning. In the first experiment we conducted a set of trials to see if minnows were attracted to conspecifics. A focal fish's location was tracked while four demonstrator fish were left in the pool during the trial. In the second experiment to test for local enhancement, the demonstrator fish were removed once the acclimation period ended. We found that the focal fish were significantly oriented towards the demonstrator fish when left in the pool, but they did not exhibit local enhancement if the demonstrators were removed. These results suggest that minnows exhibit strong social tendencies but visual signals from conspecifics may be necessary for schooling tendencies.

An additional 100+ new spider distribution records in Indiana (Poster)

Lucas Frandsen, Marc Milne, Brodrick Deno, Janise Acosta, and Alison Sobczak, University of Indianapolis; Brian Foster, Indiana State University; Julian J. Lewis, Lewis & Associates LLC

Spiders are understudied in Indiana even though they play a critical part in almost every state ecosystem. However, to understand the important roles spiders play in Indiana's natural habitats, the most basic question must first be answered: What species are present in Indiana? To better answer this question, we re-identified spiders collected from Indiana housed in The Field Museum of Natural History in Chicago, IL. Additionally, we collected spiders from various sites around the state, including Yellowwood State Forest, Morgan-Monroe State Forest, Indiana Dunes National Lakeshore, McVey Memorial Forest, Goose Pond Fish and Wildlife Area, Hoosier National Forest, White River Woods, Glacier's End Nature Preserve, and various private properties in western Indiana. Spiders were caught using pitfall traps, sweep netting, berlese funnels, and by hand. Herein, we report over 100 new distribution records to the state. Indiana now has over 550 known spider species throughout the state.

Collaborating for Conservation: Utilizing zoo resources for scientific advancement

Maraiah Russell, Fort Wayne Children's Zoo

Local zoos have much to offer in the area of conservation research. Zoos can offer the knowledge and experience from passionate individuals with expertise in a variety of areas. Zoo keepers and veterinary staff possess skills that could benefit many disciplines of scientific research. The skills of most interest to researchers might include experience in the areas of animal husbandry, collecting and processing biological samples, and providing medical care to wild/exotic animals. One project that demonstrates the value of these skills is the involvement of Fort Wayne Children's Zoo (FWCZ) staff in hellbender care and research at Purdue University. Currently FWCZ houses 59 juvenile hellbenders as participants in the Purdue University captive rearing program. FWCZ zoo keepers provide daily husbandry and water quality testing for this population. FWCZ zoo veterinarians provide medical care for this population, as well as assistance with the care and medical procedures of the population at Purdue University. This collaboration developed further in the summer of 2016, when veterinary staff was invited to assist with a Ph.D. research project which required collecting and processing serial blood samples from captive juvenile Eastern Hellbenders *(Cryptobranchus alleganiensis alleganiensis*). The purpose of this study was to investigate the effects of pre-exposing captive hellbenders to environmental microbes in river water, in an effort to increase their survival after release from captivity. Two veterinary technicians assisted in establishing and training Purdue staff in blood collection techniques, creating and staining blood smears, and evaluating white blood cells on a subset of slides to compare with the results of the researcher. Veterinary staff traveled to Purdue regularly, as well as to the release site in Southern Indiana to assist with these procedures. Fort Wayne Children's Zoo is excited to participate in new research opportunities and develop partnerships with scientists conducting conservation efforts locally.

Growth Factors of Nile Tilapia (*Oreochromis niloticus*) Fed Isoflavone-Supplemented Feed Under Stress (Poster)

Destin Furnas, Austin Petras, and Ahmed Mustafa. Biology Department, Indiana University-Purdue University Fort Wayne

Nile tilapia (*Oreochromis niloticus*) is one of the major species of fish reared in aquaculture. The aquaculture environment produces many stressors not found in the normal environment of Nile tilapia. As energy is diverted to cope with these stressors, it is typical to see a reduction in growth in stressed fish. This study examines the effects of two isoflavones (puerarin and genistein) on enhancing growth in chronically stressed Nile tilapia. Nile tilapia were randomly distributed into one of four groups: 1) non-stressed with commercial feed, 2) stressed with commercial feed, 3) stressed with puerarin-supplemented commercial feed, and 4) stressed with genistein-supplemented commercial feed. Weight and length were recorded weekly for seven weeks. At the end of seven weeks, stressed Nile tilapia fed puerarin- and genistein-supplemented commercial feed had mild improvements in growth compared to those fed non-supplemented commercial feed. Isoflavones, such as puerarin and genistein, may be effective feed supplements for growth in the aquaculture environment.

Habitat Correlations for Indiana's Critically Imperiled Mayfly Species (Poster)

Stephanie F. Baumgartner and Luke M. Jacobus , Indiana University Purdue University Columbus

Of the 169 recorded mayfly species in Indiana, 19 are considered state critically imperiled. Nearly all of these critically imperiled species are known from only 1 occurrence in the state. Five of these species are of conservation interest because they are at the edge of their geographic range of distribution. They include *Siphlonurus alternatus, Stenacron carolina, Ephemerella subvaria, Drunella walkeri,* and *Attenella attenuata. Traverella lewisi* is a big river species, but all of its historical locations have been drastically altered. *Tortopsis primus* is another big river species that has unique clay-bank burrowing behavior. *Ephemera varia* is a substrate burrower whose biology in Indiana is poorly understood. Notably, the remaining 11 species can be divided into 2 groups based on their habitat requirements: sand dwellers, and those that live in cobble-pebble bottomed streams.

Identifying an unknown *Agyneta* sp. via DNA sequencing (Poster)

Brodrick Deno and Marc Milne, University of Indianapolis

The identification of spiders to species, especially lesser known groups, is notoriously difficult. The genus *Agyneta* (Araneae: Linyphiidae) is a widely distributed, largely North American, group of small (1-3 mm) spiders. We collected unknown specimens of *Agyneta* from multiple habitats in Indiana in 2016 and 2017. In order to properly identify these specimens, we attempted to sequence the COI gene (barcode) from these specimens and compared the sequence to an online barcode database. We determined that the species was *Agyneta angulata*, a species native to the Eastern United States and one that was recently discovered in

Indiana. This study shows the effectiveness of using DNA barcodes to accurately identify spiders that may be difficult to identify with more classical morphological techniques.

Interactions between predatory dragonflies and early Eastern newt developmental stages

Theresa E. Wrynn, and Brian G. Gall, Hanover College

We examined the predator-prey relationship between predatory dragonfly (*Anax junius*) nymphs and early developmental stages of the Eastern newt (*Notophthalmus viridescens*), some of which may contain the potent neurotoxin, tetrodotoxin (TTX). First, we conducted a palatability study to determine which newt life-history stages were palatable to dragonflies. We also tested the metamorphosis and survival rates of larval newts when exposed to predatory dragonflies in small mesocosms. Finally, we tested the predator avoidance behavior of larval newts in response to kairomones from a control (water), food stimulus (blackworms), and predatory dragonflies. All life-history stages (young and old larvae, and recent metamorphs) were palatable to dragonflies. In mesocosm trials, we found that newt larvae had a lower chance of surviving and transforming when dragonflies were present compared to a control. Finally, newt larvae decreased movement significantly when exposed to predatory dragonfly stimulus compared to either a control or food stimulus. These results suggest that dragonfly nymphs are efficient predators of newts from hatching through metamorphosis, but these larvae likely possess behavioral avoidance mechanisms that reduce the risk of predation.

Intraspecific Phylogeography of the Southern Redbelly Dace *Chrosomus erythrogaster*

Rex Meade Strange, University of Southern Indiana

The Southern Redbelly Dace (*Chrosomus erythrogaster*) is broadly distributed across the North American Central Highlands with a few relict populations on the Great Plains. The species was originally described by Rafinesque in 1820 from specimens collected from the Kentucky River system (the type locality), while distinct populations on Crowley's Ridge (Arkansas) and Caney Fork (Tennessee) appear to be different than the nominate form. I examined sequence variation of the mitochondrial cytochrome *b* gene from specimens collected from across the species' distribution to better understand the historical relationships among the populations currently recognized as *C. erythrogaster*. Phylogenetic analysis revealed two major clades within *C. erythrogaster*: one clade represents the nominate form and includes populations occurring in the Eastern Highlands and Western Plains. Interestingly, one of the samples from the Caney Fork system was basal to all other members of the nominate clade. A second (Ozark) clade consists of populations from Missouri, Arkansas, and tributaries of the upper Mississippi River system. The sample from Crowley's Ridge was closely related to samples collected from the southern Ozarks. These data suggest that differences in color pattern among isolated populations are not indicative of cryptic speciation, but represent phenotypic plasticity within one or two well-defined species. Subsequent morphological analysis will be needed to assess whether the Ozark clade should be recognized as taxonomically distinct from *C. erythrogaster* sensu stricto.

Palatability and predator avoidance behavior of salamanders in response to the Virginia opossum (Poster)

Shelby L. Hart, Theresa Wrynn, and **Brian G. Gall**, Hanover College; Mackenzie M. Spicer, The University of Iowa; Trevor L. Chapman, East Tennessee State University

Our understanding of interactions between salamanders and potential mammalian predators is primarily restricted to small carnivorous species (i.e. shrews). We conducted a series of investigations to determine whether the Virginia opossum (*Didelphis virginiana*) is a potential predator of various salamander species [*Desmognathus fuscus*, *Plethodon dorsalis*, and *Notophthalmus viridescens* (adult and eft)] from the eastern United States. First, we fed three individuals of each species/phase to opossums to test their general palatability. Next, we conducted behavioral assays with each species and life-history stage to determine whether they possess predator avoidance behaviors in response to opossum kairomones. All species, including toxic eft newts, were palatable to opossums. These results suggest opossums may be an active predator of salamanders. While aquatic adult newts did not modify activity in response to opossum kairomones, each terrestrial salamander exhibited avoidance responses. These results suggest opossums may be an important predator on terrestrial salamanders and that each species with significant risk of mortality has evolved mechanisms to minimize this predation risk.

Probiotics in Aquaculture- I: Fish Growth

Kenneth Saillant, Ashley Baum, and Ahmed Mustafa, Indiana University-Purdue University Fort Wayne

Stress presents a significant problem in aquaculture. Stress over time leads to a reduction in immune response and, therefore, a decrease in the ability to fight against diseases. Current aquaculture techniques, therefore, involve the use of antibiotics and other chemicals in order to reduce disease and mortality within the crops, which is not good for fish, their consumers, and the environment. In order to provide solutions to the problems of aquaculture and to provide quality protein to the consumers without the use of potentially harmful substances, we are looking into the use of nutraceuticals. For this experiment, we have used probiotics in order to decrease stress responses, increase immune responses, increase growth, and increase the nutritional value of tilapia, reared in recirculating aquaculture systems. Over a 4 week long experiment, we have measured Specific Growth Weight (SGR), Absolute Feed Intake (FIABS), Feed Conversion Ratio (FCR), Protein Energy Retention (PER), Protein Production Value (PPV), and Fulton Condition Factor (K). Apparently, fish fed probiotic-supplemented commercial feed have better indices than fish fed sham-supplemented commercial control feed in terms of all growth parameters.

Probiotics in Aquaculture- II: Fish Health (Poster)

Ashley Baum, Kenneth Salliant, Mckenzie Comby, and Ahmed Mustafa, Indiana University-Purdue University Fort Wayne

Stress presents a significant problem in aquaculture. Stress over time leads to a reduction in immune response and, therefore, a decrease in the ability to fight against diseases. Current aquaculture techniques, therefore, involve the use of antibiotics and other chemicals in order to reduce disease and mortality within the crops, which is not good for fish, their consumers, and the environment. In order to provide solutions to the problems of aquaculture and to provide quality protein to the consumers without the use of potentially harmful substances, we are looking into the use of nutraceuticals. For this experiment, we have used probiotics in order to decrease stress responses, increase immune responses, increase growth, and increase the nutritional value of tilapia, reared in recirculating aquaculture systems. Over a 4 week long experiment, we have measured blood glucose, packed cell volume, and plasma protein for changes in fish physiology; and spleen-somatic index and macrophage phagocytic capacity for fish immunology. Apparently, fish fed probiotic-supplemented commercial feed has better indices than fish fed sham-supplemented commercial control feed in terms of physiological and immunological responses.

Regulation of Reproduction and Gastrointestinal Development in the Rice Rat (*Oryzomys palustris*)

Kent Edmonds, Indiana University Southeast

Environmental factors and hormones can regulate the development of various physiological systems in many species. Photoperiod, melatonin, and thyroid status are known to affect significantly the reproductive system in seasonal breeders, but effects on the GI tract have not been as extensively studied. The present studies examined whether constant light (which inhibits melatonin release), oral melatonin administration, and hypothyroidism affect reproductive and gastrointestinal (GI) development in juvenile male rice rats. Rice rats were subjected, in separate experiments, to 14L:10D or constant light (24L:0D) photoperiods, the administration of oral melatonin, or the administration of oral propylthiouracil (PTU; 0.06%) to induce hypothyroidism from 21-56 days of age. The following masses were examined: body, testes, seminal vesicles (SV), Harderian glands (HG), spleen, and wet (W) and dry (D) masses of the stomach (St), small intestine (SI), cecum (Ce), and colon (Co). In addition, small intestine and colon lengths were measured. Constant light significantly reduced only SV and HG masses. There was no effect on the masses of the body, testes, spleen, or any GI variable examined. Oral melatonin administration reduced body, testes, SV, HG, WSt, WCo, DCo masses and the SI and Co lengths. PTU-induced hypothyroidism reduced masses of the body, testes, SV and HG, while causing decreases in only the WCe and DCe masses. These data show that melatonin most dramatically affects growth, reproduction, and GI development in males, but that constant light and hypothyroidism were without effect on nearly all GI endpoints. It was hypothesized that changes in the gut may be a necessary mechanism for coping with likely seasonal changes in metabolic requirements.

The Bird Cherry-Oat Aphid, *Rhopalosiphum padi* Circadian Rhythm and the Impact on Feeding Behavior (Poster)

Travis Isaacs, Sarah Moh, Vamsi Nalam, and Punya Nachappa, Indiana University-Purdue University Fort Wayne

The circadian rhythm is an internal biological clock that affects the feeding behaviors of aphids. This mechanism is regulated by internal and external stimuli. Daylight is an important factor in the regulation of this cycle. Daylight regulates the circadian cycles of most organisms as a timing mechanism. Most organisms have a specific time at which they feed or intake their food in the highest quantities, with many animals being specific time-of-day feeders. The circadian rhythm also regulates geological factors such as seasonal and local settings which tell an organism what location is best for survival. The aphid circadian rhythm isn't well understood or tested but in a select few species. The feeding behaviors of aphids are difficult to test and determine except with very specific scientific tools. An Electrical Penetration Graph (EPG) is a way in which to measure the feeding of insects such as aphids. When the aphid probes into the plant, the circuit connecting the roots of the plant to the aphid is recorded by the resulting electrical signal, showing the feeding habits of the aphids. Taking data from day and night trials using this method, comparisons are done to see the different feeding behaviors of aphids by analyzing rates of probing. Waveforms of probing, feeding from the xylem, and phloem are seen with the use of the EPG.

Impact of neonicotinoid insecticide on *Aphis glycines* population on water-stressed soybean plants (Poster)

Asif Mortuza, Valeria Castro Salazar, Chengyu Bi, and Punya Nachappa, Indiana University-Purdue University Fort Wayne

The soybean aphids are a serious pest of soybeans in North Central United States. The insect is native to Asia and exotic to North America. They reproduce both sexually and asexually and give live birth. Due to recent outbreaks of aphids in North America, the use of insecticide and pesticide has risen. Often the seeds of the plants are coated with neonicotinoids (insecticide) to provide protection to the newly germinated plants. However, water stress can determine how chemicals travel through the plant and at what rate. The goal of this study is to investigate the interaction between water stress and the efficacy of the neonicotinoid insecticide seed coat on the soybean aphid population. In order to do so, we applied three seed treatments: 1) CruiserMaxx (Insecticide+ fungicide), 2) ApronMaxx (fungicide; control to CruiserMaxx) and 3) AG3334 (No seed coat; control to ApronMaxx) and two water stress levels 1) drought and 2) well-watered control. Soybean plants subjected to drought stress and well-watered conditions were populated with soybean aphids, and aphid numbers were counted after 7 days. We hypothesize that the effectiveness of the neonicotinoid seed treatment on soybean aphids will be reduced under drought stress conditions. Results of our study will increase our understanding of the efficacy of neonicotinoid seed coats in controlling aphid population under water-stress conditions.

Hover fly diversity in the Yellowwood State Forest Back Country Area (Diptera: Syrphidae) (Poster)

Zoë Bachmann and Glené Mynhardt, Indiana Forest Alliance

Few studies have focused on the diversity of Diptera (flies) in Indiana, particularly in a deciduous forest ecosystem. In collaboration with the Indiana Forest Alliance (IFA), which is focused on understanding and protecting the fauna and flora of Indiana state forests, the objective of this study is to estimate the diversity of flies, with an emphasis on the hover flies (Diptera: Syrphidae). Hover flies are well known for morphologically and behaviorally mimicking various bees and wasps (Hymenoptera), and often serve as pollinators as adults. As larvae they play a major role as predators of economically destructive insects, including aphids. Adult specimens were collected using passive Malaise traps throughout the Yellowwood State Forest Backcountry Area. Samples were collected between May and October of 2016. A total of 38 unique species in 20 genera were collected, and represent all three subfamilies of syrphids. Among those species collected, the majority are generalist pollinators with peak activity during the months of May through June. In addition to these findings, it would be beneficial to identify potential hymenopteran models and study their effects on syrphid mimic diversity. Further work should include larval sampling to ensure a more robust estimate of the diversity of hover flies in the sampling area.

Diversity of Beetles in the Yellowwood State Forest Back Country Area (Insecta: Coleoptera) (Poster)

Samuel Bowman Stryker and Glene Mynhardt, Hanover College

Currently, our knowledge of the Coleoptera (beetles) of Indiana is limited mainly to the work of Willis Stanley Blatchley (1910). Since his publication, other works have added to our knowledge, but few studies have focused entirely on the diversity of the order Coleoptera. Through collaboration with the Indiana Forest Alliance (IFA), a non-profit organization dedicated to the protection and well-being of Indiana's native forests, the main objective of this study is to estimate Coleoptera diversity within the Yellowwood State Forest Back Country Area. Since sampling efforts have begun in 2014, at least 240 unique species of Coleoptera have been identified within the sampling area. This study focuses on those specimens collected by Malaise traps between May and October of 2016. The data on the family Cerambycidae (longhorn beetles) are more complete than for other families of Coleoptera, so this family is the taxon of interest. For 2016 alone, 25 species of Cerambycidae have been identified to species level, with 20 species being newly found within the sampling area, and 5 species being previously recorded between 2014 and 2015. In total, 37 Cerambycidae have been identified to species level since 2014. These new data indicate that there are many species that have not been previously catalogued as present in the sampling area, thus suggesting that collecting efforts should continue in order to assist in completing our knowledge of the diversity of Coleoptera in the area.

Diurnal Oviposition of Blow Flies in Different Aged Carrion (Poster)

Raenah Bailey, Janelle Bouman, Cecelia Frankenwich, Monique Le Donne, Shelby Leucuta, Lauren Smith and Kristi Bugajski

Blow flies (Diptera: Calliphoridae) are among the first insects to oviposit (lay eggs) on carrion. The timing of blow fly oviposition is critical for determining a postmortem interval (PMI) estimation, which is the time that has passed between death and corpse discovery. The objective of this investigation is to gain more information about the timing of blow fly oviposition so that a more accurate PMI can be calculated. Past research in our lab has shown that blow fly oviposition occurs an average of 4.75 hours after sunrise. This year's research expanded on previous studies by placing six piglets in a remote, wooded area an hour after sunrise. Three of the piglets had been thawed for 15 hours, and the other three were thawed for approximately 55 hours to see if there was a difference in the timing of oviposition related to the age of the carrion. The piglets were checked once an hour until oviposition occurred, and it was recorded whether flies and eggs were present each hour. Egg masses were collected and reared to the third larval instar stage for identification using taxonomic keys.. The timing of oviposition, in hours after sunrise, was analyzed with respect to temperature, humidity, and light intensity. The research was repeated six times in the fall of 2017. There was no significant difference found in the timing of oviposition between treatments. The most common flies found ovipositing were Diptera: Calliphoridae, Phormia regina (Meigen) and Diptera: Calliphoridae, Lucilia coeruleiviridis (Macquart). This research has importance in both the scientific and forensic communities, as a more accurate PMI can assist with the validity of a forensic investigation.