Abstract Submissions

Anthropology

Presentation

Description and Quantification of Sexual Dimorphism of Pubic Body Shape in Hispanic Populations Using Elliptical Fourier Analysis

Erica Cantor, Krista Latham, and Stephen Nawrocki, University of Indianapolis

Sex determination is important in the creation of a biological profile by forensic anthropologists, as personal identification relies on an accurate reconstruction of the decedent’s biological traits. The pubic bone is cited as one of the best indicators of sex due to the effects of childbirth, with its length being significantly greater in females than in males. Current methods that use the pubic bone for sex determination, however, rely on poorly-defined and subjective observations that are subject to inter- and intraobserver error. Additionally, many of the methods currently in use are based on Euroamerican and Afroamerican populations and thus do not necessarily model the variation seen in other populations. The aim of this study is to gain a better understanding of variation in pubic bone shape in Hispanic populations by examining the different influences of sex, ancestry, and age at death. A total of 164 pubes from US Hispanic and Chilean individuals were compared to 287 pubes from individuals of US Euroamerican ancestry, using elliptical Fourier analysis of photographs, principle components analysis, and ANCOVA. The first principal component (PC1) explains 78.7% of the variation in the shape of the pubic body and appears to depict differences in the bone’s height-width ratio. This principal component is significantly affected by sex, with male pubes being taller and thinner and female pubes being shorter and wider. PC1 is also significantly affected by ancestry: individuals of Latin American descent tend to have taller, thinner pubes, while individuals of Euroamerican descent tend to have shorter, wider pubes. Due to a small US Hispanic sample size, however, future studies utilizing a larger Hispanic North American sample are needed to further elucidate whether these differences can be attributed to either ancestral or continental (environmental) effects.
Using multivariate techniques to analyze human skeletal variability: Pelvic scars as indicators of parity and sex

Stephen Nawrocki, University of Indianapolis, and Helen Brandt, Binghamton University

Anthropological studies of skeletal features have traditionally relied on univariate and bivariate statistics to sequentially examine a range of variables (sex, age at death) that might affect or control the expression of those features. Unfortunately, these approaches to testing hypotheses are generally weak because they fail to control for the effects of all independent variables simultaneously. As a result, uneven sampling can produce spurious results, and interactions between variables cannot be assessed. Previous studies of pelvic scars and their relationship to parity (births) are marred by poor statistical control, producing conflicting results. This presentation uses analysis of covariance, ordinal regression, and logistic regression to clarify the determinants of pelvic scarring in a well-balanced sample, demonstrating the power and utility of these multivariate statistical approaches. The presence and degree of expression of dorsal pubic pitting, the pubic tubercle, the preauricular sulcus, the interosseous groove, and retroauricular surface rugosity was scored for 530 recent, identified Euroamerican coxae from U.S. collections. Parity, sex, ancestry, and age at death was known for all specimens, and coxa height was used as a proxy for body size. Results indicate that parity has a low-level but significant effect on the expression of dorsal pubic pitting only; no other scar or feature is influenced by the number of births. However, most scars are strongly affected by sex, with females displaying higher frequencies. Controlling for sex and parity, the pubic tubercle increases with increasing pelvic (body) size, and retroauricular surface rugosity increases with increasing age at death. In sum, the utility of these skeletal features lies primarily in their ability to predict the sex of the decedent, while retroauricular surface rugosity can be used to estimate age at death. These findings would not have been as clear had traditional statistical methods been used.

Sexual dimorphism in the shape of the auricular surface of the ilium

Madeline F. Parker, Stephen P. Nawrocki, Krista E. Latham, University of Indianapolis

The coxa is considered to be the most sexually dimorphic bone in the body and one of the most useful for sex determination from the skeleton. The auricular surface of the ilium preserves well and is often present in archeological samples and forensic cases. While the shape of the auricular surface has been analyzed with both metric and
nonmetric techniques, these studies tend to simplify the feature’s complex shape or else rely on the use of a number of arbitrarily-defined landmarks and semi-landmarks, which can limit the researcher’s ability to describe subtle shape differences or to elucidate the causal effects of profile variables. In the current study, the auricular surfaces of 964 individuals from the United States and Portugal were photographed, outlined, and subjected to elliptical Fourier analysis, principle component analysis, and ANCOVA, to determine which factors influence its shape. The first principal component (PC1) explains 38% of the variation in the shape of the auricular surface and reflects the curvature of the posterior border; it is not influenced by any of the tested demographic variables. PC2 explains 18% of the variation in the shape of the auricular surface and appears to depict the extension of the apex from the central axis and the curvature of the cranial and caudal limbs. This principal component is significantly affected by sex, with male surfaces being straight and “L-shaped” and females being more curved and “C-shaped”. The size of the pelvis is also significant, where wider iliac breadths correspond to a more ‘feminine’ auricular surface. PCs 3 through 5 depict other minor shape differences and are variously affected by the age at death of the individual.

Presentation
Craniometric Variation and Sexual Dimorphism in a Chilean Population

Rachel M. Kreher, Krista E. Latham, and Stephen P. Nawrocki, University of Indianapolis

In forensic anthropology, an understanding of human variation can be used to predict the biological profile (sex, age, and ancestry) of an unidentified decedent. Generating these predictive methods first requires an appropriate reference sample that is used to model the population at large. In the U.S., many forensic anthropology labs utilize FORDISC (Ousley & Jantz, 2005), which uses metric (measurement) data to predict sex and ancestry. However, the reference samples in FORDISC are primarily White and Black, with much smaller samples of Hispanics and other subgroups. This limitation reduces the accuracy of the method when applied to other subgroups. As Hispanics are one of the largest minority groups in the U.S., further investigation into their skeletal traits is warranted. The current research examined craniometric variation in a Chilean sample in order to assess (1) whether sexual dimorphism is similar to other documented samples, (2) the effects of age at death on cranial size, and (3) how Chileans differ from other Hispanic reference samples in FORDISC®v.3.1. Measurements taken from 250 individuals in the Cementerio General Collection located in Santiago, Chile were entered into FORDISC 3.1 to determine how the program would classify them. Additionally, IBM SPSS was used to conduct ANCOVAs on the measurements to determine the simultaneous effects of sex and age. The most dimorphic measurements were then used to create a discriminant
function specific to this Chilean sample. FORDISC’s classifications were only 32% correct for both sex and ancestry, and a disproportionate number of males were misclassified as females. The new discriminant equations correctly classified sex in 81% of cases. Results suggest that Chileans are smaller but more dimorphic than the Hispanic reference sample in FORDISC, thus necessitating population-specific discriminant functions.

Poster

The expression of osteoarthritis in a modern cadaver sample

Chou, Haller, & Nawrocki
University of Indianapolis

Osteoarthritis (OA), a degenerative disease of the synovial joints, is one of the most commonly-observed pathological conditions in human and nonhuman skeletons. While OA has been well-documented in numerous archeological and historic samples, few studies address its occurrence in recent populations, and thus the utility of OA in determining age at death in forensic casework is largely unverified. This study looks at the expression of OA in the shoulders and knees of 38 anatomical cadavers of known sex and age at death (43 to 96 years). Arthritis was scored at each joint using 4-point semi-continuous ordinal scales measuring (1) the severity of peri-articular lipping, (2) the total circumference of lipping, (3) the degree of hyaline cartilage destruction, (4) the degree of subchondral bone exposure, and (5) the degree of subchondral eburnation (‘polishing’). Traits (3) and (4) are not normally available on dry bone specimens. Each trait was assessed individually and also summed with all other traits for each joint to obtain a global arthritis indicator. Because both sides were not available for all decedents, left and right sides were averaged. ANCOVA and ordinal regression were then used to determine the simultaneous effects of sex and age on the severity of arthritis. Results indicate that for both the shoulder and the knee, age is a significant predictor of arthritis severity, with sex playing little or no role. The correlation between age and arthritis severity is much stronger in the right limbs than in the left limbs, indicating a connection with preferential handedness and footedness. Traits that include measures of soft tissue (cartilage) destruction (3 & 4) are more sensitive to increasing age than those traits that reflect primarily bony changes, suggesting that predictions of age from dry bones will only ever be moderately accurate.
The Identify Indiana Initiative: Combining Outreach, Education and Service to Reinvestigate Cold Human Remains Cases in the State of Indiana

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The Identify Indiana Initiative grew from the proposition that many unidentified cold cases in Indiana could benefit from the application of new technologies, such as missing persons databases, DNA profiling, and isotope analysis. A team of volunteer specialists including forensic anthropologists, law enforcement personnel, DNA laboratory technicians, coroners, and National Missing Persons Data System (NamUs) administrators was assembled to reopen these cases, which date back to 1974. The first step in this process is inventorying the unidentified cases in the state. Locating some of the unidentified individuals was straightforward since they are curated at the University of Indianapolis Human Identification Center, while others require exhumation from cemeteries across the state. Skeletal and dental analyses were conducted on the exhumed remains that were buried before forensic anthropologists were practicing in the state. The information from the skeletal examination was combined with data from law enforcement records to create an entry in the Unidentified Persons section of the NamUs database. Skeletal samples were then collected for molecular investigations, including DNA profile generation and isotope analysis. The collaboration process, utilization of new technologies, and challenges faced by the team will be discussed within the context of several examples in which unidentified remains were exhumed and reanalyzed for this initiative. As the scientific tools and technologies of forensic investigations evolve, unidentified human remains should be reinvestigated to increase the chance of identification. Outreach to state county coroners and offering a basic scientific education on new molecular technologies has been essential for team members to facilitate positive identifications on 16% of its cold human remains cases to date.

Poster
Analyzing Soil Fungus in Decomposing Sus scrofa Burials

Arden Mower, University of Indianapolis, Gillian Fowler and Nicola Crewe, University of Lincoln, U.K.

Forensic mycology is a new field that utilizes fungal evidence in forensic investigations. Fungal spores are unique forms of trace evidence that can be recovered from crime scenes, suspects, and victims. In addition, a knowledge of postmortem fungal growth can help to determine the postmortem interval for decomposing remains located on the surface or in a burial. In theory, a different fungal profile
should be obtained for each progressive stage of decomposition. In the current study, an experiment was designed to test the latter application. Four boxes were filled with soil collected from an English woodland. Fully-fleshed, semi-fleshed, and macerated pig legs were buried in these boxes; the macerated leg was cleaned one year prior to the start of this research. These boxes represent the active decay, advanced decay/early stage skeletonization, and late stage skeletonization decomposition stages, respectively. The remaining box was a control, containing no remains. After six months (May to October), the soil was analyzed for the presence of fungal growth. Fungi were isolated from each box and examined under a microscope. Species of interest were noted based on their forensic relevance. One species in particular (class: Zygomycetes) varied in abundance in proportion to the amount of flesh that had decayed. This species therefore has the potential to be a biomarker species for locating decomposed soft tissues, or it could aid in determining the postmortem interval by measuring its biomass and determining how much time it would take to become prominent in the soil with the nutrients present.

Poster

**Skeletal Comparisons of the Upper Limb of Pan troglodytes, Homo sapiens, and Papio papio**

Samantha L. Beck, and Stephen Nawrocki, University of Indianapolis

Comparative atlases of subadult skeletal remains within the Primate order are important but lacking in the fields of anthropology and zoology. Compared to subadults, adult bones contain more identifiable shapes and features that can be used to distinguish species. These features are often poorly expressed in infancy and childhood, so one must use more subtle indicators to differentiate subadults of closely-related species. The aim of this study is to analyze and describe the unique morphological features of the upper limb bones of the chimpanzee (*Pan troglodytes*), baboon (*Papio papio*), and human (*Homo sapiens*) in order to assist with identification in the field and to make interpretations about their functional importance at a young age. A partly-fleshed, articulated infant chimp was processed and compared to baboon and human specimens of similar developmental age. Contrary to what might be expected based on their genetic relatedness, the current study demonstrates that the infant baboon’s and human’s antebrachium and forepaw bones are more similar to each other than either is to the chimpanzee’s. Specifically, the chimp’s forearm and hand bones are more strongly curved even at this early age. In addition, the infant chimp’s humerus displays a wider mix of features: its curvature is more similar to that of humans, but its narrower olecranon fossa is more baboon-like. It is likely that the greater range of extension of the human elbow is stabilized by a broader olecranon. It may be that the chimpanzee’s unique, knuckle-walking locomotor style makes them very different from both baboons and humans, and any
similarities that would be expected from the more arboreal adaptations of non-human primates are superseded by the extreme elongation and compressive loading of the chimp’s forelimb.

Poster
Zooarchaeology Analysis and Contending with Variation in Natural History Collections

Amanda Burtt and Larisa DeSantis

Zooarchaeologists use modern comparative skeletal collections to identify and quantify animal remains from archaeological sites, using modern animal skeletons as analogs for their ancient animal counterparts. Environments have changed and are changing, and this has real implications when we assume that modern animals are exact replicas of ones that existed on long ago landscapes. Zooarchaeologists working with natural history collections must consider animal communities change over time, sometimes within generations for a variety of reasons, including anthropogenic ones. This poster explores this dilemma by examining the dietary behavior of a sample of gray wolves (Canis lupus) culled from Yellowstone within the last 15 years via their dental microwear, measured with Dental Microwear Texture Analysis (DMTA). These microwear data are compared to a similar sized sample of gray wolves from various North Americans localities, from Chihuahua, Mexico to the Northwestern Territories in Canada, all culled decades before the Yellowstone wolves. Research presented here shows that these specific animal communities exhibit dietary behavior variations that are significantly different from each other. Making their use as baseline data for species specific dietary behavior problematic. This project serves as caution for analysts’ that rely on modern animal representations to interpret animal communities in the past.
Botany

Presentation
The Effects of Salicylic Acid on the Hypersensitive Response of Physcomitrella patens

Matthew Hathaway, Butler University
Philip Villani, Butler University

Reactive Oxygen Species (ROS) were originally believed to be toxic to plants and induce apoptosis in cells. New findings have led to the suggestion that ROS act as a secondary signaling pathway for cells and play an important role in generating a hypersensitive response (HR), which is crucial for dealing with cellular infections. The hypersensitive response will lead to a plantwide immune response known as a systemic acquired response (SAR). This discovery warranted further study by observing how plants, specifically mosses, generate ROS in the presence of hormones like salicylic acid (SA). *Physcomitrella patens* is a model species for non-vascular plants and was selected for this study. Elevated levels of hydrogen peroxide indicated that a hypersensitive response was occurring when *P. patens* was placed in SA solution. Using a colorimetric assay, we were able to quantify hydrogen peroxide levels and establish an elevated response curve. This curve showed that there were statistically significant levels of hydrogen peroxide present at several instances during treatment. Utilizing PCR, we were able to see if there was any variance in the gene expression of P. patens species exposed to SA solution.

Poster
Screening *N. crassa* Knockouts for Genes Important to Cold Adaptation

Michael Watters, Jacob Nava, Cody Holmgren, Valparaiso University

Many organisms go through an adjustment in order to adapt themselves to changes in the environment. The fungus Neurospora normally maintains a fixed distribution of branching at extremes of temperatures, while select mutants show altered branching at temperature differences. This helps reveal genes important to Neurospora’s maintenance of homeostasis in the face of changing temperatures. In addition, both Neurospora and E. coli go through transient changes in response to cold shock, providing a second window on the maintenance of homeostasis. Identifying genes important to the maintenance of morphological homeostasis will help improve our understanding of this process. Three groups of knockouts were selected to be screened: genes previously suspected to be involved in hyphal development as well as knockouts resulting in morphological changes; transcription factors; and genes
homologous to E. coli genes known to alter their expression in response to cold shock. These have all previously been examined for their morphological response to cold shock. We conducted a screen of these same mutants for any morphological responses to steady-state growth in the cold vs. standard incubation temperatures. The results, especially compared with the previous cold-shock results, provide an additional window on branching homeostasis in fungi. A total of 344 knockout strains were screened for morphological differences in growth at different temperatures. We report here the morphologies of strains subjected to this screen.

Poster

The Cold Shock Response of *Neurospora crassa*: Influence of light vs temperature

Michael Watters, Kirsten Treptow, Allison Pariso, Megan Kelly, Valparaiso University

The filamentous fungus *Neurospora crassa* exhibits a characteristic, temporary change in morphology when shifted from warmer incubation temperatures into the cold. This change was termed the “cold shock response.” Other workers have suggested that the temporary morphology changes observed were not due to the temperature shift, but were instead a result of changing light conditions. A series of experiments were conducted in order to distinguish between these two competing hypotheses. Eight samples of *N. crassa* were grown at 33°C. Four of the eight were subject to constant light, and the other four were subject to constant dark. These cultures were then shifted to 4°C while maintaining their respective light/dark exposure. The cold shock response of both the “light” and “dark” samples were similar to both conventional cold shock and each other, supported the hypothesis that the cold shock morphological response is due to temperature rather than light changes. These competing hypotheses were further tested by subjecting *Neurospora crassa* growth to a gradient of temperature shifts. Identical stains of *N. crassa* were grown first at temperatures ranging from 37°C to 20°C, before shifting to 4°C. The spacing and appearance of the apical branching characteristic of the cold shock response correlated with the temperature shift despite the lighting conditions being constant. These results further support the conclusion that the previously described cold shock response of *N. crassa* is indeed the result of the temperature shift and not the changing light conditions.
Presentation

It takes a village: Dendrochronological analysis of the Wabash and Erie Canal Village in Delphi, Indiana

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Christopher Baas, Associate Professor, Department of Landscape Architecture, Ball State University
Darrin L. Rubino, Professor, Biology Department, Hanover College

The annual rings of temperate trees provide a long-term record of the conditions in which a tree grew. The conditions affecting a tree over its lifetime produce unique tree-ring patterns that can be assigned precise calendar dates. The method for assigning these calendar dates is known as crossdating and has been used to determine timing of human settlement. We have used tree rings to date six buildings from the Wabash and Erie Canal Park in Delphi, Indiana (Carroll County). This living history village incorporates structures salvaged from Carroll and surrounding counties in the northern region of Indiana. A total of 45 timbers were sampled across all structures with 35 being assigned precise calendar dates. American beech (Fagus grandifolia) and White oak (Quercus alba) were the most commonly sampled (n = 15 timbers), followed by tulip poplar (Liriodendron tulipifera; n = 7) and red oak (Quercus rubra; n = 4). The oldest structure was the Reed-Case House, an ornate Federal-style home that dates to 1844. The youngest structure in the village is the Kuns Cabin, dating to 1866. This structure was the least ornate of the buildings included in the village and was built with red oak and white elm (Ulmus americana). In addition to determining felling dates of the timbers, tree rings can be used to better understand the dynamics of both humans and forests that are no longer on the landscape. For example, the stylized Reed Case house was actually built prior to the more plain Kuns Cabin. By using tree rings to date these buildings we are able to solidify the timeline of construction events and improve the interpretation of the history of these structures. Historic structures such as these are the last remaining repositories of pre-settlement forest information and provide a means of better understanding historic forest functioning.
Tree-ring dating of the Foxx/Jones House, an Italianate farmhouse from the DePauw University Campus (Greencastle, Indiana)

Darrin L. Rubino, Hanover College
Jennifer Everett and James Mills, DePauw University
Christopher Baas, Ball State University

Tree-ring analysis offers a unique opportunity to date the construction of historic buildings of unknown age. The goal of this investigation was to determine the build and modification(s) dates of the Foxx/Jones House which stood on the DePauw University Campus Farm. This unique structure shows evidence of multiple nineteenth century renovations including a transformation to embrace the Italianate architectural style. Since the house underwent extensive modifications, timber samples were obtained from throughout the building (sills, wall studs, roofing members in all rooms). We focused sampling on timbers with a bark or wane edge (rounded outer surface of a timber from which bark has sloughed off or was removed) since the outermost ring in these samples indicates the year in which a tree was felled and subsequently used for timber. We collected samples from five different timber species: tulip poplar, sycamore, red pine, black cherry, and white pine. Crossdating, comparing the tree-ring growth patterns in samples of unknown age with regional reference chronologies (series of accurately dated and measured tree-rings), was performed to accurately assign calendar dates to the tree rings in each timber. We successfully crossdated 49 tulip poplar samples from 45 different timbers to create a 5330-ring chronology that spans from 1589 – 1877. Cutting dates were clustered in 1846, 1851, and 1878. Given the distribution of cutting dates throughout the building, we hypothesize that the house was originally erected as a two-room structure in 1846 and that subsequent modifications in 1851 and/or 1878 led to its final nineteenth-century configuration. The data collected from this project represents a valuable addition to our regional tree-ring database since we were able to create a robust chronology for a previously unstudied geographic region of the state.

Climate-Growth Relationships of Sugar Maple (Acer saccharum Marsh.) along a Latitudinal Climate Gradient in its Western Range

Aaron Rudolph, David LeBlanc, Ball State University

Sugar Maple is an ecologically and economically important species that is widespread throughout eastern North America. As climate change is predicted to modify temperature and precipitation regimes throughout the range of the species, it is important to understand how climate has impacted the growth of sugar maple in the
past to predict how it may be affected in the future. However, little research has been conducted in the western half of the range of sugar maple to determine how climate will affect the radial growth of the species. A dendroecological study was conducted in Michigan, Indiana, Kentucky, and Missouri to determine how past temperature and precipitation regimes have affected sugar maple growth, and how that relationship has changed both spatially and temporally. Sugar maple radial growth correlated strongest with maximum temperature, precipitation, and Palmer Drought Severity Index during the summer period of May-August across all sites. Sugar maple at southern sites correlated stronger during the May-June period while northern sites correlated stronger with the June-August period. The strength of these correlations was also found to change over the last century with certain climate variables becoming stronger influencers of radial growth, while others have weakened. Climate has a stronger impact on sugar maple radial growth in the western part of its range compared to the east. However, differences in climate-growth correlations were not particularly evident across a latitudinal gradient.

Poster
**Using Unmanned Aerial Vehicles to characterize maize canopy reflectance responses to experimental treatments in field-scale research**

Ana Morales, Robert Nielsen, and James Camberato, Purdue University

Collecting data to characterize maize response to fertilizer treatments is an important component of applied field research. When field-scale trials are conducted (experimental areas ranging from 8 to 40 ha), traditional methods of data collection can be challenging. Large plot sizes make it difficult to adequately represent variability in the field without numerous observations and/or samples, which increases the cost of an experiment substantially. The overall goal of our project was to assess the potential of using unmanned aerial vehicles (UAV) to acquire aerial imagery to characterize treatment effects via differences in crop reflectance. To achieve this goal, an experiment examining maize response to sulfur fertilization was used as a case study. Sulfur deficiency reduces plant size and chlorophyll concentration, which alters reflection of red and near infrared wavelengths. The trial, conducted in a 24-ha maize field located in southeast Indiana, included six different sulfur rates, four with sulfur (+S) and two without sulfur (–S) replicated 5 times in a randomized complete block design. Aerial imagery collected from flights at two growth stages (R2 “blister” and R4 “dough”) was used to create whole-field images representing four vegetative indices (VI). Digital values for the VI were extracted and averaged for each experimental unit. Analysis of variance was performed on the data and treatment contrasts were calculated. Three of the VI effectively discriminated between +S and -S treatments at both growth stages, and only one VI did not detect treatment differences at the R2 growth stage. The differences among the S treatments detected
by VI correlated to differences in yield at harvest, with the +S treatments yielding about 1.25 Mg ha-1 greater than the -S treatments. Aerial imagery acquired by UAV was effective in characterizing differences among treatments that translated into grain yield differences at harvest.

**Poster**

**Characterizing the antimicrobial and anticancer activities and several associated bioactive compounds of *Argemone mexicana***

**Teodora Najdeska** - Valparaiso University  
Tj Lefeber - Valparaiso University  
Kelly Davidson - Valparaiso University  
Danielle Orozco-Nunnelly - Valparaiso University

Commonly called the Mexican prickly poppy, *Argemone mexicana* is a stress-resistant member of the Papaveraceae family of plants that has been used in traditional medicine for centuries by indigenous communities in Mexico and Western parts of the United States. This plant has been used to treat a wide variety of ailments, including skin diseases and intestinal infections, with reported antimicrobial and anticancer properties. However, these properties are poorly understood, with no associated bioactive compounds yet identified. Herein, we describe the germination conditions of *A. mexicana* and preliminarily characterize the antimicrobial and anticancer activities of different parts (seeds, leaves, inner vs. outer roots) of the plant. We show that when comparing 1 mg of each sample normalized to background solvent alone, the *A. mexicana* methanol outer root and leaf extracts possess the strongest antimicrobial activity, with greatest effects against the gram-positive bacteria tested, and less activity against the gram-negative bacteria and fungi tested. Additionally, we report that when using the MTT colorimetric assay, the outer root and leaf methanol extracts and the seed hexane extract have pronounced inhibitory effects against T84 human colon cancer cells. Using normal-phase column chromatography and subsequent mass spectrometry analysis of the outer root and leaf methanol fractions, we have begun to chemically characterize several candidate antibacterial compounds. These preliminary results warrant further research into defining the bioactive chemicals produced in the roots, leaves and seeds of *A. mexicana* and are especially significant given the growing global concern of antibiotic-resistant ‘superbugs’ and lack of new antimicrobial and anticancer drug discovery.
Plants respond to wounding with both local and system wide reactions to prepare non-damaged regions to produce defenses. An increase in calcium ions and reactive oxygen species will work both locally and systemically to activate defense hormones in the plant. The production of the defense hormone, jasmonic acid aids in preventing future attacks. This research explores the immune response of a soybean leaf when the opposite leaf is mechanically wounded. Using an abiotic stress responsive transgenic reporter line of soybean, we demonstrate the timing of systemic immune response. In these transgenic lines the promoter driving the reporter protein GUS, responds to jasmonic acid. Thus, as jasmonic acid increases, reporter activity will increase. Soybean unifoliate leaves are mechanically wounded using a standard one-hole holepunch. One leaf receives two holes that are parallel to each other on opposite sides of the midrib. Both the wounded leaf and the opposite, unwounded unifoliate leaf are collected over a time course to determine the timing of systemic immune signaling. Leaves were flash froze in liquid nitrogen and the sample tissue was crushed using a plastic pestle dipped in liquid nitrogen. Soluble protein was collected by homogenization of the leaf powder in GUS extraction buffer. GUS enzymatic activity was measured via fluorometric analysis. Preliminary data demonstrates an increase in GUS activity in the non-wounded leaf between the 24 and 48 hour mark. Finer scale resolution of the timing of this response is being investigated.
In a living cell, protein degradation is as important as protein synthesis. Protein degradation is necessary to recycle proteins that are only needed for a short time (such as proteins involved in DNA replication), discard damaged or misfolded proteins, or remove proteins that behave aberrantly. One way in which proteins behave aberrantly is by persistently engaging a cellular channel called the translocon. The translocon is a protein channel that allows proteins to cross the endoplasmic reticulum membrane. Hrd1 is a ubiquitin ligase that targets proteins that clog the translocon for degradation by ligating multiple copies of a protein called ubiquitin to clogging proteins. This process is conserved in yeast and man. Ubiquitin ligases rarely function without the assistance of additional proteins. Further, loss of Hrd1 does not completely prevent the destruction of translocon-clogging proteins. Therefore, we hypothesized that other proteins function with Hrd1 or in parallel mechanisms to unclog the translocon. A previous master’s student performed a yeast growth-based genome-wide screen to identify genes required for protein degradation at the translocon. This screen identified ~150 genes that behave similarly to HRD1. Because large-scale screens may yield false positives, we performed small-scale growth assays to validate roles for identified genes in protein degradation. Our small-scale growth assays support potential roles for 28 genes in regulating abundance of translocon-clogging proteins. We are currently performing cycloheximide chase and western blot analyses to determine abundance and degradation kinetics of translocon-clogging proteins in yeast lacking the validated genes, as compared to wild-type cells and cells lacking HRD1. Since a protein found in low-density lipoproteins (“bad cholesterol”) clogs the translocon under certain circumstances, validated genes may represent therapeutic targets for elevated cholesterol.
The impact of different forms of ER stress on quality control at the translocon

Courtney L. Broshar (Ball State University), Bryce W. Buchanan (Ball State University), Mark Hochstrasser (Yale University), Eric M. Rubenstein (Ball State University)

Endoplasmic reticulum (ER) stress arises when ER-resident proteins misfold or when ER lipid homeostasis is perturbed. Cells respond to these conditions by activating multiple mechanisms which reduce ER stress, including the unfolded protein response (UPR), the ER surveillance (ERSU) pathway, the stress-induced homeostatically regulated protein degradation (SHRED) pathway, and the rapid ER stress-induced export (RESET) pathway. The ubiquitin-proteasome system (UPS) is responsible for the majority of degradation of misfolded and aberrant proteins at the ER (such as those arising during ER stress) through ER-associated protein degradation (ERAD).

Multiple diseases are associated with elevated levels of ER stress, including some forms of cancer, neurodegeneration, and heart disease. Understanding how the cell responds to different sources of ER stress may inform the treatment of such diseases. We investigated the effects of ER stress caused by protein misfolding and disruption of lipid homeostasis on ERAD of translocon-associated proteins (ERAD-T) in Saccharomyces cerevisiae. Addition of the chemicals dithiothreitol (DTT) and tunicamycin induce ER stress by causing proteins to misfold. By contrast, inositol limitation induces ER stress by altering the lipid composition of the ER membrane. We observed that DTT and tunicamycin impaired ERAD-T. On the contrary, ERAD-T was unaffected by inositol limitation. We tested whether known ER stress-sensing pathways are required for ERAD-T or its impairment during ER stress. However, none of the characterized ER stress-sensing pathways tested were required for ERAD-T or its impairment by ER stress. These results suggest the existence of a novel ER stress-responsive mechanism that impairs ERAD-T.

Immunostimulatory Effects of Basil on the Proliferation of Spleen Cells in Mice and Fish

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Basils, native plants of central Africa and Southeast Asia, have been used in traditional medicine and in worship for hundreds of years in Indian traditions. There are mentions of them in various religious scriptures and traditions. They are known for their anti-tumor, anti-bacterial, and immune cell proliferating abilities. On the other hand, Thai basil, a specific member of the basil group, is primarily known for its
usage in cooking all over the world. Its medicinal use is not as well explored. Due to the rise in human population, there is a high demand for protein. This leads to the usage of antibiotics as a preventative medication in farming protein producing animals. The over use of antibiotics could lead to bacterial resistance which could cause hazards for human health. Nutraceuticals are, therefore, being currently used as alternatives to drug treatments. Different species of basils, as nutraceuticals, can be grown in an aquaponics system and can be used as food for human consumptions as well as immunomodulators for fish. Basil mixed fish food would help the fish immunologically and the basil plants would keep the water clean for the fish thus eliminating the ammonia from the rearing system making aquaponics farming more efficient. In this experiment, we therefore, wanted to see the immunomodulatory effects of basils (Holy basil and Thai Basil) by measuring the proliferation of spleen cells from mice and fish (Nile tilapia) in-vitro using different concentrations of the plant extracts of these two species. We hypothesized that there would be a greater proliferation when the spleen cells are exposed to Holy Basil extracts compared to the Thai Basil extracts. This is due to the fact that, Holy basil has a history of being used in medicine and Thai basil is mostly used for culinary purposes. Results are now being compiled and will be presented at the meeting.

Poster

**Loss of STE24 sensitizes yeast to defects in protein quality control**

**Molly K Dolan, Avery M Kirshbaum, Eric M Rubenstein**

From yeast to humans, regulated protein degradation is as important as protein synthesis. Conditions ranging from hypercholesteremia to premature aging are linked to ineffective protein degradation. The endoplasmic reticulum (ER) is an epicenter for both protein synthesis and degradation in eukaryotic cells. Enzymes involved in protein synthesis and degradation are present at the ER membrane. Proteins entering the ER pass through a channel called the translocon. Cells have developed multiple quality control mechanisms at the translocon, whereby proteins stalling in the channel are removed and degraded. The ubiquitin ligase Hrd1 targets a subset of proteins that stall in the translocon for proteasomal degradation. Likewise, the protease Ste24 also cleaves other translocon-stalled proteins. Despite the important roles of Hrd1 and Ste24, individually deleting the genes that encode either has no effect on yeast growth rate. However, combined loss of Ste24 and Hrd1 slows growth. Since both enzymes target stalled proteins at the translocon, we hypothesized that this growth phenotype was due to loss of Hrd1 and Ste24 function at the translocon. However, because Hrd1 and Ste24 have other roles, it was also possible that the phenotype was due to loss of Hrd1 and Ste24 function outside the translocon. To determine if the phenotype is dependent on Hrd1’s translocon-associated function, yeast were engineered to lack both Ste24 and Usa1, a cofactor needed for Hrd1 to target aberrant ER luminal or
transmembrane proteins but not translocon-associated proteins. To determine if compromised quality control generally impairs growth when Ste24 is absent, we created strains lacking Ste24 and either protein quality control enzyme Doa10 or Rkr1. The slow growth phenotype persists in strains that lack Ste24 and any of these quality control proteins. Thus, loss of Ste24 sensitizes yeast to defects in multiple protein quality control pathways.

Poster

**Rifampin and ML141 Cotreatment Enhances Clearance of Intracellular Staphylococcus aureus**

**Melissa D. Evans**, Robert E. Sammelson, and Susan A. McDowell, Ball State University

*Staphylococcus aureus* is a leading cause of infective endocarditis, skin and soft tissue infections, and bacteremia worldwide. *S. aureus* can invade host cells and reside within the intracellular environment, providing protection from antibiotic therapy and host immune defenses. Most first-line antibiotics are not readily able to penetrate host cells to kill intracellular bacteria, making treatment of intracellular *S. aureus* infections particularly difficult. ML141 is a small molecule that decreases intracellular *S. aureus* infection by inhibiting invasion of *S. aureus* into host cells. Rifampin is an antibiotic capable of killing intracellular and extracellular bacteria, but high dose treatment is associated with rapid development of antibiotic resistance and adverse side effects, such as thrombocytopenia. We hypothesized that ML141 would reduce the number of bacteria invading host cells and therefore a lower concentration of rifampin would be needed to clear the bacteria that do invade the host cells. To study this hypothesis, we determined the concentration of rifampin needed to decrease the number of viable *S. aureus* by 50% (IC50). We then assessed the use of ML141 with rifampin. Cotreatment with ML141 and rifampin decreased the number of intracellular bacteria relative to using ML141 or rifampin alone. We plan to investigate whether the decreased number of intracellular bacteria is due to the cotreatment decreasing host cell viability or ML141 enhancing the bactericidal activity of rifampin. If cell viability is maintained and bactericidal activity is not enhanced, then our hypothesis is supported, and the data indicate a novel approach for treating intracellular *S. aureus* infections.
G protein-coupled receptors (GPCRs) regulate diverse cellular processes. FSHR-1 is a conserved GPCR controlling metazoan gonad function, but is also expressed in nervous systems. FSHR-1 regulates signaling at the *C. elegans* neuromuscular junction (NMJ) where a balance of excitatory acetylcholine (ACh) and inhibitory GABA signaling controls muscle contraction. Loss-of-function *C. elegans* fshr-1 mutants exhibit reduced muscle contraction, which is rescued by fshr-1 re-expression in either neuron type. However, the cell type(s) where FSHR-1 is required and the intracellular pathway 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 for neuromuscular signaling. We hypothesized that in *C. elegans* GABA and ACh neurons, FSHR-1 controls muscle contraction using the GSA-1/ACY-1/PKA pathway to regulate synaptic vesicle release. To test cell type-specific differences, we performed RNA interference to knock down expression of FSHR-1 pathway components in *C. elegans* GABA or ACh neurons and analyzed effects on muscle contraction. Knockdown of pathway components in ACh neurons caused decreased muscle contraction with statistically significant decreases seen with knockdown of the synaptic scaffold UNC-10. Knockdown in GABA neurons caused increased muscle contraction with statistically significant increases seen with knockdown of UNC-10 or ACY-1. These data suggest the FSHR-1 pathway promotes synaptic vesicle release in either neuron type. We are now assessing requirements for pathway components in cell type-specific FSHR-1 overexpression backgrounds. Results will provide insight into how FSHR-1 controls NMJ function, aiding understanding of GPCR biology and neuronal signaling.
A general role for the Hrd1 ubiquitin ligase in regulating the abundance of translocon-associated proteins

Avery M. Kirschbaum, Ball State University
Molly K. Dolan, Ball State University
Eric M. 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, translocon-associated quality control mechanisms have not been fully elucidated. A better understanding of the mechanisms that clear aberrantly clogged translocons could lead to improved therapeutic approaches for various diseases, including hypercholesterolemia. In yeast, the ubiquitin ligase Hrd1 ubiquitylates the model translocon-clogging protein Deg1-Sec62, thereby targeting the protein for degradation. A mammalian homolog of Hrd1 targets the cholesterol-related protein apolipoprotein B (apoB) for degradation when it persistently engages the translocon. However, the generality of Hrd1 function at the translocon is not clear. We investigated the role of Hrd1 in the degradation of an engineered translocon-clogging protein that is structurally unrelated to Deg1-Sec62 and apoB. We found that Hrd1 promotes the turnover of this engineered protein. Further, under particular conditions, yeast lacking Hrd1 exhibit a growth defect, which is rescued by translocon perturbation. These results are consistent with the important and general role of Hrd1 in maintaining proteostasis. Further, a larger number of translocon-associated substrates of Hrd1 will facilitate future investigations of the molecular details of Hrd1-mediated quality control.

Poster
Investigation of G protein-coupled receptor FSHR1 in regulation of UNC-10 RIM synaptic localization in C. elegans

Morgan Buckley and Jennifer R. Kowalski
Butler University

Regulation of neuronal signaling is essential for nervous system function. 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 and regulate neurotransmitter release. Follicle Stimulating hormone receptor (FSHR) is a conserved GPCR present in the
nervous system of *Caenorhabditis elegans*, where it regulates neuromuscular signaling. Inhibition of *fshr-1* expression causes reduced muscle contraction and synaptic vesicle accumulation at motor neuron presynapses in worms, likely reducing neurotransmitter release. The specific pathway activated by FSHR-1 to affect synaptic transmission is unknown. UNC-10 (RIM in mammals) is a candidate FSHR-1 effector that regulates synaptic vesicle release at the *C. elegans* neuromuscular junction. To determine whether FSHR-1 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 (GFP), were imaged on a fluorescence microscope and the level of UNC-10 abundance measured. *fshr-1* mutant worms exhibited elevated levels of UNC-10::GFP intensity and increased axonal fluorescence (p = 0.02, 0.02) at cholinergic presynapses. However, UNC-10::Cherry expression in GABAergic neurons of *fshr-1* mutants showed no significant differences from those of wild type animals, in (p>0.05). Together, these data suggest *fshr-1* may be required to specifically restrict the localization of UNC-10 to synapses in cholinergic neurons. Given the conservation of *C. elegans* and human genes and nervous system organization, determining the function of FSHR-1 may aid understanding of human neurological disorders.

Poster

**In-vivo studies on slowing the progression of Idiopathic Pulmonary Fibrosis**

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Fenghua Zhang, Department of Chemistry, Purdue University  
Philip S. Low, Department of Chemistry, Purdue University

Idiopathic Pulmonary Fibrosis (IPF) is a chronic lung disease characterized by scarring of the lungs. The pathogenesis of IPF is not completely understood which makes it difficult to find a cure for the disease. Nintedanib and Pirfenidone are two FDA approved drugs which show some positive effect on the treatment but do not completely cure IPF. Macrophages play an important role in fibrosis. It is commonly believed that the phenotype of profibrotic macrophages are like the M2 subtype, whereas the anti-fibrotic population are more like M1. Based on previously conducted in-vitro studies, the Toll like receptor-7 agonist was successful in reprogramming the M2 macrophages to the M1 subtype. The in-vitro study results led to in-vivo studies being conducted using the TLR-7 agonist. This agonist targets the folate receptor beta + macrophage to slow down the progression of Pulmonary Fibrosis in a C576L6 mice model. Black male mice are injected intratracheally with a single dose of bleomycin, inducing lung fibrosis, and then treated with the TLR-7 agonist. The main end points include survival rate and progression of fibrosis. By comparing with the disease control and healthy control, our compound can increase the survival and decrease the progression of fibrosis determined using the hydroxyproline assay, H&E staining and
trichrome staining. Ongoing in-vivo studies are experimenting on drug dosage and frequency, to receive the optimal regimen for the treatment of IPF. These further studies could lead to a potential treatment for IPF.

Presentation

**Determining the Relationship of XIAP and TAL-1 in T-Cell Acute Lymphoblastic Leukemia Chemotherapeutic Resistance**

**Joseph Klahn** and James Olesen

T-cell acute lymphoblastic leukemia (T-ALL) accounts for 15-25% of all acute lymphoblastic leukemia cases, with an estimated 5,960 new cases in 2018. T-ALL patients have a 68.1% survival rate, yet treatment options are still limited. T-ALL cells resist chemotherapy treatments, and the cellular mechanism behind this resistance is unknown. One protein that may be involved in this resistance is TAL-1, which is a mutated and ectopically expressed transcription factor in T-ALL. Additionally, X-linked inhibitor of apoptosis (XIAP) is another protein that may play a role in the malignant progression by preventing apoptosis. It is suggested that this protein may have altered expression levels, but it is unknown if it is involved in the resistance to chemotherapeutic drug treatments, or is potentially influenced by TAL-1. This research aims to quantify the relative expression levels of XIAP in Jurkat cells resisting chemotherapy treatments, determining if the removal of this protein decreases cell viability, and if there is a localization change in this protein. To determine the levels of XIAP from cells treated with various concentrations of etoposide, a Western blot analysis will be completed. Additionally, silencing XIAP and quantifying its relative expression levels in relation to the silencing of TAL-1, will help determine if XIAP is influenced by TAL-1 misexpression. Furthermore, immunofluorescence will confirm the localization and expression patterns of XIAP and TAL-1. Classifying the oncogenic activity of TAL-1 has been relatively under-researched thus far, providing a platform to research if TAL-1 affects XIAP expression, thus preventing apoptosis. This research is the first attempt to characterize this relationship and could identify XIAP as a biomarker for earlier diagnosis of T-ALL, or even suggest TAL-1 or XIAP to be potential treatment options in the future.
Examining the role of FSHR-1, PKA, and potential downstream components in controlling neuromuscular signaling at the NMJ in *Caenorhabditis elegans*.

**Abigail Shores**, Jennifer Kowalski, Butler University

A tightly regulated balance of excitatory and inhibitory (E:I) neuronal signals is essential for nervous system function. E:I imbalances occur in many diseases like epilepsy and autism. One way neurons regulate this balance is via G-protein coupled receptors (GPCRs). One GPCR, FSHR-1, controls E:I balance at the neuromuscular junction (NMJ) in *Caenorhabditis elegans* roundworms. At this synapse, excitatory acetylcholine (Ach) signaling and inhibitory gamma-aminobutyric acid (GABA) signaling control muscle contraction. Our previous data suggest FSHR-1 activates a Ga/ACY-1/Protein Kinase A (PKA) pathway in motor neurons to promote synaptic vesicle release and muscle contraction. PKA regulates many proteins by phosphorylation; however, the specific targets of PKA in the context of FSHR-1 NMJ signaling are unknown. We hypothesized that PKA promotes signaling at the NMJ downstream of FSHR-1 by activating synaptic proteins that may include Ca2+ and/or K+ channels, which are required for synaptic vesicle release. We used behavioral assays to measure the effects of mutations in candidate synaptic PKA targets on net signaling for muscle contraction. Loss of function (lf) mutants in *egl-36* K+ channels showed increased muscle contraction, whereas *unc-2* (lf) Ca2+ channel mutants showed decreased contraction. This suggests the corresponding genes for these proteins are essential for normal muscle contraction and E:I balance. Current experiments are using double mutants analyses to test whether these potential PKA substrates act in a genetic pathway with *fsshr-1*. Given the similarity between worm and human genes and nervous systems, this research provides insight into the molecular mechanisms by which GPCRs like FSHR-1 control neuronal signaling.
Coordination of G4R1, RNAPII, and AGO proteins to regulate the transcription of developmental genes and proto-oncogenes

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Regulation of developmental gene and proto-oncogene transcription is a complex process involving a diverse array of proteins. These proteins tightly regulate developmental gene and proto-oncogene expression to prevent uncontrolled, cancerous cell growth and division. One of the many layers in which the expression of these genes is regulated is through G-quadruplex (G4) structures. G4 structures form within guanine-rich regions of DNA and RNA. The guanines self-associate into vertically-stacked planar tetrads, stabilized via Hoogsteen hydrogen-bonding and coordinate bonding with a monovalent cation. The human genome contains 716,310 putative G4 motifs, which are present in upwards of 40% of gene promoters. The degree of enrichment of promoter G4 structures has been shown to correlate with the function of the gene. For example, proto-oncogenes and developmental genes are especially enriched with G4 structures in contrast to tumor suppressor and housekeeping genes. Promoter G4 structures negatively regulate transcription via impeding the transcriptional machinery. The cell can overcome this barrier by using G4 helicases such as G4 Resolvase1 (G4R1) (aliases DHX36 and RHAU), which unwinds G4 structures. AGO2 and RNAPII are also key proteins involved in transcriptional regulation and are binding partners with G4R1. We hypothesized that G4R1, RNAPII, and AGO2 function together to regulate the transcription of developmental genes and proto-oncogenes. We tested this hypothesis using a chromatin immunoprecipitation on chip (ChIP-chip) with antibodies specific to G4R1, RNAPII, and AGO proteins (AGO). We found that G4R1, RNAPII, and AGO binding sites overlap with genes involved with developmental processes, pattern specification processes, and cell fate commitment. Furthermore, under serum starvation, the three proteins co-localized to the enhancer of C-MYC. These data suggest that G4R1, RNAPII, and AGO function together to regulate the transcription of developmental genes and proto-oncogenes.
Posters

**Lippia origanoides extract significantly decreases the viability of MDA-MB-231 triple negative breast cancer cells by inhibiting metabolic pathways**

**Rodrigo Mohallem Ferreira** [1], Vishak Raman [1], Ignacio Camarillo [1,2]

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Breast cancer is the most common type of cancer worldwide in women, affecting about 1 in 8 females in the U.S. The most aggressive subtype of breast cancer, triple negative breast cancer (TNBC), is typically resistant to conventional therapies that target hormone receptors. Previous studies have shown that a natural extract from *Lippia origanoides* possesses significant anti-cancer properties. In this study, we confirm the concentration-dependent decrease in viability of MDA-MB-231 cells treated with L42, as well as a significant increase in cleaved caspase 8 in MDA-MB-231 cells treated with L42, demonstrating the activation of the extrinsic pathway of apoptosis, and thereby explaining the decrease in cell viability. We also show that the main metabolic pathways were being targeted by the extract. In this study, we provide evidences that L42 is a potential source of bioactive compounds that could be used for alternative treatment for TNBC.

**Presentation**

**Localization and functionality of the SUMO conjugating enzyme UBC-9 in C. elegans neuromuscular signaling**

**Elly B. Mawi**, Victoria A. Kreyden, Kristen M. Rush, Morgan K. Harrison, and Jennifer R. Kowalski, Butler University

The nervous system relies on tight regulation of proteins to maintain a balance of excitatory to inhibitory signaling to ensure proper function. SUMO (small ubiquitin-like modifier) polypeptides are attached to proteins by the UBC-9 conjugating enzyme to regulate processes including neuronal signaling. Multiple neuronal proteins are SUMOylated, including several implicated in neurodegenerative diseases; yet, the mechanisms by which SUMOylation affects the balance of neuronal signaling remain unknown. We are investigating how SUMO enzymes control synaptic transmission in *Caenorhabditis elegans* at the neuromuscular junction (NMJ), a specialized synapse where a balance of excitatory (acetylcholine) and inhibitory (GABA) signaling from presynaptic motor neurons controls postsynaptic muscle contraction. We hypothesized that UBC-9 acts in presynaptic motor neurons to control signaling via its SUMO-conjugating activity. We used behavioral assays, loss-of-function and overexpression studies, and fluorescence imaging to examine how UBC-9 acts in
specific neuronal cell types to control muscle contraction. Neither overexpression nor inhibition of UBC-9 in cholinergic neurons affected neuromuscular activity. However, knockdown or overexpression of UBC-9 in all neurons or exclusively in GABA neurons caused muscle hypercontraction. Thus, proper UBC-9 levels in GABA neurons appear required to control NMJ signaling. Worms expressing a catalytically inactive mutant of UBC-9 in GABA neurons also displayed increased muscle contraction, suggesting SUMO-conjugating activity may not be required for UBC-9's effects. Imaging results indicate that both green fluorescent protein tagged UBC-9 (GFP::UBC-9) or SMO-1 (GFP::SMO-1) can localize with red synaptic vesicle proteins at inhibitory neuromuscular presynapses. Current studies are investigating the role of UBC-9's catalytic site for UBC-9's effects at the NMJ. Given the similarities between C. elegans and mammalian nervous systems, understanding how SUMO enzymes control the balance of normal neuronal signaling may provide critical information related to human neurobiology and diseases of imbalanced signaling.

Poster
Investigation of ccar-1 and lst-4 as stem cell Notch target genes

Nicholas Fox, Hunter Jones, Erika Sorensen-Kamakian

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 is the only known pathway that functions to maintain germline stem cells (GSCs) in an undifferentiated and totipotent state (capable of differentiating into all cell types) by promoting the expression of target genes that function in stem cell maintenance. The maintenance of the GSC population is critical because they ensure that a continuous supply of gametes (eggs and sperm) are made which can give rise to the subsequent generation. Therefore, GSCs are directly tied to an animal’s fitness. Although numerous studies support the role of Notch signaling in GSC maintenance, the identification and characterization of critical downstream target genes and how they function to support animal fertility remains largely unexplored. Here we report an initial characterization of apoptotic (cellular suicide) genes ccar-1 and lst-4 as Notch target genes. Loss-of-function analyses, using RNA interference or genetics mutants, indicate that these genes are important for maintenance of proliferative germ cells (where stem cells reside), normal brood size, embryonic viability, and effective egg-laying. We propose that expression of ccar-1 and lst-4 via Notch signaling is critical for the creation of functional eggs. Given that germline apoptosis is an important mechanism in egg development in many animals including humans, we propose that C. elegans may present a simplified model for how Notch signaling and its effectors regulate egg development.
Presentation

**Determining Potential Roles of G-quadruplex Helicases in ALS**

**Antonio Chambers, Philip J. Smaldino**

Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disease. This disease is relentlessly progressive, leading to the inability to eat, speak, move, and breathe. There is no cure, and 80% of all people with ALS will succumb to the disease within 5 years of diagnosis. A better understanding of the molecular underpinnings of ALS is necessary for the development of effective future therapies. A dramatic increase in the number of GGGGCC-repeat units in the first intron of the C9orf72 (C9) gene causes the most common subset of familial ALS. Healthy individuals harbor between 2 and 30 repeats, while C9 ALS patients typically have >30 and often hundreds to thousands of the repeat unit. The guanine (G)-rich nature of the C9-repeat sequence allows it to fold into secondary DNA structures, termed G-quadruplexes. Expansion of C9-repeat DNA leads to transcription of truncated C9 RNAs that aggregate into toxic foci and sequester essential RNA-binding proteins. C9-repeat RNAs are translated into toxic dipeptide repeats, and the net effect is neuronal death. Therefore, C9 ALS is foundationally a DNA G-quadruplex disease, and identifying the proteins that modulate these structures is essential for understanding and treating C9 ALS. The enzyme that accounts for the majority of all G-quadruplex helicase activity is G4 Resolvase 1 (G4R1) (aliases: RHAU, DHX36). We hypothesize that G4R1, as the major G-quadruplex helicase, is upregulated in C9 ALS. We are testing this hypothesis by comparing G4R1 expression in ALS patient cell lines compared to age-sex matched controls. Our results indicate a significant increase in G4R1 expression in C9 ALS cell lines compared to matched controls. These findings demonstrate a potential role for G4R1 in C9 ALS and further investigation is being conducted.

Poster

**Depletion of lin-9 and mcm-6 cell cycle genes causes Notch signaling loss of function phenotypes**

**Joseph Ballard, Sophieara Koy, Erika Sorensen-Kamakian**

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 while most components in known biological signaling pathways are conserved. Given their small size, rapid life cycle, and transparent body, C. elegans is an excellent system for gene discovery. C. elegans exists primarily as a hermaphrodite that can generate both gametes (oocytes and sperm). The germline (the lineage of cells responsible for producing gametes) 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. A specialized microenvironment, a niche, maintains stem cells in an undifferentiated, self-renewable, and totipotent (capable of giving rise to all cell types) state. In *C. elegans*, niche-initiated Notch signaling is the only pathway known to maintain totipotent GSCs and functions to maintain normal ovulation. However, key molecular effectors are largely unknown. In the study, we investigated if cell cycle genes *mcm-6* and *lin-9* function as Notch signaling molecular effectors in GSCs and the somatic gonad by analyzing their expression pattern and loss-of-function phenotypes via RNA interference. Loss of these putative effectors results in smaller germlines, reduced number of proliferative cells (where stem cells reside), polyploid oocytes, embryonic lethality, and defective egg laying. We propose that Notch effectors *mcm-6* and *lin-9* have multiple roles in fertility: the maintenance of the GSC pool to generate gametes and roles in the somatic gonad to promote normal ovulation.

Presentation

**Investigation of SYD-2 Liprin-alpha Ubiquination by the Anaphase-Promoting Complex in C. elegans Motor Neurons**

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Daniel Lester, Butler University  
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Proper nervous system function relies on a tight balance of excitatory to inhibitory (E:I) signaling, which is regulated by the activity of numerous synaptic proteins. Ubiquitination (ub) is one process used by all cells to control protein levels and activity. Ub and E:I signaling imbalances occur in several neurological disorders. We use the neuromuscular junction (NMJ) of *Caenorhabditis elegans* roundworms as a model to study E:I balance. At the *C. elegans* NMJ, excitatory signaling (Ach) causes muscle excitation and inhibitory signaling (GABA) causes muscle relaxation. We previously used aldicarb assays, which measure overall muscle contraction, to characterize the role of a conserved ubiquitin ligase, the Anaphase-promoting complex (APC), in controlling E:I balance in *C. elegans* GABA motor neurons. Our data support a model in which the APC indirectly promotes GABA release, preventing excess muscle contraction by down-regulating an intermediate substrate. SYD-2 is one potential APC substrate that localizes to presynaptic sites and contains APC recognition sequences (D-boxes). Loss-of-function (lf) *syd-2* mutants, as well as double *apc; syd-2(lf)* mutants, have reduced muscle contraction, suggesting SYD-2 works downstream of the APC. Imaging experiments demonstrated GFP::SYD-2 accumulation at GABA neuron pre-synapses in *apc (lf)* mutants, consistent with negative regulation of SYD-2 by the APC. Synaptic levels of GFP::SYD-2(delta Dbox) were unchanged between wild-type and *apc(lf)* animals. Hence, in the absence
of the APC or D-boxes, SYD-2 levels increase. Current experiments are determining whether SYD-2 is ubiquitinated by the APC at the NMJ. If so, ub-SYD-2 levels should fall and overall SYD-2 abundance should increase in *apc(lef)* mutants. Thus far, a Ub-SYD-2 band has been identified, but additional controls are needed to ensure the band is specific. Biochemical experiments are underway using GFP-Trap Beads and Tandem Ubiquitin Binding Entities (TUBES) to isolate ub-GFP::SYD-2 and Western blotting for protein identification.

Poster

**Defining Critical Differences between Epigallocatechin Gallate and Matcha Tea as Therapeutic or Dietary Supplements in Cancer**

Meghan VanWanzeele, Keeley Cleghorn, Catherine E. Steding, Indiana State University

*Epigallocatechin Gallate* (EGCg) is the active ingredient of Matcha tea which allegedly has the ability to fight against the growth of cancer cells by inhibiting proliferation and inducing apoptosis. Though there have been extensive studies supporting the ability of EGCg to affect cancer cells, little has been done putting it in the context of the whole tea with other ingredients such as caffeine. Doing so allows for a more complete understanding of the tea’s effects on cancer growth, and not just EGCg. With the over 600,000 people expected to die from cancer within this next year, it is important to tests these claims to insure that the fears of a deadly disease are not being used to take advantage of vulnerable people. The study of EGCg in conjunction with other tea ingredients will determine the drink’s potential as an anti-cancer method that can be adopted by those wishing to reduce their risk. While preliminary results utilizing isolated EGCg show promise in terms of the antioxidant’s ability to reduce growth, the levels far exceed that which is physiologically relevant for average tea consumption. This study indicates that there are potentially significant limitations to the consideration of Matcha tea as a dietary supplement in cancer therapeutics.

Poster

**The Essential In Vitro World of Cancer Cells**

Nykara Brown, Danielle Muse, Keeley Cleghorn and Catherine E. Steding, Indiana State University

Cell culture remains of critical clinical significance. *In vitro* analysis provides the framework for both understanding and combating several key features of cancer cell behavior and growth. The artificial model system that culture techniques are centered on functions to supply cancer cells with the nutrients and environment necessary to
survive extensive periods outside of the body. As such, the techniques we use in culture have the ability to directly impact the results we analyze, and serve as a primary, and in some instances only, foundation for in vivo expectations. While the widespread use of “favorable conditions” allows us to achieve maximal results, there are many potential consequences for culture conditions that have yet to be fully defined. Work in our laboratory has demonstrated differential short-term and long-term effects of utilizing variations in growth conditions previously thought to be relatively innocuous. The short-term behavioral changes observed included: altered rate of growth, overall viability, and rate of colony formation. These specific changes become indistinguishable after longer periods of maintenance in differential serums. Other changes have been shown to be maintained and to significantly impact overall cellular behavior. These results suggest a potential role for differential gene expression and modified cellular activity capable of allowing the cells to acclimate to various culture conditions. Determining the factors involved in this acclimation has the potential to provide researchers with a better understanding of cellular behavior, the discrepancy between in vitro and in vivo results, and the very nature of cancer development and progression.

Poster

Protein Expression and Apoptotic Activity of HSP27 in Jurkat Cells

Emily Horning and James Olesen, Ball State University, Muncie, Indiana 47306

T-Cell Acute Lymphoblastic Leukemia (T-ALL) is a type of blood cancer that emerges first in lymphocytes, a type of white blood cell that is central to the immune system. Clinically, this aggressive cancer spreads rapidly in patients and there is little known of its cause. T-ALL is thought to arise from abnormal expression of TAL-1, a protein transcription factor. The impact of ectopic expression of TAL-1 is not completely understood but it has been suggested that other genes and their protein products are also affected. To this end, expression of heat shock protein (HSP27) was analyzed. HSP27 is a heat shock protein that is upregulated due to cellular stress and has also been shown to block or inhibit apoptosis or cell death. HSP27 has anti-apoptotic activity, which can lead to the survival and potential over-proliferation of cells actively expressing this protein. In this study, whole cell lysates were isolated from untreated and etoposide-treated (1uM, 5uM) Jurkat cells. Additionally, Jurkat cells were transfected with a siRNA molecule against HSP27, effectively silencing HSP27 within the cell. Expression levels of HSP27, either prior or after silencing, were then assessed by Western blot analysis and immunofluorescence. Immunofluorescence was completed to observe any changes in protein localization and expression patterns within the cells. If altered expression of HSP27 results in decreased cell viability across various etoposide concentrations, this may suggest HSP27 as a potential therapeutic target to help combat T-ALL.
Role of NADPH oxidases in growth and guidance of zebrafish retinal ganglion cells

Saron Bhoopathy, Aslihan Terzi, Haley Roeder, and Daniel M. Suter

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The NADPH oxidase (Nox) family of enzymes is composed of membrane-bound, multi-subunit proteins which produce reactive oxygen species (ROS). Though ROS are typically associated with oxidative stress, there has been emerging evidence that ROS regulate neuronal development. It is, however, presently unknown whether Nox enzymes act downstream of growth and guidance cue receptors. Previous work in the lab has shown that neurite growth and guidance in response to specific guidance cues is abolished when Nox enzymes are inhibited or mutated. In order to test the hypothesis that growth cone ROS levels are altered in response to guidance cues, we will express the ROS biosensor roGFP2-orp1 in developing zebrafish embryos. We will then culture retinal ganglion cells (RGCs) from these zebrafish embryos and treat them with the guidance cues netrin-1, BDNF, and slit2. It is hypothesized that guidance cues alter ROS levels in the RGCs and that this would be hindered when Nox enzymes are inhibited.

Effect of Estrogen on Morphological Change in Candida albicans and Neurospora crassa

Idalia Zachara, Paige Camp, Hamza Hasan, Chase Jones, Michael Watters and Patrice Bouyer. Valparaiso University

Introduction:
Within the human intestine, opportunistic pathogens, like Candida albicans (C. albicans), live in symbiosis with the host. Under certain stresses, C. albicans can change its morphology from yeast to filamentous and invade the body. During sepsis, estrogen levels dramatically increases, and are associated with poor patient outcome. We hypothesize that high estrogen levels may cause filamentous growth in C. albicans. Morphological changes in the filamentous fungus, Neurospora crassa, have been well characterized therefore, in addition to C. albicans we are using Neurospora as a model to test gut environmental factors on morphology.

Methods:
C. albicans, was grown on liquid media YEPD, whereas Neurospora was grown on agar plates. Estrogen (1 µM, 1 nM, and 0.1 nM) was added to either the liquid media...
or to the plate and morphological changes were assayed by light microscopy.

**Results:**
Our preliminary data show that we are able to induce morphological change in *C. albicans* with N-acetyl-D-Glucamine, a positive control. In one experimental series, we tested increasing concentration of estrogen (0.1 nM, 1 nM and 1 µM) on filamentous growth in *C. albicans* and did not find a significant effect of estrogen compared to control (ANOVA P = 0.8, n = 8). Exposing *Neurospora* to 1 µM and 1 nM of estrogen caused *Neurospora* to grow more densely and erratically. Conversely, 0.1 nM estrogen caused *Neurospora* to grow not as dense but branch out farther onto the plate (triplicate experiment).

**Conclusion:**
We have established experimental conditions to test the effect of estrogen on both *C. albicans* and *Neurospora*’s morphological changes. This will allow for rapid screening of other factors that may be prone to causing morphological changes, such as inflammatory cytokines.

**Poster**

**Role of microenvironmental stress in increased breast cancer risk**

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The cell microenvironment is essential for tissue architecture that controls cell signaling, survival, and proliferation. These features which, when altered, can lead to cancer phenotypes. Besides stromal cells (e.g., fibroblasts and immune cells) and elements with mechanical impact, such as the extracellular matrix (ECM), the microenvironment also comprises chemical components that include reactive oxygen species (ROS). The metabolism of environmental factors like cigarette smoke, alcohol, high fat diet, and heavy metals in drinking water, can lead to the accumulation of ROS in the microenvironment. High levels of ROS may lead to oxidative stress (OS) and are observed in aggressive cancers including that of the breast. ROS interact with stromal components, change ECM stiffness, and may play a role in carcinogenesis. However, the role of ROS-microenvironment interaction in cancer onset is unknown. Interestingly, ROS has been reported to alter nuclear morphometry (shape and size) itself under the influence of mechanotransduction from the ECM that is known to control gene expression.

Our central hypothesis is that the density of ECM, influenced by ROS, increases the risk of cancer via an altered nuclear morphology controlling gene expression. We have used a three-dimensional (3D) cell culture model that comprises non-neoplastic HMT 3522 breast epithelial S1 cells embedded in collagen 1 matrix with stiffness
(800 Pa) similar to that of real tissue, and a higher stiffness of 1500 Pa which correlates with the density of tissues-at-risk for cancer. To induce acute OS, cells were exposed to 250 μM H2O2 for four hours on day 10 of culture. Using immunostaining we have assessed for the distribution of ZO-1, a marker of apical polarity. Polarity is a major characteristic of normal epithelial differentiation, if altered cells will exit quiescence. The nuclei were stained with DiAmidinoPhenyIndole (DAPI) and imaged. We measured a significant loss of polarity in the population of phenotypically normal breast glandular structures exposed to ROS compared to the control group. Moreover, cells cultured in the stiffer matrix of 1500 Pa, exhibited less circular and bigger nuclei compared to those cultured in the matrix of 800 Pa. They also displayed significantly less amount of ZO-1, meaning there was a loss in polarity. These results confirm that a microenvironment associated with cancer onset controls the maintenance of a normal phenotype of the breast epithelium. To further understand the significance of ROS in altering the microenvironment and influencing cell phenotype, in the next step we will study induced-changes in matrix stiffness under ROS by incorporating fibroblasts in the culture model.

Poster

**Inflammatory Characteristics of Peripheral Immune Cells in Migraineurs and Matched Controls**

**Hongtao Li,1,2 Kamaira H. Philips,2,3 Can Cui,4 Keturah (Kim) Faurot,2 Xuewei Zhu,5 Susan A. Gaylord,2 Christopher E. Ramsden,6 John Douglas Mann,7 Lishan Su8**

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**Background/Purpose:** Migraine is a neurological condition characterized by chronic inflammation. However, much is still unknown about the impact of chronic inflammation on innate and adaptive immunity.
**Methods:** 15 migraineurs were recruited chronologically from an on-going RCT on Nutrition for Migraine (NCCIH 5R01AT007813-05) and matched by age, gender and BMI to 15 non-migraine, healthy volunteers (control). Blood samples were obtained from both groups and processed as follows: 1) Antibody-labeled whole blood was analyzed by 8-color FACSCanto™ II and FlowJo software to detect different immune cell populations; 2) Peripheral Blood Mononuclear Cells (PBMCs) were stimulated with LPS in vitro, and 3) Nine cytokines from supernatants were examined by R&D multiplex system.

**Results/Outcomes:** CD4+ (Helper) T cells and the CD4+CD25+ regulatory T cell population from migraineurs were significantly lower than controls. The percentages of intermediate (CD14++CD16+) and non-classical monocytes (CD14+CD16++) were significantly decreased and a trend toward an increase in classical monocytes (CD14++CD16-) in the migraine group. CD8+ killer T cells demonstrated decreased expression (mean fluorescence intensity) of Integrins CD49d and CD18. Cytokine analysis with 24-hour LPS stimulation revealed no significant difference in inflammatory cytokine levels (IL-B, TNF-a, IL-6) between migraineurs and controls.

**Conclusions:** The diminished helper T cell population, especially CD4+CD25+ regulatory T cells, indicates that chronic migraine may be correlated with impaired immune control and incomplete inflammation resolution. Decreased non-classical and intermediate monocytes demonstrated less macrophage differentiation via the M2 alternative pathway, suggesting reduced ability to resolve inflammation at the cellular level. Reduced integrin CD49d and CD18 expression on CD8+ killer T cells suggest these T cells may be involved in chronic inflammation in migraineurs. Cytokine analysis warrants further study with more detailed time points, and specific cell types, to closely examine the process of inflammation activation and resolution.

**Presentation**

**Notch activates the expression of different pericardial genes using distinct permissive and instructive mechanisms in order to specify cardiac cell subtypes.**

**Manoj Panta,** Andrew J. Kump, John M. Dalloul, Kristopher Schwab and 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. Our previous work showed 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. Using cis- and trans-assays with enhancer-reporter constructs for a PC-specific gene, *Holes in muscle*(Him), we demonstrate 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. However, using identical approaches with enhancer-reporter constructs for another PC-specific gene, Zn finger homeodomain1 (Zfh1), we show that Notch signaling activates the expression of Zfh1 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 indicate that, in the case of Zfh1, upon Notch signaling, the Notch intracellular domain must bind 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.

Poster

Utilization of a Zebrafish Visual Motor Response Assay to Discover FDA Approved Drugs to Treat Retinitis Pigmentosa

Rebecca James, Purdue University; Logan Ganzen, Purdue University; Yuk Fai Leung, Purdue University

Retinitis Pigmentosa (RP) is a degenerative eye disease that causes night blindness, loss of peripheral vision, and loss of central vision in approximately every 1 in 3500 people. RP targets the photoreceptors of the retina and there is currently no effective treatment. This project aims to identify novel drugs that can be used to treat RP by utilizing the model organism zebrafish. The fish expresses a truncated Q344X human Rhodopsin transgene which causes significant rod photoreceptor degeneration by 7 days post fertilization (dpf). These zebrafish larvae are used in a visual motor response (VMR) assay to investigate the effects of over 1,400 FDA approved drugs on vision. This VMR assay utilizes a dim light to analyze scotopic vision. The VMR apparatus isolates the larvae from external light and sound. After one hour of dim light adaptation, the light turns off and the swimming activity of the larvae is quantified. An infrared camera is used to track the changes in larvae movement. By 7dpf, Q344X larvae experience a significantly diminished VMR at light offset. At 5dpf, Q344X fish are treated with 10µM of a designated drug. After two days of drug exposure, the larvae are tested for a VMR at 7dpf. For drug treated fish, a response is considered positive if there is a significant increase in displacement at light offset indicating rod function.
Upon completion of the drug library, some of the drugs displayed significant peaks with displacements as high as 0.4cm. Therefore, a percentage of the drugs screened appear to have a positive effect on vision. As the next step of this project, embryos treated with positive drugs will be rescreened for validity. The retinace of positive drug treated fish will also be analyzed histologically for increased rod presence.

Poster

**Using Next Generation Sequencing to Identify Cancer Transcripts in Mantle Cell Lymphoma.**

**Zachery Todd**, Chioniso P. Masamha, Butler University

Mantle Cell Lymphoma, MCL, a subtype of Non-Hodgkin’s Lymphoma, has no standard treatment and is declared incurable upon relapse. MCL is characterized by the abnormal expression of Cyclin D1, which is not normally expressed in B-cells. Cyclin D1 is associated with increased chromosomal instability, further increasing chromosomal translocations. This together adds to the chromosomal instability B-cells already have through VDJ recombination. Increased instability experienced by B-cells leads to more translocation events and thusly the formation of novel transcripts and fusion genes. A major limitation of commonly used second generation sequencing is that the short reads produced may exempt novel splicing events like fusion genes and result in underrepresentation of the 3’UTR. Our goal is to identify cancer-causing transcripts in MCL for molecular targeting. We used third generation, full-length PACBIO Iso-sequencing to sequence a MCL cell line. Further, we have developed an algorithm in R to identify over 7,700 transcripts from PACBIO and mapped them to the human reference genome. One of the transcripts we identified were from the gene CD74. CD74 has been characterized as a membrane protein linked with B-cell survival and cell proliferation. Current studies have noted an elevated expression of CD74 within some different types of cancer including MCL. We have developed CD74 specific amplicons to identify and map CD74 within a MCL cell line and a control. Future work will involve evaluating expression differences between MCL and control cell lines and developing anti-sense oligonucleotides for molecular targeting of CD74.
Microtubule assembly is necessary for dynein-mediated microtubule translocation and neurite elongation

Jessica Stone, K. McElmurry, D. Suter, Purdue University

Axonal navigation is driven by highly motile structures located at the distal tips of elongating axons, called growth cones. These sensory vehicles detect substrate and chemical cues and transduce them towards the dynamic cytoskeleton that drives their motility. Axonal elongation has been previously shown to require both microtubule assembly and dynein-mediated microtubule translocation; however, the relative contributions of these processes to axonal elongation are not fully understood. To address this, we investigated the relationship between microtubule assembly, microtubule movement, dynein activity, and axonal elongation in Aplysia neurites using phase contrast, differential interference contrast and fluorescent speckle microscopy as well as immunolocalization. We found that pharmacologically inhibiting either microtubule assembly or dynein activity resulted in reduced neurite growth and microtubule translocation. Simultaneously suppressing both processes produced similar results, but to a lesser degree. Our results indicate microtubule assembly is necessary for dynein-mediated microtubule translocation and neurite elongation.

Poster

In vivo and in vitro studies of gonadotropin-releasing hormone neuron function in obesity to measure obesity-related cellular stress and identify therapeutic natural products

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Because obesity and its co-morbidities (such as diabetes and cardiovascular disease) affect over 30% of the US population, understanding cellular mechanisms underlying this disease is crucial to identifying potential therapies. Thus, we have used in vitro and in vivo models to determine how obesity generates one of these co-morbidities, infertility, and to study the mechanisms of potential therapeutic natural products. Our work focuses on hypothalamic gonadotropin-releasing hormone (GnRH) neurons that govern fertility. First, since neuronal endoplasmic reticulum (ER) stress and inflammation are known to increase in diet-induced obesity in rodent models, we measured expression of three genes (Ddit3, Grp78, Il6) in hypothalami from high- and low-fat diet fed rats to determine if ER stress and cellular inflammation correlate with diet-induced changes in Gnrh1 expression. Previous research from our laboratory showed in vitro treatment of a GnRH-producing cell line
with the saturated fatty acid palmitate repressed *Gnrh1* at high doses but increased *Gnrh1* expression at low doses, while ER stress and inflammation increased as palmitate concentration increased. In vivo, there was a trend toward reduced *Gnrh1* expression with high-fat diet while the cytokine gene *Il6* was, unexpectedly, significantly lower in high-fat diet-fed rats compared to low-fat diet. Expression of the ER stress markers *Grp78* and *Ddit3* did not differ significantly between groups. Because IL-6 can promote or inhibit inflammation depending on cellular context, future studies will measure levels of other pro- and anti-inflammatory cytokines such as IL-10 and IL-1b. To begin to identify natural products that may target inflammation in obesity, we have begun testing the anti-inflammatory properties of maca root, a traditional Peruvian medicine used to enhance fertility, in our in vitro model. We hope to employ cell-based screening of macamides (alkamide compounds found only in maca root) to identify potential modulators of obesity-related cellular stress *in vivo*.

**Poster**

**Investigating Inhibition of Apoptosis as a Novel Treatment for Retinitis Pigmentosa using a Transgenic Zebrafish Model.**

**Emre Coskun** - Purdue University

Retinitis pigmentosa (RP) is the most common cause of night blindness, and it affects 1 in 3,500 people around the world according to the National Eye Institute. RP is a heterogeneous disease because it can be inherited through many mutations, including the Q344X mutation in the human rhodopsin gene. This mutation causes the truncation of the rhodopsin, which leads to its incorrect trafficking and causes rod degeneration. To discover an effective treatment for patients with RP, we use a transgenic zebrafish model expressing the Q344X mutation. The Q344X larvae show onset of rod degeneration at 5 days post fertilization (dpf) which progresses significantly by 7 dpf. The extent of diminished vision in the 7 dpf Q344X larvae can be assessed with a Visual-Motor Response (VMR) assay. The VMR assay acclimates the Q344X larvae to dim light preceding a dark flash, which does not induce a significant behavioral response. This suggests that the behavioral phenotype of Q344X is due to rod degeneration. Previous studies indicate that degenerating rods undergo apoptosis. Inhibition of apoptosis to decrease rod degeneration is investigated further with our model by screening the Selleckhem anti-apoptotic compound library with the VMR assay. The Q344X larvae are treated with these compounds between 5 – 7 dpf. The dim light condition of the VMR assay is adjusted to the larval rod range. Therefore, compounds that decrease rod degeneration would be expected to show increased behavior in the VMR. 82 of the 142 compounds in the anti-apoptosis library have been screened and analyzed with the VMR assay, and 7 showed an increase in behavior. In the future, the extent of apoptosis in rods in these treatment groups could
be verified using TUNEL staining of retinal cryosections. Drugs that produce consistent results can be reimplemented toward an effective and operational treatment for RP.

Poster

**Palmitate signaling in gonadotropin-releasing hormone neurons induces inflammation and endoplasmic reticulum stress in a TLR4-independent manner**

**Rithy Sakk Heng**, Michael Tanchevski, Heidi E. Walsh
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Hypothalamic gonadotropin-releasing hormone (GnRH) neurons are the final common pathway controlling fertility, and transcription of the gene encoding this peptide hormone (Gnrh1) is repressed by obesity-related cellular stressors such as endoplasmic reticulum (ER stress) or excess saturated fatty acids. To determine the intracellular signaling pathway utilized by the saturated fatty acid palmitate (PA) in GnRH neurons, we measured the expression of toll-like receptor 4 (TLR4) mRNA in an immortalized GnRH-producing cell line (GT1-7). In other cell types, TLR4 and its accessory protein MD2 can bind to PA and activate downstream inflammatory NF-kappaB signaling; thus, we tested whether a TLR4 inhibitor (TAK-242) could block the effects of PA in GnRH-producing cells. Neither repression of Gnrh1 expression nor activation of ER stress and inflammation-related genes were prevented by TLR4 inhibition, potentially supporting a role for PA metabolites such as ceramides in the regulation of GnRH neuron function by saturated fatty acids.

Presentation

**Obesity-related cellular stressors regulate gonadotropin releasing hormone gene expression via c-Fos/AP-1.**

Noah J. Levi, Christopher W. Wilson, Graham A.J. Redweik, Nathan W. Gray, Cody W. Grzybowski, Joseph A. Lenkey, A. Warren Moseman, Alec D. Bertsch, Nhien Dao, **Heidi E. Walsh**
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Obesity is a risk factor for infertility, but mechanisms underlying this risk are unclear. Fertility is regulated by hypothalamic gonadotropin-releasing hormone, encoded by the Gnrh1 gene. Because obesity promotes endoplasmic reticulum (ER) stress, we sought to determine how tunicamycin-induced ER stress affected Gnrh1 gene expression in the mouse hypothalamic cell line GT1-7. Tunicamycin repressed expression of Gnrh1 in a PKC- and JNK-dependent manner, while upregulating expression of a known Gnrh1 repressor, Fos. Obesity is associated with increased
circulating free fatty acids, and exposure to palmitate promoted ER stress and inflammation. Fos expression increased with palmitate dose, but Gnrh1 expression was upregulated with low-dose palmitate and repressed with high-dose palmitate. Using a small molecule inhibitor, we determined that AP-1 was required for Gnrh1 repression by high-dose palmitate or tunicamycin-induced ER stress. These findings suggest that hypogonadism driven by decreased hypothalamic GnRH may be a component of obesity-related infertility.

Presentation

**Antagonism between Bone Morphogenetic Protein and Activin signaling pathways in osteoprogenitor cells**

**Madeline Totten, Sydni Yates, Kelli Jestes, Sylvia Chlebek, Jordan Newby, Jon Arthur, Jonathan W. Lowery**, Marian University College of Osteopathic Medicine

Osteoporosis is a disease characterized by low bone mineral density due to the rate of bone resorption exceeding that of bone formation. Substantial evidence indicates that the Bone Morphogenetic Protein (BMP) pathway promotes bone formation through action of the effectors SMAD1/5/8 while the Activin pathway negatively influences bone mass through action of the effectors SMAD2/3. Recent studies suggest that BMPs and Activins regulate bone mass in a see-saw-like mechanism. Here, we seek to test this hypothesis *in vitro* via signaling responsiveness assays using pathway-specific western blot analyses in the osteogenic murine bone marrow stromal cell line W-20-17. We first confirmed that W-20-17 cells exhibit basal activation of SMAD1/5/8 and SMAD2/3 under serum-restricted conditions. Moreover, treatment with Follistatin, which sequesters Activin ligands in the extracellular environment, leads to an increase in BMP pathway activation. To determine the molecular mechanism allowing for this relationship, we treated W-20-17 cells with SB431542, which is an intracellular inhibitor of Activin signaling that functions downstream of receptor engagement, and found no effect on BMP pathway activation. In contrast, treatment of W-20-17 cells with BMP pathway inhibitor Noggin had no effect on Activin pathway activation despite robust inhibition of BMP signaling. Collectively, our results suggest Activin-mediated repression of BMP signaling in these cells is ligand-dependent but occurs upstream of SMAD2/3 activation. Gene expression analyses indicate that W-20-17 cells express Activin A and its receptors ALK4, ACVR2A, and ACVR2B. Given that ACVR2A and ACVR2B also have high affinity for BMP ligands, this raises the possibility that Activin-mediated repression of BMP signaling may occur via competition for a shared pool of receptors. Over-expression studies coupled with osteoblast activity assays are currently underway to examine this hypothesis. Collectively, our work seeks to elucidate the mechanism(s) that regulate antagonism of BMP and Activin signaling pathways in the osteoblast lineage to identify novel opportunities for treating low bone mass in humans.
Elucidating the molecular signatures associated with elevated bone formation rate

Kelli Jestes, Krista Jackson, Jonathan Lowery; Marian University College of Osteopathic Medicine

Osteoporosis is a disease of decreased bone density that occurs when bone resorption exceeds bone formation, thereby placing individuals at greater risk of fracture and disability. We previously reported that deletion of the Bmpr2 gene in embryonic skeletal progenitor cells causes substantially elevated bone density in young adulthood and reduced age-related decline in bone density, likely due to elevated bone formation rate. Thus, these mice may serve as a novel model in which to explore the mechanisms regulating bone formation in the aging skeleton. Here, we performed transcriptome profiling and identified a concise gene signature associated with elevated bone formation rate in Bmpr2 mutant mice, with 120 transcripts up-regulated and 131 transcripts down-regulated. Candidate-driven qRT-PCR provided secondary confirmation of this dataset. Notably, only 8 of these differentially-expressed transcripts have been previously implicated in bone physiology (Pak4, Rpl38, B2m, Fgf1, Nmu, Phospho1, Smpd3 and Inhbe), thus representing potentially novel regulators of osteoblast function in the aging skeleton. Additionally, we sought to examine the cell communication events that are associated with elevated bone formation rate. Using small quantities of protein from bones of control and Bmpr2 mutant mice, we took advantage of recent advancements in high-throughput phospho-profiling antibody arrays, which allow simultaneous detection of roughly 300 targets. These results indicate that the phosphorylation status of several signaling molecules is differentially regulated in Bmpr2 mutant mice as compared to control littermates, including proteins known to regulate osteoblast differentiation and/or activity (TAK1, AKT1, PAK1, Rho/Rac and PI3K). Collectively, our work highlights novel factors associated with elevated bone formation rate and may identify new opportunities for treating low bone density in humans.

Host-Targeted Therapeutic for Bacterial Infection: A Potential Alternative to Antibiotic Treatments

Nathan C. Hahn, Ball State University

An ongoing treatment challenge for patients with tonsillitis are relapsing, recurrent infections. Recurrent Streptococcus pyogenes infections occur regardless of the susceptibility of infectious strains to antibiotics. These persistent infections are caused by the ability of pathogenic S. pyogenes to invade host cells and escape common
antibiotics. Invasion of host cells is mediated by human protein CDC42. In our current work, we demonstrate that simvastatin, a commonly prescribed therapeutic for hypercholesteremia, and ML141, a novel small molecule inhibitor with specificity for human CDC42, limit host cell invasion by *S. pyogenes*. This is mediated by limitation of actin stress fiber depolymerization and limiting binding to host glycoprotein fibronectin. This work demonstrates a novel application of simvastatin and the potential of CDC42-targeted therapeutics for limiting intracellular *S. pyogenes* infection to prevent recurrent infection.

Poster

**Relationship between Reactive Oxygen Species (ROS) and the rate of neurite growth in *Aplysia californica* neurons**

Halie Szilagyi, Purdue University
Daniel Suter, Purdue University

In the field of neurobiology, the role of the neuronal growth cone is relatively well understood: located at the tip of neuronal processes, it is a highly motile structure that uses cell surface receptors to detect the surrounding environment. However, what is less known are the underlying biochemical mechanisms of signal transduction which the growth cone employs during directional migration. The goal of this research, specifically, is to understand the role of reactive oxygen species (ROS) in neuronal growth in *Aplysia californica*. ROS are derived from molecular oxygen in the atmosphere; however, mitochondria and NADPH oxidase (Nox) enzymes are the main sources of intracellular ROS. ROS have long been considered as a toxic molecule and an unavoidable side effect of these cellular processes for several years. However, recent studies have revealed that ROS should be regarded as second messengers in numerous signaling pathways in health and disease. ROS have been linked to neurogenesis, polarization, and maturation of neurons. Yet, this needs to be distinguished from oxidative stress, which can compromise homeostasis. Therefore, the source, timing, localization, and targets of ROS are important when evaluating its effects on neural physiology. Previous literature results indicated a slight positive relationship between growth cone ROS levels and neurite growth rate. However, this data was analyzed as average values and was not correlated at the individual growth cone level. The goal of this research is to correlate the growth cone advance rate more closely to the actual ROS levels at the time of neurite growth rate determination and to do this on an individual growth cone basis. This experiment requires precise determination of neurite advance rate and growth cone ROS levels using live cell imaging and fluorescent dyes. Currently, data from this research is still be compiled in an attempt to elucidate this trend. However, a better understanding of ROS and their physiological roles will allow for improvement of antioxidant treatments of neurodegenerative diseases and injuries in the nervous system.
Presentation

The FDA-approved Drug Carvedilol Improves Vision and Retinal Morphology in a Zebrafish Model of Retinitis Pigmentosa

Logan Ganzen, Purdue University; Rebecca James, Purdue University; Truc Kha, Purdue University; Yuk Fai Leung, Purdue University

Retinitis pigmentosa (RP), also known as night blindness, is an incurable disease which affects approximately 1 in 4000 individuals globally. Since there are no effective treatment options for RP, the goal of this project is to identify novel drug treatments that can prevent or slow the disease progression. To accomplish this, a visual-motor response (VMR) assay was utilized to assess the diminished vision of a transgenic zebrafish RP model carrying a human Rhodopsin transgene with Q344X truncation mutation, and we used this line to screen compound libraries. This line experiences significant rod death and diminished VMR by 7 days post-fertilization (dpf). One drug that was identified to improve the visual function and retinal morphology of the Q344X zebrafish model was the FDA-approved drug Carvedilol. In our screen that identified Carvedilol, we treated the Q344X mutant with 10 μM drug from 5 to 7 dpf. Treatment of this drug ameliorated the VMR of the Q344X zebrafish. Also, the rescuing effects on rod number and morphology was evaluated through fluorescent imaging and in situ hybridization. Carvedilol-treated Q344X zebrafish had a significant increase in rod-positive area on 7 dpf. Treated larvae had a larger average area of fluorescent signal near the marginal compared to the untreated larvae. Treated larvae also had a larger average area of fluorescent signal in the ventral patch compared to untreated larvae. Similarly, in situ hybridization revealed that Carvedilol increased staining of nr2e3 and rhodopsin near the Q344X ventral patch and marginal zone. We conclude that Carvedilol is a beneficial drug to the Q344X RP model. Fluorescent imaging and in situ hybridization indicate that rod-positive area near the marginal zone area and ventral patch were expanded with treatment. This finding implies that the drug potentially increased rod numbers and in turn improved the vision of the Q344X RP model.

Poster

PKC activation induces ubiquitination of the Na-K-2Cl cotransporter 1 in the human colonic epithelial cells T84


Gut clearance (i.e., fluid secretion) is one of the first lines of defense of the intestine against toxins and opportunistic bacteria. In the colon, fluid secretion is coupled to chloride secretion. Previous work has demonstrated that the basolateral Na-K-2Cl...
cotransporter 1 (NKCC1) represents a site for regulating fluid secretion independently of the apical chloride channels such as the cystic fibrosis transmembrane regulator. In addition, the lab has shown that protein kinase C (PKC) activation causes internalization of NKCC1, which in turn blocks chloride secretion. To date, the post-translational signal responsible for NKCC1 internalization during PKC activation remains unknown. Similarly, the fate of NKCC1 in the endocytic pathway has not been elucidated. In the present study, we investigated the role of ubiquitin as post-translational signal responsible for NKCC1 internalization. Experiments were performed on the human colonic T84 epithelial. Cells were incubated with or without 100 nM phorbol 12-myristate 13-acetate (PMA), a PKC activator, or in presence of 100 µM carbachol, a M3 muscarinic receptor agonist, for 15 or 30 min. NKCC1 ubiquitination was tested by western blot after immunoprecipitating NKCC1. Our preliminary results show that PMA and carbachol induced an increase of NKCC1 ubiquitination compare to control. In addition, blocking the lysosome with 5 µM NH4Cl did not prevent NKCC1 degradation during PKC activation by PMA. Our results suggest that PKC induces NKCC1 internalization through a ubiquitin-dependent pathway and may target NKCC1 for degradation in a lysosomal independent-manner.

Poster

Inhibition of Breast Cancer Cell Proliferation by the Dietary Polyphenols Ellagic Acid and Luteolin

Jessica Rivosecchi-Fulton and Kimberly M. Baker, University of Indianapolis

Epidemiological studies have demonstrated a positive correlation between the consumption of polyphenolic compounds and cancer prevention. Ellagic acid and luteolin, dietary polyphenols found in a variety of fruits and vegetables, have been shown to inhibit cancer cell proliferation and motility and induce apoptosis in vitro. In this study, we evaluated the antiproliferative effects of ellagic acid and luteolin, individually and in combination, using MCF-7 breast cancer cells. We found that both ellagic acid and luteolin inhibited MCF-7 cell proliferation in a dose dependent manner. Furthermore, we found that co-administration of ellagic acid 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 ellagic acid and luteolin may be an effective chemotherapeutic strategy against breast cancer.
**Poster**

**Suppression of Breast Cancer Cell Proliferation by Benzyl Isothiocyanate and Luteolin**

Laura McKee, Zacharias Raptopoulos, and Kimberly M. Baker  
University of Indianapolis

Epidemiological studies suggest that a high dietary intake of fruits and vegetables protects against the development of cancer. Benzyl isothiocyanate, found in cruciferous vegetables, and luteolin, found in a variety of fruits and vegetables, have been shown to exhibit anticancer activity *in vitro*. In this study, we evaluated the effects of benzyl isothiocyanate and luteolin, individually and in combination, on cell proliferation in MCF-7 breast cancer cells. We found that both benzyl isothiocyanate and luteolin inhibited MCF-7 cell proliferation in a dose dependent manner. Furthermore, we found that co-administration of benzyl isothiocyanate and luteolin led to a greater inhibition of cell proliferation. These results suggest that combinatorial treatments using benzyl isothiocyanate and luteolin may be an effective chemotherapeutic strategy against breast cancer.

**Presentation**

**Bringing Attention to Lesser-known Bone Remodeling Pathways**

Mehdi Shadmand; Marian University, College of Osteopathic Medicine

Abstract Osteoporosis, a disease of low bone mass, places individuals at enhanced risk for fracture, disability, and death. In the USA, hospitalizations for osteoporotic fractures exceed those for heart attack, stroke, and breast cancer and, by 2025, the number of fractures due to osteoporosis is expected to rise to nearly three million in the USA alone. Pharmacological treatments for osteoporosis are aimed at stabilizing or increasing bone mass. However, there are significant drawbacks to current pharmacological options, particularly for long-term management of this chronic condition. Moreover, the drug development pipeline is relatively bereft of new strategies. Consequently, there is an urgent and unmet need for developing new strategies and targets for treating osteoporosis. Casual observation led us to hypothesize that much of the bone remodeling research literature focused on relatively few molecular pathways. This led us to perform bibliometric analyses to determine the relative popularity of bone remodeling pathways in publications and US National Institutes of Health funding of the last 10 years. In this review article, we discuss these findings and highlight several less-examined signaling pathways that may hold promise for future therapies.
Hot Topics
A brief history of CRISPR and potential roles for genome editing in human health

Doug Bernstein, Assistant Professor, Ball State University Department of Biology

*Clustered regularly interspaced short palindromic repeat* (CRISPR) mediated genome editing continues to revolutionize molecular biology. Genome editing 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. Since CRISPR’s genome editing capability was realized noteworthy experiments investigating human disease have been performed in mammals. In this hot topic we will discuss some of the foundational discoveries made by scientists that have paved the way for these advances. We will discuss the required components of CRISPR genome editing systems and the role these components play in genome editing. In addition, we will examine the next generation of CRISPR mediated genome editing techniques that are being developed and how these advancements could advance our ability to edit genomes. Finally, we will discuss how CRISPR mediated genome editing is impacting human health and potential consequences of editing the human genome.
Chemistry

Presentation

Toehold Mediated Shape Transition of Nucleic Acid Nanoparticles

Jordan Hartung, Nathan McCann, Kheiria Benkato and Emil F. Khisamutdinov

Development of novel materials possessing structural transformations in response to an external stimuli such as environmental changes (pH, temperature etc.), presence of particular proteins or short oligonucleotides are of great interest for a variety of applications ranging from medicine to electronics. The dynamic operations of most Nucleic Acid (NA) devices including circuits, nano-machines, and biosensors rely on networks of NA strand displacement processes in which an external or stimulus strand displaces a target strand from a DNA or RNA duplex. The rate of strand displacement can be greatly increased by the use of “toeholds,” single-stranded regions of the target complex to which the invading strand can bind to initiate the reaction forming additional base pairs that provide a thermodynamic driving force for transformation. Herein we developed a highly robust nanoparticle shape transition approach, sequentially transforming DNA polygons from one shape to another using toehold-mediated DNA strand displacement technique. The shape transformation was confirmed by agarose gel electrophoresis and atomic force microscopy. Furthermore, we demonstrate that our approach is applicable for RNA shape transformation from triangle to square, which can be detected by fluorescence emission from Malachite Green binding RNA aptamer. Using gel-shift and fluorescence assays, we demonstrated that transformation is highly effective and occurs at isothermal conditions (37 °C) that can be implemented within living cells as reporter molecules. This work is intended to provide simple, cost-effective and straightforward model for the development of biosensors and regulatory devices in nucleic acid nanotechnology.

Presentation

Flexible Tetra-U/T Helix Linking Motif Underlies the Design of Therapeutic Nucleic Acid Nanoparticles

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RNA nanotechnology has traditionally relied upon synthetically modified ribonucleic acid (e.g. 2’-fluorine, 2’-amine, 2’-methyl) for its therapeutic ability and to engineer stable nanostructures. Unmodified RNA has long been sought to be incorporated into
RNA nanotechnology, but has been prevented because of its immunogenicity and susceptibility to enzyme degradation. Nucleic acid nanostructures may offer the ability to deliver unmodified therapeutic RNA’s without threat of immune system or ribonuclease detection. Herein, we propose a tetra-uracil (tetra-U) motif that can be incorporated into these nanostructures to allow for efficient and stable assembly. This motif underlies the foundation of a recently developed therapeutic nucleic acid nanoparticle library. The goal of the Khisamutdinov lab has been to design and investigate the physical and chemical properties of these nanoparticles. This project is a part of a larger collaboration between other universities and research institutions that seeks to investigate how structure and composition of these nucleic acid nanoparticles define immunorecognition.

Poster

Structure and composition define immunorecognition of nucleic acid nanoparticles

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Nucleic acid nanoparticles (NANPs) have evolved as a new class of therapeutics with the potential to detect and treat diseases. Despite tremendous advancements in NANP development, their immunotoxicity, one of the major impediments in clinical translation of traditional therapeutic nucleic acids (TNAs), has never been fully characterized. Here, we describe the first systematically studied immunological recognition of 25 representative RNA and DNA NANPs selected to have different design principles and physicochemical properties. We discover that, unlike traditional TNAs, NANPs used without a delivery carrier are immunoquiescent. We show that interferons (IFNs) are the key cytokines triggered by NANPs after their internalization by phagocytic cells, which agrees with predictions based on the experiences with TNAs. However, in addition to type I IFNs, type III IFNs also serve as reliable biomarkers of NANPs, which is usually not characteristic of TNAs. We show that overall immunostimulation relies on NANP shapes, connectivities, and compositions. We demonstrate that, like with traditional TNAs, plasmacytoid dendritic cells serve as
the primary interferon producers among all peripheral blood mononuclear cells treated with NANPs, and scavenger receptor-mediated uptake and endosomal Toll-like receptor signaling are essential for NANP immunorecognition. The TLR involvement, however, is different from that expected for traditional TNA recognition. Based on these results, we suggest that NANP technology may serve as a prototype of auxiliary molecular language for communication with the immune system and the modulation of immune responses.

Poster

**Synthesis and Electrochemical Characterization of Copper(II) Bis(thiosemicarbazone) Catalysts for Hydrogen Evolution Reactions**

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Bis(thiosemicarbazone) (BTSC) metal complexes are well known in the literature with applications in medical imaging, therapeutics, and electrocatalysis. The parent ligand, *N,N*-bis(methylthiosemicarbazanato)butane-2,3-diimine (ATSM) is highly functionizable in the backbone and pendant positions. Transamination of the ATSM derivatives provides an attractive method to vary the terminal –CH3 substituent(s). In this work, we present a series of new Cu(II) complexes based on the N2S2 chelates, *N,N*′-(dimethyleneaminothiosemicarbazanato)-4-(methylthiosemicarbazanato)butane-2,3-diimine (H2L1) and *N,N*′-bis(dimethyleneamino-thiosemicarbazanato)butane-2,3-diimine (H2L2), which have been synthesized and characterized by spectroscopic, electrochemical, and single crystal X-ray diffraction methods. Complexes 1–4 are analogues of the well-known *N,N*′-bis(methylthiosemicarbazanato)butane-2,3-diimino copper(II) (CuATSM), which contains a similar N2S2 donor core with terminal non-coordinating amines. Substitution of the methyl group(s) of the terminal amines of ATSM with *N,N*-dimethyleneamidine followed by alkylation generates a charged quaternary amine in the ligand framework. This charged site tunes the redox potentials of the complexes with minimal changes in their physical and electronic properties. Electrochemical studies reveal significant anodic shifts in the CuL/CuL− and CuII/I reduction potentials of the alkylated vs non-alkylated species. The effect of single charge vs double charge of the quaternary amine of the ligand in 1–4 will be discussed along with their catalytic activity as hydrogen evolution reaction (HER) catalysts.
Poster

**Toxic Effect of Chitin to Cells**

**Chayli Brock**  
Indiana University Purdue University Columbus

Chitin is a major component in the exoskeletons of arthropods that has numerous medical applications. Chitin is already important in pharmaceuticals, but it is almost exclusively obtained from the shells of lobsters or crab. Since lobster and crab are already highly sought after, cicada shells were used in the following experiments. Cicadas are abundant and readily available at no cost, but the toxicity to cells of chitin derived from cicada shells is unknown. To determine toxicity, fibroblast cells from a mouse were cultured and then treated with varying concentrations of chitin. A cell counting kit was employed to determine how many cells survived the treatment based on a colorimetric scale. The expected outcome is that the source of chitin does not bear any toxic effects to cells of a model organism.

Poster

**Electrospinning with Chitosan Based Polymers**

**Adrian Morales**  
**James Mendez**  
Indiana University Purdue University Columbus

Chitin, a structural polysaccharide responsible for the shell strength of various decapods and insects, is a readily available biopolymer with many novel uses. It and its deacetylated counterpart chitosan have previously been used in the creation of such dissimilar products as electrical conductors, bone grafts, and suture material. To optimize its potential surface area, the technique of electrospinning has been used to create nanofibers. While electrospinning has been around since the late 19th century, techniques have abounded. In order to determine optimal conditions under which to spin, several variables were manipulated, including distance to collection plate, voltage, syringe pump speed, and polymer solution composition. A carrier polymer, polyvinyl alcohol, was used to ease the process of collection.
Poster
Heavy metal water filtration by chitin with varying degrees of acetylation

Stephanie F. Baumgartner - Indiana University Purdue University Columbus
JD Mendez - Indiana University Purdue University Columbus

Chitin is a biopolymer found in the exoskeletons of arthropods (lobster, shrimp, insects…) and some fungi (mushrooms). These exoskeletons are composed of chitin, along with other organic materials and metals such as calcium. The high propensity of chitin to bond to metals makes it an excellent candidate for use as a water purification system. Since lobster and shrimp are in high demand commercially, we used cicada exuviae as the source for our chitin. Chitin was extracted from cicada sloughs with varying degrees of acetylation. As the percent of deacetylation increases, so does the solubility of the product which is why chitosan is more able to form films than is chitin. However, the product becomes less able to form hydrogen bonds leading us to hypothesize that as the percent of the degree of acetylation decreases, so does lead uptake.

Poster
Development of a Novel Epoxide-Containing Trimethylenemethane Precursor for Palladium-Catalyzed Cycloadditions

Mara Paterson and Kevin Jantzi, Valparaiso University

Pd(0)-catalyzed trimethylenemethane (TMM) cycloaddition reactions have been used extensively to generate disubstituted 5-membered rings with high levels of regioselectivity, chemoselectivity, and stereoselectivity. We aim to improve upon existing methodologies by introducing an epoxide into the TMM precursor, which should yield more highly functionalized products. A three step synthesis of a TMM precursor has been achieved in 20% overall yield. Efforts are underway to determine the reactivity of this precursor in Pd(0)-catalyzed cycloaddition reactions with substituted alkenes.

Presentation
Synthesis of an Unnatural Fluorescent Amino Acid

Moriah Carmel, Valparaiso University
Hannah Laatsch, Valparaiso University

The long-term goal of this project is to chemically synthesize an unnatural fluorescent amino acid (UFAA) that can later be used to build glow-in-the-dark proteins. UFAAs allow the visualization of a single protein in an otherwise transparent living cell. The
specific objective of this project is to synthesize a 4-(N,N-dimethylamino)phthalimide-based environment-sensitive fluorescent amino acid. The first part of this synthesis was the preparation of an anhydride (4-Methyl-Aminophthalic Anhydride), which was then coupled with commercially-available Boc-Dap-OtBu Hydrochloride. Finally, trifluoroacetic acid was used to remove the protecting groups, yielding the desired product. The product was characterized using 1H and 13C NMR, and Liquid Chromatography-Mass Spectrometry (LC-MS). The remaining tasks include improving the purification and percent yield.

Poster

**Streamlining the synthesis of folate mimics as potential antifungal agents**

**Zachary Bennett, Kassidy Grumbles, Anna Bockman, Jeff Pruet**
Valparaiso University

*Methionine Synthase* (MetSyn) is an enzyme that creates the amino acid methionine, which is essential for all organisms. There are key differences between the B12-independent fungal MetSyn enzyme and the B12-dependant mammalian form, especially with regard to the proximity of the two active sites. Taking advantage of these differences, an antifungal drug could be developed to exclusively bind the fungal enzyme and inhibit fungal growth while leaving the host (patient) unaffected. As MetSyn is a folate-dependent enzyme, we are currently exploring the synthesis of various pterin-based molecules as these mimic the essential folate substrate. We have developed optimized paths to allow for rapid generation of new folate-mimics and other pterin derivatives. We have expressed the MetSyn enzyme and begun testing these molecules for activity in a fluorescent assay for monitoring MetSyn activity.

Presentation

**Theoretical Investigation of Non-covalent Interactions Between Selected Lactams and Fullerenes**

**Shannon Lieb**, Butler University (*emeritus*) and Joe Kirsch, Butler University

There has been an explosion of papers based on C2n allotropes since 1985 when C60 was first discovered. Areas of research stimulated by C2n allotropes are actively pursued in medicine and agriculture, nanosensors (pharmaceutical and biomedical analysis), development of green chemistry processes, solar cell efficiency enhancement to name a few. In order to develop a complete picture of this broad area of research, both theory and experiment are needed. X-ray structures of C60, C70 and other C2n structures have been accomplished, yet the aforementioned areas of research require a co-crystallization of the fullerene and a ligand. Because of the difficulty of such an undertaking, other experimental techniques such as Fourier
Transform Infrared (FTIR) and Nuclear Magnetic Resonance (NMR) need to step up to augment this undertaking. In addition, a theoretical interpretation of these spectral techniques must accompany the experimental measurements to round out the full picture of these investigations.

In previously published research by Joe Kirsch, et. al., the infrared shift of the carbonyl stretching frequency of 2-pyrrolidinone (g–lactam) and d–valerolactam (d–lactam) due to the non-covalent interaction between these lactams and the fullerenes, C60 and C70 has been investigated. Theoretical modeling of these particular combinations of lactam/fullerene studies draws out structural details as well as relative strengths of these interactions. This presentation is the theoretical modeling of these experimental results to lay the groundwork for other experimental/theoretical investigations of non-covalent interactions.

Poster
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**Reveal the Structural Complex of Benzoic Acids at Electrode-Electrolyte Interfaces Using Electrochemical STM**

*Kuo-Hao Chen, Cody Leasor and Zhihai Li*

In the present work we employed electrochemical scanning tunneling microscopy (EC-STM) in combination with electrochemical cyclic voltammetry (CV) techniques to explore the adsorption and phase formation of benzoic acids (BZAs) at Au(111)/electrolyte interfaces. Our experiments show how electrolyte, molecular concentration, electrochemical potential, co-adsorption of aqueous ions affect the adsorption and self-assembly of BZA molecules. It is found that the BZA molecules are not assembled into long range ordered structures in the presence of sulfuric acid electrolyte due to the strong competing co-adsorption of sulfate ions on a gold electrode. BZA molecules can form flat-oriented ordered adlayers in perchloric acid electrolyte (containing weakly adsorbed ClO4- ion) at a negatively charged surface only when the concentration of molecular solution reaches above 12 mM. Below 12 mM, the CVs of BZA on Au(111) in 0.1 M HClO4 shows only one pair of adsorption/desorption peaks. When the BZA concentration increases to 12 mM, the voltammogram exhibits 3 pairs of peaks, corresponding to the structural transformation of a disordered phase I, linear stripe pattern (phase II), zigzag pattern (phase III) and upright packing pattern (phase IV), revealed by STM within 4 electrochemical potential regions.
Poster

**A Catalytic Challenge: Can We Degrade Pharmaceuticals in 5 Minutes?**

**Ruth E. Nalliah**, Huntington University

The accumulation of excreted pharmaceutical substances in the environment has been documented for decades as an emerging problem. The long-term objective of our work is to find a catalytic substance which can be added to automatic toilet bowl cleaners to begin degrading excreted pharmaceuticals safely, within the timeframe of a toilet flush. The Terrence Collins group at Carnegie Mellon University has engineered a homogeneous Fe-TAML catalyst which mimics the biological behavior of enzymes and has been shown to catalyze the oxidative degradation of a variety of organic pollutants and endocrine-disrupting substances with hydrogen peroxide. In a first-year chemistry laboratory, we tested TAML catalyst FeB to estimate the extent of degradation of five food coloring dyes and five over-the-counter pharmaceuticals within 5 minutes using UV-visible spectroscopy, comparing results from aqueous hydrogen peroxide added to a basic solution vs. hydrogen peroxide released from added solid sodium percarbonate. The FD&C food coloring dyes for which the absorption peak was observed to mostly or completely disappear within 5 minutes include Blue 1, Blue 2, Red 40, and Yellow 5, while Red 3 showed much less degradation under the conditions of this preliminary study. The pharmaceuticals appear to be more resistant to degradation than the dye molecules, for which the structures are typically larger. Pharmaceuticals for which the absorption peak was observed to partially disappear within 5 minutes include salicylic acid and acetaminophen, while caffeine, ibuprofen, and quinine showed little evidence of degradation within that timeframe under the conditions used here. The relative effectiveness of using hydrogen peroxide solution vs. solid sodium percarbonate varied depending on the substance being degraded. Future studies include pH-dependent studies as well as studies involving mixtures of hydrogen peroxide solution and sodium percarbonate.

Poster

**Formation of 1D water wires in two isostructural Cu(II) and Ni(II) complexes: Synthesis, characterization, and thermal analyses**

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Hydrogen bonding (HB) interactions are well known to impact the properties of bulk water and within hydrated materials. Most notably, 1D water wires are involved in
proton and water transport. Therefore, the design and synthesis of efficient proton conductors in terms of both cost and performance are of significant interest. In this work, two isostructural related Cu(II) and Ni(II) complexes based on the chelate containing $N,N'$-(ethane-1,2-diyl)bis(1-methyl-1H-imidazole-2-carboxamide) (H2L) have been synthesized and fully characterized by single crystal X-ray diffraction, spectroscopic methods, and thermal analysis. Both complexes crystalize in the monoclinic space group $P2_1/c$. The asymmetric unit of $\mathbf{1}$ and $\mathbf{2}$ contain two equivalents of CuL or NiL and four molecules of water of hydration. In the solid state packing of both complexes, the extended hydrogen bonding (HB) networks form 1D helical zig-zag chains of water along the crystallographic $b$-axis. For both complexes, the identical ligand framework, coordination geometries, and solid state packing generates similar HB expansion motifs, which result in analogous thermal stabilities and phase transitions at elevated temperature.

Hot Topics

**Molecular Mapping of Solid Samples by Mass Spectrometry Imaging: From Plants to Pharmaceuticals**

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This hot topic will discuss the current state of mass spectrometry imaging (MSI), various ionization sources available for MSI, and our recent work mapping active ingredient crystallization and agglomeration in pharmaceutical tablets by electrospray laser desorption ionization (ELDI) MSI. While separations in the form of chromatography commonly precede detection by mass spectrometry, important spatial information about the distributions of compounds is lost. The spatial distributions of compounds in their natural biological matrices contain important information for the fields of lipidomics, metabolomics, and proteomics. Similarly, biomedical research benefits greatly from accumulation information of drug-uptake in animal tissue. Recently, MSI has emerged as a viable source of spatial distribution data utilizing the sensitivity and versatility of mass spectrometry. Since MSI is a non-targeted technique it is not limited to detection of a single compound of interest but can concurrently provide valuable molecular distribution data for different classes of compounds. This has led to an explosion of applications and ionization mechanisms for MSI in the past decade. Plant metabolomics is one such application, which shows how MSI can benefit an entire field. Our current study applies ELDI-MSI to interrogate tablet formulations for the spatial distributions of ingredients. This is the first application of
ELDI for tablet imaging, and the first MSI method to detect agglomeration and diagnose crystallinity in formulations. As crystallinity alters the bioavailability of active pharmaceutical ingredients (API) it is important to prevent crystallization in the tablet formulation process. Currently, there is not a reliable analytical method to detect API crystals in the tablet form. ELDI-MSI has demonstrated potential to detect low amounts (<5% drug load) of crystallinity in tablets and has shown agglomeration effects as a direct result of crystalline API. This is at the detection level which the Food and Drug Administration has called on pharmaceutical companies to report.

Poster

**Synthesis of Monolayer Protected Gold Nanoparticles for the Study of Electrochemical and Mechanical Properties**

**Yifei Chen**, Andrew Riley, Tykhon Zubkov, and Zhihai Li*

Department of Chemistry, Ball State University, Muncie, IN 47304

In this study, hexanethiol–gold *nanoclusters* are synthesized via the modified Brust-Schiffrin method. Synthesized nanoparticles were characterized with various analytical techniques such as TEM, scanning tunneling microscopy (STM), UV-Vis and electrochemical techniques. We have systematically varied the molar ratio of gold to protecting agent to discover the effect of gold-to-ligand ratio on the size of gold clusters. It is found that the average particle size decreases from $4.28 \pm 0.83 \text{ nm}$ to $1.54 \pm 0.67 \text{ nm}$ as the gold-to-ligand molar ratio changes from 1:1 to 1:9. When the ligand-to-gold ratio become small, gold atoms tend to form larger particles, and eventually make it impossible to form “nanoscale” particles when ligand-to-gold ratio is lower than 33% mole ratio (gold:thiol is equal to 3:1). Further, we also explored the electrochemical application of synthesize nanoclusters in terms of quantized charge at electrode-electrolyte interfaces, and capacitance of monolayer-protected nanoclusters were calculated.

Poster

**Graphene Supported Ni4 Catalysts for Propane Dehydrogenation**

**Joshua Duensing**-Valparaiso University

**Jacob Allred**-Valparaiso University

Stan Zygmunt-Valparaiso University

Propane dehydrogenation (PDH) is used to produce propene, which is the primary building block for many commercial plastics. The catalyst most commonly used for this reaction is platinum. Because of rising demand for propene, an alternative catalyst...
is being sought due to platinum’s high cost. For this purpose, we have performed a computational study of the PDH reaction with a 4-atom nickel cluster (Ni4) catalyst on a graphene support. We have computed the equilibrium structures of the Ni4 cluster on both single- and double-vacancy sites and have calculated the complete PDH reaction pathway for each case. This allowed us to study the effect of the graphene support on catalytic activity. These results help clarify the relationship between the PDH activation energy and the propane binding energy and overall reaction energy, and may aid in the design of new potential catalysts for the PDH reaction.

Poster

**Using 3D printing technology to recreate historically accurate teaching models from the 1800s**

**Carrie Caldwell** and James Mendez, IUPUC

The future of 3D printing provides endless possibilities to innovate, create, and modify how learning can take place in a classroom. Recently, we have been attempting to design and recreate crystalline structure representation models from the 1800s. These models will be used to represent the crystalline structures, or ordered arrangement of atoms, ions or molecules as well as to provide a historical context. This is significant because it allows us to see how various materials are formed based on their crystalline structure. For example, “The reason that graphite and diamond are so different from each other is because the carbon atoms are stacked together into two different crystal structures” (ASU, 2018). We have been striving to recreate 10+ different structures from the wooden block sets originating from the 1800s. These structures include:

1. The basic cube
2. Octahedron
3. Rhombic Dodecahedron
4. Tetrahedron
5. Pentagonal Dodecahedron
6. Tetragonal Pyramid and Tetragonal Prism and Base
7. Hexagonal Pyramid and Hexagonal Prism and Base
8. Rhombohedron
9. Rhombic Pyramid and Rhombic Prism and Base
10. Triclinic Pyramid

**Educational Impact:**

We plan to integrate this technology in introductory chemistry courses as well as potentially using it with younger students to provide a tangible way of learning about
the different crystalline structures, and how they make up different objects that we utilize daily. These models will create a hands-on way to get students involved with the learning process and allow different stages of chemistry courses to see how different structures and molecules are being formed by their internal structures. The hope for this 1800’s renovation is that students are provided with an easier method to learn this style of chemistry that will help them along their path and through their field of study. These models would be introduced during lessons associated with them so that they can physically see what the crystalline structures appear to be and understand the importance of how they shape the world.

Poster

Light-activated Vitamin-Chlorin Conjugates for Triple-Negative Breast Cancer

Dewana M. Hammonds and Meden F. Isaac-Lam, Department of Chemistry & Physics, Purdue University Northwest, Westville, IN 46391

Breast cancer is among the most common types of cancer occurring in women. Triple-negative breast cancer (TNBC), representing 15-20% of breast cancer cases, is an extremely aggressive subtype of breast cancer due to absence of potential therapeutic targets. Photodynamic therapy (PDT) is a binary therapeutic method with the potential to target the most difficult subtypes of breast cancer. This procedure involves the selective uptake of a photosensitizer (PS) into targeted cells, which are then irradiated with red light, leading to a photochemical reaction producing singlet oxygen and other reactive cytotoxic oxygen species causing direct tumor cell death. PDT is highly selective, reducing damage of normal cells near the tumor. PDT causes direct cytotoxicity on tumor cells, damage to the tumor vasculature, and local inflammatory response. Specific localization of PS can be achieved by targeting receptors that are over-expressed on certain tumor cells and not on normal cells. Rapidly growing cells such as tumor cells require more vitamins, so they produce more receptors for taking in these nutrients. PS such as chlorins (chlorophyll derivatives) can be chemically bound to vitamins allowing for increased cell uptake. Chlorins are chlorophyll derivatives that show enhanced tumor localization than normal tissues. Synthesized photosensitizers used in this experiment involve a chlorin conjugated to a pantothenic acid (Vit B5) or lipoic acid including its zinc and indium complexes. Molecular structure characterization was performed using 1D/2D NMR, UV-visible and mass spectrometric techniques. Photodynamic efficacy of the synthesized photosensitizers will be evaluated in vitro using triple-negative breast cancer cell line and will be compared to triple-positive breast cancer cell line.
Light-Activated Degradation of Glyphosate by Palladium-Decorated m-BiVO4/BiOBr Nanosheets

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Glyphosate (\(N\)-(phosphonomethyl)glycine) is one of the most widely used herbicides in the United States and was found to be persistent in the environment. Recently, glyphosate was classified as a potential carcinogen and was related to the disruption of gut bacteria in honey bees. In this poster, we report the light-activated catalytic degradation of glyphosate by Pd/m-BiVO4/BiOBr photocatalyst. We prepared the ternary composite photocatalyst Pd/m-BiVO4/BiOBr according to our previously reported one-pot synthesis. The photocatalytic activities of the bare m-BiVO4/BiOBr and the Pd-decorated particles, with various Pd content, were elucidated via the degradation of rhodamine B. These materials were employed in the photocatalytic degradation of glyphosate and subsequently aminomethylphosphonic acid (AMPA), under simulated sunlight irradiation. The kinetics of the photocatalytic degradation reaction was studied by monitoring the concentration of glyphosate and its degradation products by ion chromatography. Complete degradation of glyphosate by Pd/m-BiVO4/BiOBr was achieved within 5 minutes of the reaction. Such results could lead to the development of sustainable catalytic technologies to minimize the global impact of glyphosate.

Synthesis, Characterization and Electrochemical Properties of Monolayer Protected Gold Nanoparticles

Zhihai Li
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Controlling the size of nanoscale entities is important because many properties of nanomaterials are directly related to the size of the particles. In this study, hexanethiol–gold nanoclusters are synthesized via the modified Brust-Schiffrin method. Synthesized nanoparticles were characterized with various analytical techniques such as TEM, scanning tunneling microscopy (STM). We have systematically varied the molar ratio of gold to protecting agent to discover the effect of gold-to-ligand ratio on the size of gold clusters. It is found that the average particle size decreases from 4.28 ± 0.83 nm to 1.54 ± 0.67 nm as the gold-to-ligand molar ratio changes from 1:1 to 1:9. Further, we also explored the electrochemical application of synthesize nanoclusters in terms of quantized charge at electrode-electrolyte interfaces, and capacitance of monolayer-protected nanoclusters were
calculated. The stability of Au nanoparticles is also studied and found the particles are stable on platinum surface, but disintegrated into atomically gold islands on Au(111), indicating that the instability and disintegration of nanoclusters are related to the properties of substrate electrode materials.

Poster
A Study on the Gating Properties of a Mutant Connector from Bacteriophage Phi29

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The connector from bacteriophage Phi29 is a channel protein where genome DNA can pass during DNA packaging. As DNA is packaged into the prohead, its shell acts as a semipermeable membrane which causes Donnan potentials to build up across it. Due to high concentration of DNA in the prohead at the late stage of DNA packaging, high pressure acted upon the shell arises. No technique available today can be used to study what roles the signals, pressure and Donnan potentials can play in how they affect functions of the channel protein in DNA packaging, instead planar bilayer membrane technology is used as a tool to study the gating properties of ion channels.

Using this, a study on the wild-type connector was conducted. Results suggested that a physical model could be used to explain roles of the connector at the late stage of DNA packaging. In the model, protein used an inherent Brownian ratchet to unidirectionally transport DNA into the prohead. In turn, Donnan potentials and internal pressure may correlate to fluctuations of the channel so that it can produce an asymmetric, periodic force onto the DNA to ratchet Brownian movement into the capsid.

In this study, we used a mutant connector for the planar bilayer membrane measurement. To avoid non-specific interactions with channel proteins, potassium chloride was used to replace a hydrophilic polymer creating osmotic pressure. We compared the gating properties of the mutant connector under different voltages and pressures. We found that the voltage required to fluctuate and gate the channel can be lowered when the channel is placed in a membrane with certain osmotic pressure. In addition, we found that the mutant connector can fluctuate at a higher-voltages than the wild-type, suggesting the mutant connector could be a potential ideal candidate protein channel for biological nanopore sensor.
Poster

**Photodegradation of variously substituted tetrazolethiones**

**Ethan Gibson**, Sundeep Rayat*
Ball State University

*Tetrazole* and its derivatives have a wide range of uses in many fields. These can be used for the treatment of hypertension, allergic reactions, bacterial infections, convulsions, cancer and AIDS. In agriculture, tetrazoles are used as plant growth regulators, herbicides and fungicides. These are also used in imaging technology as photostabilizers. These molecules have very interesting photochemical properties and can be exploited to afford synthetically useful photoproducts. Rayat group has studied the photochemistry of 1-methyl-4-phenyl-1H-tetrazole-5(4H)-thione and 1-(3-methoxyphenyl)-4-methyl-1H-tetrazole-5(4H)-thione in acetonitrile at 254 and 300 nm. The expulsion of molecular nitrogen and sulfur from these compounds resulted in the formation of corresponding carbodiimide as the sole photoproduct. Currently, the photochemistry of 1-(4-chlorophenyl)-4-methyl-1H-tetrazole-5(4H)-thione, 1-(4-nitrophenyl)-4-methyl-1H-tetrazole-5(4H)-thione, 1-(4-methoxyphenyl)-4-methyl-1H-tetrazole-5(4H)-thione, 1-(4-trifluoromethylphenyl)-4-methyl-1H-tetrazole-5(4H)-thione, and 1-(3-nitrophenyl)-4-methyl-1H-tetrazole-5(4H)-thione is being studied in acetonitrile at 254 nm via 1H NMR and UV-Vis spectroscopy, to study if the electronic effect of different substituents influences the photodegradation. The resulting 1H NMR and UV spectra provides evidence in support of the formation of a single product. Product confirmation studies will determine the identity of the photoproduct.
Ecology

Presentation

Habitat Type use of Migrating Northern Saw-whet Owls: A Second Year of Study

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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 in Indiana, but efforts have not been widespread. Christmas tree farms contain the coniferous trees preferred by these owls in their breeding habitat, but has the potential to be too disturbed. This study examines if there are any differences in capture rates between these two habitat types. 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. The past three seasons (two falls, and one spring) have yielded 29 owls, 15 at Whitetail, 14 at Ginn Woods. Of these owls, only 2 were captured in the spring, and only 5 were either male or unknown sex. This presentation will also include data on the second spring capture season.

Presentation

Effect of parasites on white-throated sparrow (Zonotrichia albicollis) song.

Zoé Delefortrie & Rusty A. Gonser

Secondary sexual characteristics in birds, such song and plumage color, are associated with sexual selection behaviors such as same sex competition and mate choice. Generally, secondary sexual traits are energetically costly to express or maintain, therefore variation in these traits can serve as an honest signal of an individual’s quality. The Hamilton & Zuk Hypothesis supported that females choose their mates based on their parasite resistance. This resistance could influence sexual selection through the expression of secondary sexual characteristics. The objective of my research is to understand the influence of parasitism on secondary sexual characteristics involved in sexual selection. A number of studies have investigated the effect of haemosporidians on songbird behavior showing that parasite loads influence
reproductive success and singing behavior. The white-throated sparrow (Zonotrichia albicollis) is a passerine with a unique genetic polymorphism that is linked to behavioral and color polymorphism (white and tan). The purpose of this research is to understand whether these birds transmit information about their fitness status, such as resistance to parasites, through their song and if this information differs between the two white-throated sparrow morphs. It has been shown that tan morphs had higher parasite loads than white morphs. We are comparing the song of parasitized birds before and after anti/protozoal treatment to investigate if there are perceptive differences between parasitized and non-parasitized songs. We expect that the parasite loads will influence the song quality and that non-parasitized individuals will have a higher song rate, as shown in other studies. This research will increase our understanding of the impact of parasites on behavior and set the foundation for future studies investigating the genes underlying sexually selected behavior.

**Poster**

**Will the intrusion of an avian nest predator illicit a change in the behavior of a canopy-nesting passerine?**

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Kamal Islam, Department of Biology, Ball State University, Muncie, Indiana 47306-0440, USA

The Cerulean Warbler (Setophaga cerulea), a small, neo-tropical migrant, has gained significant attention in recent years as a species that has declined faster than any other North American songbird. Since 2007, we have been monitoring Cerulean Warbler populations within the Morgan-Monroe and Yellowwood state forests of Southern Indiana. Previous studies on cavity nesting and communal cup nesting species suggest that parents will adjust their behavior in response to an avian nest predator, to either avoid detection, or to fend off the predator. These behaviors include mobbing, selective provisioning of nestlings, and increased vigilance and aggression on the nest. Studies suggest that forest fragmentation positively impacts nest predators, and in-turn leads to an increase in nest predation. The objective of this study is to determine if and how Cerulean Warblers change their behavior when a common avian nest predator, Blue Jay (Cyanocitta cristata), is in the vicinity of the nest. In the summer of 2018, we conducted a preliminary study on seven Cerulean Warbler nests found at our study sites. During the observation period, three of the nests were randomly assigned to a control group, and subjected to Ovenbird (Seiurus aurocapilla) vocalizations, and the other four nests were assigned to a treatment group, and subjected to Blue Jay
vocalizations. We videotaped parents at the nest during the observation period to analyze their behavior. The results of this preliminary study will be presented. This study is the first to analyze the behavior of an open-cup, canopy nesting species in response to an avian nest predator. Data collected from this study will determine if Cerulean Warblers alter their behavior to avoid detection of their nest, and may help shed some light on the causes of poor nesting success in this species.

Poster
**What is the Relative Influence of Bed Sediment Composition and Water Chemistry on Aquatic Macroinvertebrate Metrics in Agricultural Headwater Streams?**

Tyler C. Shuman, Robert B. Gillespie, Purdue University Fort Wayne, Fort Wayne, Indiana. and Peter C. Smiley Jr.2. USDA-ARS, Soil Drainage Unit, Columbus, Ohio.

Macroinvertebrates of channelized headwater streams in agricultural landscapes are exposed to pesticides and altered bed sediments. We hypothesized that macroinvertebrate assemblages inhabiting sediments with a low percentage of gravel and low organic content are less diverse than those with greater gravel and organic content. We further hypothesized that agricultural chemicals in the water column would have less influence on macroinvertebrate assemblages than bed sediment characteristics. We analyzed particle-size distribution, percent organic content, and agricultural chemical concentrations of three channelized headwater streams and one reference site. Water samples were collected weekly, sediment samples were collected three times from May-September, and macroinvertebrate assemblages were sampled twice. Taxon richness was significantly greater at the reference site (mean = 13; 11.33-14.67) than that at channelized sites (mean = 8.4; 7.00-10.67). The percent of sediments with particles greater than 16.0 mm (gravel) was significantly greater at the reference site (mean = 12.2%; 4.70-18.20 %) than that at channelized sites (mean = 2.53%; 0.00-10.33 %). However, organic content did not differ significantly between channelized (mean = 2.2%; 1.1-3.1 %) and reference (mean = 2.8%; 1.7-4.8%) streams. Additionally, mean total concentrations of pesticides did not differ between channelized sites (mean = 0.80 ppb; 0.10-2.06 ppb) and reference sites (mean = 0.07 ppb; 0.02-0.15 ppb). These data support the hypothesis that sites with greater gravel content in sediments have greater macroinvertebrate taxa richness. However, it appears that organic content of sediments had no influence on macroinvertebrate richness. Although, as predicted, pesticide concentrations in water were not associated with taxa richness, it is possible that pesticide concentrations in sediments could negatively influence the diversity of macroinvertebrate assemblages. Multivariate analyses of these independent variables and diversity metrics could determine the relative importance of physical and chemical sediment characteristics on macroinvertebrate diversity of channelized headwater streams.
Poster

Sustainable Urban Agricultural Systems in Indianapolis

Becca Lewis, Butler University
Sean Berthrong, Butler University

Urban agriculture is capable of restoring ecosystem services such as recreation, food production, and clean soil and water. Urban farms can help relieve pressure for areas with limited food access, also known as food deserts. This is especially important to the community of Indianapolis because the city is surrounded by one of the largest food deserts in the US. To help a community, an urban farm must have healthy, nutrient rich soil. Nitrogen is a limiting nutrient for plants when it comes to growth and development. Plants cannot produce nitrogen; they acquire the mineral by external fertilizers or internal N-fixing bacteria. The goal of this project is to test if urban farming practices are increasing the abundance and variety of N-fixing bacteria. Because the farms under investigation utilize symbiotic legume-N-fixing plants, it is hypothesized that there will be more copies and variety seen in the bacteria of soils that have been farmed longer. Analysis of the results of this project focus on DNA-based methods of quantifying and identifying N-fixing bacteria as well as investigate common soil health indicators such as organic matter composition, pH, and quantities of key elements. This project coincides with longstanding, ongoing research conducted at Butler University that focuses on the overall health of urban farms. The soils used were collected from multiple sites around the city; therefore, the data collected can be analyzed individually and in the context of the larger project with the goal of helping farms across Indianapolis improve overall sustainability.

Presentation

Effect of Garlic Mustard (an Invasive Plant) on Survival and Mycorrhizal Fungal Abundance in Hydrophyllum appendiculatum (a Native Understory Plant)

Heidi Porod, Catherine Vaerewyck, Deborah Marr, Indiana University South Bend

*Hydrophyllum appendiculatum* is a native biennial understory plant that has declined over 100-fold between 2003-2018 in St. Patrick’s County Park, St. Joseph County, Indiana. We tested two hypotheses regarding whether *Alliaria petiolata* (garlic mustard) is contributing to the decline of *H. appendiculatum* (HA). First, we tested whether presence of garlic mustard affects leaf production or leaf mortality in first-year HA in a greenhouse experiment. HA leaf mortality was lower with GM present (mean 44%), but there was no difference in survival between allyl isothiocyanate and control treatments (72% and 68% respectively). Second, we compared mycorrhizal abundance in HA roots collected from forests with and without GM. Garlic mustard has been shown to decrease mycorrhizal spore abundance in soil by releasing a
secondary compound, allyl isothiocyanate (AITC) into the soil. There was no difference in amount of mycorrhizal fungal structures in HA roots collected from forested areas where garlic mustard was absent compared to areas where garlic mustard was common. These results suggest that garlic mustard had minimal effects on leaf production and ability of mycorrhizae to colonize roots in first-year *H. appendiculatum*. Experiments testing longer-term exposure and effects on second-year plants are needed to determine whether garlic mustard is contributing to the decline of *H. appendiculatum*.

Poster

**Indiana Master Watershed Stewards**

Leslie Dorworth Purdue University Northwest

**Vanessa S. Quinn** Purdue University Northwest

The Indiana Master Watershed Steward (IMWS) Program is a pilot project, developed in partnership with the U.S. Forest Service, Urban Waters Initiative, The Nature Conservancy, Indiana Dunes National Park, the Lake Michigan Coastal Program, Purdue University Extension, Purdue University Northwest, Northwestern Indiana Regional Planning Commission, and Illinois-Indiana Sea Grant. The program offers communities the opportunity to get involved at the local level to protect their waterways and associated water quality. The program grew out of the efforts of various organizations in northwest Indiana concerned with improving and protecting the Lake Michigan Watershed in Indiana. The purpose of IMWS is to recruit and instruct volunteers who will work on various water quality and pollution prevention and control projects in their own communities. The Pennsylvania Master Watershed Steward Program was used as a model for the development of the program. Program materials cover the following topics: (1) Fundamental of Watershed Hydrology; (2) Water Quality and Quantity; (3) How Land Use affects Water Quality and Quantity; (4) Weather and Climate Vulnerability; (5) soil and Geology; (6) Green Infrastructure; (7) Trees and Forestry; (8) Agricultural Best Management Practices; (9) flooding and Stormwater; (10) Wildlife in the Watershed; (11) Benthic Macroinvertebrates; (12) Invasive Species; (13) Stream and River Recreation; and (14) Citizen Science and Volunteers. There topics address such issues as habitats, food webs, watershed management, agriculture, pollutants and climate variability. The materials represent issues across the state and is therefore transferrable across the state. In order to become a steward, one needs to complete 35 hours of volunteer time with an approved partner. To continue to be a steward once certified, the individual needs to complete extra training annually.
Influence of a lowhead dam on macroinvertebrate assemblages of an Indiana stream.

Deanne E. Jensen and Robert B. Gillespie, Purdue University Fort Wayne

The presence of a lowhead dam has been shown to modify a stream’s hydrogeomorphology, with increased siltation upstream and substrate scouring downstream. This shift in habitat structure may result in longitudinal differences in biological assemblages. Our objective was to provide preliminary qualitative data on macroinvertebrate assemblage diversity and habitat structure upstream and downstream of a lowhead dam traversing Cedar Creek in the St. Joseph River watershed of northeastern Indiana. This survey was developed in part to inform decisions regarding a dam removal project proposal. We predicted that macroinvertebrate diversity would be higher upstream of the lowhead dam compared to downstream as a result of variations in hydrogeomorphology and habitat availability. Macroinvertebrates were sampled within 50 meter reaches upstream and downstream via dip nets and Surber samplers. Hydrogeomorphological and habitat features were measured at three transects within each reach. The upstream reach had a greater mean depth (60.2 cm), a lower mean velocity (0.08 m/s), and higher abundance of fine sediments (75% sand/silt) compared to the downstream mean depth (56.5 cm), velocity (0.11 m/s) and substrate composition (70% sand/gravel). A Shannon Diversity Index calculated a H’ value of 1.94 for the upstream reach versus a H’ value of 2.46 for the downstream reach, indicating greater diversity in the downstream reach. Upstream assemblage evenness (0.58) was also lower compared to downstream evenness (0.77). These results are contrary to our prediction. However, the presence of a two-lane bridge in the upstream reach appears to have influenced available habitat and subsequent assemblages. Our results suggest that the lowhead dam within Cedar Creek has altered the physical and habitat characteristics of the stream, thereby contributing to differences in macroinvertebrate assemblage diversity above and below the dam. Further investigation is warranted to quantify the effects the lowhead dam has on instream habitat and macroinvertebrate community metrics.
Investigation of Community Structure in the St. Joseph River: Connecting Plant Diversity with Macroinvertebrate and Fish Diversity

Steven Swanson - Indiana University South Bend
Daragh Deegan - City of Elkhart
Andrew Schnabel - Indiana University South Bend

We surveyed plant, macroinvertebrate, and fish diversity in the St. Joseph and Elkhart Rivers in order to estimate diversity at multiple trophic levels, to better understand interactions between plant and animal communities, and to identify nonnative species. We sampled from two sites along the St. Joseph River in South Bend IN and two sites along the Elkhart River in Elkhart IN and Goshen IN. Plant sampling used a stratified random design with 50-m transects running parallel to the shoreline in areas of vegetation. Plants were harvested at three random intervals along each transect. Macroinvertebrate samples were collected at the same sampling points as for plants. Fish were sampled using seines and backpack electrofishers along 2-3 transects per site. In total, we sampled 17 plant species, 28 fish species, and 34 macroinvertebrate families, with mean (± SD) richness per site of 11.7 ± 2.5, 15 ± 0, and 16.7 ± 4.6, respectively. Species composition varied between the two rivers, most notably in macroinvertebrates. Gastropods were found exclusively in the St. Joseph River, and 93% of those gastropods came from just one of the two sites. The Elkhart River had 182% more gammarids than did the St. Joseph River. Of the 28 fish species identified, nine were common to both the St. Joseph and Elkhart Rivers, with four of those species being found at each site sampled. Of the 17 plant species identified, three were non-native (Cabomba caroliniana, Myriophyllum spicatum, and Potamogeton crispus), with C. caroliniana and M. spicatum being found only in the St. Joseph River. Additional sites along both rivers should be sampled to gain a better understanding of the differences between species composition by location and to identify other possible non-natives.

Animating beetle community dynamics

Jeffrey D. Holland, Purdue University

The communities of longicorn beetles found in forests are altered by forest disturbance and the subsequent plant community dynamics. The Hardwood Ecosystem Experiment in south-central Indiana is exploring these biotic responses to timber harvest in a large-scale, long-term, controlled experiment. This presentation uses three-dimensional animations to visualize the alterations in the beetle community to explore hypotheses of community change and resilience.
Poster

*Houttuynia cordata*, an exotic and invasive plant species in your garden

Xianzhong Wang, Patricia Clark

Indiana University-Purdue University Indianapolis

A relatively little known garden species, *Houttuynia cordata*, has been found to be highly invasive and extremely difficult to control once established. Here we describe the morphological characteristics of the species and report the effectiveness of using selective and non-selective herbicides in controlling this species in a greenhouse study. *H. cordata* is a perennial herbaceous species that typically grows prostrate, but can grow to 0.8 m tall. Leaves are heart-shaped, alternate and sometimes variegated. Inflorescences are spikes with four conspicuous white basal bracts. Flowers are green-yellow and inconspicuous. Seeds ripen in summer and are rarely seen because of their small size. *H. cordata* adapts to a wide range of environmental conditions but prefers moist and nutrient-rich habitats. It propagates both sexually and asexually with asexual reproduction being the main mode of reproduction. In our study, application of herbicides caused leaves and stems to die back, but had little effect on the rhizomes, which re-sprouted in four weeks. Results from our study and observations from gardeners across the U.S. on the tenacity and aggressiveness of this species demonstrate that *H. cordata* is a highly invasive species and could pose serious threat to the ecological integrity of native ecosystems in the United States.

Presentation

Bat summer habitat use 9 years after experimental timber harvest at two Indiana State Forests

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

The Hardwood Ecosystem Experiment (HEE) is the most comprehensive study of forest management impacts to 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 multi-faceted project has included acoustic monitoring of bat activity in HEE research areas at Morgan-Monroe and Yellowwood State Forests. The HEE research area consists of three replicated treatments: two treatments harvested using even-age and uneven-age silvicultural methods, while the third treatment remains unharvested mature forest as a control. Even-age treatments consist of 10-acre clearcuts and 3-stage shelterwoods within a matrix of unharvested mature forest; each uneven-age treatment included eight 1-5 acre patch cuts within a matrix harvested using single-tree selection. Findings from the post-harvest period spanning 2009-2017 will be presented. Bat calls were collected using ultrasonic
detectors (Anabat II/ZCAIM and SD2) and identified to species based on call characteristics using the automated classification software BCID (version 2.7d), followed by manual verification of individual auto-classified calls. Multiple-season occupancy models were used in program Presence to estimate site occupancy (i.e. “use”) and detection probabilities. During the study period, white-nose syndrome (WNS) was introduced to bat populations in Indiana. This devastating disease has caused significant declines in bat populations among species that had previously been common on State Forests. Findings will be discussed as they relate to species-specific timber harvesting effects, as well as the impact WNS appears to be having on bat occupancy at these State Forests.

Poster

Influence of crop fields on fish assemblages within channelized streams.

Gabriel L. Curtis, Darren J. Shoemaker, and Robert B. Gillespie. Purdue University Fort Wayne

An important factor regarding habitat restoration is determining which environmental variables most critically impact aquatic communities, so managers may effectively focus their restoration efforts. One such variable is mean distance to nearest crop field. This variable can provide insight into how likely a stream is to be disrupted by agricultural activities. Here, we examine if that distance may have an influence on fish assemblages. Riparian habitat at these sites is typically adjacent to agricultural row crop a few meters from the water’s edge. Agricultural activity such as plowing, driving, and harvesting can expose streams to runoff, erosion, organic wastes, and disrupt canopy cover. Our study area comprised three ditch sites along Cedar Creek in Waterloo, Indiana and one reference stream in Hillsdale County, Michigan, all within the St. Joseph River Watershed. Fish surveys were conducted along 125 meter transects. Across two years, we surveyed fish assemblages with electrofishing and seine netting, and measured distance from water’s edge to crop field every 25 meters at each transect. We hypothesized that sites with greater distance would have a higher richness, evenness, and Shannon Diversity Index within fish communities. Mean distances to crop field were calculated for ditch sites (10.75 meters) and the reference site (1605.23 meters). Shannon Diversity, species evenness, and species richness were calculated for both ditch and reference sites, respectively: mean Shannon Diversity (1.82; 1.71), mean evenness (0.70; 0.66), and mean richness (13.5; 13.5). Preliminary analysis of these data suggest that fish assemblage metrics do not vary significantly between ditch and reference sites, and do not appear to be influenced by distance to crop field. We propose that impacts on fish assemblages cannot be attributed to a single variable, and that future analyses should incorporate multiple variables to identify factors which most influence those communities.
Pollinator Diversity and Abundance in Neighboring Urban Wetlands

David Mitchell, Jessica Jones, Andrew Schnabel, Deborah Marr
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Ecological studies help us understand the complex relationships organisms have with one another. One major aspect of the ecosystem revolves around the intricacies of plant and pollinator relationships. Pollinators such as bees, flies, and other insects are vitally important, because they aid in seed production. Learning about pollinators in restored habitats in urban areas will allow us to better understand what is needed to preserve these important relationships. We observed insect visitors to flowers of *Asclepias incarnata*, *Iris virginica*, *Penstemon digitalis*, *Physostegia virginiana*, *Silphium perfoliatum*, and *Tradescantia ohiensis* at two restored wetlands on the IU South Bend campus. For all six species, we estimated rates of insect visitation and recorded the diversity of potential pollinators. We also applied fluorescent dyes flowers of four species to test for pollinator movement between wetlands, and we performed supplemental pollinations on two species to test for pollen limitation of fruit production. Visitation rates were similar between wetlands for all species, but showed high variation from day to day and between time periods within days. *Asclepias incarnata* and *I. versicolor* had a greater diversity of visitors than did the other four species. Bees and flies were dominant visitors to all species, except to *A. incarnata*, where beetles were also frequent visitors. We found no evidence of pollinator movement between the two wetlands, which were separated by about 250 m of mowed lawn. Supplemental pollination produced small and insignificant increases in fruit set and fruit mass compared to natural pollination in both *P. digitalis* and *T. ohiensis*. Our wetlands are small and recently created, but have fairly high insect visitor diversity, and plants do not appear to be limited by low levels of pollen. Connectivity between wetlands might be increased by replacing the current lawn grass with native plantings in the areas between wetlands.

Poster

Habitat effect on bird diversity in two commonly found habitats in Central Indiana: a small urban park and a wooded patch habitat.

Tanner Langley, Jennifer Weideman, and Lina Rifai, Indiana University Kokomo

In an attempt to better understand how to preserve local bird communities and biodiversity in central Indiana, we compared two commonly found habitats in this region: a small urban park and a wooded patch habitat surrounded by agricultural fields. We selected Jackson Morrow Park, in Kokomo, as the urban park site and a site
30km southeast of the park as the wooded patch habitat. We conducted point counts to collect data on bird diversity from these two sites. Our results showed that species richness as well as species diversity of birds was higher in the urban park habitat. Both habitats had two invasive species present, the European Starling (*Sturnus vulgaris*), and the House Sparrow (*Passer domesticus*). Approximately one third (32.3%) of the bird abundance observed in Jackson Morrow Park was due to these two species. However, biodiversity indices indicated a more diverse bird community, with and without including these invasive species in our calculations, at Jackson Morrow Park. The higher species richness and diversity in Jackson Morrow Park can most likely be attributed to the fact that it represents a diversity of habitats including patches of native grassland habitat. Grassland habitats are declining rapidly and typically offer support for a wide variety of bird families. The wooded patch habitat from this study represents a relatively small area within an agricultural region, which is known to create an edge effect, in which only a small number of bird species are known to be able to survive in (indeed when removing invasive species from our calculations, diversity indices dropped dramatically here). Understanding these habitat effects on diversity can help us find ways to mitigate the rapid decline of bird populations across the country.

Presentation

**Assembly of Arthropod Communities at Restored Prairie, Old Field and Monospecific Exotic Grass (*Phalaris arundinacea*) Stand in Northwestern Indiana: A Functional Perspective**

**Eric Kelleher**, Purdue University Northwest

Effects of enhanced diversity of plant species on arthropod diversity was investigated. In doing so, a total of 35,408 arthropods belonging to 13 taxa in the restored prairie (RP1 and RP2), old field (OF) and monoculture stand of *Phalaris arundinacea* (reed canary grass – RCG) sites, located in Gabis Arboretum, Valparaiso, Indiana, were captured, counted and compared. The enhanced diversity of plant species caused by restoration of prairie vegetation did not appear to promote the diversity of arthropod taxa. However, the restoration led to a more balanced composition of arthropod functional groups and thus elevated the diversity of functional groups. As a result, the arthropod assemblages in the three sites diverged clearly, according to my canonical correspondence analysis (CCA) ordination. Pollinator abundance was greatest at RP sites and least at RCG, positively correlating with greater forb diversity, suggesting greater potential for nectar feeding and pollination potential at RP sites. Herbivore abundance was also greatest at the RP sites, positively correlated with increasing plant species diversity. Predator abundance was significantly greater at the RCG site compared to the OF and RP sites; it was positively correlated with greater C3 grass cover, a characteristic of the structurally homogenous RCG site, and negatively
correlated with increasing plant diversity and forb cover, a characteristic of the diverse and more structurally complex RP sites. Given the apparent non-random distribution of arthropods among the field types, our results suggest plant species composition has a significant effect on arthropod assembly. The monoculture grass stand was found to have a predator dominated arthropod community supported by a small, diverse herbivore community. It is concluded that the prairie restoration has resulted in alteration of arthropod communities supporting greater pollinator and herbivore abundance and a more balanced ratio of herbivores to predators due, in part, to increased plant structural diversity.

Poster
The effect of fire on spider assemblages in central Indiana forests

J. Gonsiorowski, N. Tuft, J. Acosta, L. Frandsen, and M. A. Milne, University of Indianapolis

Disturbance, including fire, can dramatically change the animal communities living within the affected environment. The Hardwood Ecological Experiment (HEE) is a 100-year study that encompasses experiments run at Yellowwood State Forest and Morgan-Monroe State Forest in central Indiana. The HEE measures the ecological effects of different types of forest management practices. These effects include the resilience of communities, whereby communities, in response to a disturbance, slowly shift back to a pre-disturbed state. Specifically, our study measured the effect of prescribed burns on the diversity, richness, and resiliency of spider communities in these forests. We collected spiders within 24 sites in both forests before and after burns for ten months a year for four years using pitfall traps, sweep nets, and berlese funnels. We found that spider communities significantly changed after a fire in both abundance and composition, but not in diversity. These spider communities were also quite resilient, as evidence of recovery from the disturbance appeared after only two years. Future studies of spider assemblages in and outside HEE will help to determine spider resiliency and how forest management practices can adjust in order to maintain natural spider communities.

Poster
The Genetic Contribution of Small-Mouthed Salamanders to Sympatric Unisexual Ambystoma

Minh T. Le, and Mark A. Jordan, Purdue University Fort Wayne

Unisexual reproduction should be unfavorable in nature due to the limited genetic variation that is passed on to the next generation. Nevertheless, the
unisexual *Ambystoma* is an all-female Amybstomatid salamander that can reproduce asexually and was derived from a hybridization event between two bisexual species: *Ambystoma jeffersonianum* and *Ambystoma laterale*. Genetic evidence has demonstrated that the unisexual lineage has persisted for five million years, making it the most ancient unisexual vertebrate species to date. The key to this persistence in the face of limited genetic variation is that the unisexual *Ambystoma* does not strictly reproduce asexually. It can mate with males of other bisexual *Ambystoma* species within a community, incorporating the male’s haploid genome into its own genome—a reproductive mode called kleptogenesis. An incorporated haploid genome is known as a subgenome, and it usually replaces a pre-existing subgenome.

A community consisting of unisexual *Ambystoma* and *Ambystoma texanum* is found at the Eagle Marsh Natural Preserve, Allen County, Indiana. The subgenomic pattern in the unisexual population suggests that *A. texanum* had historically contributed its genome to the unisexual’s gene pool. However, it is unknown at what rate kleptogenesis is still occurring in this community, if any. At this stage of the research, 16 microsatellite loci were tested on *A. texanum, A. jeffersonianum*, and *A. laterale* using PCR and gel electrophoresis to find microsatellites that uniquely amplify *A. texanum* genome. This is to distinguish the *A. texanum* subgenome from the other species when we examine the unisexual genome later on. The results show that 10 out of 16 microsatellite loci amplify differently in *A. texanum*. This research will provide insight into the ecological dependency of the unisexual *Ambystoma* on the *A. texanum*, providing new knowledge on unisexual reproduction and supporting conservation effort on this species.

Poster
**Herpetological Surveys of Red Tail Land Conservancy Properties**

*Maurice Dantzler* and Kamal Islam
Department of Biology, Ball State University, Muncie, IN 47306

Reptiles and Amphibians are excellent bioindicator species that provide a benchmark for the condition of an ecosystem. Amphibians use cutaneous respiration which allows for direct intake of any liquid or gas they may encounter in an environment. Reptiles, being a group of species that have a wide range of diets from small mammals to vegetation, will display a drop in population size if their food resources are limited. During the summer of 2018, a search and seizure was used for the capture reptiles and amphibians found on four Red Tail Land conservancy properties: Reber Woods, McVey Woods, White River Woods, and Fall Creek Woods. The objectives for these surveys were to 1) obtain information specific to species diversity, 2) species abundance, and 3) environmental/weather dependent factors that may affect the behavior of reptiles and amphibians. In addition, Geographic Information System (GIS) maps were created to display where species were located on the properties.
These surveys are the first to record field data for these four Red Tail Land Conservancy sites. Data from these surveys will provide Red Tail Land Conservancy information on amphibians and reptiles for species-specific management on the properties.

Presentation

**Interseeding forbs in a grass-dominated prairie restoration in northeast Indiana: Year six results.**

Adam R. Thada, and Robert T. Reber, The Center at Donaldson

Many tallgrass prairie restorations fail to match the level of floral diversity found in undisturbed prairie remnants. Restorations often become excessively dominated by warm-season grasses at the expense of forbs due to the lack of historical disturbance regimes. Interseeding new species often requires a disturbance mechanism to aid establishment of new plants. In April 2013, five native forb species were seeded into a restoration dominated by *Andropogon gerardii* (Big Bluestem). Test plots were treated either once or twice during the growing season with a grass-specific herbicide in order to examine effects of this disturbance on the dominance of *A. gerardii* and the establishment of interseeded forbs. Treatments were completed for the first three growing seasons. Plots were sampled again in year six (June 2018). Aerial coverage and above-ground biomass of *A. gerardii* was significantly lower in both once and twice-treated plots in comparison to control plots. The abundance and coverage of some species of interseeded forbs were also significantly greater in herbicide-treated plots, starting in year one and continuing through year six. Where other management options such as grazing or mowing are limited, grass-specific herbicide application could constitute a cost-effective tool for land managers interested in increasing floral diversity.

Poster

**Use of agricultural fields by bats, and its implications for pest control**

Morgan Chaney, William Mitchell, Indiana State University

Insectivorous bats in the Midwest rely on agricultural land and adjacent forest for foraging, travel, and roosting. I plan to determine how Indiana’s bat species are utilizing this landscape. Specifically, I am investigating whether the size and isolation of agricultural fields, and insect density, affects how bats choose to utilize agricultural areas. I hypothesize that overall bat activity decreases with distance from the tree line, but the rate of decrease varies among species; that is to say, some species will stay near corridors while others will utilize the entire field due to habitat preferences and ecomorphology. Insect samples and sonogram recordings will be taken in Vigo,
Vermillion, Owen, and Sullivan County, Indiana from mid-June until mid-September. I will analyze samples using generalized linear mixed models (GLMM) with bat acoustic activity as the response variable and closest tree patch, field size, insect density, distance to water, and temperature as predictor variables. Analysis so far suggests that bats are avoiding open field in favor of areas closer to forests edges and tree lines. This result suggests that larger, homogenous (tree-less) agricultural landscapes receive less pest control benefits than landscapes with smaller patches of agricultural fields. If my results are representative, then bat foraging patterns could directly influence how field owners in the Midwest manage their field boundaries to promote natural pest control. The re-establishment of fence row vegetation could be necessary if homogenous landscape owners wish to benefit from their natural predators.

Poster

Effects of predator presence on aphid (*Lipaphis psuedobrassicae*) reproduction

Meghan J. Wright, Lanae J. Singleton, and F. Collin Hobbs, Department of Biology, Huntington University

When preyed upon, aphids release alarm pheromones such as β-farnesene to inform nearby aphids of the predation threat. Other studies have shown that aphids exposed to β-farnesene exhibit stress behaviors and are more likely to produce winged morphs to aid in dispersal. However, few studies have examined the effect of alarm pheromones on aphid reproductive rates. We hypothesized that exposure to alarm pheromones released during active predation would result in decreased reproductive rates in our study species, the false cabbage aphid (*Lipaphis psuedobrassicae*).

Our experiment consisted of a predation treatment and a non-predation control with six replicates each. Each replicate consisted of a sealed, clear plastic arena containing 50 aphids and a supply of radish seedlings for a food source. In each predation arena four ladybugs (*Hippodamia convergens*) were placed in a smaller, screened feeding chamber that also contained aphids. Non-predation replicates were identical except that they did not contain ladybugs. Ventilation was used to prevent cross contamination of alarm pheromones between arenas. The experiment ran for 10 days, after which the aphids in each replicate were counted. Predation arenas had, on average, 1,881 (standard error = 115.6) aphids per arena at the end of the experiment, while non-predation arenas had on average 1,550 (standard error = 143.1). Despite limited replication, a one-tailed t-test was nearly significant at an alpha of 0.05 (p=0.051). Our results indicate that alarm pheromones may actually increase the reproductive rate of aphids, suggesting an alternative strategy for dealing with predation stress. More replication of the treatments, as well as testing different concentrations of alarm pheromones, may provide more conclusive evidence.
Results of a pilot study examining the space-use relationship between the territory and home range of Cerulean Warblers (*Setophaga cerulea*) in Indiana

Brandon Connare and Kamal Islam, Department of Biology, Ball State University

The Cerulean Warbler (*Setophaga cerulea*), a once common neo-tropical migrant throughout its breeding range, is one of the fastest declining North American wood-warblers. Listed as state-endangered in Indiana and a species of conservation concern across its range, this small songbird has recently been the subject of much research throughout the Eastern United States and Canada. Over the past 10 years, we have been monitoring Cerulean Warbler breeding populations at Yellowwood and Morgan-Monroe state forests in Southern Indiana as part of a larger 100-year project, the Hardwood Ecosystem Experiment. This long term study intends to determine the effect of a variety of forestry practices on local plant and animal communities. Understanding the impacts of forest management practices on Cerulean Warbler populations is reliant upon accurate estimations of the size of home range and territory. Previous research conducted at our study sites and elsewhere mapped Cerulean Warbler territories by following males and recording song-perch locations, which is difficult due to this species’ tendency to stay high in the forest canopy. For similar reasons, studies examining the size of this species’ home range outside of its defended territory have not been previously conducted. Our objective is to determine the size and seasonal plasticity of Cerulean Warbler territories and home ranges throughout the nesting cycle. In the summer of 2018, We conducted a pilot study on four male Cerulean Warblers using radio-transmitters. We tracked and recorded each bird’s location throughout its home range and territory. We present preliminary results from our study. A better understanding of Cerulean Warbler movements in its territory and home range is vital for the management and conservation of this declining species.
Commercial watermelon (*Citrullus lanatus*) production in the Midwest typically relies on neonicotinoid insecticides to manage insect pests, particularly for striped and spotted cucumber beetles (*Acalymma vittatum* and *Diabrotica undecimpunctata howardi* Barber, respectively). Common cucumber beetle predators include coccinellid beetles found on plants, ground-dwelling carabid beetles (Coleoptera: Carabidae), and lycosid spiders on the soil surface. However, these predators and the ecosystem services they provide are at risk from pest management practices used without regard to economic thresholds. The role of arthropod predators in managing cucumber beetles is poorly understood, and data on the effects of insecticides on predators in watermelon production is scarce. Our study compared predation and production yields under two treatments: 1) watermelons treated with weekly neonicotinoid insecticide applications surrounded by corn with neonicotinoid treated seeds (Conventional), and 2) watermelons treated only when pest populations reached economic thresholds following integrated pest management (IPM) practices surrounded by corn with untreated seeds. Spotted cucumber beetle eggs and waxworm (*Galleria mellonella*) larvae were used as surrogate prey to measure field predation in replicated 24-hour assays. Although insecticide inputs significantly decreased cucumber beetles in the Conventional field, predation and yield were higher in the IPM field. While the source of the field predation observed in these studies remains to be determined, the ultimate goal is to provide management recommendations to growers that maximize insecticide efficacy while minimizing their negative impacts on natural enemies and their ecosystem services.

Keywords: watermelon, neonicotinoids, natural enemies, predation.
Presentation

A survey of spiders for the Ecoblitz in Morgan-Monroe/Yellowwood State Forests in Indiana

Leslie Bishop, Earlham College, Marc Milne, University of Indianapolis, and Brian Foster, Indiana State University

Spiders, as both predators and prey, are important components of forest ecosystems. As part of the Indiana Forest Alliance Ecoblitz within 900 acres of the Back Country Area of Morgan-Monroe/Yellowwood State Forests, we collected 127 species of spiders between 2014-2018. Of these species collected, 31 are new distribution records for Indiana. Of the total number of species collected, 62% were collected in the bottomland habitat, 60% on slopes, and 19% on ridges. Only 10% of the total species were found in all three habitats. In pair-wise comparisons of habitats, we found that the species composition differed between habitats even when species richness was similar. Similarly, we found that our collection of spider species during the day differed in composition from those collected at night with only 26% collected during both periods. These data emphasize the benefits of a multi-year survey, such as the Ecoblitz, and the importance of sampling in multiple habitats as well as during the day and the night. The high number of new distribution records in our sample reinforce that spiders as a group are underrepresented in scientific studies of forests in Indiana.

Presentation

The Genus Camelobaetidius Demoulin 1966 (Insecta: Ephemeroptera: Baetidae) in eastern North America

Alan Barradas-Zarate, Luke M. Jacobus
Division of Science, Indiana University Purdue University Columbus

The mostly Neotropical genus Camelobaetidius is present but seldomly collected in eastern North America. Three species may be found in this region, including C. musseri (Traver & Edmunds 1968), C. waltzi McCafferty 1994, and C. warreni (Traver & Edmunds 1968). Of these three species, C. waltzi is the most widespread within the region. Camelobaetidius warreni is a mostly western species that might be considered extralimital, and C. musseri also is a western species but with an enigmatic record from an uncertain location in North Carolina. The seasonal, geographical, and ecological distributions of the species are reviewed, based on data from published records and materials housed at Purdue University and Clemson University.
Update on the Bee Surveys in the Ecoblitz Area of Morgan-Monroe and Yellowwood State Forests in Indiana

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Pollinators are important for the ecological services they provide and because many species are suspected of declining. Bees are among the most important pollinators due to the pollination services they provide, because they are species rich, and because several species have recently been listed under the Endangered Species Act. However, little research on bee abundance and distribution in forest ecosystems, especially in Indiana, have been performed. To expand this knowledge, bees were surveyed in a designated Ecoblitz Area of Morgan-Monroe State Forest in Monroe County in 2018. Additional areas were surveyed in Monroe County and Yellowwood State Forest in Brown County in 2016 and 2017. The survey sampled bees using three techniques: netting at flowers, blue vane trapping, and bowl trapping. Bowl trapping involved setting up transects comprising 30-12-ounce bowls (10 each of fluorescent blue, fluorescent yellow, and white). Blue vane trapping used three blue vane traps suspended from trees and separated by 50 m. Netting was conducted on twenty dates between 12 April and 13 September 2018 and bowl trapping at nine transects (not simultaneously) occurred on 11 dates between 12 April and 19 September 2018. 1,014 bees representing 110 species in 21 genera including all five common bee families in Indiana were collected. The most common bees were Lasioglossum coriaceum, L. subviridatum, Augochlora pura, Osmia taurus, Ceratina calcarata, and Bombus impatiens. Forests provide nesting, overwintering, and floral resources which are requisite to support these species. Four species collected (two percent) were non-native to Indiana suggesting a fairly intact native bee fauna, with two of those species, Osmia taurus and O. cornifrons, being a fairly recent invasion. The survey provides initial baseline data on bees using forests in Indiana and represents the first season-long collection and systematic sampling of bees in Indiana forests. These surveys included five state and 83 county records of bees. The survey also provides evidence of multiple bee species using and relying on forest resources even in times of low floral availability. Further sampling and pollinator monitoring of forests in Indiana will likely reveal the presence of additional species, contribute to the knowledge of floral and habitat use of bees in Indiana, and help determine how forest management affects bee and floral communities.
**Poster**

**Death Stinks: Characterizing the Volatiles that Attract Blow Flies During Decomposition**

**Raenah Bailey, Janelle Bouman, Shelby Leucata, Monique Le Donne, Lauren Smith, Heather Wendland and Kristi Bugajski**
Valparaiso University

Blow flies (Diptera: Calliphoridae) are attracted to the volatile compounds (organic compounds that easily become airborne) given off by decomposing matter. Baits are often used as human substitutes in forensic entomology, and a bait should approximate the volatiles given off by decomposing human bodies as closely as possible. This fact has led us to investigate the different volatiles, and how the volatiles change throughout the process of decomposition in chicken liver. Based on this, we will assess how well chicken liver approximates the process of decomposition in a human body. Chicken liver was placed in a sealed mason jar for gases to accumulate in the headspace. A small hole was punctured in the lid of the mason jar and gases were drawn into one of three different solid-phase microextraction (SPME) fibers (100μm polydimethylsiloxane, 65μm polydimethylsiloxane/divinylbenzene, 75μm Carboxen/polydimethylsiloxane). After the SPME fiber was left in the jar for five minutes, a Gas Chromatography-Mass Spectrometry was used to analyze the volatiles. Preliminary results that show the most common compounds being released from the liver are methyl disulfide, dimethyl trisulfide, dimethoxyflavone, and 1-(3-hydroxypropyl)-2-piperidinone. The results are being compared with published literature to identify important substances in gases that attract blow flies and which sample gives the most reliable results in comparison to a human cadaver. The knowledge gained will be used to assess the appropriate amount of time to age baits to best simulate human cadaver volatiles found in the literature.

**Poster**

**Diversity of Ichneumon Wasps in the Yellowwood State Forest Back Country Area (Hymenoptera: Ichneumonidae)**

**Jordan Craven** and Glené Mynhardt, Hanover College

No recent studies have focused on studying the diversity of Ichneumonidae in Indiana, especially in temperate deciduous ecosystems. Ichneumonids are found worldwide and are common parasitoids of all life stages of other insects that include Coleoptera, Lepidoptera, and other Hymenoptera. This project focuses specifically on establishing baseline data on the diversity of ichneumonid wasps in the Yellowwood State Forest Back Country Area in south-central Indiana. Adult specimens were collected using Townes-style Malaise traps between May and October of 2016. A total of eight
sampling locations differing in topology, shade, and vegetation density were identified. Ethanol-preserved specimens were collected six times throughout the sampling period, and then curated and identified using existing subfamily-level keys. For many taxa, no lower-level keys are currently available. Thus far, 100-115 unique morphospecies from 12 subfamilies have been identified. The most abundant subfamilies include the Ichneumoninae and Cryptinae. While identification of this taxonomically challenging group of insects is still ongoing, this study provides the first baseline data on ichneumonid diversity in one of Indiana’s public forests, and highlights the need for taxonomic training and insect diversity studies in the state.

Poster

Male competition in the leafhopper *Erythroneura bistrata* may involve female-like decoy signals

Dalton Collins, Jesse Moberly, and Randy Hunt, Indiana University Southeast

Mate recognition and localization in insects that use vibrational signals involves duetting between males and females. Several studies have shown that male competitive interactions involve signals that disrupt male-female duets. Disruptive signals emitted by males generally overlap a female’s response to another male’s mating signal. Disruptive signals are either similar to the normal male signal or a unique signal not emitted in the absence of competing males. We observed that some male *Erythroneura bistrata* leafhoppers emit a putative disruptive signal that follows a female’s response to a competing male. Interestingly, the signal structure is similar to the female signal. The aim of this study was to determine whether we could elicit this behavior in males using recordings of female signals played to two males in the absence of a real female and to quantitatively compare female signals to the female-like male signals. Our results show that playback of a female signal to the first male to call frequently causes the second male to emit the female-like response. Results of the comparison of male signals to female signals will be presented. Optimization of playback procedures will allow us to design experiments aimed at determining conditional strategies used by males when competing for females. Female mimicry as a conditional mating strategy has been documented in a few animals, but not in insects that use vibrational communication.
Comprehensive studies focused on insect diversity, species richness, and taxonomy in the state of Indiana remain limited. More specifically, the existing knowledge of arthropods inhabiting Indiana’s forests is based on a few select taxa and outdated species lists. Since 2014, local academic and non-profit institutions have partnered with the Indiana Forest Alliance (IFA) to gain an understanding of the flora and fauna inhabiting Indiana’s temperate deciduous forests. This study aims to provide baseline insect diversity estimates from the Morgan-Monroe and Yellowwood State Forest Backcountry Areas during the summer of 2016, when more directed efforts of sampling of specific groups of insects were initiated. Insects were collected using passive Townes-style Malaise traps, which collect a large diversity of insects over short periods of time without requiring daily retrieval of specimens. Traps were deployed in six sampling locations differing in topology, vegetation density and shade, and collecting vessels exchanged and replaced with fresh 70% ethanol every four to six weeks between May and October of 2016. Identification efforts thus far have focused on holometabolous insect orders including the Coleoptera (beetles), Hymenoptera (wasps), and Diptera (true flies), although other orders were also collected in smaller numbers and remain unidentified. Preliminary estimates using only Malaise traps include a total of 41 beetle families, 38 fly families, and 25 wasp families. Continued efforts towards genus- and species-level identification are still ongoing. Even though Malaise traps were the only collecting method employed in 2016, the data provide useful baseline information for faunistic comparisons between the current and other potential forest tracts in Indiana.

Current knowledge of the Coleoptera (beetles) of Indiana remains limited, and the most comprehensive taxonomic lists of beetles focus on wider geographic ranges. Thus far, few studies have focused specifically on beetle diversity in Indiana's forest ecosystems. In collaboration with the Indiana Forest Alliance (IFA) and as part of a robust Ecoblitz that attempts to catalog all fauna and flora in the area, the main objective of this study was to provide an estimate of beetle diversity within the
Yellowwood State Forest Back Country Area. Taxa collected by Townes-style Malaise traps, which are passive flight-intercept traps, are presented. Traps were deployed between May and October of 2016 in six unique ecological zones within the area. Malaise traps have captured a conservative estimate of 91 genera that were not collected using other methods (hand collecting, funnel traps, panel traps, light traps) between 2014-2015 sampling periods, as well as 43 genera that were previously captured and identified without the use of Malaise traps. Of those genera identified, 115 specimens have been identified to the species level. A total of 41 families were identified from the 2016 sampling period, with 54 families identified in total. A total of 11 families not collected before were identified from Malaise trap samples from 2016. While efforts to identify specimens are still ongoing, this study suggests that Malaise traps are an effective means of collecting the majority of major beetle families typically found in deciduous forest ecosystems, especially when active collecting techniques are not feasible. In addition, a combination of collecting methods should be employed to arrive at closer estimates of total beetle diversity in the sampling area.
Engineering

Hot Topics
Biofuels and Biochemicals: Where are we today?

Irene Reizman
Rose-Hulman Institute of Technology

In the early 2000s, increasing oil prices drove strong investment in companies developing new production routes for biofuels and bulk biochemicals. With changes in the global economy and energy prices, some of the economic pressures for developing alternatives to crude oil have eased today. However, other drivers for innovation are motivating new progress in production of biofuels and biochemicals, whether to reduce carbon dioxide emissions or to provide supplies of chemicals difficult to synthesize from crude oil feedstock. This Hot Topic presentation will provide an overview of the current production routes and markets for biofuel, focusing on the impact on agriculture and industry in Indiana, and will explore new technologies being developed for advanced biofuel production and production of specialty biochemcials.

Presentation
Analysis of Carbohydrates, Dicarboxylic Acids and Fatty Acids from Fermentation Broth by HPLC-DAD and HPLC-RID

Xin Tang, Irene M Reizman, Gregory T Neumann
Rose-Hulman Institute of Technology, Terre Haute, Indiana

Production of long chain dicarboxylic acids (diacids) through biotechnology has gained great interest. They are widely used in the production of plastics, polyesters, and other chemical products. In industry, diacids have been produced from chemical conversion processes. However, chemical processes have some limitations such as long chain diacids are difficult to synthesize, the variety of diacids produced is limited, and the feedstock is not renewable. In contrast, with biochemical approaches, various long chain diacids can be produced through environmental-friendly alternatives. For effective bioprocess development, a stable and fast method for analysis of carbohydrates, long chain dicarboxylic acids, and long chain fatty acids (FAs) is critical. Although many high-performance liquid chromatography (HPLC) methods have been developed for analyzing FAs, few have been applied to analyze underivatized long chain FAs or diacids. Among those few methods, even fewer are able to achieve the goal with common detectors like a refractive index detector. As shown in this work, we developed a fast and stable alternative method for
simultaneous analysis of long chain FAs and diacids. Both a refractive index detector (RID) and a diode array detector (DAD) were used for analysis. The optimized method utilized a C18 column with a mobile phase of methanol: water: acetic acid (90:9.9:0.1, v/v/v), a flow rate of 1 mL/min, a column temperature of 35°C, and a RID temperature of 35°C. All analytes eluted within 25 min. Meanwhile, a HPLC-DAD method for analysis of carbohydrates was also developed, using a Hi-Plex H column with a mobile phase of 0.005 M sulfuric acid, a flow rate of 0.5 mL/min, a column temperature of 60°C, and a RID temperature of 52°C. All analytes eluted within 10 min. All calibration curves of analytes showed good linearity (r² > 0.987). This study provides a simple and fast method for quantification of substrates and products from fermentation broth, which can be applied to studies involving bioconversions of fatty acids.

Presentation
Landfill History Revealed Through Site Characterization for CSO Underground Storage Tank, Lafayette, Indiana

Erin O’Connor and Terry R. West, Earth, Atmospheric, and Planetary Sciences, Purdue University

Lafayette Indiana, like many cities in the U.S., is actively involved in reducing combined sewer overflow problems under an edict by USEPA. In the past, sanitary effluent and storm water runoff were combined into a single sewer system which reaches full capacity during heavy rainfall events. To relieve pressure, overflow of the untreated water is dumped directly into rivers and lakes yielding contamination. In Lafayette, a 4.2 million gallon underground storage tank, roughly the size of a football field, is under construction. This storage will delay flow to the sewage treatment plant and reduce the CSO problem. Two of the nine overflow points into the river will be eliminated. The site, adjacent to the Wabash River, is a former municipal solid waste landfill that closed in about 1970. Sand from the river was pumped to the site for daily cover and closure. Eleven exploratory borings 75 feet deep indicate a landfill thickness up to 28.5 feet and groundwater depth ranging from 20 to 38 feet. Below the fill, the soil consists of a fine to medium sand. About 18 million gallons per day is pumped from the construction site so the tank can be built in the dry. To build the tank approximately 100,000 cubic yards of solid waste and soil were excavated. Because of the high GWT, the tank was built to resist uplift pressure. Eight hundred and sixteen auger cast piles, 25 to 55 feet deep, were used to anchor the tank. Eight thousand cubic yards of concrete and 750 tons of rebar were used in the tank floor, walls and roof slab.
Presentation

**Engineering a Fluorescent Biosensor for the Detection of Herbicide Glyphosate**

**Pierre-Emmanuel N’Guetta**, Maggie Fink, and Shahir S. Rizk
Department of Chemistry and Biochemistry, Indiana University South Bend

Glyphosate is a phosphonate that is the active ingredient in the herbicide RoundUP used to kill weeds by blocking pathways essential to plant growth. Currently, glyphosate is the most popular herbicide used around the globe. It has been recently classified as a probable carcinogen and it also has made recent headlines for its widespread use on genetically modified seeds with research that links it to antibiotics resistance and hormone disruption. Several states are planning to restrict its use, while it has already been banned in the state of California. Hence, there is a need for the development of reliable detection methods for glyphosate in the soil, rivers and drinking water. This can help determine its concentration and effect on the environment. A probable macromolecule that could serve as a biosensor for glyphosate is PhnD or Phosphonate-binding protein from *E. coli*. When *E. coli* is in an environment that is phosphorus-deficient, the pho regulon in the bacteria will turn on transcription of the *phn* operon which codes for a number of proteins including PhnD, a periplasmic-binding protein. This protein is able to bind many phosphonates and allows their uptake by the bacteria for use as a phosphorus sources. Naturally, PhnD binds to 2-Aminoethyl Phosphonate with very high binding affinity, however previous research found that PhnD could bind to other phosphonate molecule such as glyphosate but with very low affinity, in fact the *Kd* was around 650 µM. The *Kd* (dissociation constant) is an equilibrium constant for the dissociation of a complex into its components. A low *Kd* value indicates high affinity and vice versa. Thus, our goal is to use site directed mutagenesis to engineer PhnD mutants with high affinity for glyphosate.
Environmental Quality

Presentation

**Historic HOLC Redlining in Indianapolis and the Legacy of Environmental Impacts: A research model for other redlined Indiana cities**

Donovan J. Moxley, **Burnell C. Fischer** Indiana University-Bloomington

Redline mapping, as practiced by the Home Owner’s Loan Corporation (HOLC) in the 1930s, has been criticized for evident racial discrimination in addition to financial risk assessment. Modern urban tree canopy cover has been demonstrably lower in historically redlined zones of four US cities already. This project focused on Indianapolis, IN, where a HOLC map was developed in 1937. Analysis using a geographic information system (GIS) was conducted to detect evidence of an ecological legacy of redlining. Using this method, evidence of relatively high-intensity development, low greenspace and forest cover, and disproportionately high incidences of brownfield sites, Superfund sites, industrial waste sites, and Interstate highways were detected in historically redlined zones in Indianapolis. While opportunities for further and more detailed research are considered, the method employed here should be developed such that it is replicable for other cities with redlining histories in Indiana and other surrounding states with comparable spatial datasets.

Presentation

**Active Indiana municipal urban forestry programs: How are they addressing sustainability/environmental change?**

**Stephanie Freeman-Day,** Burnell C. Fischer, Charlie B. DeVoe, Donovan J. Moxley – Indiana University

Both urban forest ecosystem services and threats to urban forest sustainability are well known. Urban forest management is more than individual tree care; it is managing the whole urban forest sustainably. Adding complexity is the concern for environmental change and identifying urban forest management strategies to both adapt and transition toward the future. To explore how Indiana urban forestry programs are addressing sustainability and environmental change we identified all Indiana cities that had been or are currently involved at least minimally in urban forestry from a variety of sources. These sources include IDNR CUF Program’s Tree City USA records and their grant awards (1992-2016), tree inventory consultancy projects, Indiana Arborist Association certified city arborist list, etc. We then used the list of active cities identified (74) and conducted a city-by-city website search. Collected
information included tree ordinances, tree inventory reports, urban forest plans and related documents, citywide plans, etc. We downloaded each document to conduct a keyword analysis, identifying cities addressing urban forest sustainability and/or environmental change. Prominent keywords were diversity (species and age), monitor, stormwater, pests, sustainability and climate. We found 16 city urban forestry programs that are beginning to address sustainability/environmental change. Lead cities are contracting inventories with resultant reports and management plans that include monitoring and taking action around tree species and size diversity and assessing for urban forest environmental change impacts. Additionally, they are realizing the benefits trees have in reducing stormwater and heat island effects. We also identified cities on the verge of addressing urban forest sustainability/environmental change. Clearly, in the near future Indiana will have a group of model urban forestry programs for addressing sustainability/environmental change.

Poster

Soil Lead Bioavailability in an Urban Community Severely Impacted by Legacy Pollution

Brenda Lower, Jennifer Latimer, and Hannah Veldhuizen, Indiana State University

Some urban areas, identified as environmental justice communities, have been disproportionately impacted by pollution. These communities are often industrial and characterized by a high proportion of minorities and overall lower socio-economic status. These communities also often suffer from a higher prevalence of disease, for example higher rates of childhood lead poisoning compared to suburban areas. East Chicago, Indiana, is one of these environmental justice communities. In 2009, the US Environmental Protection Agency declared the former USS Lead site a Superfund Site. In 2016, residents of the West Calumet Housing Complex, which was located across the street from USS Lead, were forced to relocate, and the complex was folded into the Superfund site. Active community groups in East Chicago have been working diligently to better understand their situation and the scope of the lead pollution in the city. The purpose of this project was to collect surface soil samples on playgrounds throughout East Chicago. We collected lead data using a portable X-ray Fluorescence (XRF) Analyzer. Samples were dried and powdered and re-analyzed using the XRF. Subsamples are also being reacted with a simulated gastric solution to evaluate lead bioavailability during digestion. We will use this data, along with previously collected data, to create predictive maps, using the geospatial technique of kriging for XRF-based total lead and estimated bioavailability from the simulated gastric extraction. This data will help the residents of East Chicago better understand their potential exposures to lead while also helping us better understand the factors that contribute to soil lead bioavailability.
Bioaccumulation of legacy pollutants in fresh water systems using *Lontra canadensis* (North American river otters) as a bioindicator

**Kathryn Mudica** and Jennifer Latime, Indiana State University

While environmental quality has improved significantly over the last 40 years, assessing water quality alone cannot predict the ecological impacts of metal pollution. In order to quantify these impacts, bioaccumulation and biomagnification in organisms must be studied. Because of their long environmental residence times, metals can remain in an environment for decades where they may be or may become bioavailable. Once introduced into a food web, these metals may bioaccumulate and biomagnify and can reach toxic levels very quickly. For example, fish tested in this study had average Pb = 3 to 6.5 ug/dL depending on trophic level. Fish maintain equilibrium with the water they inhabit, and they are pH, temperature, and most importantly, pollution tolerant. This allows fish to carry metal burdens throughout their lifespan.

Keystone species are often used to assess bioavailability of pollutants. The North American river otter, a piscivorous keystone species in fresh water systems, is an ideal choice as a surrogate for fish and a proxy from human fish consumption. As a pollution sensitive, apex predator, river otters hold the potential for both bioaccumulation and biomagnification. The purpose of this research is to evaluate the metal content of whole livers collected from otters that were surrendered to IN DNR during the 2017-2018 trapping season. Analysis of the livers revealed Pb levels 4Xs the levels found in fish, with average Pb = 23 ug/dL, suggesting bioaccumulation of Pb is occurring in freshwater systems in Indiana. In addition, elevated liver Pb concentrations were found in all counties where otters were trapped and surrendered.

Comparing lead results in the river otters to surface water monitoring by Indiana’s Department of Environmental Management indicates much of the lead contamination is recycled in the food web of freshwater, exposing all organisms in the food web. The results of this research will provide an alternative assessment of potential metal exposure to humans through fish consumption.
Particulate Matter Phytoremediation: The Effects of Leaf Morphology.

Taylor J. Mikulyuk and Thomas A. Sobat Ivy Tech Community College, Columbus Indiana 47203.

Particulate matter (PM) is a common air pollutant found in high concentrations in urban and industrial areas. PM has been shown to have significant effects on human health, causing an increase in the occurrence and severity of cardiopulmonary and respiratory illnesses and a marked decrease of lifespan in individuals suffering from heart or lung diseases. PM also affects the environment by creating acidic conditions in aquatic systems, depleting nutrients in soil, damaging crops and forests, and diminishing biodiversity. PM emissions are of anthropogenic origins such as fossil fuel and wood burning, road dust, and vehicle exhaust. Despite increasing regulations, these emissions often exceed permitted values, and once released into the atmosphere, options for reducing PM concentrations are limited. One unusual method for reducing atmospheric PM and improving overall air quality is phytoremediation; using live plants, especially trees, to reduce environmental contaminants. The PM of foliage was quantified to assess whether leaves with compound structure will collect more PM than those with simple leaf structure. Sample surface area was computed, then the sample was washed with water to dislodge surficial PM, and the resulting mixture was subjected to a fractionated filtering process which separated particulates into respective size classes. A secondary organic solvent wash was performed to dissolve the epicuticular wax layer and release PM trapped there, followed by a fractionated filtering process. Filter pre-weights were compared to post weights to determine the mass of PM per size class in each sample. Preliminary results from this study suggest a difference and will be the focus of this presentation.

Presentation
Cesium-137 Radioactive Fallout Findings in Indiana and Implications for Future Undergraduate and Graduate Research

Richard T. Whitman, Indiana University and Purdue University School of Public and Environmental Affairs and IvyTech Community College.

The era of Atmospheric Atomic Testing extended from 1945 until 1992. During that time more than 500 detonations puerized, activated, and then lifted significant quantities of debris into the stratosphere for worldwide distribution by the jetstream. Cesium-137 with a half-life of 30 years remains the principal isotope remaining after many years have passed. Research in 2017 found Cesium-137 in the soil in all 92 Indiana Counties due in large part to the clay in Indiana soil clay content. Significant opportunities using the deposited Cesium-137 to examine erosion, soil chemistry, and
deposition in soil, stream beds, lakes, and elsewhere could provide opportunities for undergraduate and graduate students for several decades. This presentation will show how gaps in the historic record, previous research elsewhere, and make this an attractive subject area spanning many science subjects.

Hot Topics
THE PAST, PRESENT AND FUTURE OF ENVIRONMENTAL PROTECTION-- THREATS AND OPPORTUNITIES

Janet McCabe, Indiana University McKinney School of Law; and Environmental Law and Policy Center

The news is filled with stories about environmental programs the Trump Administration is delaying or eliminating, and how science is, or is not, factoring into those policies. At the same time, we hear every day about new studies describing the impacts of climate change on public health, ecosystems, and the environment around the world. This talk will explore the ways in which the current Administration is changing direction on climate change and environmental policy, the short and long term impacts those changes may have, and how individuals can get involved.
Earth Science

Presentation

The Paleocene foraminifera of Foralgal reef Bed – Salt Mountain Formation, Alabama; Biostratigraphy and Paleoecology

Belkasim Khameiss, Ball State University & Richard Fluegeman, Ball State University

Fourteen foraminifera have reported and described on the Foralgal Reef Bed-Salt Mountain Alabama Section. The total thickness of Foralgal Reef Bed Mountain Alabama Formation is 18 m, it is mostly grainstone to boundstone, white to yellow colors, medium hard, and it is highly fossiliferous limestone by red coralline algae, coral fragments, ostracods, echinoderms, and foraminifera. The planktonic foraminifera have been reported are Parasubbotina pseudobulloides, Eoglobigerina fringe, Subbotina triangularis, Subbotina cancellate, Subbotina velascornsis, Subbotina gortanii, Glogerinatheka kugleri, Globigrinatheka micra, Turborotalia possagnoensis?, Zeeuvgirina zelandica. The Benthic foraminifera included in this section are Anomalinoideas acutus, Anomalinoideas midwayensis, Discocylina cookie, Ranikothalia catenula.

In this study, the stratigraphic occurrences of the larger foraminifera were correlated to biozonations based on planktonic foraminifera P4 (Thanetian) Late Paleocene age, and the rare percentage of benthic foraminifera with low diversity is marked accelerated global warming during the Middle to late Paleocene (Greenhouse).

Presentation

Tidal Marsh Vulnerability to Rising Sea Level Along the Southern Coast of North Carolina: A 30-Year Record of Change

Elena Solohin, Indiana University Bloomington
Taehee Hwang, Indiana University Bloomington
Christopher B. Craft, Indiana University Bloomington

Tidal marshes 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 impact wetland’s ability to keep pace with rising seas in the future. We used a combination of in-situ biomass measurements, and remote sensing techniques to estimate how tidal marsh biomass health and resilience has changed over time in a Southeast estuary of Cape Fear River, North Carolina. Aboveground biomass and marsh elevation were measured at 10 sites across the estuary. We established a relationship between in-situ biophysical variables and biomass estimates,
derived from remote sensing data. The study created a time series analysis of Normalized Difference Vegetation Index (NDVI) and then correlated it to climate variables of temperature and precipitation, temperature–seasonality, as well as historical sea-level data, and sediment supply.
Non-parametric trend analysis using the Theil-Sen slope revealed statistically significant negative trends in biomass and in the retreat of marsh margins. Largest decline in biomass occurred between 1997 and 2002 and between 2005 and 2015. This decline correlated with drought frequency and severity trends. Additionally, declines in marsh biomass were positively correlated with decreasing sediment supply, indicating that human activity plays a role in declining primary production. The loss of marsh area through landward marsh edge erosion ranged from ~ 0.6 m to 3 m yr⁻¹. Our 30-year analysis of NC tidal marshes is supported by real time kinematic global positioning system elevation data, which suggest that NC marshes have lower elevation in tidal frame, relative to many SE Atlantic tidal marshes, making them more susceptible to sea level rise. 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.

Presentation

**Body Size and Stride Length in Bipedal Dinosaurs: Comparing Non-Avian Theropods and Birds**

**James O. Farlow** (Purdue University Fort Wayne)
Lisa G. Buckley (Peace River Palaeontology Research Centre, British Columbia)
Dan Coroian (Purdue University Fort Wayne)
Philip J. Currie (University of Alberta)
Martin G. Lockley (University of Colorado Denver)

Theropod dinosaurs are one of the most successful groups of terrestrial vertebrates, including among their ranks both birds and a host of small to gigantic Mesozoic non-avian forms. We compared aspects of the locomotion of typical non-avian theropods with Mesozoic and Cenozoic striding birds using fossilized and modern footprint sequences (trackways). “Main sequence” trackways (presumably made by normally walking animals) of both non-avian and avian theropods show a curvilinear positive correlation between stride length and footprint length; bigger theropods take relatively shorter strides than smaller theropods. Mesozoic and Cenozoic fossil bird trackways show a similar stride length: footprint length relationship. For footprints of a given length, bird trackways show shorter strides than trackways of non-avian theropods. For Mesozoic trackways, the difference in the stride length: footprint length relationship between non-avian theropods and birds may reflect differences in the limb length: toe length (toe length serving as a proxy for footprint length) relationship between the two groups.
Trackways of large Cenozoic walking birds have relatively shorter strides for a given footprint length than do trackways of non-avian theropods, even though big ground birds have relatively longer legs for a given toe length. The difference may be due to differences in hindlimb kinematics between non-avian theropods and birds. Even so, maximum stride/footprint length ratios of running animals are comparable between non-avian theropods and big ground birds.

Poster

**Detailed Phosphorus Geochemistry of Sediments Collected From Lake Tanganyika**

**Ravin Gaines, Garrett Carr, Jennifer Latimer and Jeffery Stone, Indiana State University**

Lake Tanganyika is among the world’s oldest, deepest, and largest freshwater lakes. As a transboundary water body, it is also a vital resource for Tanzania, Democratic Republic of Congo, Burundi, and Zambia. Future climate change scenarios predict that Africa will experience drastic changes in surface temperatures and water resources. To better predict how climate change may influence natural resources such as those associated with Lake Tanganyika, we can study past intervals of climate change using long lake records. One approach to understanding past biogeochemical cycling in the lake and the relationships between productivity and nutrient availability is to study detailed phosphorus geochemistry. Today, Lake Tanganyika productivity is co-limited by nitrate, phosphate, and iron. The lake experiences a semiannual seasonal shift of wet and dry. Wet season being experienced from October to April and dry season through the months May to September. Experiencing higher phosphorus concentrations in the wet season over the dry season. During the wet season, new phosphorus is introduced to the lake via runoff, but during the dry season internal P recycling is important. We hypothesize that this seasonal relationship will be amplified by wet and dry climate patterns. To test this hypothesis, we are analyzing 200 samples at a 1 cm resolution from the Kalya Slope on the eastern margin of Lake Tanganyika using a sequential phosphorus extraction. This extraction isolates phosphorus associated with oxides, minerals, and organic matter. Through sequential extraction we can accurately show the concentration of detrital, authigenic, organic, and phosphorus absorbed and in oxides. The results will ultimately be compared to changes in diatom assemblages over known intervals of past climate change to elucidate lake responses to past climate variability leading us to having better ability predicting how future climate variation will affect lacustrine productivity and help the bordering countries maintain and take advantage of shifting prosperity.
Presentation

**Building with Beauty--Geodes as Construction Materials**

**Nelson R. Shaffer**, Nannovations

Geodes are enigmatic but common geologic objects that occur in select areas including southcentral Indiana. They are fascinating and useful objects. Indiana geodes are subspherical, bumpy, masses of quartz contained within Mississippian age carbonates, mainly the Harrodsburg Limestone. Geodes are very robust and many have withstood millions of years of weathering and transport. They became concentrated in soils and streams of the outcrop areas, even into “useful” deposits.

Besides their natural beauty, people collect geodes for their unusual and often well-formed secondary minerals. Geodes have also been used to build, or embellish, all sorts of manmade structures. Geode structures occur all over southern Indiana. They range from simple landscape items to entire buildings, artistic structures, even religious shrines. Geode collecting is a geotourism business in some states.

Buildings usually have size-matched geodes from about 4 to 12 inches set in cement. Most are decorative, but a number of load-bearing applications occur.

Poster

**Blood Lead (Pb) Levels in the City of Muncie, East Central Indiana**

**Benjamin Azar**, Carolyn Dowling, Jessi Haeft, Ball State University

Approximately 7 percent of residents tested in Delaware County had elevated blood lead levels, which is more than double the national average. Lead (Pb) exposure is known to cause serious adverse health effects stemming from its capacity to replace calcium in bodily processes, leading to cell death and damage to the nervous system. Potential sources of lead in the environment include lead-based paint, lead leachate in drinking water, and lead contamination in soil.

During the fall semester of 2018, a Ball State immersive learning class constructed blood lead level maps for the City of Muncie within Delaware County, using GIS ArcMap and a dataset provided by Regenstrief Institute. Preliminary mapping of elevated blood lead levels resulted in a clustering effect for levels greater than or equal to 25 micrograms per deciliter (Blood Lead Poisoning). The clustering was prominent south of the White River in the City of Muncie. An area of interest to note in the south was identified along a major thoroughfare in the industrial zone. A possible influence for the elevated levels in this zone could be historic use of leaded gasoline by vehicles traveling this roadway.

To best assess the presence of lead in soils from possible sources such as legacy
manufacturing sites or leaded gasoline, a comprehensive, biased sampling approach was taken. Soil samples were collected from at-risk areas as indicated by blood lead level maps. Soil sample analysis will be conducted via EPA method 3050B for bio-available lead and confirmed by an external lab for quality assurance. A map of soils containing bio-available lead will be generated and overlapped on the blood lead levels map. The final map and associated data can be utilized in the development of a remediation plan for the Delaware County area, should the soils contain significant contamination.

Presentation

Energy Lost and Found

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The use of groundwater aquifers as a source of low-enthalpy geothermal energy for heating buildings and other applications is widespread globally, although minor relative to fossil-fuel technologies. Spurred by the need to reduce global greenhouse gas emissions, significant gains have been made in harnessing the vast potential of low-enthalpy geothermal resources via direct use and closed-loop heat pump systems. A particularly promising approach is the use of anthropogenic groundwater heat reservoirs where the “urban heat island effect” has elevated aquifer temperatures beneath cities. Despite only modest increases of a few degrees Celsius documented in such aquifers, research suggests that this resource could meet the entire heating demand of buildings in major cities in North America, Europe, and Asia. At the Bloomington campus of Indiana University, we’ve recently discovered an exceptional anthropogenic heat reservoir having temperatures elevated more than 10 degrees C above ambient values in the shallow carbonate aquifer, and anomalously high temperatures extending to more than 60 m depth in portions of the aquifer. We attribute this remarkable energy resource to long-term heat loss from the university’s district energy system, which heats more than one million square meters of building space via 34 km of buried steam and condensate piping. In this paper, we present initial results from borehole logging, surface thermal imaging, soil and rock core analyses, and model calculations to estimate the magnitude and distribution of the university’s geothermal resource. In addition, we discuss opportunities for Indiana University to recover this energy for beneficial use through an innovative campus-
wide R&D initiative. With 23 additional district energy systems installed across Indiana and nearly 700 across the United States, our discovery at Indiana University could have significant implications for expanding clean energy technologies throughout the state and nation.

**Poster**

**Assessing Irrigation Suitability of the White River Water under the Context of Climate Change and Water Availability**

Shahin Alam, Bangshuai Han

Natural Resources and Environmental Management, Ball State University

The quality of irrigation water largely determines crops’ productivity, quality, and effect of human health. Indiana's Upper White River Watershed has encountered extreme drought issues amid harvesting seasons and shrinking groundwater aquifers because of the climate change and irrigation use. This pattern is anticipated to continue, turning White River into another irrigation water source. A hydrochemical assessment of the White River water will be conducted in order to characterize, classify and evaluate the suitability for irrigation. This research will evaluate six water quality parameters as primary data over the harvesting period between April to October in 2019 from five locations biweekly and analyze secondary long term (2002-2018) monitoring data through weighted regressions on time discharge and season (WRTDS), hierarchical spatiotemporal analysis, cluster analysis, and principle component analysis. Irrigation water’s suitability will be determined through pH, total dissolved solids (TDS), electrical conductivity (EC), and sodium components. Sodium may hamper penetrability, influence fruitfulness and reduce crop yields. Sodium adsorption ratio (SAR), adjusted sodium adsorption ratio (SARadj), and soluble sodium percent (SSP) will be analyzed for judging irrigation water suitability. Based on the statistical, spatial and temporal analysis of primary and secondary data of the water, irrigation suitability will be justified.

**Presentation**

**Extensive Fern Prairies were grazed by dinosaurs during the Later Mesozoic**

David Dilcher, Maximillian Scott, Department of Earth and Atmospheric Science, Indiana University

The Mesozoic is The Age of Ferns. Ferns are abundant and undergo a great deal of evolution and modernization. Part of this evolution may be the product of their co-evolution with dinosaurs. Many modern fern families, and even some genera, can be traced back to the Cretaceous. Many of the late Paleozoic and early Mesozoic ferns
grew as medium to small trees. Their growing shoots at the tip of these trees were exposed to predation by the new Mesozoic dinosaurs. If the cells of the shoot apex were chewed away the tree would die or drastically change it growth habit. Many successful Mesozoic ferns grew short fleshy herbaceous stems that branched frequently. Many kept their shoot apices close to the ground or below ground level. From this level they produced abundant leaves that could be easily grazed and readily regrown. We can see a similar evolution has occurred in grasses during the Cenozoic that are grazed and continuously replace the leaves eaten by herbivorous mammals. This pressure on ferns resulted in the presence of extensive fern prairies in central and western North America during the Cretaceous. This is evidenced by the increase of fern spores in the dispersed pollen and spore record of Cretaceous sediments.

Presentation

Diversity and Digestion: Herbivorous Dinosaur Food Intake

Maximilian Scott, David Dilcher, Department of Earth and Atmospheric Science, Indiana University

Large herbivorous dinosaurs of the early to middle Cretaceous were eating machines. Evidence suggests that the majority of them were likely mesothermic. Mesotherms fall in between endotherms and ectotherms. Mummified dinosaur remains from the Cretaceous revealed them that some of them had four chambered hearts. They have been found living in colder environments such as Alaska during the Mesozoic, which had a climate similar to that of coastal Washington state. Sustaining survivable body temperatures as well as keeping the energy to stay alive in such a large animal would require a large food intake. Many sauropods from the middle Jurassic to middle Cretaceous did not hold their heads up high, but rather extending them out for grazing a wide area. Today, there are large grass prairies capable of feeding millions of large herbivores. Grass did not become abundant until some time about 55-66 million years ago. Keeping that in mind, one must look at the digestive system of large herbivorous dinosaurs. Sauropods had peg-like teeth that could not chew, but rather were ideal for stripping leaves from stems. The gastroliths in their gizzards ground the ferns up, and their size made digestion of tough plant matter easier. In the absence of grass, the primary candidate to populate the vast prairies and feed the diverse, dominate, and often massive herbivorous dinosaurs could have been ferns. Ferns are low, adaptable, and tough to digest. The adaptations of sauropods in the Cretaceous were ideal for fern digestion. While some herbivorous coprolites from the Cretaceous contain wood fragments which suggest browsing on the vegetation of woody plants. The hypothesis that fern prairies dominated the plains of the Cretaceous provides a food source that the grazing dinosaurs needed.
Microstructural study of mylonitic rocks from SW Sweden

Aranzanzu Pinan-LLamas, Dessiree Hurst, Reed Hathaway, Purdue University Fort Wayne

We present preliminary structural and microstructural data obtained from samples collected within the Mylonite Zone, a prominent ~10 km wide and >450 km long Sveconorwegian tectonic boundary that separates the Eastern Segment from allochthonous units to the west within the Sveconorwegian orogen. The Sveconorwegian orogeny was active in our research area between 1140 and 900 Ma, and has been traditionally related to a mountain-building event that led to the assembly of the supercontinent Rodinia. The studied samples were collected from the southern segment (Bua) and the frontal ramp (~30 km east of Gothenburg) of the Mylonite Zone. The main goal of our study is to document the Sveconorwegian microstructures preserved in Mylonite Zone, since structural data from this area are still scarce. In the Bua segment, metagranodioritic samples are characterized by a gneissic banding formed by coarse-grained plagioclase, quartzofeldspathic ribbons, elongated K-feldspar grains, quartz, green amphibole and biotite. Quartz shows interlobate grain boundaries consistent with high-temperature grain boundary migration (GBM); minerals and microstructures present in these samples are indicative of deformation at upper greenschist facies conditions. Near from Gothenburg, the analyzed mylonites are generally formed by plagioclase, K-feldspar, quartz, biotite and/or muscovite, opaque minerals, zircon and epidote. The foliation is defined by fine-grained biotite-rich (or muscovite-rich) and quartzofeldspathic bands. Some samples contain quartz that displays bulging recrystallization (BLG) and subgrain rotation recrystallization (SGR). The mineralogy present in the samples and the quartz and feldspar microstructures are consistent with low-grade (greenschist facies) metamorphic conditions during mylonitization. Strain analysis was conducted on a mylonite sample to estimate the mean kinematic vorticity number (Wm) using the Passchier, Wallis, and Rigid Grain Net methods. Wm values for the Mylonite Zone sample range from Wm= 0.52 to Wm=0.55 which suggests a significant component of pure shear during deformation.

Winter and Fall Crops on Soil Fertility and Sweet Corn Yield

Jessi Haeft and Carson Wright

The over application of herbicide and fertilizer is a problem that plagues the environment and agricultural systems. The over application of herbicides has led to herbicide resistant weeds and environmental degradation. The study objectives are to
examine the effects of a winter triticale and hairy vetch cover crop mix on weed biomass and available soil nutrients. After spring planting sweet corn, sunn hemp was interseeded into the standing corn. The sun hemp’s effect on available nutrients and the yield of the sweet corn was examined. This study was conducted in northeastern Delaware County, Indiana through the fallow season months of Fall 2017/2018 through Spring 2018/2019. Soil analysis for nutrients included the Mehlich 3 extraction method for potassium, calcium, sodium, and magnesium. For phosphorus the Strong Bray method was utilized. 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. Statistical variance was analyzed using ANOVA.

Presentation

**Stable Isotope evidence of human-induced modifications to the water cycle in southern Indiana**

Paul K. Doss and Sarah A. Hostetler-Shull
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The Ground Water Monitoring Laboratory at the University of Southern Indiana houses a deep-shallow piezometer nest in the Inglefield Sandstone Aquifer, a locally important domestic aquifer. Deep and shallow groundwaters and local precipitation, including rain and snow from diverse sources, were sampled for stable isotopes of oxygen and hydrogen over year-long periods in 2005/2006 and 2015/2016. Isotopic data from ‘15-'16 are measurably different from ‘05-'06 data, with a general shift to more depleted Delta-18O and Delta 2H compositions. We argue that the significant decadal change in stable isotope composition of a bedrock aquifer system in Southern Indiana is a response to observable changes in local land use and regional climate variables. Changes to isotopic composition of groundwater in the Inglefield Sandstone from 2005 to 2015 correlate to an expansion of the public water supply in Evansville, IN, which is sourced from the Ohio River. Over the past 10 – 20 years, public water has replaced hundreds of previous domestic well water withdrawals in Western Vanderburgh County Indiana. However, the municipal sewer system has not mirrored the supply expansion. Recently connected public water supply users now recharge groundwater with Ohio River water through domestic septic system infiltration, largely, and other pathways. This localized recharge of bedrock groundwater with surface water from the Ohio River has generated an observable shift in isotopic composition. Moreover, a decadal increase in local precipitation is consistent with large-scale regional assessments, and correlates to decadal changes in the isotopic composition of local precipitation. The shift to
isotopically-heavier precipitation also appears to correspond to decadal increases in mean annual temperature. The decadal change in groundwater isotopic composition reflects both processes, recharge from a new source of isotopically lighter river water, and recharge from isotopically heavier local precipitation.

Presentation

**Patterned Ground in the Central Wabash Valley**

**Darrell G. Schulze**, Purdue University; **Michael Konen**, Northern Illinois University; **Darryl Granger**, Purdue University; **Carolyn Olson**, US Geological Survey

For at least 1,000 years after Wisconsin glacial ice retreated from central Indiana the land was a frozen tundra similar to the Arctic today. Ice wedge polygons (patterned ground) formed in the permafrost as the result of seasonal freezing and thawing. Today, the evidence for these ice wedge polygons manifests itself most prominently in polygonal growth patterns of soybeans and corn during drought years. Crops grow taller over relict ice wedges in drought years because roots penetrate deeper and plants access more moisture. Aerial photos taken during the 2012 drought year show polygonal patterns of taller crops late in the growing season in soybean and corn fields in northwestern Indiana. Polygonal patterns also occur in bare earth aerial photography. The 1992 black and white, leaf-off aerial photography of Tippecanoe County shows prominent polygonal patterns over much of the Wea Plains southwest of Lafayette. Direct observations of known relict ice wedges are difficult. The features extend at least 2 meters below the soil surface, are probably one or two meters across laterally, and the contrast between the filled wedges and the interiors of the polygons is subtle and has little or no surface expression.

Presentation

**State and Federal Geological Surveys Conducted from New Harmony, Indiana in the Mid-Nineteenth Century**

**William S. Elliott, Jr.**, University of Southern Indiana

New Harmony was established in 1814 by the Harmonists, a religious group led by Johann Georg Rapp. In 1824, the Harmonists relocated to Economy, Pennsylvania, and subsequently sold the town of New Harmony to Robert Owen, a Scottish social reformer. William Maclure, Father of North American Geology, partnered with Robert Owen in 1825 to begin an experimental Utopian society. Artists, educators, natural scientists, and others arrived on a keel boat named “Philanthropist” in January 1826; noted passengers included Madame Marie Louise Duclos Fretageot (educator), Charles Alexandre Lesueur (artist and zoologist), Balthazar Obernesser (educator and artist), Gerard Troost (geologist), and Thomas Say (zoologist). Although the Owen-
Maclure Utopian experiment dissolved in 1827, New Harmony served as an important intellectual center for scientific studies of conchology, entomology, geology, ichthyology, and paleontology. In particular, the approach and methods of conducting state geological surveys were refined by the geologists of New Harmony, and resulted in several published geological surveys: Tennessee (1836) by Gerard Troost; Indiana (1838-1839; 1859-1860), Kentucky (1853-1857), and Arkansas (1857-1860) by David Dale Owen; and Illinois (1851-1855) by Joseph Norwood. Moreover, David Dale Owen was appointed federal geologist by Congress in 1839 to lead the first geological survey of northern Illinois, Iowa, and Wisconsin, and again appointed by Congress in 1847 to add Minnesota and parts of Nebraska to the federal survey (1847-1851). The methods developed by New Harmony geologists in conducting geological surveys proved paramount to identifying economic natural resources vital to westward expansion in the mid-nineteenth century. In addition, the New Harmony community of thinkers in the mid-nineteenth century led to progressive educational views, establishment of public libraries through the Working Men’s Institutes, and recognition of the importance of art to documenting scientific observations and natural history.

Poster

**Characterization of the mineralogy and deformation in a mylonitized granite from NW Argentina- a SEM study**

**Joseph Kline** and Aranzazu Pinan-Llamas
Purdue University Fort Wayne

The Eastern Sierras Pampeanas (northwestern Argentina) record an Early to Middle Ordovician magmatic and deformational episode associated with the Famatinian orogeny. This orogeny predated the formation of the present-day Andes and involved the formation and deformation of an extensive magmatic arc along the southwestern margin of the supercontinent Gondwana. The magmatic arc localized Famatinian deformation along N-S and NW-SE trending shear belts. As a result of this deformation, the intrusive rocks exhibit a variety of shear fabrics that range from highly-strained ultramylonitic bands to less deformed anastomosing granitic lenses. We present petrographic and Scanning Electron Microscope (SEM) data from five oriented granitic samples collected along a 1-km long transect across one of these Famatinian shear zones. The samples have similar mineralogy but show various degrees of deformation, which took place under greenschist-grade metamorphic conditions.

The main goal is to document textural and compositional changes as well as to constrain the mechanisms of deformation with increasing degree of deformation. Based on petrographic and SEM/EDS analyses we describe the less deformed granite
as a porphyritic granite with orthoclase phenocrysts embedded in a groundmass mostly composed of quartz, biotite, muscovite, orthoclase and plagioclase. The plagioclase is mostly oligoclase, although andesine is also present. Observed accessory minerals are zircon, ilmenite and apatite. In more deformed samples, the large K-feldspar porphyroclasts show a gradual size reduction by intense brittle deformation (microfracturing). As a result, they are transformed into asymmetric kinematic indicators enclosed in a main foliation defined by mica-rich domains depleted in quartz and enriched in iron oxides and quartzofeldspathic ribbons. Although the plagioclase is still oligoclase, slight changes in composition are observed in newly recrystallized biotite grains (in the feldspar pressure shadows) that generally show a lower K, Mg and Fe content with respect to the large biotite grains in less deformed samples.

Poster

**Observed and global climate model based changes in wind power potential over the Northern Hemisphere during 1979-2016**

Tian Qun, Purdue University

Using an observed dataset, we study the changes of surface wind speeds from 1979 to 2016 over the Northern Hemisphere and their impacts on wind power potential. The results show that surface wind speeds were decreasing in the past four decades over most regions in the Northern Hemisphere, including North America, Europe and Asia. In conjunction with decreasing surface wind speeds, the wind power potential at the typical height of a commercial wind turbine was also declining over the past decades for most regions in the Northern Hemisphere. Approximately 30%, 50% and 80% of the stations lost over 30% of the wind power potential since 1979 in North America, Europe and Asia, respectively. In addition, the evaluation of climate models shows their relatively poor ability to simulate long-term temporal trends of surface winds, indicating the need for enhancing the process that can improve the reliability of climate models for wind energy assessments.
History of Science

Hot Topics

Fifty Years Since the First Moon Landing: Commemorating the History and Achievement of NASA

Chris Edwards
Fishers High School
Fishers, IN

On July 20, 1969, the Apollo 11 lunar module touched down on the Moon. The American desire to land astronauts on the Moon began as a way to check the Soviet Union's advancement in rocket technology. However, as the U.S. entered into Vietnam and the Civil Rights movement, the moon landing took on different connotations. Chris Edwards is the author of the forthcoming Young Adult book titled All About the Moon Landing (Blue River/Cardinal Press) and will be sharing the history and science of the moon landing on this anniversary year.

Microbiology & Molecular Biology

Presentation

Investigating the Variable Mutational Bias Hypothesis using informatics techniques

Tony J. Smith & Sridhar Ramachandran, Indiana University Southeast, New Albany, Indiana

Genetic diversity is generated by mutagenesis, which causes changes in the DNA sequence. Darwin distinguished three kinds of mutations: advantageous, deleterious, and neutral. The selectionists (neo-Darwinians) believe that if diversity is essential to survival, and if mutagenesis is required to generate such diversity, perhaps mutagenesis has been positively selected for throughout evolution. Whereas advantageous variations expand in the progeny (by positive, or Darwinian, selection), the deleterious ones tend to disappear (by negative, or purifying, selection), and the neutral ones may come out of their limbo to be fixed (like the advantageous variations), or to disappear (like the deleterious ones). Does Darwinism explain mutagenesis, or is there more to it than natural selection? A new influential theory called the neutral theory of molecular evolution was introduced by Motoo Kimura in the late 1960s and early 1970s. Kimura’s revolutionary proposal started a neutralist/selectionist debate, which is still going on and which concerns the important issue of the role of chance in evolution. The research work discussed in this research
investigates on the open question (Variable Mutational Bias Hypothesis) in the neutralist/selectionist debate. The Variable Mutational Bias (VMB) Hypothesis is an open question in genomics that is now testable on the human genome with the availability of the human genome data on the NCBI and Sanger sites. The VMB hypothesis states the existence of varying mutation rates across a genome and attributes these varying local mutational rates as biases in the mutagenesis providing support to the selectionists’ view of evolution. The neutralists argue that the pattern of neutral evolution is determined by the pattern of mutation, so unless a whole-genome investigation is not conducted we may falsely reject neutrality. In this presentation we shall discuss our findings from investigating the VMB hypothesis in the human genome using informatics tools and techniques.

Poster

**Bacterial Growth in Coconut Oil and Listerine: Testing the validity of "natural" remedies**

**Genevieve Phillips**: Ivy Tech Community College-Kokomo  
Breannah Vogel: Ivy Tech Community College-Kokomo  
Tammy Greene: Ivy Tech Community College-Kokomo

The purpose of this experiment was to compare the antibacterial properties of coconut oil and Listerine. Oil pulling has been around for decades but there has not been conclusive testing to signify that oil pulling is effective at killing oral bacteria in comparison to Listerine. To test this, we used samples of human saliva. This was done by using pre-sterilized disposable inoculating loops. The inside of the mouth between the gum and cheek was swabbed for saliva from two human subjects. The saliva samples were put in test tubes that contained a sterile liquid media of nutrient broth and coconut oil or nutrient broth and Listerine. Next, the samples were transferred from the test tubes to agar plates and the bacterial growth was compared. A Kirby-Bauer test was also conducted to compare the zone of inhibition between the coconut oil and Listerine. It was observed that Listerine does provide a larger zone of inhibition than the coconut oil. Coconut oil is not as effective as Listerine at killing oral bacteria. However, when examining the samples from the agar plates for number and types of colonies, results were inclusive. More testing needs to be done to develop conclusive results.
Restriction Digest Screening facilitates efficient identification of UME6 Mutants

Ben Evans, Olivia Smith, Ethan Pickerill, Doug Bernstein, Ball State University

Alteration of amino acid sequence is an important tool in assessing protein function. Using CRISPR-mediated genome editing a residue change can efficiently be introduced into an organism. DNA sequencing of suspected transformants is traditionally the preferred method of mutation confirmation. When dealing with many potential transformants, sequencing can become expensive and time consuming. We have developed a method which utilizes only colony PCR and restriction digest to efficiently confirm mutation. Using CRISPR we introduced a variety of restriction sites coding for desired amino acids into Ume6, a known filamentation regulator of the human fungal pathogen Candida albicans. Success of transformation was highly variable among restriction sites, however, restriction digest screening allowed efficient identification of mutants. Alterations to the conserved zinc binding domain of Ume6 resulted in decreased filamentation of C. albicans. Furthermore, ume6 strains showed decreased virulence in Galleria mellonella larvae. To test if the zinc finger domain was sufficient for filamentation, truncations were introduced just downstream in the UME6 C-terminal region. Truncation of the C-terminal tail indicate the zinc binding domain is required, but not sufficient for hyphal extension. Our data indicate restriction digest screening as an efficient way to identify mutants, and the C-terminal region of Ume6 is important for filamentation.

The development of CRISPR-mediated genetic engineering in Candida viswanathii

Elena North - Ball State University
Kierra Adams - Ball State University
Doug Bernstein - Ball State University
Irene Reizman - Rose-Hulman Institute of Technology

Candida viswanathii is a fungal species that efficiently produces long-chain dicarboxylic acids, which are useful in petrochemical production. It is closely related to Candida tropicalis, a prominent human fungal pathogen. Because CRISPR has been implemented in other Candida species, particularly Candida albicans, we hypothesized that a similar CRISPR-mediated genome editing method can be developed in C. viswanathii. We have sequenced the C. viswanathii genome and identified guide sequences for the C. viswanathii ADE2 gene. We cloned these sequences into a vector that also expresses the Cas9 nuclease. After generating these constructs, we co-transformed these vectors along with a 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 to evaluate if these transformants have incorporated the repair template. We will then analyze the implications of genetic engineering in *Candida viswanathii* and work in collaboration to determine why and how this yeast species makes long-chain dicarboxylic acids.

**Poster**

**Investigating the effects of PUS5 deletion on mitochondrial encoded protein expression in Candida albicans and Saccharomyces cerevisiae**

**Jazmin L. Marks-Burns**, Allyson R. Morris, Doug 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 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. We find *Saccharomyces cerevisiae PUS5* deletion is more sensitive to drugs that inhibit oxidative phosphorylation such as oligomycin, suggesting they have a defect in mitochondrial function. We will investigate the roles of Pus5 mediated pseudouridylation on mitochondrial protein expression. We will use mass spectrometry to determine if mitochondrial rRNA pseudouridylation is required for wild type mitochondrial gene translation. 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.

**Presentation**

**Identification of Pseudouridine Synthase 7 (Pus7) Function in Pathogenic Yeast Candida albicans**

**Ethan Pickerill** (Ball State University), Rebecca Kurtz (Ball State University), Munni Begum (Ball State University), Douglas Bernstein (Ball State University)

*Candida albicans* is the most common human fungal pathogen. However antibiotic resistant *C. albicans* isolates are prevalent, highlighting the need for the identification
of new therapeutic targets. To identify novel drug targets, we must understand what differentiates fungi from humans at the molecular level. Pseudouridine is the most prevalent RNA modification. Pseudouridylation, the process of creating pseudouridine, occurs when a pseudouridine synthase cleaves a uridine glycosidic bond and reattaches the uracil at the C5 position. Pseudouridine is abundant in tRNA and rRNA where it plays roles in translation. Bacterial and mammalian pseudouridine synthase 7 (Pus7) homologs modify tRNAs, where Saccharomyces cerevisiae Pus7 modifies numerous substrates including tRNAs, snRNAs, rRNA, mRNAs. This work has identified Pus7 dependent residues in C. albicans tRNA E(CUC), and has also identified phenotypic changes in C. albicans that occur in response to PUS7 deletion. Pus7 knockout C. albicans has defects in growth, virulence, filamentation, rRNA processing, and biofilm production.

Poster
Multi Drug Resistant Plasmid Transfer Between Salmonella and Two E. Coli Strains

Jand Pangburn and Samina Akbar, Marian University, College of Osteopathic Medicine.

Multiple drug resistance is becoming increasingly problematic in the U.S., where antibiotics are overused both in agriculture and healthcare. One mechanism by which bacteria can develop this resistance is through acquisition of antibiotic resistance genes either on their chromosomes or on plasmids. A plasmid is an autonomous extrachromosomal DNA structure that replicates independently of the bacterial chromosome. Plasmids can be horizontally spread across different bacterial species through conjugation, transformation or transduction allowing multiple bacteria the ability to select for a resistant, advantageous phenotype. Salmonella infects around 400,000 people a year. Children, elderly, and the immunocompromised are the most at risk. Recent Salmonella and Escherichia coli clinical isolates have been found to carry antibiotic resistance plasmids. This limits treatment options even for healthy individuals. Salmonella, like other members of the Enterobacteriaceae family, have been found to carry more than one type of beta-lactamase genes, such as the blaCMY2 gene or blaCTX-M gene. To determine if horizontal transfer of these plasmids is possible, two different E. coli lab isolates were transformed with plasmids from Salmonella clinical isolates. The goal of the research project was to isolate these plasmids and analyze them for their incompatibility groups and their antibiotic resistance profiles. Using an extraction kit specifically designed for larger plasmids, isolates were analyzed in order to form a more complete picture of the Inc plasmids transferred to the E. coli strains. These strains are currently being tested for their antibiotic susceptibility profiles to determine which antibiotic resistance genes each plasmid carries. This study will advance our knowledge concerning the development
and dissemination of multiple antimicrobial resistance among \textit{Salmonella} spp. and other enteric bacteria in nature.

**Poster**

**Characterization and Dynamics of the R2 Retrotransposon in the Least Brook Lamprey, \textit{Lampetra aepyptera} (Abbott 1860)**

Laura Unfried and Rex Strange, University of Southern Indiana

R2 is a retrotransposable element that inserts itself into a specific location within 28S rDNA. Although R2 is well-studied in Arthropods, little is known regarding its insertion activity in vertebrates. Studies with \textit{Drosophila} have demonstrated that R2 activity correlates with the formation of inactive truncated fragments. We examined patterns of R2 truncation in two populations of the least brook lamprey (\textit{Lampetra aepyptera}) to better understand the activity of R2 in vertebrates. Most of the individuals from our Indiana population have a full-length and presumably functional R2 retroelement; however, no full-length R2 sequences were present in our specimens from Arkansas. Similarly, more truncation variants were found in the Indiana specimens (n=14) than in the Arkansas population (n=3). Examination of the sequences downstream of the R2 elements in the Arkansas specimens revealed divergences between the interrupted rDNA and the functional rDNA. However, the downstream sequences of the Indiana specimens were identical to the functional rDNA gene, suggesting recent transpositions. Our observations of \textit{L. aepyptera} suggest that although R2 is active in some populations, functional R2 elements have been lost in others.

**Poster**

**Biodegradation of Nylon-11 using Soil Bacterial Isolates**

Jocelyn Gatz-Schrupp and Hisako Masuda, Indiana University of Kokomo

Nylon-11 is synthetic polymer used commonly in many manufacturing industries. Because of its highly crystalline structure, it is resistant to chemical and physical decomposition. Therefore, when released into environments such as then ocean, it persists without degradation for a long period of time. This poses a significant threat to human and animal health. In this study, we show that pre-treatment of nylon-11 with organic solvents was effective in increasing the bioavailability of nylon-11. 16S rRNA gene sequencing of nylon-11 enrichment cultures show that strains belonging to genus \textit{Pseudomonas} become abundant when nylon-11 was the sole source of nitrogen. We successfully isolated a \textit{Pseudomonas} strain that can grow on media with nylon-11 as a sole source of nitrogen. In this presentation, we will discuss our current
progress in the characterization of these bacteria and the mechanism of nylon degradation in these strains.

Poster

**Survey of Antibiotic Resistance and Associated Genes in Natural Communities of Bacteria in an Urban Wetland Ecosystem in Indiana**

Megan Desrosiers\(^1\), Jane Pangburn\(^2\), Bridgette Antolin\(^1\), Samina Akbar\(^2\), and Azeem Ahmad\(^1\).

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Approximately 2 million infections from antibiotic resistance (AR) bacteria occur annually in the United States, resulting in a minimum of 23,000 deaths. These infections pose risk of more severe complications resulting in hospitalization and even death in the elderly and the very young. According to the Center for Disease Control (CDC), spread of AR among gram-negative bacilli (GNB) is arguably the most worrisome. This is because of limited treatment options, the ease of plasmid-mediated transfer of resistance genes among GNB, and the widespread distribution of *Enterobacteriaceae* in our environment as part of the human microbiome. Though there is an urgent need to develop new and useful antibiotics, importance of comprehensive AR surveys in the environment cannot be ignored. Much research has focused on antibiotic resistance from specific sources leading to good understanding of its occurrence in the environment but a lot less is known about antibiotic resistance in urban wetlands and its dispersal to opportunistic pathogens in natural habitats. In this study, we have targeted a pristine wetland ecosystem (Nina Mason Pulliam EcoLab) in the city of Indianapolis for the survey of AR bacteria and genes. We hypothesized that even nonpathogenic bacteria could serve as a reservoir of resistance genes as a result of introduction (and progressive accumulation), in the environment, of antimicrobial agents etc. The least understood and the most important aspect of AR is its spread among natural bacteria. Spatial and temporal detection of these resistant bacteria will help in source-tracking of antibiotic resistance and identifying *in situ* factors for selection of resistance genes and may even identifying a hidden “resistome” in natural environments. In this study, we first isolated antibiotic resistant bacteria of the *Enterobacteriaceae* group and analyzed their antibiotic resistance profiles against 20 currently used antibiotics in health clinics. Our initial results show multidrug resistance (MDR) prevalence (65% of natural bacteria belonging to various genera) in this ecosystem, and suggest widespread mixing, evolution and dispersal of antibiotic resistant bacteria. Molecular methods including PCR, sequencing and
DNA/RNA hybridization have been employed for the detection of specific resistance genes from each group of bacteria isolated from these environments. This study will allow comprehensive analysis of antibiotic resistance aimed at an undisturbed natural water ecosystem in Indiana.

Presentation
**Inactivation of a panel of recombinant respiratory syncytial virus (RSV) strains by cathelicidin (LL-37) and human beta-defensins**

Nathan A. Junod, Katelyn R. Castiglia, Caitlin E. Haas, Karina N. Latsko, Andrew T. Jacob, and Christopher C. Stobart (Butler University, Indianapolis, IN)

Antimicrobial peptides are small proteins that play a critical role in the human immune response. Human beta-defensins and LL-37 are antimicrobial peptides that provide defense and limit the growth of bacterial, fungal, and viral pathogens in the respiratory tract. Respiratory syncytial virus (RSV) is an upper and lower respiratory pathogen which can cause serious lung infection and death in young infants and the elderly. RSV has a legacy of physical instability, poor immunogenicity, and vaccine-enhanced disease, hindering the development of a vaccine for RSV. Therapeutics used after onset of symptoms are limited for RSV and the host events associated with RSV clearance remain unclear. Several studies have shown that LL-37 was able to inactivate the RSV laboratory strain A2. However, these studies were limited to one strain and appreciable differences in the structure and replication among RSV strains are known to exist. In this study, we evaluated the efficacy of LL-37 and beta-defensins on inactivation of a panel of recombinant RSV strains that express the attachment and fusion proteins of different RSV strains. We show that RSV is susceptible to inactivation by both LL-37 and human beta-defensin regardless of the strain. These studies demonstrate that RSV is susceptible to inactivation by several important antimicrobial peptides and highlight new targets for the development of RSV therapeutics.
Presentation

Evaluation of the Susceptibilities of a Panel of Recombinant Respiratory Syncytial Virus (RSV) Strains to Inactivation by Variations in Temperature, pH, and UV light exposure

Darby M. DeFord1, Jenna M. Nosek1, Katelyn R. Castiglia1, Christina A. Rostad2,3, Martin L. Moore2,3, Sean T. Berthrong1, and Christopher C. Stobart1 (1Butler University, Indianapolis, IN; 2Emory University, Atlanta, GA; 3Children’s Healthcare of Atlanta, Atlanta, GA)

Respiratory syncytial virus (RSV) is a leading cause of infant mortality worldwide and intensive efforts are being made to develop a vaccine. However, study of RSV biology and advancement of a safe and successful vaccine have been delayed by a history of vaccine-enhanced disease, poor viral immunogenicity in target populations, and hereditary and physical instabilities. Natural infection with RSV does enhance disease, which has supported the advancement of a live-attenuated RSV vaccine for infants, yet historic physical instabilities of RSV may limit development of viable virus preparations for vaccine use. Little is known regarding RSV strain-specific differences on physical stability. We hypothesize that the viral fusion (F) surface glycoprotein, which mediates viral entry through an active conformation change, is the primary contributing factor to RSV stability. In this study, we describe the characterization of a panel of recombinant laboratory and clinical RSV A and B strains that differ in viral attachment glycoprotein (G) and/or F protein expression. Using assays to evaluate thermal stability and susceptibility to inactivation by variations in pH and UV exposure, we observed significant differences in the thermal stability and limited variations to pH inactivation among several tested strains. We saw that strain A2-line19F showed significant enhanced thermal stability over strain A2. No differences in susceptibility to UV inactivation were observed. These studies provide the first insight into the physical stability of various strains of RSV, identify a key strain associated with enhanced thermal stability compared to the most commonly used lab strain A2, and support the idea of RSV F mediating virus stability.

Poster

Determination of the Role of the Spike Attachment Protein of Mouse Hepatitis Virus on the Stability of the Virus

Maham Nadeem, Emily F. Hasik, and Christopher C. Stobart (Butler University, Indianapolis, IN)

Mouse hepatitis virus (MHV) is often used as a model to study coronaviruses. In the past, differences among MHV variant strains have been noted in terms of infectivity
and the ability to establish syncytia. Previous studies looking at other viruses have proposed that the spike attachment protein may be a factor controlling for viral stability. In terms of coronaviruses, the differences in stability between different strains is unknown. Through this study we propose that the spike attachment protein of coronaviruses mediates environmental stability. In order to test this, we compared MHV-A59, MHV-2, and a chimeric virus, MHV-2S, which expresses the spike protein of MHV-2 in the genetic background of MHV-A59. These experiments highlight a unique function of the spike attachment protein in mediating environmental stability while also showing how diversity in spike protein structure and function can impact infectivity.

Presentation
Identification of a New Critical Regulatory Region within the Nonstructural Protein 5 (nsp5) Protease of Mouse Hepatitis Virus (MHV)

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

Current research of human coronaviruses aims to better understand and treat a variety of pathogenic systems ranging from the common cold to severe acute respiratory syndrome (SARS). Mouse hepatitis virus (MHV) is a coronavirus that remains an important and safe model to study coronavirus biology. The coronavirus protease, nsp5, remains a critical area of research in the field due to its vital role in virus replication. We hypothesize that the nsp5 interdomain loop (IDL), which connects domains 2 and 3 of the protease and has yet to be studied, plays a structural or catalytic role in protease activity due to its close proximity to the active site. Multiple amino acid positions on the IDL were mutated via site-directed mutagenesis and recovered viral mutants were evaluated for debilitated growth, altered protease activity, and increased sensitivity to temperature. Predictive modeling was also used to show the change in nsp5 structure before and after mutation. We show that the IDL is an important regulatory region for nsp5 function and identify novel regions and residues within the protease which may be targeted for future coronavirus inhibitor design efforts. These studies provide new insight into the structure and function of a key protein in the coronavirus genome and may advance efforts at development of effective therapeutics against these viral infections.
Evaluation of strain-specific physical stability among human metapneumovirus (hMPV) clinical isolates

Jenna M. Nosek1, Karina N. Latsko1, Darby M. DeFord1, Molly K. Roe1, John V. Williams2,3, and Christopher C. Stobart1

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Human Metapneumovirus (hMPV) is negative sense, single-stranded RNA virus from the newly formed family of Pneumoviridae. hMPV is a major cause of acute lower respiratory tract infections in young children and the elderly. Despite a global clinical burden of this virus, there remain no vaccines for hMPV and little is known of strain-specific differences among clinical isolates and laboratory strains. We recently demonstrated using respiratory syncytial virus (RSV) that appreciable differences in replication and physical stability exist among RSV strains. We hypothesize that significant differences in stability will also be observed among diverse hMPV strains. To test this hypothesis, we evaluated the replication kinetics and physical stability of a panel of hMPV clinical isolates. These studies provide the first detailed analysis of strain-specific differences among hMPV isolates and have implications for improving public health practices to limit the spread of and providing guidance on selection of strains for the development of future live-attenuated vaccine preparations targeting hMPV.

In Search of the Syk Phosphorylation Mechanism

Jacob J. Kinnun, Chao Feng, Duy Hua, and Carol B. Post, Purdue University

Spleen tyrosine kinase (Syk) has a central role in the transmission of activating signals within B-cells of the adaptive immune system. Abnormal function of Syk is implicated in many hematopoietic tumors and autoimmune diseases. Therefore, the activation mechanism of Syk is of particular biological and pharmacological interest. Syk is an enzyme composed of two SH2 domains separated by a linker region. Nuclear magnetic resonance spectroscopy (NMR) and molecular dynamics simulations (MD) have revealed that there is an increase uncoupling and dynamics of the two SH2 domains when Syk is phosphorylated. This suggests a novel phosphorylation mechanism which is entropically driven. The question remains how phosphorylation in the linker region induces domain separation. MD results suggest,
upon phosphorylation, the linker region has increased interaction with a few basic residues (R45, R67 & K164) of the SH2 domains. To test if these interactions are important, we substituted these residues with alanine (a small uncharged amino acid). If these interactions are important; mutations should affect the uncoupling mechanism with phosphorylation mimics of Syk. However, we still observed domain uncoupling in NMR measurements. One potential way to prevent interaction after phosphorylation is to increase salt concentration, which screens the charge introduced in phosphorylated residues. The results of introducing salt to uncovering the phosphorylation mechanism of Syk will be presented and future directions discussed.

Poster

High-throughput gene expression analysis of in vitro and in vivo mammalian cardiogenesis identifies shared developmental gene expression signatures

Andrew Williamson, Joseph Dalloul, Dwayne Tally, AJ Farmer, Laura Cochran, Hayden Fell, Garett Oxford, Rusty Gonser, Shaad Ahmad, Jeff Kinne, and Kristopher Schwab, Indiana State University

Mammalian heart development is regulated by an evolutionarily conserved genetic network that has been defined from numerous studies of model organisms and the genetic investigation of human congenital heart defects. Significant advancements in pluripotent stem cell technology have established new in vitro experimental systems allowing for the investigation complex developmental mechanisms within a single tissue culture dish. Of note, an impressive procedure has been developed that uses the modulation of developmental signaling pathways using small molecules to robustly and efficiently differentiate human pluripotent stem cells (hPSCs) into cardiomyocytes in a manner that recapitulates early embryonic heart development. This procedure allows both the experimental study of gene function in cardiomyocyte differentiation and a potential therapeutic treatment for heart disease.

Although high-throughput gene expression analysis has described the “transcriptome” of early mammalian heart development and in vitro hPSC-cardiomyocyte differentiation, a thorough comparison of developmental gene expression signatures of both data sets has yet to be completed. We first utilized several readily available bioinformatics tools to identify differentially expressed genes within the early developing mouse heart and then clustered these genes creating unique developmental gene expression signatures. Next, we analyzed the in vitro hPSC-cardiomyocyte differentiation data using the gene expression profiles obtained from the early developing mouse heart and identified genes that share similar gene expression profile during cardiomyocyte differentiation. These commonly shared genes include previously identified developmental regulatory genes, including pluripotency and cardiac genes, as well as uncharacterized genes that remain to be investigated. In
conclusion, we have developed a unique set of developmental expression signatures that characterize both the differentiation of hPSCs into cardiomyocytes by referencing the gene expression profiles of the early stages of mouse heart development. These analyses are essential for the future investigation of gene function utilizing the hPSC-cardiomyocyte differentiation procedures.

Poster

Characterization of Microbial Communities in the Rhizosphere Soil of an Invasive Shrub Amur Honeysuckle (*Lonicera maackii*) in Indiana

**Azeem Ahmad**, Marian University, 3200 Cold Spring Road, Indianapolis, IN 46222

Indiana has more than 2,000 species of vascular plants, approximately 25 percent of these are non-native to Indiana. *Lonicera maackii* (Amur honeysuckle), is an invasive shrub specie with devastating ecological and economic impacts in northern and central parts of the state. Despite the widespread negative effects of *L. maackii*, soil biological factors critical for its establishment and dispersal are still not known. The least understood and perhaps the most important aspect of establishing invasive plants is the interaction with the rhizosphere bacteria. In Indiana, the studies covering the ecological role and importance of these bacterial communities in the establishment of Amur honeysuckle are non-existent. Hence, an improved understanding of the rhizosphere community could play an important role in evaluating these interactions. The objective of this research is to specifically evaluate the functional role of rhizosphere bacteria associated with invasive Amur honeysuckle at the Nina Mason Pulliam EcoLab (NMPE)-an urban wetland ecosystem near Indianapolis. Here we constructed soil rDNA libraries and characterized bacterial community structure in the rhizosphere of Amur honeysuckle and compared it with the bulk soil. Total DNA is extracted from rhizosphere and bulk soil and then amplified with V4 region of the 16S rRNA gene. PCR amplicons are sequenced and analyzed using online bioinformatics tools. Temperature, pH, and soil moisture have been measured at sampling sites as well. Preliminary data from our laboratory suggest that specific bacterial phyla inhabit Amur honeysuckle rhizosphere that may be implicated in the generation of soil conditions conducive for its growth, establishment and dispersal. This study may identify novel plant-associated taxa while broadening our knowledge of the diversity of invasive plant-microbe associations.
**Poster**

**Purification and Imaging of Bacteriophages Obtained from Environmental Samples**

*Caleb M. Pinney*, Pamela L. Connerly  
Indiana University Southeast

A bacteriophage, or phage for short, is a type of virus that specifically infects and kills bacteria with a limited host range. The desired phages in this case are ones that infect *Caulobacter crescentus*. In short, four samples had the desired phages present out of the ten samples tested, were then purified and imaged by TEM. The purification began by filtering the water samples through a sterile 0.2 µm filter into sterile conical tubes, and spot plated on a lawn of bacteria. Five μL of the phage samples are placed on the top agar containing bacteria, and are allowed time to incubate and diffuse into the agar. When the raw phage did not form plaques, it meant that the desired phages were not at a high enough concentration to form plaques or that there were no phages present that were specific to *C. crescentus*. An enrichment process was done to the raw filtrates in order to increase the desired phage concentration if any were present which were then spot plated again showing four viable samples out of the ten samples analyzed. After this was done, the mixture was filtered again and another spot plate was done at three different concentrations in order to see the ideal concentration to make an infection plate to where individual plaques could be seen and then picked to purify the sample if there were phages present. The picks were then be placed in phage buffer and followed by another spot plate. This process was then done a minimum of five times in order to obtain a purified bacteriophage sample. The samples used a high titer stock of the purified samples for TEM imaging to determine phage morphology. The newly obtained morphological information of the phages from the samples can then be compared with known *C. crescentus* phages. The next step will be to compare the genomes of these phages with the genomes of known phages of *C. crescentus*.

**Poster**

**Isolation, Purification, and Morphological Characterization of Phages Found in Local Water Samples.**

*Danielle K Watt*, Pamela L. Connerly, Indiana University Southeast

Bacteriophages (phages) are ubiquitous viruses that infect bacteria with their DNA, replicate, and typically lyse the host cell in order to survive. Phages are estimated to be the most abundant biological entities on the planet and are specific to the bacterial hosts they infect. To help characterize and understand phage-host interactions, we are isolating, purifying, and characterizing bacteriophages that infect the nonpathogenic...
bacterium, *Caulobacter crescentus*, which is typical in freshwater sources. Water samples were collected and filtered through a 0.2-micron filter that allowed water and phage through but not bacteria and other contaminants. A phage filtrate enrichment was conducted to increase the population of any phage that infected *C. crescentus*. The enrichment was then plated with host bacteria and a soft top agar on 1% PYE agar and incubated overnight at 26°C. Plates were observed, and tiny spot clearings or plaques in the agar indicated the presence of an infecting phage. A 5-step dilution and reinfection method was used to purify each phage sample. Four *Caulobacter crescentus* infecting phages were isolated and purified. A high titer was produced for each phage sample to be used for TEM imaging, DNA extractions, restriction enzyme digests, and genome sequencing. Sequencing and comparative studies of phage genomes will provide a better understanding of phage genetic diversity and function.

**Poster**

**Isolation and characterization of bacteriophage in Hanover/Madison, IN**

**Breanna Amelunke, Leonardo Carnicelli,** Winter 2019 Bio334 Virology, and Natalia B. Hubbs  
Department of Biology, Hanover College, Hanover, IN

Bacteriophages (phages), viruses that infect bacteria, are some of the most abundant viruses in the biosphere. The host for many of these phages can be responsible for numerous diseases that have hazardous effects in human health, such as bacteremia, shigellosis, and urinary and respiratory tract infections. Just recently, there was an *E. coli* outbreak that was spread by Romaine lettuce, which affected 11 states and had a significant impact in human health by causing symptoms that ranged from diarrhea to kidney failure. The primary focus in this work was to isolate and characterize phages from the surrounding Hanover/Madison, IN areas, specifically from the Madison Wastewater Treatment Plant (MWWTP). The MWWTP processes sewage into safe water before releasing it into the Ohio River. In this work, phages were isolated from four locations in the MWWTP (from the raw sewage, 1° effluent, and before and after UV treatment stages) and grown on the following hosts: *E. coli*, *Shigella*, *Salmonella*, and *Citrobacter*. Phages were isolated and characterized by performing enrichment techniques, plaque assays, and gel electrophoresis. In the future, we plan to expand our sampling locations and hope to image our isolated phage, and eventually, sequence the genomes of the phage. By characterizing the phages in Hanover and Madison, IN, we can determine the hosts present in the area, which can provide clues about the health of the environment and give us insights into human health.
Poster

**Role of lipid droplets in prostaglandin E2 production during *Coxiella burnetii* infection.**

**Morgan E. Harrison, Minal Mulye**
Marian University College of Osteopathic Medicine, Indianapolis, IN

*Coxiella burnetii* is an obligate intracellular bacterial pathogen and the causative agent of Q fever. Chronic Q-fever manifests as potentially fatal endocarditis weeks to years after initial infection, suggesting *Coxiella*'s ability to live long-term inside the host. Our overall goal is to understand *Coxiella*'s long-term survival in the host. While *Coxiella* initially infects alveolar macrophages, it is also found in lipid droplet (LD)-containing foamy macrophages in cardiac valves of endocarditis patients. Our previous studies demonstrate that *Coxiella* manipulates LD homeostasis via the Type 4 Secretion System (T4SS), a major virulence factor which secretes bacterial effector proteins into the host cell cytoplasm to manipulate host cell processes. Further, inhibiting LD breakdown almost completely blocks bacterial growth. Since LD breakdown releases lipids, our data suggests LD-derived lipids are critical for *Coxiella* intracellular survival. LD breakdown releases arachidonic acid, a precursor of prostaglandin E2 (PGE2), which promotes an immunosuppressive environment in alveolar macrophages. Hence, *we hypothesize* *Coxiella* achieves long-term intracellular survival by manipulating host cell LD metabolism to establish a PGE2-mediated immunosuppressive environment. To test this hypothesis, we quantified gene expression of PGE2 synthesis enzyme cyclooxygenase-2 (cox-2) in differentially infected alveolar macrophages. Compared to uninfected cells, cox-2 was upregulated in *Coxiella*-infected macrophages but not T4SS mutant-infected cells. Further, quantitation by ELISA showed increased PGE2 levels in *Coxiella*-infected cells compared to uninfected cells. These studies indicate *Coxiella* T4SS actively manipulates cox-2 expression thus resulting in increased PGE2 in *Coxiella*-infected cells. Ongoing studies are identifying the direct correlation between LDs and PGE2 production in *Coxiella*-infected cells. Future studies will determine the potential of blocking PGE2 production as a supplemental therapy for *Coxiella* endocarditis.

Presentation

**Study to Decipher Cellular Targets of Lytic Protein YdfD on the *Escherichia coli* Genome**

**Hisako Masuda**, Indiana University Kokomo and Chun-Yi Lin, and Masayori Inouye, Rutgers University

Bacteria carry a number of genes that cause cell growth arrest or cell lysis upon expression. Some of the toxic genes constitute a Toxin-Antitoxin (TA) system, which
are found in all free-living bacteria. Antitoxins neutralize the toxins’ activity under favorable growth conditions. Degradation of antitoxins during stress conditions causes the inhibition of essential cellular functions by toxins, which ultimately leads to growth arrest or cell death. We identified a novel lytic gene on the E. coli genome called ydfD. YdfD lysed 99.9% of cells within 2 hours of expression. The co-expression of the upstream cognate antitoxin gene, dicB neutralized YdfD-induced cell lysis. Toxicity was also neutralized by the co-expression of another cell division inhibitor, SulA, indicating that YdfD-induced lysis is a cell division dependent event. We further characterized that the soluble C-terminal domain alone can exert toxicity when expressed in cytoplasmic space. Therefore, YdfD must compromise an essential cellular process occurring in cytoplasm. In this presentation, I will discuss our recent findings regarding potential cellular targets of YdfD and lysis mechanism.

Poster

**Contribution of lipid droplet breakdown to Coxiella burnetii infection.**

Rachel Silliman*1, Stacey D. Gilk2, Minal Mulye1
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*Coxiella burnetii* is an obligate intracellular bacterium and causative agent of culture-negative endocarditis. Although *Coxiella* initially infects alveolar macrophages, it is found in lipid droplet (LD)-containing foamy macrophages in endocarditis patients. LDs are host lipid storage organelles containing cholesterol esters (CE) and triacylglycerols (TAG). Our previous studies show that *Coxiella* actively manipulates host LD metabolism via its Type 4 Secretion System (T4SS), which secretes bacterial effectors in the host cell cytoplasm to manipulate cellular processes. Further, specifically blocking adipose triglyceride lipase (ATGL)-mediated LD breakdown inhibits *Coxiella* growth suggesting importance of LD-derived lipids for bacterial growth. However, how *Coxiella* regulates LD breakdown and the composition of LD-derived lipids is unknown. Our preliminary fluorescence microscopy studies using CRISPR knockouts and LD inhibitors indicate presence of TAG-rich LDs in *Coxiella*-infected cells. ATGL-mediated breakdown of TAG-rich LDs releases arachidonic acids, precursors for lipid immune mediators important for immunomodulation during bacterial infections. Hence, we hypothesize that *Coxiella* manipulates ATGL via its T4SS to initiate TAG-rich LD breakdown and subsequently modulate the immune response to promote bacterial survival. To test this hypothesis, we analyzed ATGL gene expression in differentially infected cells using qRT-PCR. Compared to uninfected and T4SS-infected cells, *Coxiella* infection increased ATGL expression indicating T4SS-dependent regulation of ATGL. Ongoing studies are elucidating the *Coxiella* T4SS-ATGL interaction. To identify cellular CE and TAG levels and the
breakdown products at different times post-infection, we are performing thin layer chromatography (TLC). Completion of our studies will identify the LD breakdown-derived lipids and how *Coxiella* regulates LD breakdown of to promote its intracellular survival.

**Mathematics**

Presentation

A Time-Lapse Visualizer for Pre-Computed Physiologically Based Pharmacokinetic (PBPK) Models

**Joel Stauffer**, Kimberley Grobien, Beomjin Kim, Stephen Coburn, Douglas Townsend, Purdue Fort Wayne

Physiologically Based Pharmacokinetic (PBPK) models are used to mathematically model the absorption, distribution, metabolism and excretions (ADME) of animals and humans. They allow for a researcher to approximate how an animal’s ADME systems process a particular chemical compound or compounds. These PBPK models typically produce time-indexed data, and the data volume can be quite large when a PBPK model is complex and processed in small time increments. The focus of this project resides in creating a time-lapse simulation supplemented with visualization that allows for a researcher to improve comprehensive understanding of a PBPK model (or more generally, compartmental models). The developed application (or computer system) is superior to traditional compartment graphical analysis considering it takes advantage of an individual’s ability to process changes and progression of state, and correlate intensity of color with chemical density. By compiling PBPK model data into this single cohesive time-lapsed animation, it assists the researcher’s understanding of the entire data leading to greater efficiency and accuracy in the analysis phase. The system design of the application is a single page web application (SPA) using the React framework since a SPA provides a great potential for accessibility and easily facilitates distribution. The application is designed to be flexible enough to facilitate the majority of PBPK models by allowing any number of compartments and fluxes, importing data from excel spreadsheets following a simple formatting standard, allowing user customization of the color, expected maximum compartment value and expected maximum flux value, and allowing the user fine control over the time-lapse animation speed. This application, currently licensed under the MIT license, should prove to be an effective free tool for researchers utilizing PBPK models. Ideally, this program will provide further insight into complicated PBPK models accompanied by volumes of data too large for traditional graphical analysis.
Breast cancer is a recurrent disease among women not only in the USA but also all over the world. There are several factors that can influence the multiple recurrences of this disease, which have been studied and recognized in various studies. With the fact that these factors are known, one cannot always select variables objectively and accurately. Very few attempts had been made to systematically detect the variables that are appropriate for recurrent event data. In this research, we explore an objective way of illustrating Cox-proportional hazards type models using two key features such as clinical and genomic information. The objective of the paper is to first identify most influential genes and clinical variables that are associated with higher risk of occurring single tumor among breast cancer patients from a high dimensional feature space (TCGA) using penalized Cox proportional hazard model. The next step is to explore if the genes and clinical factors associated with single tumor occurrence are also influential for the recurrent tumors. Our objective variable selection process reduces the total number of variables to eight clinical factors and sixty-five genes. Among these, metastasized extended near other parts of the body of a patient, ER-negative and PR-positive level associated with altered genes such as TNFSF4, DRP2, PLEKHH2, ST3GAL3, GFAP, NCRNA00095, HYMAI, CCDC123, BHLHE41, CCDC74B are the most significant covariates that influence the occurrence of single tumor among breast cancer patients. Chemotherapy and primary biopsy associated with altered genes HSD17B6, SDS, C9orf172, TMCO1, IDH2, RIMS4, TBCE, PLEKHH2, WHSC1, C1orf230 have an overall impact on the occurrence of recurrent breast tumors.

Presentation

Classification of Breast Cancer Treatment by Statistical Learning Approaches

Md. Monzur Murshed, Masudul Islam, Michal D. Lazar, Munni Begum

Classifying treatment plays a crucial role in cancer medication. It is strongly related to patient safety. Incorporating gene information is one of the effective approaches to
classify treatments before medicating breast cancer patients. Still, the accuracy of
gene combinations by treatment classification is a new approach in biomedical
research. On a large scale, it is undertaking to exhibit supervised statistical learning-
based approaches to compute potential gene combinations for treatment classification.
The main objectives of this paper are as follows: (i) Comparing four types of kernels
for the SVM-based model. (ii) Comparing SVM-based kernel models to LDA, QDA,
bagging, boosting and random forest classification models. (iii) Establishing a
benchmark breast cancer treatment classification method as a standard against others.
In this research, breast cancer genomic and treatment information are taken from the
Cancer Genome Atlas (TCGA) data portal.
The comparison with five traditional classifiers (LDA, QDA, Bagging, boosting and
Random Forest) under training-test partitions exhibit the superiority of SVM-based
RBF kernel model, which achieves under the boxplot, AUC and ROC curves.
Moreover, our statistical learning method shows the SVM-based kernel model, in
particular, the RBF model achieves the highest ACC (71.05), SE (88.12), PPV
(73.55), NPV (61.29), and AUC (73.80) among other methods. So, we proposed the
SVM-based RBF kernel model as a standard for modeling treatment through genomic
information.

Poster

**Bayesian Predictive Modeling for Personalized Treatment Selection for Breast
Cancer Patients**

Md. Monzur Murshed, Munni Begum
Department of Mathematical Science, Ball State University

Cancer is a complex biological phenomenon and will become the major cause of
deaths in the near future. Selection of treatment for cancer patients is still in its early
stage. Further research is needed in this important area of biomedical research, in
particular on treatment selection for the cancer patients. Treatment selection for
cancer patients depends on both clinical and biological information. An effective and
personalized treatment selection is not possible without combining these two sources
of information. In addition, each patient is biologically different from another patient.
In this research, we consider combined patient information along with the extent to
which historical patient is similar to the new patient (i.e., between patient variation).
This rigorous approach has the potential for selecting precise and effective treatment
for a new patient. We first use the Cox proportional hazard model to calculate the
treatment utility weight from clinical data. Next, we identified differentially expressed
genes. Then we use clustering algorithms to find the homogenous cluster for those
differentially expressed genes. Those homogenous genes serve as historical
information in Bayes predictive model to calculate the predictive probability for
treatment. Finally, we combine predictive probability and utility weight to
personalized treatment for a new patient and check whether assigned treatment improves the treatment outcome. We apply this approach to The Cancer Genome Atlas (TCGA) breast cancer data. Our result showed that instead of random assignment of treatment if we use Bayesian predictive modeling, treatment outcome improves by 3% for the breast cancer patients.

**Physics & Astronomy**

Presentation

**Numerical Simulations of Accretion Induced Collapse in a Double White Dwarf Binary**

Patrick M Motl
Indiana University Kokomo

We present a simulation of dynamically unstable mass transfer and merger of a double white binary. The binary components have initial masses of 0.6 and 0.9 solar masses so that the merged object will have a mass exceeding the Chandrasekhar mass. The simulation proceeds from an equilibrium binary system that is driven into contact and evolved with a fully three-dimensional fluid code that also solves Poisson's equation to incorporate the self-gravity of the fluid. While this simulation does not include nuclear burning and its associated injection of energy, we explore this initial simulation to determine the temperature and angular momentum structure of the merged object as well as characterize the material ejected from the system.

Presentation

**Measurement of lipid vesicle charge in solutions of zwitterions**

Azam Shafieenezhad, Rania Ousman, Ryan Z. Lybarger, Bruce D. Ray, Horia I. Petrache, IUPUI

Binding of water-soluble molecules to biological membranes is an important step in cellular signaling. We investigate the physical nature of such interactions using model lipid membranes. In particular, neutral lipid vesicles can acquire electrostatic charge by binding free ions from solution. Interestingly, they can also acquire charge from *zwitterionic molecules* that have dipole character. Examples are amino acids, *pH buffers* such as *MOPS* and *HEPES*, as well as *adenosinetriphosphate (ATP)* and its hydrolysis products. By using x-ray scattering we have determined that the presence of zwitterions affects the lattice spacing of multilamellar phosphatidylcholine vesicles in a manner that is consistent with the presence of electrostatic repulsion between neighboring membranes. The remaining question is how to quantify the surface electric charge and, equally interesting, how to determine
the sign of the charge conferred by zwitterions on membranes. Here we report measurements by dynamic light scattering (DLS) and by diffusion of phosphatidylcholine lipid vesicles in constant electric fields for a class of zwitterionic solutes. We find that the measured electrostatic charging can qualitatively account for the swelling of multilamellar structures and is consistent with NMR measurements of molecular binding. However, the results also show that more theoretical work is needed for quantitative analysis of electrostatic and van der Waals forces in these systems and for explanation of zwitterion affinities to biological membranes.

Presentation

**Structural and Magnetic Properties of Thermally Annealed Nd–Fe–B thin films**

**Saeed Yazdani**, Aaron Mosey, Joseph Soruco, Ashley S. Dale, Thomas Bsaibes, Ricardo Decca, Ruihua Cheng, Indiana University-Purdue University-Indianapolis (IUPUI)

In this investigation annealing and cooling processes of the substrate during the DC magnetron sputtering of the Nd-Fe-B thin film were investigated. In order to achieve high quality interface in multilayer structures, the Nd-Fe-B thin films were fabricated using a vacuum compatible heater during the sputtering. We have developed an in house vacuum compatible heater using high thermal conductive ceramic materials. The substrate heater is attached with a thermocouple in order to simultaneously measure the substrate temperature up to 700 oC. A series of Nd-Fe-B samples with a Ti wetting layer were fabricated at different temperatures and various thicknesses. In addition, the effect of Fe diffusion effect on NdFeB crystallization is studied. The samples were characterized by x-ray diffraction (XRD) and magnetometer measurements.

Poster

**EXPLORING THE ACOUSTICS OF 3-D PRINTED REEDS AND MOUTHPIECES**

**Justin Yoder**

Panayioti Panayi

Mark F. Masters

Purdue University Fort Wayne

3-D printing is a revolutionary manufacturing process with several applications. We endeavor to apply 3-D printing to manufacture clarinet reeds and trumpet mouthpieces that are acoustically identical to natural reeds and metal trumpet mouth pieces. Additionally, this process allows us to explore modifications to the reed structure and
shape of the mouth piece to see how it affects the sound. Thus far we have been successful in 3-D printing reeds and mouthpieces that sound close to the real thing. In our analysis we record a single note played by a human player and then perform a Fourier Transformation to determine the acoustic spectrum of the note. Unfortunately, the human player introduces a variable that makes comparing data from one day to another difficult as the player may be unable to play their instrument the same every time. The next step in our research is to eliminate the human player and replace them with a mechanical one that can play the notes in a consistent manner. With these human analogs we will be able to accurately test the acoustical properties of the reeds and mouthpieces using Fourier analysis.

Presentation

**Improving Short Range Gravitation Limits Using Cylinders**

**Thomas Bsaibes** - IUPUI
Luis Pires - Universidade Federal do Rio de Janeiro, Instituto de Física, IUPUI
David Czaplewski - Argonne National Laboratory
Daniel Lopez - Argonne National Laboratory
Ricardo Decca - IUPUI

Non-Newtonian interactions are predicted from both unifications theories and some extensions to the standard model. Either discription of these non-Newtonian interactions lead to Yukawa-like potentials, \( V(r) = Gm_1m_2 \alpha e^{-r/\lambda} r \), where \( \alpha \) is the relative strength, \( \lambda \) is the characteristic length of the interaction, and \( r \) is the distance separating two-point masses \( m_1 \) and \( m_2 \). Previous studies have placed upper bound limits on \( \alpha \) of \( \sim 10^{-3.5} \) for \( \lambda \sim 10^{-2} \) m. Limits on \( \alpha \) for \( \lambda < 10^{-5} \) m, however, are poorly constrained with the current best limits being \( 10^7 \) for a \( \lambda \sim 10^{-6} \) m.

Current efforts are being taken at IUPUI to improve limits on \( \alpha \) for interaction ranges below \( 10^{-5} \) m. The goal is to implement a new system consisting of a cylindrical test mass with radius of \( \sim 150 \mu \text{m} \) and planar source mass. Initial estimates show that the new system will improve the limits on \( \alpha \) by two orders of magnitude over previous measurements. The cylinder-plane system has some technical challenges that need to be overcome. Two main challenges this talk will focus on are aligning the cylinder and manufacturing the cylinders.
A Multi-Band Examination of the Spotted Short-Period Variable Star NSVS 2827877

Alexander Thomas
Ball State University
Robert C. Berrington
Ball State University

This study presents a photometric analysis of the eclipsing binary star NSVS 2827877. The images utilized to study this system were taken in May and June of 2018 using the Ball State University Observatory 20-inch (0.5 m) telescope in three different filters. The filters chosen for this analysis include the Bessel prescription of the Johnson B and V filters, and the Cousins R filter. The images received from the telescope were examined with the program AstroImageJ to find the differential ensemble magnitude of the system. The magnitude of the system found from the data was then further looked at using a program called PHysics of Eclipsing BinariEs (PHOEBE). PHOEBE is a program that was used for the purpose of generating a synthetic light curve from a physical model of the stellar system. The best-fit stellar models from this process were then determined from the observed data. NSVS 2827877 is shown by research to be an over-contact eclipsing binary star in thermal contact with surface temperatures $T = 5599 \pm 231$ K. The binary star system was found to have an orbital period of $0.3293 \pm 0.0006$ days, and a mass ratio of about 1.09 between the stars. At this point a period analysis was performed by comparing the Observed minus Calculated (O-C) times of minimum. Our analysis confirms the period of $0.3293 \pm 0.0006$ days.

Near-Earth Asteroid Monitoring Program at the NIRO Observatory

Adam Rengstorf, Purdue University Northwest

The Northwest Indiana Robotic (NIRO) Observatory has been monitoring near-Earth asteroids since 2012 and has had the ability to contribute positional data to the Minor Planet Center (M.P.C.) since 2013. Since that time, 132 observations of 19 different asteroids have been accepted for publication by the M.P.C. Even well-known and much-studied asteroids need to be continually monitored and tracked, as minor perturbations in their established orbits can cause drastic and potentially harmful (to Earth) changes in their future trajectories. Every semester, this program selects asteroids with the appropriate position in space and minimum brightness to be observed from NIRO. With the help of undergraduate researchers from physics and engineering, we observe the asteroids, calibrate and analyze the digital images,
determine their precise positions on the sky at the times of observation, prepare the data for submission, and add to the M.P.C.’s database, which is used to periodically update the asteroids’ orbits and publish new trajectories. This poster will summarize the work done to date and highlight the particular asteroids we have observed so far.

Poster

**Photometric Study of the Spotted W-Ursae Majoris Variable Star NSVS 896797**

Kyle Koeller, Ball State University

We present new differential ensemble photometry of an eclipsing binary star candidate NSVS 896797. All data was acquired using the Ball State University Observatory 20-inch telescope during February and May 2015. Then the stars brightness magnitude was compared with other stars of known brightness. I observed the variable star in three band-pass filters, Johnson B, Johnson V, and Cousins R and all images were reduced using the Image Reduction and Analysis Facility (IRAF). The differential ensemble photometry was performed using AstroImageJ (AIJ). The light curves were analyzed using the Physics of Eclipsing Binaries (PHOEBE) software. Our analysis shows that the binary system is consistent with an over-contact binary in thermal contact with a surface temperature of 5110±304 K. With this model the orbital period of the star was found to be 0.31297±0.00032. An analysis of an orbital period of the system was performed using an observed minus calculated (O-C) times of minimums. We report best-fit model parameters that were determined using the PHOEBE program.

Poster

**NSVS 3792718, a Spotless W Ursae Majoris Star**

Alexander J. Neal
Robert C. Berrington

Ball State University

We present a photometric analysis of the eclipsing variable binary star NSVS 3792718. A total of 644 images were taken in the Johnson B, V and Cousins R band passes. All images were acquired using the Ball State University Observatory 20-inch telescope atop the Cooper Science Complex in September, October and November of 2015. All images were reduced with the Image Reduction and Analysis Facility (IRAF). Differential ensemble photometry was performed using the AstroImageJ (AIJ) program. The system was modelled using PHysics of Eclipsing BinariEs (PHOEBE) program. With PHOEBE, a simulated light curve was compared to the observed light curve to determine best-fit model parameters. Our analysis shows that
NSVS 3792718 is consistent with an over-contact binary in near thermal contact with a surface temperature of $6470 \pm 290$ K and an orbital period of $0.4381 \pm 0.0007$ days. Observed times of minimum light were determined and compared to a calculated time of minimum light.

**Poster**

**Optimization of ferroelectric beta-phase in PVDF thin films**

**Ashley Dale, Aaron Mosey, Joseph Soruco, Ruibua Cheng, Indiana-University Purdue-University Indianapolis**

Precise control of the ferroelectric properties exhibited in poly (vinylidene fluoride) (PVDF) polymer thin films is of high interest due to its potential applications in electronic devices. Past research has indicated that pairing PVDF with a monomer hexafluoropropylene (HFP) and annealing the thin film at high temperatures induces the optimal beta-phase crystal structure for ferroelectric behavior. However, some of the applications for PVDF-HFP based multilayer structure devices prohibit the process of high temperature annealing. In this study, we demonstrate that it is possible to achieve high quality beta-phase crystal structure in PVDF-HFP thin films using Langmuir-Blodgett fabrication with thicknesses as low as 120nm without post annealing and this is demonstrated by x-ray diffraction data.

**Presentation**

**Optical refrigeration on CdSe/CdS quantum dots**

**Muchuan Hua and Ricardo S. Decca**

Indiana University-Purdue University Indianapolis

Cadmium selenide (CdSe) quantum dots (QDs, also known as nanocrystalines) are well known for their size tunable absorption and emission spectra within the visible range. Realizing optical refrigeration (OR) on them provides an active, non-contacting cooling method, in principle, by using sunlight. Here, we report the first time OR been achieved on zinc-blende CdSe/CdS (core/shell) structure QDs. The sample was synthesized with the method reported by Peng's group, which provides complete surface passivation. During our experiment, the sample was optically cooled by 0.3 K in an ambient environment about 298 K. While the average cooling efficiency of a single QD in the experiment was estimated to be close to $10^{-11}$ W per dot (only $10^{-15}$ mol dots were participated in the cooling processes), suggesting great potential of this technique (ideally, approximately 0.1 W per nmol). Details of the experiment will be discussed in the presentation.
Poster

Photometric Analysis of Asteroids (418) Alemannia and (4911) Rosenzweig

Richard D. Gorby, Adam W. Rengstorf
with Ball State University and Purdue University Northwest respectively

We present a new photometric analysis of the minor planets 418 Alemannia and 4911 Rosenzweig from September 11 to October 30 2017. All images were taken at the Northwestern Indiana Robotic (NIRo) Observatory, maintained by the Department of Chemistry and Physics at Purdue University Northwest. Images were reduced using the MaxIm DL software package. The light curve for 418 Alemannia showed a rotational period of 4.670 ± 0.006 h with a magnitude of 0.16 ± 0.02 which agrees with previously established light curves. The light curve for 4911 Rosenzweig showed a rotational period of 9.75 ± 0.01 h at a magnitude 0.08 ± 0.02. This result is in disagreement with two independently reported results stemming from data taken over a similar date range. Attempts to fit the data to either result produced an unconvincing light curve. We discuss possible reasons for the discrepant observations.

Poster

Bound Free Emission Spectra of Sodium Potassium Alloy

Anna Patterson
Mark Masters
Purdue University Fort Wayne

We are currently pursuing a deeper understanding of the experimental results of the bound free-emission of the sodium potassium alloy as it compares to our predicted models. This provides us with an opportunity to apply concepts from Quantum Mechanics to a physical system. Our first priority is maintaining the experimental apparatus. The essential component of the apparatus is the heat pipe oven. It allows us to heat the sodium potassium alloy into its gaseous state, which gives us the opportunity to analyze the compound without it reacting to the water in the atmosphere. The heat pipe oven has two main branches. Along one branch, a laser is used to excite the molecules from their ground state into the next electronic energy level. After the molecules undergo radiationless decay, a narrow bandwidth of photons is emitted as the molecule either dissociates or returns to its ground level. The energy distribution from the emitted photons will be compared with our predictions in order to determine the accuracy of our models.
**Poster**

**Development of a Low-Cost Single Photon Detector**

Spencer Kelham, Mark Masters, Purdue University of Fort Wayne

In physics, quantum optics is an important and fundamental field as it deals with the particle nature of light, the photon. A number of experiments have been used over the years to probe this aspect of light, all using single photon detectors. Traditionally, avalanche photodiodes are used as detectors in these experiments due to their high sensitivities. However, commercially available single photon avalanche diodes (SPAD’s) and the detectors that use them are very expensive, making implementation of quantum optics experiments difficult. These detectors usually consist of the avalanche photodiode as the sensor, a discriminator circuit to pick out detections caused by photon arrivals from electronic noise, and a quenching circuit that resets the avalanche photodiode into its reverse biased state after a photon is detected. These detectors also incorporate temperature control to reduce thermal noise in the detector. Our goal is the development of lower cost single photon avalanche detector. To test our discriminator and quenching circuits, we are using LED’s as photodetectors. Light emitting diodes or LED’s, have similar semiconductor junction designs to avalanche photodiodes, and some LED’s have been shown to detect single photon arrivals when placed in reverse bias, and can be used as cheap and expendable sensors for our preliminary designs. Additionally, this research has educational significance in the field of instrumentation, and could increase access to these experiments for undergraduate students.

**Poster**

**Multi-Band Ensemble Photometry of the Eclipsing Binary Star NSVS5196635**

Nathaniel Sparrow
Robert C. Berrington
Ball State University

Multi-band aperture photometry in the Johnson B, V, and Cousins R filters for eclipsing star candidate NSVS 5196635 from the Northern Sky Variability Survey is presented. All multi-band images were obtained by the Cooper Science Rooftop Observatory 20-inch telescope between April and May 2017. All images were reduced using the ccdred images reduction package in the Image Reduction Analysis Facility (IRAF) software suite. Ensemble multi-aperture photometry was performed with AstroImageJ software. Measured light curves are modeled by the Physics of Eclipsing BinariEs (PHOEBE) software package. Our analysis shows that the system has an earlier spectral type consistent with an A-type W Ursae Majoris system and an orbital period of 0.41166 ±0.00001 days.
Presentation

Harnessing spin crossover phenomena: thermodynamically stable voltage control of spin states at the ferroelectric interface

Aaron Mosey Indiana University Purdue University Indianapolis, Ashley Dale Indiana University Purdue University Indianapolis, Guanhua Hao University of Nebraska, Alpha N'Diaye Advanced Light Source Lawrence Berkeley Labs, Peter Dowben University of Nebraska, Ruihua Cheng Indiana University Purdue University Indianapolis

Thermal constraints and the quantum limit will soon put a boundary on the scale of new micro and nano magneto-electronic devices. This necessitates a push into the limits of harnessable natural phenomena to facilitate a post-Moore’s era of design. Thermodynamic stability at room temperature, fast (Ghz) switching, and low energy cost narrow the list of candidates. Molecular electronic frontier orbital structure of some d-block transition metal ions in octahedral fields will deform in response to the local energetic environment, giving rise to the eg and t2g suborbitals. The energetic scale between these two orbitals yields an S=0 low spin diamagnetic state and an S=2 high spin paramagnetic state. When placed in a polar environment, spin crossover complex [Fe(II)(H2B(pyz)2(bipy))2] will show locking of its spin state well above the transition temperature with an accompanied change of conductivity. We deposit molecular thin films of [Fe(II)(H2B(pyz)2(bipy))2] on a ferroelectric polyvinylidene fluoride hexafluoropropylene substrate and show voltage controllable, room temperature, stable locking of the spin state with the corresponding conductivity change. This opens the door to the creation of a thermodynamically stable room temperature multiferroic gated voltage device.

Presentation

Magnetically Levitating Magnet over Elliptical Hole in Superconductor

Grace Mattingly and Ricardo Decca, Indiana University-Purdue University Indianapolis, USA
Jordi Prat-Camps, University of Sussex, UK
Adrián Ezequiel Rubio López and Oriol Romero-Isart, Institute of Quantum Optics and Quantum Information, AT

It has been shown that a superconductor with a hole can trap a magnet where the gravitational force and the magnetic repulsion are in equilibrium. If this hole is elliptical, the magnet has a preferred direction along the major axis of the ellipse. The system could oscillate with a given frequency and it could be used as a highly sensitive force sensor due to its isolation from the environment. The frequency of the oscillations in three spacial (x, y, z) and two angular (α, β) directions is given by the
second derivative of the magnetic potential of the system at its minimum. This potential is characterized for a range of free parameters: height of the magnet, ellipse aspect ratios, thicknesses of the superconductor, and London penetration depths. From these, the resulting frequencies are computed. The relationship between the change in frequency and the change in the free parameters is important for selecting a trapping potential with the desired frequency. The integrity of the superconductor is confirmed with an analysis on the penetration of the magnetic field into the superconductor. Because of the prediction for very high sensitives, there are many technical applications and possible advances in precision measurement. One motivation for the system is to act as a torsional pendulum with which the gravitational constant, the least well-known constant, could be determined with greater precision.

Presentation

Use of Digital Image Correlation to Determine the Position of an Object in Motion with High Precision

Rutuj Gavankar and Ricardo Decca, Indiana University - Purdue University Indianapolis.
C.D. Hoyle and Adam Turk, Humboldt State University.

The gravitational constant $G$ is the least known fundamental physical constant. The currently recommended value is with a relative uncertainty of. There is a large discrepancy between various experimental techniques used to measure $G$. We are using a torsion pendulum setup to measure $G$. The setup involves using cylindrical attractor masses that surround a torsion balance suspended with a fiber. The angular acceleration of the balance produced by the attractor masses will be measured and used to determine $G$. We were interested in determining the drift in the position of two oscillating cylinders which would be used in the torsion pendulum setup with a certainty of 0.1 μm to determine $G$.

To do this, we are using a technique used for stabilizing the focal drift in a microscope by using image correlation. Digital image correlation is a measurement technique that involves taking images of an object and comparing the changes in the image to determine the displacement of the object. We implemented image correlation technique to determine the drift in the position of an object about a meter big with a precision of 0.1 μm.

We simulated an ideal two-dimensional case of the cylinder using a cell phone camera sensor and verified if the technique can be used to extrapolate the displacement in both dimensions with the required precision. We were able to measure the position of the object with a certainty of $\sim$0.2 μm with the current set up. Our estimates show that this technique can be scaled up and a larger image sensor can be used to measure the
object with greater precision. Future plans involve testing this method for a more realistic case involving smooth surfaces and a smaller cross-section of the cylinder.

Presentation

**Observation and Analysis of Eclipsing Binaries NSVS 4312042 and NSVS 2910034**

Wes Tobin, Indiana University East
Robert C. Berrington, Ball State University

We present new observations and analyses of two eclipsing binary systems: NSVS 4312042 and NSVS 2910034. Multiple reduction and aperture photometry techniques are discussed. Systems are modeled using the PHysics of Eclipsing BinariEs program, which utilizes the Wilson-Devinney code to determine best-fit stellar models.
Plant Systematics & Biodiversity

Workshop
Fern Identification Workshop

Roger L. Hedge, Indiana Department of Natural Resources, Division of Nature Preserves, Indianapolis, IN 46204

Indiana’s varied habitats ranging from wetlands to forests to cliff communities support a high diversity of ferns—approximately 80 species. These mostly green plants can be intimidating and challenging to identify. What they lack in showy color, however, they more than make up for in their intricate shapes and forms, from the relatively diminutive spleenworts to larger wood ferns, and even larger cinnamon fern. Geared toward beginners, this approximately 1.5-hour workshop is for those who have an interest in fern identification but lack field experience with this group of plants. Basic vocabulary, fern morphology, important field characteristics, and distribution will be covered for some common Indiana ferns. Please bring a hand lens or magnifying glass as specimens will be available to view.

Presentation
Phylogeography and population genetics of Sandmyrtle (Kalmia buxifolia, Ericaceae)

Emily Gillespie Butler University
Tesa Madsen-McQueen University of California, Riverside
Zack Murrell, Appalachian State University

Sandmyrtle, Kalmia buxifolia (Bergius) Gift & Kron, exhibits a disjunct distribution in eastern North America. Extant populations occur in the New Jersey Pinelands (NJP), the Southern Appalachian Mountains (SAM), and the Sandhills/Cape Fear Arch (SCFA) region of the Carolinas. There is no present-day geographic connection between NJP and SCFA populations, and very little between the SAM and SCFA populations. Despite relatively close geographic proximity, the SAM and SCFA populations are dissimilar in terms of habitat; SAM populations typically occur on rock outcrops and SCFA populations occur in wet pinelands that are ecologically similar to the northern NJP populations. Individual plants also vary morphologically, with SAM and NJP plants tending to be somewhat mat-forming and SCFA plants tending to be taller and spindly. Over the last 150 years, experts have recognized between one (current consensus) and three species based on a combination of morphology and habitat. We developed microsatellite DNA markers for Sandmyrtle using an Illumina sequencing dataset, sampling from three populations representing
the three main parts of the Sandmyrtle distribution. We then applied these markers to an additional eight populations in order to gain landscape-level insights into Sandmyrtle’s genetic variability and structure, deviation from Hardy-Weinberg assumptions, and phylogeographic history. Our current dataset indicates that some populations differ significantly from others with regard to these characteristics. Additionally, SCFA and NJP populations are more genetically similar to each other overall, somewhat aligning with the taxonomic boundaries indicated by JK Small in the 1930s.

Presentation

Results of the 2016 Goose Pond Fish and Wildlife Area Biodiversity Survey, Greene County, Indiana

Donald G. Ruch, Ball State University

On June 18th – 19th, 2016, a biodiversity survey (also known as a bioblitz) was conducted at Goose Pond Fish & Wildlife Area (GPFWA) in Greene County, Indiana. GPFWA, a 9033-acre restored wetland and grassland complex, is owned by the state and managed by the Division of Fish & Wildlife, Indiana Department of Natural Resources; Travis Stoelting is the property manager. Over 95 scientists, naturalists, students, staff, and community volunteers working on 17 different taxonomic teams have reported 883 taxa to date. (I am still waiting on results from the bird and aquatic macroinvertebrate teams.) Taxonomic teams included aquatic macroinvertebrates, bats, bees, beetles, birds, butterflies, fish, freshwater mussels, herpetofauna, mammals, moths, mushrooms, non-vascular plants, odonates (dragonflies & damselflies), singing insects, spiders, and vascular plants. Although this is the second bioblitz held by IAS at GPFWA (the first was in July 2010), many Greene County records have been reported, and even a few state records. The results for each group will be presented. As an example, here is a summary of the results from the vascular plant team: 417 taxa (412 to species); 318 (76%) are native; 26 potential Greene County records; one potential state record (*Typha domingensis*); seven state listed species including *Carex gravida* listed as state endangered; marsh community is still early successional with many dominant invasive species.

Presentation

Floristic Inventory of Grass Lake Nature Preserve, LaGrange County, Indiana

Scott A. Namestnik, Orbis Environmental Consulting

The 102-acre Grass Lake Nature Preserve near Plato, LaGrange County, Indiana, was acquired by ACRES Land Trust in December 2017. The preserve is largely a flat-bottomed basin surrounded by a narrow rim of degraded upland forest; the more
ecologically intact portions consist primarily of a marl beach prairie community, with smaller areas occupied by red maple-tamarack swamp forest, prairie fen, and Grass Lake itself. Prior to the 2018 study, 92 vascular plant taxa (87 native) had been documented from the preserve, including 11 Indiana endangered, threatened, rare, and watch list species. Meander surveys were conducted in the spring, early summer, and late summer of 2018, resulting in a total of 379 vascular plant taxa (334 native) identified to at least the species level. The Floristic Quality Assessment was applied to the floristic inventory, resulting in a mean Coefficient of Conservatism value of 4.2 (4.7 native) and a Floristic Quality Index of 81.8 (85.9 native). In addition, 15 Indiana endangered, threatened, rare, and watch list species were documented. An impressive 46 species said to be restricted to remnant areas that have not been damaged as a result of European settlement were recorded in the floristic inventory.

Presentation

Comparison of the composition and structure of herbaceous-layer communities within mature forests of East Central Indiana

Taylor Davis, Kemuel Badger, and Donald Ruch, Ball State University

The herbaceous layer is not a large contributor to the overall biomass of a deciduous forest making it often overlooked in terms of the influence it has on forest ecosystems. Nevertheless, this low-lying stratum is dynamic and diverse, and it maintains an important role within the forest ecosystem. Our goal was to compare the herbaceous communities within two remnant, old-growth forests of East Central Indiana. At both sites the soil and overstory compositions were analyzed in order to facilitate comparison between study areas. Ultimately there were three sample areas. Two within Ginn Woods in Delaware County and one in the Davis-Purdue Research Forest in neighboring Randolph County. A total of twenty-five 2m by 2m quadrats were placed along five transects in each of the sample areas. Every species within a quadrat was identified. The cover for each was estimated using a modified Daubenmire Cover Class system. The sites were visited a total of three times throughout the growing season in order to account for phenological changes. The quality of the sites were determined using the Floristic Quality Assessment, and the similarity of the sites were analyzed using PC-ORD multivariate statistics. Despite having different histories, the herbaceous communities at Ginn Woods and Davis-Purdue were overall similar to each other. There was some difference between sample locations in the number of total species present and the number of higher quality plants identified. This could possibly be explained by the history of grazing at the Davis-Purdue site during the early 1900s. Overall, both Ginn Woods and the Davis-Purdue Research Forest can be considered high quality sites offering reference sites to compare other potentially disturbed communities within East Central Indiana.
Recent Revision of Indiana’s List of Endangered, Threatened, and Rare Vascular Plants, with Notes Regarding Select Species Discovered Since 1980.

Michael A. Homoya and Roger L. Hedge, Indiana DNR Division of Nature Preserves

The first published list of Indiana’s endangered, threatened and rare (ETR) vascular plants was compiled by John Bacone and Cloyce Hedge of the Indiana Department of Natural Resources. It was printed in Volume 89 of the Proceedings of the Indiana Academy of Science (PIAS 1980) followed by an update in Volume 95 (PIAS 1986) authored by Jim Aldrich, John Bacone, and Michael Homoya. Other than its inclusion in the Indiana Register as a Nonrule Policy Document, no published versions have occurred since the last aforementioned PIAS volume. However, the list has been displayed online with periodic updates. Updates reflect the dynamic nature of the list, as new information can change a species’ status. It may be due to a revision in taxonomy, threats to its habitat, a shift in number of occurrences, degree of conservation concern, or the discovery of it as new (or as once present but now absent) in the state. The ETR list has proven to be a useful guide for protecting Indiana’s most sensitive flora.

Community patterns and distribution of rare species in Grand Sable Dunes, Pictured Rocks National Lakeshore

Jonathan C. Danielson and Jordan M. Marshall, Purdue University Fort Wayne

Grand Sable Dunes, as a perched dune field on the shore of Lake Superior, is a sensitive ecosystem subject to continual disturbance. Repeated natural disturbances necessitate specialized plant communities to develop. Our objectives were to quantify successional changes in the plant community over time and identify population demography changes for rare species within the dunes. Target plant community composition and structures (i.e. richness, diversity) were quantified in 2011 and 2018 across Grand Sable Dunes in 1 m² quadrats. Additionally, two relatively rare plant species (Cirsium pitcheri and Tanacetum bipinnatum) were selected to quantify demographic (i.e. flowering, non-flowering) patterns and changes over time. Samples for C. pitcheri and T. bipinnatum were acquired via circle-plots 2.5m in diameter. Population comparisons between 2011 and 2018 illustrate minimal change in community structures (richness and diversity). Composition increased slightly with eight species occurring in 2018, but not 2011. Additionally, community similarity was high (~78%) between the two years. C. pitcheri occurrence was inversely related to presence of other species. Plant community composition in eastern and western survey zones within the dunes appear to be diverging. Minor changes in the plant community
composition and structures indicate successional changes have occurred, but without major disturbance. This divergence in community composition may be related to weather event angle of incidence associated with Lake Superior and disturbance.

Presentation

The Indiana University Herbarium Digitization Project

Eric B. Knox and Paul E. Rothrock, Department of Biology, Indiana University-Bloomington

The 5-year IU Herbarium Digitization Project will be completed in a few weeks as the final details are resolved. The vascular plant specimen processing involved: 1) Creating an electronic inventory of taxa; 2) Curating all the specimens and reorganizing the collection according to current family delimitation; 3) Barcoding and imaging all specimens; 4) Transcribing the label information; and 5) Georeferencing the collection localities. Species-level resources were added by uploading: 1) Descriptions from Gleason & Cronquist’s manual for the northeastern U.S.; 2) Observations from Deam’s Flora of Indiana; 3) Standardized common names from the Biota of North America Program; 4) Photographs of live plants; 5) Creating synonym links between old names and current taxonomy; and 6) Morphological and phenological information to power the online plant identification tool for the Indiana flora. A full set of historical U.S.G.S. topographic maps were added to GEOLocate (to aid with georeferencing), various improvements were made to the underlying Symbiota platform; and detailed locality information is redacted for Indiana state and federally protected plant species. The final clean-up involves: 1) Standardizing full collector names; 2) Checking and correcting standardized author citations; 3) Creating hyperlinks between GenBank DNA sequences and IND voucher specimens; 4) Linking county voucher specimens to the Indiana species list; and 5) general error detection and correction. The relatively small IND collections of algae, lichens, bryophytes, and fungi are in dedicated Symbiota data portals for those groups, all of which can be reached through the IND webpage: https://biology.indiana.edu/research/facilities-resources/herbarium.html. These online resources are available to all other Indiana herbaria that want to digitize their collections.
Poster

Urban tree composition, structure, and health in a historic city park

Michelle Same1, F. Collin Hobbs1, Ben M. Bond2, and Sharon Cuttriss2
1Huntington University, 2City of Huntington, IN

Memorial Park is a large, historic city park in Huntington, IN. The park contains forested tracts, athletic & recreational facilities, ponds, a war memorial, and sunken gardens in a reclaimed quarry, however most of the park is an arboretum-like setting with many large, open-grown trees. In this study we quantified the biodiversity of the park’s tree community and performed a tree health assessment. We then compared our results to a previous study of the Huntington University arboretum and a local natural forest. Coordinates for each tree in the non-forested portions of Memorial Park were captured in the summer of 2017 using a Trimble Geo7x GNSS Receiver with laser attachment. Each tree was then visited again, identified to species, DBH was recorded, and health was assessed on an ordinal scale of 1-5. These data were then entered into a GIS. Forest composition and structural parameters were calculated including abundance, dominance, diameter distribution, Shannon diversity, and evenness.

Our study area covered 12.6 ha and contained 418 trees representing 48 species, with a Shannon diversity of 3.23 and an evenness of 0.82. Total basal area was 7.0 m2/ha, with black walnut, white oak, and red cedar being the three most dominant species. Of species with 5 or more individuals, Siberian elm and black cherry had the poorest average health scores. Considerable differences exist between Memorial Park, the Huntington University arboretum, and the local natural forest in patterns of species abundance and dominance, although the park and arboretum are more similar in species diversity and evenness. The data gathered in this study will help guide future management decisions regarding the tree resources at Memorial Park.
Science Education

Presentation

**Biology Indianapolis Outreach: A new undergraduate course connecting campus with community**

**Erin Gerecke** and Philip Villani, Butler University

Connecting undergraduate students with their local communities through specific coursework is a growing trend in higher education. For students studying STEM disciplines, community engagement opportunities that align with their scientific interests and potential career goals allow them to apply their learning beyond the classroom and consider their own place as individuals in the broader communities in which they live. We present the structure, student learning outcomes, and overall results of the pilot semester of our new community science engagement course—Biology Indianapolis Outreach—at Butler University, part of the university’s broader Indianapolis Community Requirement. In this course, students developed and presented hands-on, biology-themed activities for local children and families in partnership with the Indiana State Museum and Celebrate Science Indiana. Additional coursework included discussion of STEM outreach programs throughout Indianapolis, science communication strategies for different audiences, and individual reflection on the theme of community engagement.

Presentation

**Freshman Research Engagement**

**Kristi Bugajski**, Patrice Bouyer, and Michael Watters

The goal of this project is to have freshman actively involved in a research project during the academic year to improve retention in the STEM disciplines and to prepare the students for their future careers. The freshman will work directly with a faculty mentor and will have senior lab mates to supervise their work, ensuring proper lab techniques. Participants will be fully involved with the project, conducting the experiments and analyzing the results of a large-scale research project. This is much different than a traditional experience for a freshman, where they simply do an assigned project as part of a class. An important part of retention at any University is integration into campus life. Being involved in research starting in the first semester of the first year should improve their retention in the STEM discipline and at Valparaiso University. In addition, it will help students gain marketable skills and better prepare them for future careers. Starting in a lab during their first year greatly
increases the chance of having their name on a scientific publication and provides the close relationship with a faculty member that yields strong letters of recommendation. We have had eighteen students enrolled previously and have another cohort of 6 students currently. On their exit surveys, all students from the 2016 cohort communicated that the program exceeded their expectations and all are continuing with their research programs. A comment from one student said, “I think this program was a great way to integrate freshman into “doing” science” and that is a great summary of the benefits of this program.

Presentation
Introducing Scientific Literature and Scientific Writing to Freshmen Biology Majors.

Christine Barlow, Ivy Tech Community College

Expectations for freshmen biology majors to engage with and write about primary literature are increasing. Effectively meeting these standards in classrooms composed of students with varying levels of skills and experiences with scientific literature and scientific writing is challenging. In this presentation, I will present the results of two years of work developing a primary literature project for freshmen biology majors. The quality of student work has improved by incorporating resources and checkpoints that prevent common pitfalls experienced by students. This highly structured project begins with assignments that have students evaluate primary and secondary literature, provides them with resources that introduce them to literature search engines, and teaches them how to cite literature within their papers and in the literature cited page. Checkpoints include a one-on-one meeting between the student and professor early in the project, as well as a peer-review assignment near the end of the project. When finished, students summarize two biological studies from the primary literature in a 4 – 6 page paper and present their work in a professional presentation to their peers. I consider this project an essential stepping stone that builds the skills and provides the experiences necessary for students to successfully write traditional literature reviews in their future 200-level biology classes.

Poster
Improving STEM Retention and Commuter Engagement through Research, Cohorts, and Faculty Mentoring

Michael Watters, Kristi Bugajski, Valparaiso University

The Establishing Practices Integrating Commuter students (EPIC) program (NSF Award ID: 1564855) integrates commuter STEM students into the campus through shared research experiences and social activities with residential students. The unique
problems of commuter students are: transportation, multiple life roles, integrating support networks, and sense of belonging. EPIC relieves some burdens through a significant scholarship and additional support services. The keystone to EPIC is four academic years and one funded summer of research opportunities. Students participating in undergraduate research have increased retention rates and improved performance. Students also report gains in independence, motivation to learn, and active participation in subsequent coursework. EPIC students are also given academic and workforce training, such as workshops on presenting research and instruction in professional writing. Social integration activities include monthly meals, field trips, and an annual overnight retreat. The ongoing EPIC program aims to: improve student retention, especially among commuters; increase the number of STEM graduates prepared for research-intensive careers; and increase and strengthen VU’s research intensive majors.

Presentation

**Designing Botanical Teaching, Learning, and Research Spaces: A Case Study in Student Arboretum Design**

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

Arboreta are places where trees are cultivated for their scientific and educational values. They are spaces for research, teaching, and learning that often provide natural habitat for wildlife and places of passive recreation. Ball State University landscape architecture students were tasked with designing a new arboretum for the Hanover College campus. The goal for the new facility was to support Hanover College curricula and research, and to potentially be an educational outreach to the surrounding communities for programs such as tree identification, leaf collections, and other public-school science projects. Ball State students studied the history of arboreta in the US, developed case studies of landmark arboreta, interviewed university instructors to identify the character-defining features of botanical teaching and learning spaces, and assembled masterplan, designs, and details for a new arboretum. Students excelled in identifying an array of common design themes such as educational walks, unique arrangements of plants (ecosystems, Marion Jackson’s *101 Trees of Indiana*, taxonomic, natives versus exotics, food producing), interpretive signage, and digital apps communicating plant identification and botanical information. Many students proposed highly creative connections between existing land uses on the Hanover campus and their proposed designs (i.e., fine art, science, dining, athletics, and recreation). Students struggled to identify and design the ideal botanical teaching, learning, and research spaces. This presentation will report the methods and results of the course, examine student outcomes, and propose modifications in the class content to improve future arboretum design course
offerings. Examples of student arboreta design (research, programming, conceptual diagramming, final design, and design communication) will be presented.

Poster

**Improving STEM retention via early research engagement**

**Patrice Bouyer**, Michael Watters, Robert Clark, Laura Rowe, Sara Dick and Kristi Bugajski. Valparaiso University.

We present here a progress report on the FRESH (Freshman Research Engagement in the Sciences) program. The goal of the program is 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 in order to learn lab techniques but then were encouraged whenever possible to work on their own to nurture the capacity to work independently. Participants were fully engaged in an authentic research project (performing experiments, analyzing and discussing results), not 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 twenty students in three cohorts who joined the program as freshmen, eighteen are still in their major (or have graduated) and have co-authored eight different peer reviewed publications and twenty-four conference presentations to date. Freshman-to-Sophomore retention in the program has been significantly higher than among the general student population (60% retention, p=0.005). Based on these initial successes, we are now preparing to submit a proposal to fund an expanded version of the FRESH program to examine the impact of early research engagement on student retention, performance and future success.

Presentation

**Web Based Presentation Tools- The Story Map App**

**Josephine Shireen Desouza**, Ball State University

The author uses ArcGIS online in a science methods course, to design a story map app which is used as a tool supporting the teaching and learning of a science concept/topic. In this presentation, the author describes how a story map can be created that depicts the correlation of volcanoes and earthquakes including information of their impact on communities in their vicinity. The story map is commonly used as a presentation tool and does not have analytical functions. Typically, users create a story map to describe a historical event, report the importance of a place, and visualize trends and patterns of events that occur within the
perspective of a certain geography. Within the (Environmental Studies Research Institute (ESRI) gallery of story maps, various templates are presented that are suited for the presentation of photographs or videos that are labeled with captions, and are linked to an interactive map. This presentation will focus on the Story Map Cascade app template which is most suitable for a full screen scrolling effect. The narration text can be combined with interactive maps and multimedia content. By accessing the ESRI website https://storymaps.arcgis.com/en/ users can choose the most appropriate template for their narration.

Poster

Understanding Retention Pathways and Bottlenecks of STEM Majors: Implications for Student Success

William S. Elliott, Jr., Christos Deligkaris, Eric Greenwood, Adrian Gentle, Shelly Blunt, and Amy Chan Hilton, University of Southern Indiana

This project examines evidence-based student retention and instructional practices, understanding bottlenecks, and other factors impeding student progress in STEM at the University of Southern Indiana (USI). Initially, a systems map was created by a group of faculty members from chemistry, engineering, geology, mathematics, and physics to analyze retention pathways of STEM majors at USI after having read the literature and explored institutional information. Systems thinking is an effective way to understand the complexity of a topic, identify links among themes, and discover potential individualized research directions. Each faculty member involved with this project then created their own systems map to better constrain their specific area of interest. Research projects that originated from this process include: (1) comparing student attitudes towards group work implementations in introductory Physics courses; (2) evaluating the effectiveness of Pre-Calculus as a preparation for college-level Calculus; (3) exploring the impact of course repeats on student success at USI; (4) assessing the impact of an early undergraduate research program on increasing retention rates of STEM majors at USI; and (5) using a faculty learning community and systems mapping to engage faculty members with educational research. Selected student learning outcomes of these projects include: (1) improved comprehension and problem-solving skills through group work and active learning, (2) modified student pathways to improve retention and success, and (3) enriched student engagement through early undergraduate research. The results from this project will support longer-term retention initiatives and inform strategies to improve student success and retention of STEM majors at USI. In addition, these projects will better position USI to seek external funding to support student retention efforts. Finally, classroom strategies that result in improved student learning will be expanded to other sections of introductory courses in mathematics and physics at USI.
The Professional Amateur Astronomy

Wes Tobin, Indiana University East

Indiana University East is a regional (commuter) campus of Indiana University, located in Richmond, Indiana. IU East does not offer a major or a minor in astronomy, but it is located near some of the darkest skies in Indiana – a prime location for observational astronomy research. Students have the opportunity to explore observational methods in 100-level courses, with several indicating interest in attempting an observational research project. A novel approach is thereby needed, and presented here, with the purpose of providing undergraduate research opportunities in observational astronomy. Successes, limitations and challenges are examined: an ever-changing campus environment, the use of commercially available (amateur) astronomy equipment, and the creative approaches of connecting undergraduate experiences and curriculum.

Hot Topics
One Health in Action: Zoonotic and Vector-Borne Disease Investigations

Taryn Stevens, Indiana State Department of Health

This presentation will describe recent investigations in Indiana of infectious diseases that are transmitted by contact with arthropods or vertebrate animals. Effective control of these diseases requires interdisciplinary collaboration between human, animal, and environmental health professionals (a One Health approach). The role of public health agencies in the surveillance, prevention, and control of these diseases will be emphasized.

Hot Topics
The Fulbright Experience: ... an inside glance

J.D. Mendez, Indiana University-Purdue University Columbus

The Fulbright Program is a cultural exchange program that allows faculty and students to teach and do research in countries all over the world. In this informal presentation, Dr. J.D. Mendez will discuss the program and his experience as a Fulbright Scholar in Estonia. The focus will be on the basics such as how to look up awards but also some common misconceptions. For example, all of the different Fulbright programs are selective but many if not most participants are not from R1 research institutions. Student programs also exist that allow them to work on an independent research program or teach English in a variety of settings. Additionally, working in English is
fine for most countries although it doesn’t hurt to try and learn a little of the local language. Come with any questions you may have about this interesting program.

**Zoology**

Presentation

**Influence of Supplementation with Alpha Lipoic Acid on Nuclear Reduced Glutathione Levels in Kidney Cortex and Medulla from Young Rats**

**Marianna Zamlauski-Tucker** and Bingwei Ye, Department of Biology, Ball State University

Antioxidant supplementation is thought to be beneficial since it increases the level of reduced glutathione (GSH) inside cells. 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 the antioxidant alpha lipoic acid on GSH levels in the nucleus of kidney cells from young rats. Young female Lewis rats (i.e., 3 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. These findings suggest that nuclei in kidneys from young rats do respond to exogenous dietary supplementation with the antioxidant alpha lipoic acid with an increase in GSH levels.

Poster

**A Preliminary Analysis of Integumentary Features in Terrestrial Stage Northern Two-Lined Salamanders, Eurycea bislineata (Amphibia: Plethodontidae)**

**Katie L. Rust**, Mikia A. Davis, Breanna C. Davis, Kevin M. Gribbins, Department of Biology, University of Indianapolis, Indianapolis, IN 46227, USA

Studies conducted on the integument of Plethodontidae have shown that the transition from water to land results in morphological changes that permit efficient respiration via the skin. This study focuses on comparing the skin glands on the dorsal and ventral surfaces of the Northern Two-Lined Salamander, *Eurycea bislineata*. During the evolution in lungless salamanders, adaptations developed permitting efficient gas exchange across integumentary tissues. In order for gas exchange to occur, water and
surface tension control must be present at the respiratory membrane. In many amniotic vertebrates, the lungs contain pulmonary surfactant, a lipoprotein that aids in gas exchange by reducing surface tension. In amphibians, not much is known about the maintenance of surface tension when lungs are absent and the skin becomes the main regulator of external respiration. Lewis et al. conducted a preliminary investigation in Desmognathus (lungless salamander in Plethodontidae) on their skin and noted the expression of SFTPC-like, a novel surfactant protein gene associated with sites of extrapulmonary respiration. The purpose of this preliminary study is to compare the glands on the ventral and dorsal surfaces and to investigate the likelihood SFTPC-like expression ultrastructurally in Eurycea bislineata, another genus of the family Plethodontidae. We hypothesize that lamellar bodies will be found in areas where surfactant like protein may be expressed and the dorsal surfaces of adults will contain more mixed glands, which produce these bodies, than those of larva.

Poster

**Preliminary Ultrastructural analysis of spermiogenesis in Western Mexico Whiptail Lizard, Aspidoscelis costatus**

*Jaime Rhodes, Mikia A. Davis, Katie L. Rust, Kevin M. Gribbins, Department of Biology, University of Indianapolis, Indianapolis, IN 46227, USA*

To date, there are multiple studies detailing the ultrastructure of spermatozoa within the genus Aspidoscelis and within the family Teiidae. However, there are no complete data on the process of spermiogenesis within a species of Whiptail Lizard or within Ground Lizards. The purpose of this investigation is to provide the first detailed study on the entire ultrastructural process of spermiogenesis within Aspidocelis costatus. These results can then be compared to the spermatozoa data that is available for Teiidae. Testicular tissues of A. costatus were fixed, dehydrated, embedded in epon, sectioned with an ultramicrotome, and viewed under TEM. A. costatus spermatids were found to be relatively conserved to that of other squamates. This preliminary data on A. costatus spermatids will provide new species and familial characteristics that can be compared to that of the previously studied saurian taxa. The features of the spermatids that will be concentrated on in this study are: changes to the acrosome, midpiece mitochondrial morphology, the morphology of the nucleus and flagellum, and the progression of the manchete during the process of spermatid development. Recently, spermatid ultrastructural data has been suggested as a tool that could be utilized in phylogenetic analysis; however, to date too few species have successfully been studied. Thus, the new data set provided by the present study will add to the needed ultrastructural information known for lizards and provide another species of squamate that could increase our understanding of the evolutionary relationships of Ground Lizards to that of other squamates.
Analysis of the Integumentary Histology of the Cave Salamander, *Eurycea lucifuga* (Amphibia: Plethodontidae)

**Mikia A. Davis**, Katie L. Rust, Breanna C. Davis, Kevin M. Gribbins
Department of Biology, University of Indianapolis, Indianapolis, IN 46227, USA

Studies conducted on the integument of adult terrestrial Plethodontid salamanders show morphological changes that reflect the environment as they transition from water to land. This study focuses on comparing the features of integumentary glands along the dorsal and ventral surfaces of the Spotted-tail or Cave Salamander, *Eurycea lucifuga*, to those found within the Northern Two-Lined Salamander, *Eurycea bislineata*, and the Zigzag Salamander, *Plethodon dorsalis*. Although these plethodontids are found in different genera, we hypothesize that the glandular skin will be similar histologically, even if the skin gland dispersal varies between the dorsal and ventral axes. We will also be comparing the integumentary glands found in the buccopharyngeal regions between *E. lucifuga* and *E. bislineata*. The purpose of this comparative study is to investigate the features of integumentary glands on the ventral, dorsal, and buccopharyngeal surfaces and to study the potential differences and histological diversity in glandular composition among these 3 species of salamanders within the family Plethodontidae. These initial results can then be used in unison with future research that will detail the ultrastructure of these glands in a family comparison to analyze potential similarities and differences of integumentary features and functions of skin glands among the Plethodontid species studied to date.

A Familial Comparative Analysis of Integumentary Features in Terrestrial Stage Zigzag Salamanders, *Plethodon dorsalis* (Amphibia: Plethodontidae)

**Breanna C. Davis**, Katie L. Rust, Mikia A. Davis, Kevin M. Gribbins
University of Indianapolis

Studies conducted on the integument of Plethodontidae have shown mucous, granular, and modified composite multicellular glands in the skin. In amphibians, data on the morphology of skin glands is limited to a handful of studies between aquatic and terrestrial salamanders or their larvae. This study focuses on comparing the integumentary glands on the ventral and dorsal surfaces of the Zigzag salamander, *Plethodon dorsalis*, to those found on the Northern Two-Lined Salamander, *Eurycea bislineata*, and the Spotted-tail Salamander, *Eurycea lucifuga*. The purpose of this comparative study is to investigate the histological diversity of integumentary glands found on the ventral and dorsal surfaces of salamanders, as all three species in this study are found in the same family,
Plethodontidae. We hypothesize that the plethodontid salamanders will have similar morphology to that of other lungless salamanders, though distribution of these gland types may be different between species. The preliminary results of this study will then be used to do a family comparison to analyze potential similarities and differences in integumentary features among terrestrial adult. In later studies, we would like to look at zigzag embryos as they develop within the egg to see if there are any developmental changes to skin glands that we can observe.

Presentation

How do you make a messy coevolutionary witches brew? One-part snake, one-part newt, and one-part caddisfly

Brian G. Gall - Hanover College
Yiyuan Li - University of Notre Dame
Michael Pfender - University of Notre Dame
Edmund D. Brodie III. - University of Virginia
Edmund D. Brodie, Jr. - Utah State University

Coevolution occurs when two closely interacting species exert reciprocal selection upon each other. The best documented example of coevolution involves super-toxic newts (Taricha sp.) and toxin-resistant snakes (Thamnophis sp.) from the western United States. Here we show that a third species group, caddisfly larvae, are likely also involved in the selection pressure being exerted on newt toxicity. Both behavioral and genetic evidence indicates major evolutionary changes leading to Tetrodotoxin resistance in multiple species of caddisfly. These results suggest that our general model of coevolution is likely overly simplified and that multiple closely interacting species may form a web of selection acting at the phenotypic interface of coevolution.

Poster

Orientation behavior after displacement over water by semi-aquatic long-jawed orb weavers (Tetragnatha elongata)

Jakeb N. Watts - Hanover College
Courtney J. Cox - Hanover College
Tarah L. Lancaster - Hanover College
Theresa E. Wrynn - Hanover College
Brian G. Gall - Hanover College

Many organisms are capable of complex orientation through their environment using a diversity of cues including physical features, polarized light, or Earth's magnetic field. We conducted a study to determine if long-jawed orb weavers (Tetragnatha elongata) exhibit non-random orientation behavior after being displaced over water.
Spiders were displaced between 40 and 500 cm from shore and their direction of travel was recorded by an observer. We found that spiders were capable of quickly orienting directly toward shore regardless of placement distance. These results suggest that these spiders are capable of complex behavioral adjustments using unknown features of the environment, likely to minimize exposure on the surface of the water.

Presentation

**Dragonfly nymphs (Anax juniq) as predators on early developmental stages of the Eastern newt (Notophthalmus viridescens)**

**Theresa E. Wrynn** - Hanover College
Brian G. Gall - Hanover College

We observed the predator-prey relationship of larval newts (Notophthalmus viridescens) when exposed to predatory dragonfly (Anax juniq). Literature about the interactions between larval newts and their predators is sparse. The hopes of this project is to shine more light on this topic. Larval newts were exposed to different stimulus including a control (water), food (bloodworms), and predatory (dragonfly). Behavior was recorded during the pre- and post-stimulus periods for the kairomone trials. A significant difference was found for time spent moving for larval newts across the different treatments. Newt larvae decreased movement significantly when exposed to predatory stimulus compared to control and food stimulus. We also observed the metamorphosis and survival rates of larval newts when exposed to predatory dragonflies. Four control tanks and four treatment tanks containing a predatory dragonfly were constructed. Five larval newts were placed in each tank and were checked daily to see which newts successfully metamorphed. We found that newt larvae had a better chance of surviving and transforming in the control tanks compared to treatment tanks.

Presentation

**The Indestructible Insect: Predator-Prey Interactions Between Velvet Ants (Dasymutilla occidentalis) and Toads (Anaxyrus sp.)**

**Ciara J. Mergler** - Hanover College
Brian G. Gall - Hanover College

Velvet ants (Dasymutilla occidentalis) are a group of parasitic wasps that are well-known for a suite of defensive traits including bright coloration, a thick exoskeleton, and a painful sting. Previous experiments have demonstrated that while virtually all potential predators avoid velvet ants, toads (family Bufonidae) may be able to prey upon them. We conducted a series of trials to determine whether American (Anaxyrus americanus) and Fowler’s toads (Anaxyrus fowleri) prey upon velvet ants. Toad
responses toward velvet ants were variable, ranging between complete avoidance, consumption and regurgitation, and consumption and retention. In total, 11 out of 22 trials resulted in the toads eating a velvet ant. Of these, velvet ants were either regurgitated unharmed (N = 8) or retained (N = 3). There were no significant differences in responses toward velvet ants between American and Fowler’s toads. Our results indicate that toads have the capacity to consume and retain female velvet ants. However, female velvet ants demonstrate the ability to endure upwards of twenty minutes inside the stomach of a toad and survive.

Presentation

**Spiders at the Indiana Dunes National Park: New state records and undescribed species**

Marc A. Milne, University of Indianapolis

The newly-minted Indiana Dunes National Park (IDNP) is a 15-mile national park on the southern shore of Lake Michigan in northern Indiana. The park contains rare habitats within Indiana, including sand dunes, marshland, and jack pine forests. Parks such as IDNP have been decreed as being worthy of preservation, but their living contents - including spiders - are often understudied. In order to expand our knowledge of spiders at IDNP, my students and I collected spiders at the park at eight time points over the past three years. I then identified these spiders to species using various taxonomic keys. My students and I collected over 750 spiders of 101 species within 80 genera within 18 families. Ten of these species were new distribution records for the state of Indiana. One species is undescribed within the genus *Ceraticelus* in the family Linyphiidae. This study further emphasizes the need to taxonomically examine poorly studied groups in highly-visible, heavily-trafficked parks. If not properly studied, we may miss the most interesting parts of these unique habitats.

Workshop

**How to identify some common Indiana spiders**

Marc A. Milne and Emily Stern, University of Indianapolis

Spiders are ubiquitous within almost every habitat on almost every continent on earth. However, because they are arthropods and not insects, their study within the scientific community is somewhat of an enigma. Due to their unique morphology among arthropods, their identification follows different patterns than what most entomologists are used to. In this 2-hour workshop, we will teach participants about the unique anatomical structures of spiders, how to identify spiders to some common Indiana families, and how to identify some common Indiana spiders to genus. The
visualization and identification of spiders will be conducted by the workshop hosts through the display of the view of a single powerful dissecting scope at the front of the room. Participants will be given (to keep) the latest version of *Spiders of North America: An Identification Manual* (2nd ed.) and will use this guide throughout the workshop.

**Poster**

**Alternative Species for Aquaculture: Sea Urchins**

**Lindee Mason**, Asif Mortuza and Ahmed Mustafa
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Sea urchins have been used as delicate foods for many years in different regions in the world. Recently, they are also gaining importance in pharmaceuticals and in research labs as test animal models. Hence, people are trying to harvest them from the waters and depleting their population from the nature. Therefore, it is time for them to be aquacultured but due to husbandry methods in aquaculture such as, handling, salinity, and crowding, sea urchins are stressed. Since, stress is causing losses to the aquaculture, it is important to determine the stress responses and identify the way of stress modulation without doing any harm to the animals as well as to the environment. To investigate the stress responses in sea urchin, physiological and immunological responses were studied in purple sea urchins, reared in recirculatory system in the lab. Sea urchins were reared in aquaculture system in three groups – sea urchins fed sponges, sea urchins fed kelps, and sea urchins fed romaine lettuce. The experiment ran for three weeks and the animals were sampled at regular intervals. Total body weight, gonad weight, gonado-somatic index, packed cell volume, coelomic fluid protein, total and differential cell counts, and coelomocyte phagocytic capacity were observed to measure the stress and immune response. Data are being analyzed and will be presented at the meeting.

**Presentation**

**Phylogeographic structure of the Least Brook Lamprey Lamproptera aepyptera (Cyclostomata: Petromyzontidae) and the origin(s) of its non-parasitic life history**

**Rex Meade Strange**, Department of Biology, University of Southern Indiana

The taxonomic diversity of lampreys is marked by a parasitic/non-parasitic dichotomy of life history strategies in which most parasitic species are paired with a non-parasitic derivative. Parasitic lampreys are often anadromous and broadly distributed across major river systems. In contrast, non-parasitic lampreys are relatively non-vagile and typically undergo metamorphosis, spawn, and die in their natal stream. The Least
Brook Lamprey (*Lampetra aepyptera*) is a broadly distributed non-parasitic species of the southeastern United States. As the parasitic form from which it was presumably derived is extinct, it is possible that disjunct populations of *L. aepyptera* represent independent shifts in life history. I examined the phylogeographic structure of *L. aepyptera* for evidence whether the transition from a parasitic to non-parasitic life history occurred once (monophyletic hypothesis) or multiple times (polyphyletic hypothesis). Specimens were collected from across the species’ distribution and examined for sequence variation at three protein-coding regions of the mitochondrial genome. Phylogenetic analyses consistently identified three clades within *L. aepyptera*: (1) an Eastern Clade that includes samples from the East Coast, Mobile Basin, Tennessee River system, and southern portions of the Cumberland Plateau; (2) a Western Clade that consists of samples from the Ohio River Basin and Coastal Plain tributaries of the Mississippi River; and (3) a Highlands Clade that includes samples from the upper Cumberland River system and the Ozarks of Arkansas and Missouri. The Highlands Clade was consistently identified as the basal group within the species and may represent the first transition from parasitic to a non-parasitic life history. Given that contemporary non-parasitic populations appear to be incapable of long-distance dispersal, the phylogeographic structure of *L. aepyptera* supports the hypothesis that the adoption of the non-parasitic life history originated two or more times.

Presentation

**Cave spiders of Appalachia: A phylogenetic study of Liocranoides (Araneae: Zoropsidae)**

E. Hart, M. A. Milne, K. Zigler, N. C. Mann, M. E. Yancey, M. L. Niemiller, University of Indianapolis

Caves often act like islands in that organisms within them live apart from the epigean fauna and adapt to a novel environment not experienced by non-cave organisms. The evolutionary path can be different in each cave, depending on that cave’s specific microenvironmental conditions (humidity, temperature, etc.) Spiders that begin to live within cave systems may also speciate due to these unique environmental selection pressures. Speciation may change spider morphology and will certainly change spider genetics. *Liocranoides* is a genus of spiders (Araneae: Zoropsidae) that commonly live in caves throughout Appalachia. In order to determine the relationship among *Liocranoides* throughout the region, we examined morphological differences among seven specimens from seven separate caves in either Tennessee, Georgia, or Alabama. We also used molecular methods to compare the evolution of four different nucleic genes in sixteen specimens from sixteen different caves in the same region. We then assembled a phylogenetic tree and used it to hypothesize the taxonomic
relationship among these species. This experiment allows for a better understanding of spider evolution within cave environments throughout Appalachia.

Poster
A Comparison of the Axial Skeletons and Pectoral Girdles of Chiroptera and Aves

Laura Scheid, and Stephen Nawrocki, University of Indianapolis

Powered flight has been achieved by only three groups of animals throughout Earth’s history: birds, pterosaurs, and bats. The bony and soft tissue structures of the forelimb are often highlighted in discussions detailing adaptations for flight in these three groups. However, the bony structures of the pectoral girdle and axial skeleton are equally important for successful, sustained flight. The special morphology of the pectoral girdle and axial skeleton allows the animal to save muscle energy during flight, lighten the body, and provide stability against the strong forces generated by the flight muscles. The current study compares and catalogs variation in the morphology of these specific regions in a sample of bird and bat skeletons. Both groups display some convergent similarities that are adaptive to flight, including: lightweight, hollow bones; fusion of bones to minimize the need for heavy muscular stabilization and to add stability across regions; development of a keel on the sternum to anchor enlarged forelimb adductor muscles; buttressing of the rib cage; and increased robusticity of the pectoral girdle with strengthened attachments to the axial skeleton. Differences between Aves and Chiroptera include: one sternal element in birds and three sternal elements in bats; avian ribs bear uncinate processes while chiropteran ribs are wide and flat with no extra processes; bats have separate right and left clavicles while birds have a fused furculum; birds have right and left coracoid bones while bats have fused scapular coracoid processes; and bird pelvic girdles consist of one element (synsacrum) while bat pelvic girdles consist of multiple free elements. These observations can help to distinguish between species in situations where fragmentary skeletons are recovered in the field. They also help to shed light on how these two groups solved the problem of flight in different ways.