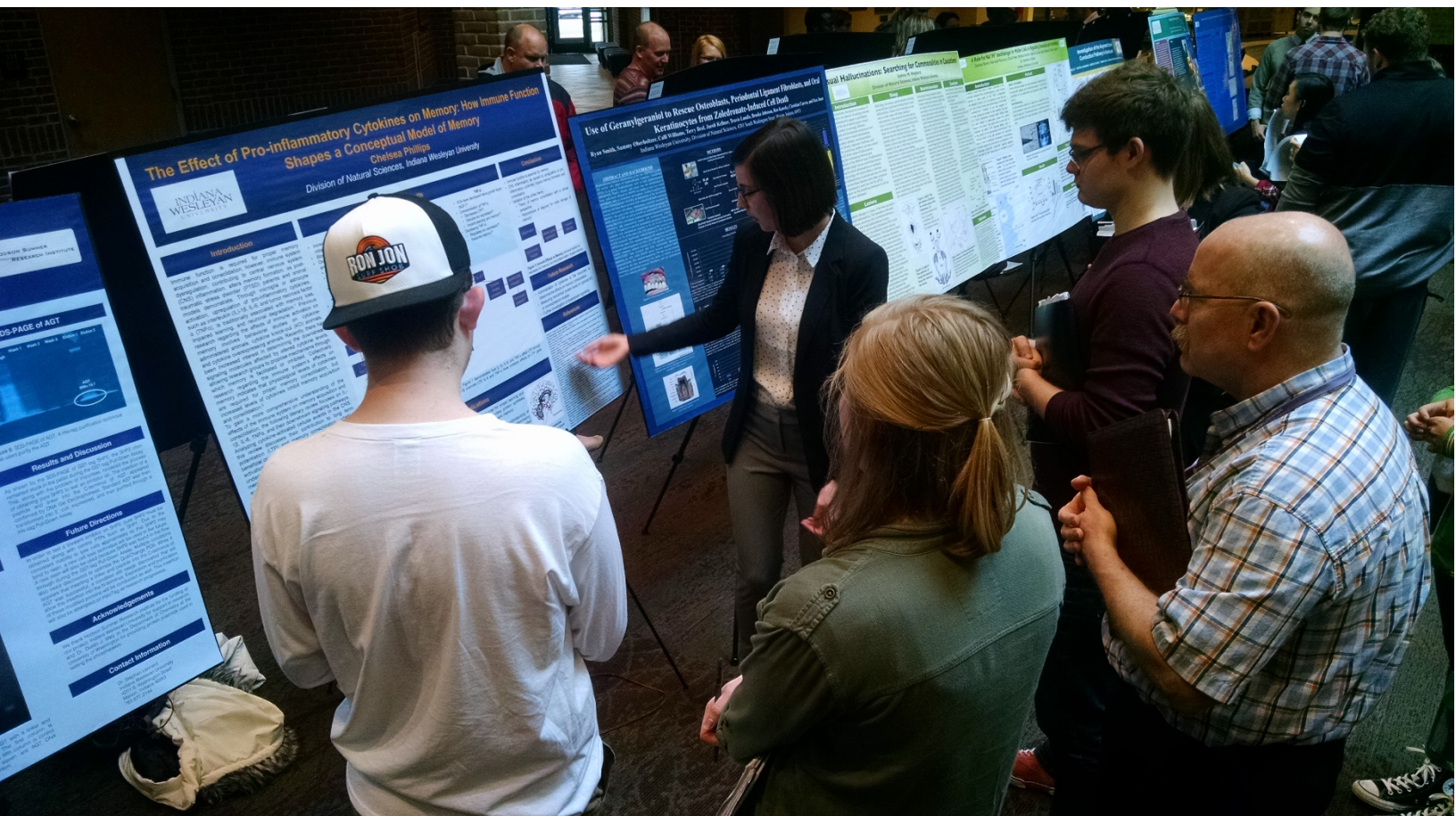


Celebration of SCHOLARSHIP



THURSDAY, APRIL 16, 2020

Poster Presentation Program

IWU'S ANNUAL UNDERGRADUATE RESEARCH CONFERENCE

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INDIANA WESLEYAN UNIVERSITY

This year's Celebration of Scholarship Poster Presentations can be engaged through two formats:

First, explore the digital repository located on Facebook where you will find copies of the research posters and have space in the comments section to post questions. The students' research posters will be available online by 2:00 pm, Wednesday April 15th.

DIGITAL REPOSITORY LINK:

<https://www.facebook.com/IWUCelebrationOfScholarship/>

Second, there will be a Zoom session for real-time conversations with the student authors of the research posters on Thursday, April 16th between 11am and 1pm. This Zoom session will have individual breakout rooms wherein you can login and ask student researchers questions about their poster presentations.

ZOOM LINK:

<https://indwes.zoom.us/j/927838727?pwd=SlFEdlNpYmFtOXY0QkFmWmRHRk5Xdz09>

Meeting ID: 927 838 727

Password: 091067

POSTER ABSTRACTS

(Listed Alphabetically by Student's First Name)

Retinal Müller glia activated by ATP cause an extracellular acidification mediated by intracellular acidification, calmodulin, and protein kinase

Adam Schantz, *Natural Sciences*

Numerous studies have shown pH regulation in the outer retina is a potent regulator of synaptic transmission. These pH dynamics have been postulated to underpin important physiological events like feedback inhibition. Previously, we have extended this regulation to include retinal glia acidifying the extracellular space when activated by ATP. We have shown extracellular ATP applied to Müller cells activates P₂Y receptors leading directly to an IP₃ mediated Ca²⁺ rise that results in a Na⁺/H⁺ exchange-dependent extracellular acidification. Here we measure intracellular pH detecting fluorescence emitted from BCECF, a pH sensitive dye, loaded in Müller cells isolated from salamander retina. These measurements demonstrated that exogenous application of ATP increases intracellular H⁺. Additionally, we show the extracellular acidification around Müller glia is mediated through calmodulin and protein kinase c pathways. H⁺ flux was measured from isolated salamander Müller cells using H⁺ sensitive self-referencing microelectrodes. Self-referencing recordings report that bath application of ATP resulted in a large extracellular acidification that was attenuated by the calmodulin antagonists, W-7, chlorpromazine, trifluoperazine. The protein kinase c antagonist, chelerythrine, also dramatically decreased the ATP-induced extracellular acidification. When used in combination the calmodulin and PKC antagonists completely abolished the ATP-induced acidification. Collectively, this work sheds additional light on non-neuronal signaling pathway through which ATP shapes extracellular pH. We hypothesize this regulation by retinal Müller cells is important in shaping visual signaling, and characterization of this glial regulatory pathway lays a foundation for understanding how glia may impact neuronal signaling throughout the nervous system.

† Should Preoperative Fascia Iliaca Block Be Used for Hip Arthroscopic Labral Repair and Femoroacetabular Impingement Treatment? A Prospective Single Blinded Randomized Study

Alison Henry, *Natural Sciences*

Purpose: To evaluate the analgesic effect of preoperative fascia iliaca block on postoperative morphine equivalent dose, pain level, and patient satisfaction for patients electing to undergo primary hip arthroscopic labral repair with osteo- chondroplasty. **Methods:** This prospective study included 60 patients (fascia iliaca block group: n 1/4 27; control group: n 1/4 33) undergoing elective arthroscopic hip surgery by a single board-certified orthopedic surgeon, fellowship trained in hip arthroscopy. Participants for the study included patients older than 10 years of age and younger than 85 years of age, American Society of Anesthesiologists classifications I to III, diagnosed with symptomatic femoroacetabular impingement, and/or hip labral tear, and/or cartilage damage, and electing to undergo arthroscopic hip surgery. Patients were randomized by surgical date to receive preoperative fascia iliaca block or control (no fascia iliaca block). Preoperative fascia iliaca block was administered by 1 of 4 board certified anesthesiologists using identical anesthetic (35-40 mL ropivacaine 0.35%). Postoperative morphine equivalent dose, self-reported pain level (visual analog scale) and patient satisfaction were measure postoperatively. **Results:** There were no significant differences between the control group and the fascia iliaca block group in sex, age, height, weight, or body mass index. There was a significant difference between the 2 groups in distribution of

† Denotes a project also included on the oral presentation schedule

American Society of Anesthesiologists classification (p 1/4 .031). There were no significant differences in postoperative morphine equivalent dose for patients receiving fascia iliaca block compared with the control group. There were no significant differences in self-reported visual analog scale pain and patient satisfaction between the 2 groups at any of the measured time points following surgery. Conclusions: Based on the results of this study, routine preoperative fascia iliaca block for elective hip arthroscopic labral repair and treatment of femoroacetabular impingement is not recommended. Level of Evidence: Level II, prospective single blinded randomized study.

Antimicrobial Use and Extraction of Essential Oils

Alyssa Nugent, *Natural Sciences*

Essential oils are increasing in popularity as an alternative method for treating various ailments from infections and illness to depression and anxiety, especially in cases of antibiotic resistance. Although they are known to have antimicrobial properties, the extent of these properties has not been well researched. It is not well known what the exact effects essential oils have on eukaryotic cells specifically, which is equally important as understanding antimicrobial properties of the oils since many are applied directly to the skin. Essential oils are obtained from plant matter most commonly through pressing or distillation, both of which are processes that could be replicated in developing countries where antibiotics are not readily available. In this study, methods were developed to collect essential oils from basil (*Ocimum basilicum*) and lavender (*Lavandula angustifolia*). These plants were selected due to their ability to grow in multiple climates and their ease of growth. Focusing on distillation, the oils from these plants were successfully extracted in three different ways. The antimicrobial properties of the essential oils were then analyzed through Kirby-Bauer Disk Diffusion, Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) against ten pathogenic and nonpathogenic microorganisms.

Role of Calmodulin and PKC in ATP-Induced Acidification of Müller Cells

Alyssa Powell, *Natural Sciences*

In recent years, an increasing number of studies have analyzed the relationship between glial cells and synaptic transmission in different parts of the central nervous system. Within visual signaling of the retina, an extension of the central nervous system, extracellular H⁺ dynamics are known to be potent inhibitors of the release of neurotransmitters. This study further characterizes the pathway through which glial cells, when stimulated by ATP, release protons into their extracellular environment. This study specifically implicates two second messengers in this ATP initiated cascade, calmodulin (CaM) and protein kinase C (PKC). CaM and PKC antagonists were added to tiger salamander retinal Müller cells and the extracellular acidifications were recorded using self-referencing. Results showed that CaM and PKC both decreased the effect of ATP, and when used together they even more potently blocked the ATP effect. Collectively, the findings reported lend further support to a cell signaling pathway through which ATP initiates a potent acidification of the extracellular space and suggests glial cells may play an important role in gain control of neuronal signaling in the retina.

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The Positive and Negative Effects CBD has on the Human Body

Ambermarie Worley, *Natural Sciences*

Cannabidiol, better known as CBD, has become a frequently used product across the nation and beyond. Cannabidiol is a cannabinoid found in marijuana, *Cannabis sativa*. In the most recent years, it has been used for recreational use, however in the past it has been used to treat pain, epilepsy, and anxiety. Several studies have been done to understand the neurological effects of this drug, including its effects on schizophrenia and epileptic patients. A narrative review was put together to summarize some of the current research that has been done, regarding the neurological and psychiatry effects of cannabidiol, as well as how it effects other systems within the body. Studies showed the CBD could lead to a more positive outcome in schizophrenic patients experiencing psychosis or drug misuse. Studies observed the effects of cannabidiol on the male reproductive system and have correlated this variety of drug misuse to a reduction in the testis size in mammals. Knowledge of every effect of this drug on the human body is not fully known, but research is being done to further the understanding of the positive and negative affects of cannabidiol.

What are the Major Factors of Cardiovascular Disease?

Brianna Bates, *Natural Sciences*

For Celebration of Scholarship, I chose to research the various factors that can lead or effect cardiovascular disease. I have not previously researched with any professors here at Indiana Wesleyan. Therefore, I am doing library research. I have found various articles that provide evidence for increasing risk factors of cardiovascular disease. This topic will provide the audience with data regarding sodium intake, blood pressure, dairy intake, and other topics that have an important relationship with cardiovascular health I believe everyone should know about. I don't think that people realize how much the food we put into our bodies effects the overall health of our bodies which is why I am presenting on this important informative topic of cardiovascular risk factors. I want to help others realize the things that they should avoid in order to live the healthiest and happiest lives they can.

Cultivation and Characterization of Icebound Microorganisms from the South Pole

Brittney Alexander and Janessa George, *Natural Sciences*

Little is known about microbial ecosystems of inland Antarctica, if indeed such ecosystems exist. Although considerable research has been carried out on microorganisms indigenous to the warmer coastal regions of Antarctica, only one species, a strain of the bacterial genus *Pseudomonas*, has ever emerged from culture-based studies of South Pole ice. Molecular studies have revealed that a diversity of bacteria exists in polar ice samples, but whether these organisms compose an active ecosystem or are simply a random assortment of microbes imported by air currents is unknown. To investigate this question, we began the process of cultivating microorganisms from ice samples collected by Dr. Michael Madigan (Southern Illinois University Carbondale). Our team has been able to successfully isolate several strains of bacteria, as well as a strain of cold-adapted yeast. Ensuing characterization of the isolates has included experiments to determine optimal growth temperature, salinity tolerance, and carbon source utilization. We have also begun phylogenetic analyses of each isolate and include preliminary data here. This information, along with further experimentation, will expand our understanding of the diversity, metabolic activities, and ecological contributions of icebound microorganisms.

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Muller Cell ATP-induced acidification is shaped by extracellular pH and carbonic anhydrase activity

Brock Goeglein, *Natural Sciences*

The study of glial cells as regulators of synaptic transmission is a budding field in Neuroscience. An especially potent and underappreciated mechanism of modulating synaptic transmission involves small alterations of extracellular levels of H⁺. Measurements of H⁺ flux from isolated Muller glial cells were made using a self-referencing approach with H⁺ selective microelectrodes. The present study builds upon a recently characterized pathway by which exogenous ATP causes a calcium-dependent extracellular acidification through activation of a GQ coupled P₂Y receptor. This acidification is directly dependent on the H⁺ gradient. Changing the H⁺ gradient by making the extracellular pH more acidic (6.8) or alkaline (8.2) attenuated or potentiated, respectively, the ATP-induced acidification. Surprisingly, in the absence of a CO₂ – HCO₃ buffer, the ATP-induced acidification was suppressed by carbonic anhydrase inhibition. This suggests H⁺ extrusion may have a close relationship with carbonic anhydrase independent of HCO₃. The present study sheds further light on a mechanism by which glial cells potentially regulate extracellular pH and by extension shape neuronal signaling.

Analysis of the complete genome of the alkaliphilic and phototrophic Firmicute *Heliorestis convoluta* strain HHT

Emma Dewey, *Natural Sciences*

Despite significant interest and past work to elucidate the phylogeny and photochemistry of species of the Heliobacteriaceae, genomic analyses of heliobacteria to date have been limited to just one published genome, that of the thermophilic species *Heliobacterium (Hbt.) modesticaldum* str. Ice1T. Here we present an analysis of the complete genome of a second heliobacterium, *Heliorestis (Hrs.) convoluta* str. HHT, an alkaliphilic, mesophilic, and morphologically distinct heliobacterium isolated from an Egyptian soda lake. The genome of *Hrs. convoluta* is a single circular chromosome of 3.22 Mb with a GC content of 43.1% and 3263 protein-encoding genes. In addition to culture-based observations and insights gleaned from the *Hbt. modesticaldum* genome, an analysis of enzyme-encoding genes from key metabolic pathways supports an obligately photoheterotrophic lifestyle for *Hrs. convoluta*. A complete set of genes encoding enzymes for propionate and butyrate catabolism and the absence of a gene encoding lactate dehydrogenase distinguishes the carbon metabolism of *Hrs. convoluta* from its close relatives. Comparative analyses of key proteins in *Hrs. convoluta*, including cytochrome *c553* and the Fo alpha subunit of ATP synthase, with those of related species reveal variations in specific amino acid residues that likely contribute to the success of *Hrs. convoluta* in its highly alkaline environment.

Testing the Efficacy of the SongSleuth App to Record and Identify Birds Based on their Vocalizations

Erik Voeller, *Natural Sciences*

This research project, overseen by Dr. Stephen Conrad, began in Fall 2019 through Spring 2020. Over the course of this academic year, I have visited nearby parks to record bird calls. These recordings are then analyzed by the app, SongSleuth. The app then tries to identify the bird whose call was recorded. In totality, there were 275 recordings, 49 of which were correct identifications, resulting in a 17.8% accuracy. During the fall, I collected 149 recordings and only 16 of which were correct. After getting the microphone, the app was able to identify vocalizations with more accuracy. A total of 126 recordings were collected; 33 of which were correct. This gives us 26.2% accuracy. This is due to the mic being able to record at superior

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quality and its ability to pick up bird calls at a greater distance. We found that the app is not particularly useful in correctly identifying birds by their calls, but it does improve with enhanced quality of audio.

New Methods in Pharmaceutical Drug Research and Development

Hannah Callaway, *Natural Sciences*

Drug development in the pharmaceutical industry has changed dramatically in recent years as technology advances and data concerning drug metabolism becomes more readily available. However, the process of formulating new medications and making them accessible to patients is not as streamlined as one might imagine. This study briefly overviews and synthesizes the effectiveness of groundbreaking new techniques and the consequential hurdles in drug development at present. All of the techniques addressed herein are found to be effective in some aspect of research or development, with the biggest hurdles that prevent their implementation being government regulations, extreme costs, and public perception.

VPAC₂R-Deficiency In Older Mice May Preserve Beta Cell Function

Hannah Hartzler, *Natural Sciences*

Vasoactive Intestinal Peptide Receptor Types 1&2 (VPAC₁R and VPAC₂R) are two G protein-coupled receptors that are linked together by two peptides, vasoactive intestinal polypeptide (VIP) and pituitary adenylate cyclase activating peptide (PACAP). To study the function of the VPAC₂R, VPAC₂R-deficient mice were created. Previous studies of these VPAC₂R-deficient mice have revealed that these mice have a different circadian rhythm compared to the wild type mouse which includes an advance in the time in which they feed. Respiratory quotient analysis of these MUT mice show that they preferentially burn fat. Since signaling through the VPAC₂R causes augmented insulin release in the presence of glucose, we wanted to investigate what effect the loss of this receptor has on glucose-stimulated insulin secretion. To do this, Islets of Langerhans (islets) were isolated from WT and MUT mice, and used in glucose-stimulated insulin secretion (GSIS) assays to determine the amount of insulin released when the islets were challenged with different glucose concentrations. Insulin levels, determined by enzyme-linked immunosorbent assays (ELISA), of the media during the different challenges generally increased as the amount of glucose in the media increased. The data is showing that the MUT islets, regardless of age, are responding to the increasing glucose levels and the older WT islets seems to be losing that ability.

Animal Behavior in Zoos

Holly Riley, *Behavioral Sciences*

There are zoos across the world and there has been a growing recognition that consistent behavioral differences between animals, and in some cases between populations of animals exist. Individual animals have different personalities and their personalities play a role in their interactions. Different interactions such as the interaction between animals and other animals, also animals and the zookeepers. Cognitive tests were studied measuring affective states of the animals. More studies demonstrate a monthly focus on a group of animals and their actions during the day. Normal and abnormal actions throughout the day. Focusing on the psychology of the animals including five descriptive traits; shyness-boldness, sociability, aggressiveness, exploration-avoidance, and activeness. One of the great advantages that zoos and aquariums have in terms of ability to conduct research into animal personality is that they know the animals as individuals. In this case animals studied in a “zoo” environment is helpful in learning their daily behaviors with each other, with human beings.

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† Somatic Embryogenesis and Regeneration of *Moringa oleifera*

Hope Blanchard, *Natural Sciences*

Moringa oleifera is a nutritious perennial native to parts of Africa and Asia that is easily propagated by seed and grafting. It possesses antibacterial and anticancer properties (Saini, Arya, & Singh, 2013). *Moringa* seeds allow for water purification with solar distillation. Previous studies have accomplished micro-propagation (Saini, Shetty, Giridhar, & Ravishankar, 2012). We present a unique form of callus induction and somatic embryogenesis.

Regeneration using tissue culture allows for mass production of plant tissue without having a seed and offers a time efficient alternative to traditional propagation. Attempts at calli formation from leaves and stems has proven successful, however, regeneration occurred through embryos. Induction of calli resulted from exposing plant material to media containing 2,4-Dichlorophenoxyacetic acid (2,4-D). Preliminary results show regeneration of embryos through the transfer of calli to media containing indole-3-acetic acid and kinetin. Calli were incubated in the dark at 25 °C for two to six weeks. The regenerated plants were subsequently exposed to 16-h light incubation at 25 °C for several weeks.

† Zoledronate-Induced Viability Loss: Determination of Rescue Agent Dosages in Human Oral Keratinocytes

Jacob Kirby, Tyler Cairncross, *Natural Sciences*

Medication-related osteonecrosis of the jaw (MRONJ) is characterized by an 8-week period of exposed necrotic bone and a lack of soft tissue regeneration in the mouth. This condition can be brought on by treatment with nitrogen-containing bisphosphonates as well as other classes of drugs. Zoledronate (ZOL) is a nitrogen-containing bisphosphonate (N-BP) used to treat post-menopausal osteoporosis and cancers that have metastasized to bone. This study used an MTT cell viability assay in normal human oral keratinocytes (NHOKs) to produce dose-response curves at 48 and 72 hours for ZOL as well as three candidate rescue compounds. Geranylgeraniol (GGOH), known to rescue NHOKs from ZOL-induced death effects in vitro, was tested as well as phosphonoformic acid (PFA), and sodium citrate. The two latter compounds were chosen based on characteristics that are thought to counteract the effects of ZOL. NHOKs were seeded into uncoated 96-well plates for growth to confluence, then treated over a time course of 48 and 72 hours to determine ZOL and rescue agent concentrations for use in future viability rescue assays. At both time points, 50 and 100 microM ZOL caused significant viability loss. The maximal doses of the rescue agents that did not cause significant viability loss at 48 hours were 10 microM GGOH, 0.5 milliM PFA, and 2 milliM sodium citrate. In contrast, at 72 hours the following doses significantly reduced viability (each $p < 0.01$ compared to the respective vehicle control): 1 microM GGOH (85% viability), 0.025 milliM PFA (83%), and 0.2 milliM sodium citrate (85%). These dose levels inform successive experiments to determine the relative efficacy of each agent in rescuing from ZOL-induced death in NHOKs.

Utilization of Fluorescence Imaging in Determining Intracellular pH Changes of Salamander Muller Cells

Jared B. Kilmer; Anna G. Phillips, *Natural Sciences*

Our lab is interested in how Muller cells shape neuronal signaling in the retina by regulating extracellular acidity. Recently we have reported that bath application of ATP results in an extracellular acidification

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around Muller cells isolated from tiger salamander retinas. Here we will describe the use of fluorescence imaging methods with BCECF-AM dye, used to correlate intracellular changes in pH with extracellular changes in pH. Collectively, we aim to use these measurements to identify the intracellular cascade mediating the ATP-dependent extracellular acidification, which may play an important role in reducing synaptic transmission in the retina, in turn reducing the likelihood of tonic glutamate release resulting in excitotoxicity.

Essential Oils', Particularly Lavender, Effects on Antifungal and Antibacterial Properties

Kennedy Rife, *Natural Sciences*

In summarizing research, it can be determined that essential oils, particularly lavender, have the highest antifungal and antibacterial properties. Essential oils were tested in scenarios including different types of fungi as well as different types of bacteria all having similar and varying characteristics. The studies were conducted by the use of freeze drying, impregnating, and simulating the lifespan. It was concluded by all research that essential oils are the most efficient method in withstanding the effect of fungi and bacteria. It was also concluded that from essential oils, lavender proved to have the greatest resistance of fungi and bacteria.

Complete Genome Sequence of the Thermophilic Purple Sulfur Bacterium *Thermochromatium tepidum* strain MCT (DSM 3771T) Compared with that of the Mesophilic *Allochromatium vinosum*

Lynn Stokes, *Natural Sciences*

The complete genome sequence of the thermophilic purple sulfur bacterium *Thermochromatium* (*Tch.*) *tepidum* is presented and contrasted with that of the mesophilic purple sulfur bacterium *Allochromatium* (*Alc.*) *vinosum*. The *Tch. tepidum* genome consists of a single circular chromosome of 2,958,290 base pairs with no plasmids, which is substantially smaller than the genome of *Allochromatium vinosum*. The streamlined genome of *Tch. tepidum* contains *nifHDK* and many other genes encoding a molybdenum nitrogenase but lacks a gene encoding a protein that assembles an Fe-S cluster required to form a functional molybdenum-iron cofactor (FeMo-co), leaving the organism phenotypically Nif⁻. The *Tch. tepidum* genome contains the necessary genes for oxidizing sulfide to sulfate as photosynthetic electron donor but is genetically unequipped to either oxidize thiosulfate as an electron donor or to carry out assimilative sulfate reduction, both of which are hallmarks of *Alc. vinosum*. Also, unlike *Alc. vinosum*, *Tch. tepidum* is obligately phototrophic and unable to grow chemotrophically in darkness by respiration. Several genes present in the *Alc. vinosum* genome but absent from the genome of *Tch. tepidum* likely contribute to the major physiological differences observed between these two species.

Association between TPH₂ Gene and Behavioral Disorders such as ADHD and MDD

Maggie Liechty, *Natural Sciences*

The purpose of this paper is to address how polymorphisms in the TPH₂ gene are associated with major depressive disorder (MDD) and attention deficit hyperactivity disorder (ADHD). Since TPH₂ is known as the rate-limiting enzyme in serotonin production, scientists have tested whether its loss of function has an effect in psychiatric disorders associated with this neurotransmitter. To analyze the relation between TPH₂ and these disorders, research studies used the method of random sampling; one group served as the control, and the other consisted of people who were diagnosed with MDD (based on a minimum Hamilton Depression Rating Scale score) or diagnosed with ADHD (according to the Diagnostic and Statistical

[†] Denotes a project also included on the oral presentation schedule

Manual of Mental Disorders). DNA of each subject was isolated, and multiple different TPH2 polymorphisms were genotyped by DNA sequencing and PCR. Single marker analysis, transmission disequilibrium test analysis, and haplotypes studies evaluated the relationship between TPH2 mutations and MDD or ADHD. Statistical analysis included using Hardy-Weinberg equilibrium tests and chi-square analysis to compare allele and genotype frequencies.

The following results were obtained for MDD in terms of TPH2: there is a significant correlation between the methylation of the TPH2 promoter and feelings of hopelessness, depression, and cognitive impairment in patients with MDD and suicidal tendencies. Additionally, significant differences in the genotypic and allelic distribution of the rs11178997, rs120074175, and rs7305115 TPH2 variants exist between the controls and subjects with MDD. Finally, the interaction between rs120074175 and rs11178997 of TPH2 and rs7997012 of 5-HT_{2A} increases the susceptibility of individuals to MDD. The following results were discovered for ADHD in terms of TPH2: significant differences in the genotypic and allelic distribution of the rs11179027 and rs1843809 TPH2 variants exist between the controls and subjects with ADHD. Additionally, several haplotypes consisting of markers rs2129575, rs1843809, rs1386493, and rs1023990 are over-represented in ADHD cases. These studies show how several different SNPs of TPH2 can affect the pathophysiology behind these psychiatric disorders, and failure to regulate this gene may cause changes in serotonin that are correlated to the susceptibility of ADHD and MDD. Future studies should focus on verifying this data using larger sample sizes and a broad range of ethnicities. Obtaining more knowledge on the regulation of TPH2 expression could also help scientists develop effective treatments.

Optimization of a Viability Rescue Assay in Human Oral Keratinocytes

Makayla Standfest, Abigail Conrad, Jessica Binkley

Zoledronate (ZOL) is used to treat bone resorptive diseases such as osteoporosis and cancer metastases to bone. ZOL is associated with medication-related osteonecrosis of the jaw (MRONJ), a condition known to cause bone destruction and maxillofacial region lesions. Geranylgeraniol (GGOH) is a potential rescue agent that provides an alternate route in the mevalonate pathway when the pathway is inhibited by ZOL. This study measured the viability of normal human oral keratinocytes (NHOKs) when exposed to ZOL and when GGOH was added under varying conditions to rescue the ZOL-induced viability loss. Viability was measured by an MTT assay in which viable cells convert water-soluble (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) into an insoluble, formazan product. Quantitation was performed by measuring absorbance of the product at 570 nm (630 nm reference) after 72 hours of treatment incubation. NHOKs grown on uncoated 96-well plates showed significantly higher corrected absorbance values ($p < 0.035$) than NHOKs grown on poly-L-lysine-coated plates; consequently, successive experiments were performed on uncoated plates. We tested rescue of NHOKs when 10 microM GGOH was added 15 minutes prior to ZOL, 24 hours prior to ZOL, or simultaneously with ZOL. In the 15 minute-prior variation, 25 and 50 microM ZOL significantly decreased viability but GGOH did not rescue. The 24 hour-prior and the simultaneous addition variations displayed significant ZOL-induced viability loss at 10, 25, and 50 microM. Only when ZOL and GGOH were added simultaneously did significant rescue occur. These results inform successive NHOK experimentation on the relative efficacy of rescue agents to rescue from ZOL-induced viability loss. This work was partially funded by an Indiana Academy of Science senior research grant.

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Characterization of Tetrahymena thermophila Promoter Regions in Induced and Constitutive Genes

Megan Kraus, *Natural Sciences*

My research projects goal was to further understand and characterize the promoter region of metabolic and constitutive genes in the species Tetrahymena Thermophila through carrying out bioinformatics analysis. This research is novel because little is currently known about the promoter regions of tetrahymena, which are Eukaryotic organisms. The characterization of the promoter regions could help to further understand these regions in other eukaryotes, such as humans.

Administration and Curriculum

Michelle Nieter, *Natural Sciences*

Administration and curriculum have been a topic discussed since the beginning of schools. Teachers want curriculum that is affective when reaching their students, and administration wants that, as well as one that is cost friendly. A survey done in 1990-1991 asked school principals how much state departments of education, school boards, teachers, parents, and the actual principals themselves influence the choosing of the school curriculum. This survey distinguished that there is a difference in where this influence in the decision comes from between public and private schools. In public schools, it seems to be evenly distributed among teachers and administration and state boards, but in private schools, the principals perceive administration boards as less influential than the teachers themselves. Administration is influential, but public and private schools differ in how influential they view it

Viability Rescue of Zoledronate-Treated Osteoblasts: A Comparison of Geranylgeraniol and Sodium Citrate

Morgan Jessup, *Natural Sciences*

Nitrogen-containing bisphosphonates (NBPs) are a common medication used to treat bone resorptive diseases. A complication that can arise from NBP use is medication-related osteonecrosis of the jaw (MRONJ). MRONJ involves deconstruction of bone and soft tissue in the maxillofacial region. Zoledronate (ZOL) is the most potent NBP. The goal of this study was to compare the ability of geranylgeraniol (GGOH) to the ability of sodium citrate in rescuing osteoblasts from ZOL-induced cell death. The study utilized MTT viability assays of MC3T3-E1 cells (a murine osteoblastic cell line). Sodium citrate is a calcium chelator and theorized to chelate ZOL-bound intracellular calcium, thus displacing ZOL from the cell. Geranylgeraniol (GGOH) rescues osteoblasts and oral keratinocytes from ZOL-induced death effects in vitro, as reported by others previously. Cells were incubated for 72 hours after the simultaneous addition of 50 microM ZOL with either 2.5, 5, or 10 milliM sodium citrate. Only at 10 milliM was a rescue observed (1.53-fold increase in viability over ZOL-treated, $p=0.08$). In parallel, cells were incubated for the same time after the simultaneous addition of 50 microM ZOL with 0.5, 5, or 10 microM GGOH. GGOH mildly rescued at all three concentrations tested ($p<0.05$, $p<0.001$, $p<0.01$ for each of 0.5, 5, and 10 microM, respectively). All concentrations were determined by dose-response curve experiments in our lab in addition to literature values. The preliminary results suggest sodium citrate may rescue from ZOL-induced death in osteoblasts; however, additional control experiments are required to confirm successful sodium citrate-mediated chelation of intracellular calcium. This work was partially funded by an Indiana Academy of Science senior research grant and a University Scholar Award from Indiana Wesleyan University.

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CULTURING AND RECORDING EXTRACELLULAR ACIDIFICATION OF CRYOPRESERVED RAT GLIAL CELLS

Rachel McKuras, *Natural Sciences*

An increasing number of studies point toward glia playing a more active role in shaping neuronal signals in the CNS including the retina. This study aims to examine whether a specific glial cell mediated regulatory mechanism first observed in the retina is also observed more broadly in the brain. Understanding the breadth of this pathway could sharpen understanding of glial-neuronal interaction and could be beneficial in the treatment of neural health conditions. Past work characterizing this regulatory pathway focused almost exclusively on retinal glial cells from tiger salamanders. This study shifts to working with cortical glial cells from rats, trying to maintain a focus on astrocytes in an attempt to see if the mechanism studied in the salamander retina extends to other glial cells of the CNS. Cryopreserved rat glial cells were received, thawed, plated, and cultured. To compare previously observed ATP-induced acidifications from retinal glia, extracellular pH dynamics from cultured glia, grown over a period of five weeks, were characterized using an electrophysiological approach called self-referencing. From preliminary trials, a clear extracellular acidification was observed which was similar to that of retinal cells studied previously. If studies continue to show consistent extracellular acidifications, like those viewed in the retinal cells, this could suggest that the mechanism is conserved throughout the CNS and increase understanding of the interactions between glia and neuronal cells.

The Effect of Chemical Solutes on the Rate of Growth of Dinoflagellate Genera Implicated in Harmful Algal Blooms

Rebekah Bruce, *Natural Sciences*

Algal blooms are a serious issue in many bodies of water, especially close to agricultural or urban areas. While it is known that both nitrogen and phosphorus are connected to these “red tides”, the exact conditions for these destructive blooms are not well known. Understanding the required concentrations of chemicals such as urea, ammonium, nitrates/nitrites, and phosphates is the first step in preventing this harmful overgrowth of algae. In this study, information found in peer-reviewed journal articles will be used to discuss the findings and commonalities of each. This study will attempt to reach a conclusion and answer the question of what causes harmful algal blooms to occur. The focus will be primarily on *Gymnodinium* dinoflagellates, but some of the information may pertain to other genera.

†Subcloning a novel hematopoietic cell kinase mutant for bacterial expression

Rebekah Drew, *Natural Sciences*

Nef is an accessory protein of HIV which increases infectivity and promotes pathogenesis. Nef does this by binding with the SH3 domain of hematopoietic cell kinase (Hck), a Src family tyrosine kinase. This complex is a promising target for anti-HIV therapies. Little is known, however, about the mechanism by which the Nef-dependant Hck activation increases HIV infectivity. In order to characterize the Nef-Hck relationship a series of Hck mutants ligation-independent cloning methods were used to insert mutants into a bacterial plasmid for subsequent expression and purification of the targeted kinase. These include K295M, PP, P2A, and Y416F mutations for modifying Hck activity. K295M abrogates Hck activity. PP carries additional prolines in the SH1/SH2 linker of Hck stabilizing the inactive conformation of the kinase. The P2A mutation keeps Hck constantly active through mutation of Hck’s polyproline motif. Finally, the Y416F mutation in the activation loop of Hck results in a mutant with little to no enzymatic activity. Insert amplification was successful, as demonstrated by agarose gel electrophoresis, and constructs were successfully transformed into bacteria.

† Denotes a project also included on the oral presentation schedule

Impact of GMOs on Human Health

Sara Wenning, *Natural Sciences*

Genetically Modified Organisms, or GMOs, have their DNA changed by taking a desired trait from one plant and inserting that gene into the other plant. The DNA changes in a plant allows more nutritious food, disease resistance, and allows for less herbicide and pesticide usage. While these advantages have made crop yield increase, many people are concerned about the health risks associated with genetically modifying food. Some concerns include antibiotic resistance, allergenicity, nutritional changes and the formation of toxins. Studies on antibiotic resistance have not been conducted enough to produce sustaining results. In regard to allergenicity, recent studies have found that *Bacillus thuringiensis* corn expresses an allergenic protein. However, assessing allergenicity in GM crops is difficult because when transferring a gene from one source to the other, it is hard to tell the allergenicity of the gene before it is introduced to the food chain. In the only study assessing toxicity and allergenicity with rats, it showed low allergenicity risk. Overall, studies have shown that GM food and feed for animals have no hazardous effects compared to non-GM diets. There is no direct harm to human health when consuming GM foods. However, more research still needs to be conducted on GMOs to come to a consensus on the risks to human health. Furthermore, research needs to be conducted to determine the long-lasting effects of GMOs to human health.

Grafting Cucumber scion onto pumpkin rootstock – increasing resistance to environmental factors and increasing ascorbic acid content.

Savannah Wall, *Natural Sciences*

Grafting of cucumbers has a long history especially in Asia, parts of Europe, and the Middle East. Interest in the U.S. has grown in hopes of utilizing vegetable grafting as a form of disease control. There has been a significant amount of research verifying the positive effects of grafting cucumbers on pumpkin rootstock including, “protect against soil-borne diseases and nematodes, against abiotic stresses such as high/low temperatures, salinity, drought or excessive soil-water content, and against elevated soil concentrations of heavy metals and organic pollutants.” However, finding the best method for grafting including which kind of graft to use, which scions and rootstock pairs work best, and ideal healing conditions for new grafts are still being tried and tested. The goal of this research is to accumulate quantitative data identifying which grafting methods are best by looking at sugar content, starch content, yield, and ascorbic acid content. The two scions being tested are Diva and Katrina, and the two rootstocks being tested are Flexifort and BabyBear. The current graft being used is the slant graft. The healing process last three days with new grafts in a flat with a misted dome covering them which is then covered by a towel to restrict photosynthesis. After three days, the towel is removed. Once the method from start to finish is perfected the afore mentioned qualities can be tested in the produced fruit.

The Effect of Coffee Grounds on Plant Life

Shelby Mowat, *Natural Sciences*

An abundance of coffee grounds is consumed worldwide thus resulting in a lot of coffee residue. Instead of allowing these coffee grounds to go to waste, a better alternative would be to recycle them. The growth of plants could benefit from the use of coffee grounds. Some articles have looked at using coffee grounds as a land amendment in place of fertilizer or an alternative to peat moss. Other studies have focused on how coffee grounds effect plant growth, carotenoids and mineral compositions. The main objective of this research would be comparing the results of coffee grounds in plants to see if coffee grounds are a viable option.

[†] Denotes a project also included on the oral presentation schedule

Viability Rescue of Zoledronate-Treated Osteoblasts: A Comparison of Geranylgeraniol and Phosphonoformic Acid

Spencer Roose, *Natural Sciences*

Zoledronate (ZOL) is a potent nitrogen-containing bisphosphonate (NBP) that inhibits osteoclastic function. It is used to treat cancers that have metastasized to bone and other conditions that reduce bone density. However, it is prone to induce medication-related osteonecrosis of the jaw (MRONJ), a side effect of the drug that manifests as prolonged cell death in the hard and soft tissues of the oral cavity. Phosphonoformic acid (PFA) is an inhibitor of the phosphonate transporters SLC20A and SLC34A, both expressed in osteoblasts. These transporters are thought to allow entry of NBPs into osteoblasts and soft tissue cells; consequently, inhibition of these transporters with PFA could potentially prevent ZOL entry and death effects. Geranylgeraniol (GGOH) is an intermediate of the mevalonate pathway, downstream of the farnesyl pyrophosphate synthase step that is blocked by ZOL. GGOH is known to rescue cells from NBP-induced apoptosis. In this study, MC3T3-E1s (murine osteoblastic cell line) were treated simultaneously for 72 hours with 50 microM ZOL and either GGOH or PFA. The resulting viability was tested via an MTT assay to measure the relative abilities of the two agents to rescue the cells. PFA was applied at concentrations of 0.05, 0.5, and 5 milliM while GGOH was added at 0.5, 5, and 10 microM. All concentrations were based on literature values and dose-response curves performed in our laboratory. No rescue of ZOL-induced viability loss occurred at any of the PFA concentrations. In contrast, GGOH mildly rescued at all three concentrations tested ($p < 0.05$, $p < 0.001$, $p < 0.01$ for each of 0.5, 5, and 10 microM, respectively). The preliminary findings suggest that PFA does not offer an alternative rescue to that provided by GGOH. This work was partially funded by an Indiana Academy of Science senior research grant and a University Scholar Award from Indiana Wesleyan University.

High Intelligence Observed in Corvids

Sydney Sylvester, *Natural Sciences*

Corvids have been thought to be extremely intelligent for years, and recent studies and experiments have only proved this to be true. Some would even compare corvid intelligence to that of primates. Corvids are a family of birds that include crows, ravens, magpies, jackdaws, and jays. Because of their high learning skills, many researchers have tested and discovered ways to show their level of intelligence. Crows have been known to recognize faces, solve puzzles, use tools, and perform social learning among their flock. Many tests focus on New Caledonian crows, one of the most intelligent species of corvids, to perform these tasks. By testing their abilities to perform different tasks, this research shows the complex diversity in corvid intelligence. The results of many of these tasks focuses on the brain activity levels and how corvids convey their responses to the stimuli they are under.

Characteristics of ATP-induced extracellular acidification from retinal Müller (glial) cells

Thomas Leuschner, *Natural Sciences*

Glial cells are anatomically well-positioned to be regulators of neuronal synaptic transmission, and increasing evidence points to their functional role in regulation of neuronal signaling. Small alterations of extracellular levels of H⁺ represent a potent and underappreciated mechanism of modulating synaptic transmission and neuronal excitability. Additionally, extracellular H⁺ dynamics are correlated with significant alterations in the release of neuro transmitter in the retina and are therefore potentially key regulators involved in shaping visual signals (cf. Thoreson and Mangel, 2012). Extensively enveloping all retinal neurons and their synaptic interconnections, Müller cells are well placed to modulate the release of

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neurotransmitter by retinal neurons, and they have recently been demonstrated to potently regulate extracellular levels of H⁺. We have previously shown that this non-neuronal cell type of the retina potently increases extracellular acidity when challenged with ATP. In this study, we use pharmacological and ion removal experiments in combination with an exquisitely sensitive self-referencing approach with H⁺-selective microelectrodes to characterize Na⁺ and K⁺ dependencies of this pathway. These findings point toward a role for the Na⁺/H⁺ exchanger mediating the ATP-induced acidification, further elucidating a glial cell signaling pathway used to regulate neuronal activity levels in the retina.

Gila Monster: Type-2 Diabetes Treatment

Vannessa Peters, *Natural Sciences*

Heloderma suspectum, otherwise known as the Gila monster, is one of the only venomous lizard species; it is with the Gila monster saliva that the Vanderbilt Diabetes Center has been able to research a new way to help treat people with type-two diabetes. Exendin-4 is the product that is extracted from the Gila monster's saliva, and it strongly resembles the glucagon-like peptide-1 analog, GLP-1, treatment. Unlike GLP-1, Exendin-4 does not get targeted by any enzymes in the bloodstream to inactivate it. This leads to the ability of Exendin-4 being able to last up to twelve hours for every injection and lacking of some of the more difficult side effects of other treatment options. The patient can now maintain a healthy weight as well as accumulate a healthy blood-glucose count within the first couple months. This method has been FDA approved.

Use of vegetative grafting to improve cold tolerance in cucumber

Zachary Biddle, *Natural Sciences*

Grafting is a technique in which parts of two different plants are fused together to create a single, functional unit. Many studies have shown grafting fruit-producing plants increases the yield compared to those that were not grafted. Additionally, grafting has been shown to increase cold-tolerance, and fungi, weed, and insect resistance. For these reasons, grafting has grown in popularity especially in Asian countries where temperatures are suboptimal and high demand calls for continuous production. The focus of this study was to identify if grafting 'Diva' cucumber (*Cucumis sativa*) scion onto 'Baby Bear' pumpkin (*Cucurbita pepo*) rootstock would increase yield of fruit in both size and total number of fruits per plant during suboptimal growing temperatures. Grafted and ungrafted cucumbers were grown on two planting dates and were harvested throughout the season. The first planting season started on May 14, 2019 when soil and air temperatures were below the recommended range. The second planting season started on May 31, 2019 when soil and air temperatures were within the recommended range. During both planting seasons the grafted produced three times the amount of fruit compared to the ungrafted. Although, the average size of the fruit was not significantly different between grafted and ungrafted plants.

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