Spring 2021

18th Annual Symposium of the School of Science, Engineering and Health

Messiah University

Message from the Dean

We in the School of Science, Engineering and Health welcome you to this 18th Annual Symposium, and our first as Messiah University. Here you will see our students, faculty and staff showcase innovation, creativity, teamwork and professionalism in our academic departments. Basic and applied research in science and health fields stem from curiosity, acquired skill, and a desire to test and improve processes from foundational principles. The outcomes of scientific research expand intellectual understanding and have tremendous impact on quality of life, environmental health, and human flourishing. We miss having you as guests on our campus but warmly welcome you to enjoy this day virtually.

Angela Hare
Dean School of Science, Engineering and Health, Messiah University

Presentation Abstracts

Biology, Cellular & Molecular

Efficacy Analysis of Zebrafish, *Danio rerio*, as a Novel Demyelination Model Through Exposure to Cuprizone-infused Food

Halle L Becker & Jennifer K Ness-Myers†

The central nervous system demonstrates its functional reliance on glial cells in support of neurological function primarily through its use of oligodendrocytes and the myelin they produce. Damage to myelin is caused by different pathways, one exemplified in Multiple Sclerosis (MS), where the immune system incorrectly attacks healthy tissue of the CNS. Most research focusing on these biological processes of demyelination has relied on rodent models. However, zebrafish, *Danio rerio*, are an ideal model for these goals because they are transparent and therefore are easy to monitor for demyelination and remyelination progression. In this study, the viability of zebrafish as a model for demyelination and remyelination research studies was evaluated. The initial steps were the development of cuprizone-infused food and the design of a cuprizone drug feeding protocol. Following treatment, myelin staining techniques were used to assess the level of myelin damage. The goal of this study is to develop a reliable new demyelination research model that will improve imaging of myelin damage and repair during demyelinating diseases.

Laboratory Culture of *Plasmodium falciparum* with Cryopreserved Human Erythrocytes

Michael L Bellerose, Mina Diep, Matthew T O'Neil, & Lawrence M Mylin†

Uninfected human blood free of anti-malarial medications is essential for the laboratory culture of *Plasmodium falciparum*, the causative agent of severe malaria, but is of limited availability in sub-Saharan Africa where research is conducted by the Macha Research Trust. Cryopreservation of erythrocytes obtained from uninfected donors in the US would provide an adequate supply. Our goal has been to identify methods of freezing that will preserve the capacity of RBCs to support asexual propagation and gametocyte formation by *P. falciparum* strain NF54 in serial asynchronous culture, as well as the shelf life of RBCs stored at -80ºC. We have investigated RBCs cryopreserved using two types of freezing solutions: 1) a mixture of hydroxy ethyl starch

(HES) and polyvinyl alcohol (PVA); 2) glycerol supplemented with human serum. Our results reveal that blood frozen with hydroxy ethyl starch and polyvinyl alcohol failed to support asexual propagation of *P. falciparum* beyond three serial passages. Blood frozen using the glycerol & serum solution was able to support asexual propagation through at least nine serial passages, and later passages were capable of gametocyte generation. NF54 parasites maintained in serial culture using glycerol/serum-frozen remained unable to proliferate when serially passaged using HES/PVA-frozen blood, confirming the absence of mutations that may have adapted the parasite to long-term culture in frozen blood. These results will be discussed with respect to essential functions of the apicoplast, a parasite organelle required for erythrocyte invasion.

Quantifying In Vitro Matrix Deposition to Elucidate the Target of Proglumide-mediated Fibrotic Inhibition in Pancreatic Cancer

Silas Buckwalter & John F Harms†

Pancreatic cancer has a 5-year survival rate of 8%, and the average prognosis has not improved in recent history. Chemotherapy is the primary clinical approach but carries poor efficacy and offers little improvement in patient survival due to the highly fibrotic collagen-rich tumor microenvironment. Our lab has previously confirmed that the gastrin receptor antagonist, proglumide, significantly decreases pancreatic tumor fibrosis *in vivo*. *In vitro* efforts to elucidate the cellular target of proglumide have been unable to demonstrate a decrease in the mRNA expression of collagen (COL1A) or its post-translational processing enzymes, in either myofibroblast-like pancreatic stellate cells or cancer cells. We hypothesize that quantification of deposited collagen matrix *in vitro* will better reflect the decreased fibrosis apparent *in vivo*. Utilizing the Sirius Red/Fast Green protein staining assay, collagen deposition was measured in cultured cells. To normalize collagen to potentially disparate cell proliferation rates, we confirmed that the alamarBlue cell quantification assay does not interfere with subsequent Sirius Red/Fast Green protein staining. Human pancreatic stellate cells (RLT-PSC), mouse pancreatic cancer cells (Panc02), and human pancreatic cancer cells (PANC-1) were stained 4 days after seeding. Significantly higher collagen was evident in Panc02 and PANC-1 wells compared to stellate cells ($p<0.05$). To test the impact of proglumide treatment, cells were treated with proglumide (200 μg/ml). Lines showed no change in normalized collagen deposition after 4 days compared to untreated controls. These preliminary data reflect the unchanged COL1A mRNA levels previously apparent in the stellate and cancer monocultures. Further inhibition studies will utilize co-cultures to address potential heterotypic intercellular communication between these cell types, characteristic of the tumor microenvironment.

Altered Heavy Metal Stress Response of Mutant *Arabidopsis thaliana* Plants in the Presence of Nickel

Annelyse M Cruzan, Michael Shin†, & Richard W Schaeffer†

While heavy metals such as zinc and nickel are necessary for several life-sustaining processes, elevated levels of these metals pose a threat to humans, plants, and other forms of life. Researchers have found that certain plants known as hyperaccumulators tolerate higher levels of heavy metals than non-hyperaccumulating plants. They can be planted in heavy metal contaminated soil to take up excess heavy metals, thus decontaminating the soil in a process

known as phytoremediation. In this research project, we will examine the altered heavy metal stress response of two Columbia-line *Arabidopsis thaliana* mutants that we hypothesize are hyperaccumulators of heavy metal divalent cations. This hypothesis will be tested using nickel tolerance and accumulation assays. The tolerance assays will compare the root length, a key indicator of plant health, of the two mutant *A. thaliana* plants against the Columbia-line wildtype plant in varying concentrations of nickel. The accumulation assay will be used to quantify the concentration of heavy metals present in the plant tissue. These analyses will allow us to determine the impact of the mutations on the nickel heavy metal stress response of the plant.

Cryopreservation of Human Erythrocytes for Laboratory Propagation of *Plasmodium falciparum*

Mina Diep, Michael L Bellerose, Matthew T O'Neil, & Lawrence M Mylin†

The cryopreservation of fresh, uninfected human red blood cells (RBCs) is fundamental in culturing *Plasmodium falciparum*, to create better preventive measures in combatting malaria. We hypothesize that human blood cryopreserved with hydroxyethyl starch and polyvinyl alcohol (HES/PVA) or glycerol and serum will support asexual parasite growth and gametocyte formation in culture following prolonged freezing at -80°C of A+ or O+ blood. These freezing techniques utilized multiple conditions to evaluate each of the ability for the culture to propagate sexually and asexually with the NF54 parasite strain. The conditions tested through serial propagation: fresh, never frozen blood, which acted as the control; short-term cryopreserved blood, which was stored at -80ºC for less than two weeks; and long-term frozen blood, which was stored at -80ºC from six months to one year. This shelf-life study found that HES/PVA cryopreservation technique supported the asexual propagation of *P. falciparum* through only two serial passages; parasites could not be rescued by subsequent addition of non-frozen blood. Blood frozen using the glycerol & serum solution was able to support NF54 propagation beyond a ninth dilution and re-plating (passage). Blood cryopreserved by either method supported the generation of mature gametocytes in the first passage. Additionally, our results suggest that the NF54 strain did not accumulate mutations that abnormally enhanced or sustained growth on cyropreserved blood.

Generation of Bacteriophage T4-specific Monoclonal Antibodies

Josiah Gonzalez, Ukamushu A Undieh, & Lawrence M Mylin†

Vaccines are an integral part of modern-day disease prevention. Healthcare professionals should be aware of the basic immunological foundations of how vaccines work. Students in past offerings of Messiah University's Microbiology course were able to carry out an exercise in which commercially obtained polyclonal neutralizing antisera (samples from simulated patients) were compared for ability to inhibit infection of *E. coli* B cells by bacteriophage T4. Unfortunately, the antiserum is no longer available for purchase. Therefore, we seek to produce similar antibodies using monoclonal technologies. Hybridoma clones capable of generating T4 neutralizing monoclonal antibodies will be produced from T4-immunized Balb/c mice. T4 virus stocks of known titer with reduced concentrations of endotoxin must be produced to vaccinate the mice. Our approach has been to reduce endotoxin concentrations in commercially obtained T4 phage stocks by mild organic extraction, followed by dialysis and concentration using Centricon centrifugal filtration units. titers of the concentrated stocks were determined by

infection of *E. coli B* cells. The endotoxin levels in the virus concentrates were measured to confirm reduction relative to viral titer.

Generation of Engineered SV40 T Antigen-Expressing Immortal Murine Kidney Cells for Vaccination Against Pancreatic Cancer

Daniel L Guevin & Lawrence M Mylin†

That pancreatic cancer is unusually aggressive and deadly has prompted extensive research into its mechanisms and options for therapy. Our research seeks to develop a vaccine that will activate the tumor-bearing host immune system against a novel protein expressed by aggressive pancreatic tumors. Our approach is to engineer immortalized syngeneic cells to express a derivative of the Simian virus 40 Large Tumor antigen protein (SV40 T ag) that contains a 20 amino acid insertion corresponding to a unique sequence found in the CCK2i4svR pancreatic-cancer associated growth factor receptor. These cells will be used as a vaccine to cause the host to develop cellular or humoral immune responses against the cancer-associated CCK2i4svR epitope. Immortalized cell lines were produced by transfection of C57Bl/6 primary kidney cells with four plasmids, each encoding a different derivative of the SV40 T antigen with respect to presence and insertion site of the CCK2i4svR 20 codon sequence. Transfected cultures were monitored for growth of dense foci which result from immortalization as a result of expression of the SV40 T ag. Foci were harvested, expanded and evaluated by indirect immunofluorescence using two SV40 T ag-specific monoclonal antibodies to confirm that they had been successfully transformed and expressed the SV40 T antigen protein derivative. SV40 T ag-positive lines were cryopreserved for characterization in subsequent immunization and tumor immunotherapy studies.

Inhibition of Muscarinic Receptor Subtypes and Effects on Oligodendrocyte Differentiation

Kylee M Kimbel & Jennifer K Ness-Myers†

Oligodendrocytes are the myelinating cells of the central nervous system, and these cells and their myelin sheaths are the autoimmune targets in multiple sclerosis (MS). Increasing the differentiation of OPCs to myelinating OLs is a promising method for treating MS. Recent clinical trials have revealed a positive effect of clemastine, an antihistamine/muscarinic antagonist, in stimulating myelin repair in patients with MS. This study is investigating the combinatorial effects of muscarinic receptor antagonists darifenacin (M1) and pirenzepine (M3) on the rate of oligodendrocyte (OL) maturation. Cultured oligodendrocyte progenitors were used to assess receptor subtype effects on myelin-specific gene expression and the rate of OL maturation during treatment for 4 days. Combination treatments were also studied in the larval zebrafish model. RNA was isolated and analyzed for changes in expression of myelin-specific genes MPZ and MAG. Several dosages of combinatorial treatment and time points were tested, but no significant changes in larval zebrafish gene expression were identified. However, muscarinic agonist, cevimeline, was shown to reduce the expression of myelin-specific genes MPZ and MAG, which supports the hypothesis of the involvement of the muscarinic pathway in myelination. Muscarinic antagonists do not appear to enhance the vigorous myelination program of zebrafish larvae.

The Effect of Stress on Drug Seeking Behavior

Christine A Milbrath, Sara C Stambaugh, Isela M. Rodriguez, & Jennifer L Thomson†

Post-Traumatic Stress Disorder (PTSD) can develop after experiencing an intensely distressing event. The condition exhibits as a maladaptation of the fear response, causing frequent activation of the sympathetic nervous system (fight and flight behavior) in response to benign stimuli. Persistent sympathetic activity results in chronic stress and precipitates other physical and psychological conditions—severely impacting the individual's quality of life. Current treatments yield low recovery rates and poor prognosis, particularly when initiated long after the original trauma. New research explores opioids as a prospective preventative medication, as morphine seems to diminish PTSD acquisition if received during the critical period of memory consolidation. However, PTSD is often comorbid with increased rates of substance abuse, including opioid addiction. Our research examined if PTSD treatment with morphine increases drug seeking behavior using a rodent model. Rats underwent Stress-Enhanced Fear Learning (SEFL) to induce PTSD like symptoms. After SEFL was completed, the rats were given two injections of morphine in a unique "Context A" and two injections of saline in "Context B". The rats were then allowed to pass between the chambers, and the relative amounts of time spent in either Context A or Context B was measured to determine if the rodents exhibited a Conditioned Place Preference (CPP). If SEFL subjected rats exhibit CPP for Context A, this indicates a correlation between receiving morphine treatment for PTSD and increased drug seeking behavior.

Cryopreservation of Human Erythrocytes for *Plasmodium falciparum* Culture

Matthew T O'Neil, Mina Diep, Michael L Bellerose, & Lawrence M Mylin†

Malaria is caused by multiple species of the parasite *Plasmodium* and disproportionately affects people living in the developing world where effective control of or protection from the parasite is lacking. This study seeks to support ongoing research at the Macha Research Trust (MRT) which is located in the Southern Province of Zambia where the virulent species, *Plasmodia falciparum* is prevalent. Our goal is to support the capacity of the laboratory at MRT to culture (propagate and preserve) locally-isolated or laboratory strains of *Plasmodium*. Laboratory cultivation of *P. falciparum* requires fresh human blood. However, it is difficult to assure the steady supply of fresh, uninfected human blood needed to sustain culture experiments at MRT because blood from local residents cannot be used, and because many visiting scientists and physicians routinely take prophylactic anti-malarial drugs which can make their erythrocytes unable to support asexual propagation of or gametocyte generation by *P. falciparum* in culture. We are investigating methods that should allow for the cryopreservation of erythrocytes obtained in the US from uninfected individuals, with subsequent shipment to Zambia. We have cryopreserved preparations of fresh, leukocyte-depleted erythrocyte suspensions using minimal aqueous volumes of solutions containing the hydroxyethyl starch (HES) and polyvinyl alcohol (PVA), or with glycerol-based solutions. This presentation will describe ongoing efforts to determine if cryopreserved RBCs can effectively support asexual propagation or gametocyte formation for the *P. falciparum* laboratory strain NF54, and how well this capacity is maintained following long-term storage at -80°C.

Stress Enhanced Fear Learning: An Animal Model of PTSD

Isela M. Rodriguez, Sara C Stambaugh, Christine A Milbrath, & Jennifer L Thomson†

Post-traumatic stress disorder (PTSD) is a stress-induced disorder that may be established after a traumatic experience. Individuals with PTSD may develop extensive fear and anxiety in non-threatening environments. The stress-enhanced fear learning animal model (SEFL) mimics the symptoms of PTSD in humans by observing the effects of an animal exposed to a prolonged stressor. SEFL is measuring the increase of subsequent learning about fearful stimuli that occurs after a severe stressor. Using the SEFL animal model, rats were exposed to two different environments, Context A and Context B. In Context A, the stressor was 15-foot shocks administered over 90 minutes induces PTSD-like symptoms. Context B, which had a different scent and noise than Context A, would be exposed to a single shock in an unfamiliar environment. The two reminder shocks were delivered in Context B, one followed the injection of morphine or saline, while the other shock was administered three weeks later. Morphine is a promising treatment for PTSD as a method to prevent the formation of the fear learning response in the animal model. The rats were recorded for seven weeks in Context B and the rats' degree of freezing was assessed. Freezing behavior in rats is indicative of a fear response. We hypothesize that morphine-treated rats would exhibit less freezing behavior than saline-treated rates following both reminder shocks.

Utilizing a Plasma Membrane Enrichment Protocol to Optimize Western Blot Detection of the CCK2 and CCK2i4sv Receptors

Noah Scholl & John F Harms†

Pancreatic cancer is currently the fourth most deadly form of cancer in the United States. One factor implicated in pancreatic cancer growth is the hormone signaling pathway between gastrin and its receptor, CCK2R. In the early 2000's, it was also discovered that pancreatic cancer cells can contain a variant ($CCK2_{i4sv}R$) of the normal receptor protein. Crucially, this longer variant has been shown to be hyper-stimulated and to drive increased cancer growth. Measuring the relative abundance of these two receptors at the protein level can help us understand their role in pancreatic cancer and may represent prognostic value as a biomarker. However, while RNA detection and measurement have been reproducible, protein detection has been problematic. Using western blot analysis, we have been able to detect the receptors in cells expressing them at high levels; however, detection in wild type and stably-transfected lines more representative of physiological expression has been unclear. Thus, we hypothesize that low, natural abundance of the receptors requires enrichment for reliable quantification. Herein, we report our initial attempt to enrich for green fluorescent protein-tagged variants, CCK2R-GFP and $CCK2_{i4sv}R$-GFP, utilizing a membrane extraction protocol based on the non-ionic detergent, Triton X-114. Following enrichment, western analysis demonstrated a significant decrease in cytosolic protein in control cells transfected with untagged GFP. Efforts to verify retention of membrane proteins in the hydrophobic fraction, and subsequent specific detection of the CCK2R variants, are ongoing.

The Effect of Morphine on the Reconsolidation of Traumatic Fear Memories: A Pilot Study

Sara C Stambaugh, Christine A Milbrath, Isela M. Rodriguez, & Jennifer L Thomson†

Post-traumatic stress disorder (PTSD) is a chronic, debilitating disorder in which the heightened fear and arousal experienced during a traumatic event generalizes to nonthreatening stimuli. Previous research has suggested that administering morphine within 24-48 hours of a traumatic event inhibits memory consolidation and prevents future development of PTSD. The goal of this study was to block the reconsolidation of previously stored stress memories by administering morphine within 24-48 hours of conscious recall of the original stressor. A rat model of PTSD called stress-enhanced fear learning (SEFL) was used, which employed two distinct contexts, Context A and Context B. The severe stressor, which mimics trauma experienced by humans, occurred in Context A, during which 15 foot shocks were administered within a 90-minute period. Two reminder shocks were delivered in Context B. The first was followed by an injection of morphine or saline, and the second was delivered three weeks later without a subsequent pharmacological injection. The animals were video recorded for seven weeks in Context B to assess for freezing behavior, a classic fear response in rats. We hypothesized that the morphine-treated rats would exhibit less freezing behavior following both reminder shocks, whereas the saline-treated rats would exhibit a heightened fear response.

Differential Efficacy of Antifibrotic and Immunotherapy on T-cell Infiltration in Murine Pancreatic Adenocarcinoma

Kally Tan, Lawrence M Mylin†, & John F Harms†

Pancreatic cancer is projected to become the third leading cause of cancer-related deaths in the United States and has a low 5-year relative survival of 10%. The poor response to treatment is attributed to the immunosuppressive and highly fibrotic nature of the pancreatic tumor microenvironment. Previous studies have demonstrated that proglumide treatment results in decreased fibrosis and may increase T-lymphocyte infiltration. Additionally, our lab has demonstrated that peptide immunization targeting the human $CCK2_{i4sv}$ receptor, a cancer-associated splice variant of CCK2R, evokes a potent T-cell response in mice. We hypothesized that combination therapy utilizing concomitant immunization and anti-fibrotic treatment will further enhance T-cell infiltration and decrease tumor burden. To address this, we first analyzed T-cell infiltration in a pilot group of orthotopic murine pancreatic tumors (Panc02) obtained from mice treated +/- proglumide. Tumors were analyzed by immunohistochemistry (n=3 per grp), and infiltrating CD-3+ T-cells were numerated in five random photomicrographs for each tumor. Tumors treated with proglumide exhibited increased T-cell infiltrate and significantly less fibrosis. To assess the efficacy of combination therapy, mice (C57Bl/6) were immunized weekly with $CCK2_{i4sv}$ and control peptides starting three weeks prior to orthotopic injection of syngeneic cancer cells (Panc02) engineered to express human $CCK2_{i4sv}$R. Both groups received oral proglumide following tumor establishment. Upon necropsy ($\leq$6 wks), mice exhibited no significant difference in tumor burden. Immunohistochemical analysis showed the number of infiltrating CD-3+ T-cells per tissue area trended higher in these proglumide-treated mice compared to the previous untreated pilot controls. However, no difference in infiltration was observed in tumors from $CCK2_{i4sv}$R-immunized mice compared to control immunization. Our data confirm proglumide treatment may enhance immune access to the tumor; however, simultaneous analysis of the circulating lymphocyte response is necessary to thoroughly determine the efficacy of combined immunotherapy on T-cell infiltration.

Genetic Control of Heavy Metal Nutrient Uptake in *Arabidopsis thaliana*

Nathan Thorne, Richard W Schaeffer†, & Michael Shin†

Plant cells uptake metals from the soil by operating specific cellular pumps to transport metal nutrients. These pumps are crucial for maintaining the healthy concentration of metal elements. Metal Micronutrients, like zinc or nickel, are essential to plant growth and their uptake is controlled by the MTP1 transporter on the vacuoles in cells. Using *Arabidopsis Thaliana* as a model organism, the effect of mutations on the MTP1 gene can provide a window into MTP1 transporter function and metal nutrient uptake pathways. Two preliminary metal tolerance experiments were conducted on Columbia line wild-type seeds with zinc and nickel contamination to learn what effects the metal ions were having on the plants' growth. Unfortunately, a problem found during a preliminary wild-type experiment highlighted problems somewhere in the experiment. After that result, the primary purpose became to troubleshooting the experimental method. Both experiments with zinc and nickel contamination yielded results that unexpectedly appeared to be hyper-concentrations of metal ions. Hyper-concentration pointed to a problem with the media, the metal ion solutions, or the seeds. The media was examined by chemical digestion and atomic absorption spectroscopy to quantify metal ion concentration. The analysis demonstrated the concentrations were not hyper-concentrated in the media meaning the problem was with the metal ion solutions. This semester began with a complete remake of metal ion stock solutions and growing a new batch of seeds that would hopefully fix the problems. Early experiments showed the same apparent hyper-concentration problem but with a noticeable consistency. The plants were reacting consistently to the presence of metal ions in a range between 20-60 uM with nickel contamination. This consistency is shown over multiple experiments and means an adjusted experiment range to can be used to test MTP1 mutant tolerance.

Titer Determination, Enrichment, and Endotoxin Quantitation in Bacteriophage T4 Stocks for Generation of Monoclonal Antibodies

Ukamushu A Undieh, Josiah Gonzalez, & Lawrence M Mylin†

Carolina Laboratory Supplies was the sole commercial supplier of polyclonal antisera specific to bacteriophage T4, an essential item used in the Phage Neutralization Lab in Microbiology courses at Messiah University. Carolina ceased production of this antiserum, necessitating discontinuation the Phage Neutralization Lab. the goal of this study is to produce monoclonal T4-neutralizing antiserum so that the Phage Neutralization Lab can be restored. The T4 suspensions used need to have confirmed titers and be depleted of lipopolysaccharide (endotoxin) which are lethal to mammals. Determining the titer of the bacteriophage suspension, purifying it, and verifying that the endotoxin levels are below lethal concentrations must be done before immunization of the mice. The successful generation of antibodies against bacteriophage T4 would enable the project to continue to the next stage, where monoclonal antiserum can be produced using hybridoma technology.

Effect of Lymphoid Cell Kinase (Lck) Inhibition on Oligodendrocyte Differentiation

Ronnie Woodring & Jennifer K Ness-Myers†

Oligodendrocytes are the myelinating cell of the central nervous system. Myelination of axons aids in the propagation of neuronal signals and are essential to the function of the nervous system. In this study, oligodendrocyte cultures from neonatal rat pups were used to analyze the signaling protein Lck and its role in oligodendrocyte differentiation. We hypothesized that Lck signaling promotes the normal development and differentiation of oligodendrocytes, thus the inhibition of Lck will result in decreased ability to express markers associated with myelinating oligodendrocytes. Differentiation of oligodendrocytes was assessed by immunocytochemistry to identify maturation-specific markers and expression of myelin-specific genes following treatment with an Lck-specific inhibitor. Untreated oligodendrocytes expressed more O1, a cell-surface marker expressed by mature oligodendrocytes, than oligodendrocytes treated with a Lck inhibitor at four and five days after treatment. No significant difference was observed in A2B5, a cell-surface marker of immature oligodendrocytes, four days after treatment. Oligodendrocyte precursor cells (OPCs) were transfected with a mammalian green fluorescent protein (GFP) plasmid and grown under conditions that either encouraged or discouraged OPC maturation to investigate the efficacy and longevity of the transfected GFP protein expression. Future studies will investigate the signaling pathways impacted by loss of Lck activity and their role in oligodendrocyte differentiation.

Biology, Organismal & Ecological

Using Drones for Rapid, Low-Impact Assessment of Panamanian Golden Frogs

Luke J Foster

The *Atelopus zeteki*, or Panamanian Golden Frog, is extinct in wild populations and only survives in captivity. Reintroduction is the future of *Atelopus*, but due to their vulnerability, a low impact method of observation and population monitoring are required. With recent advances in Unmanned Aircraft Systems (UAS: drones), a much-needed low impact solution for golden frog population survey for continued ensure post-reintroduction success. This semester we have dedicated time and planning to certify, train, and practice on drone flight. We are planning to set up experiments with frog models close to that of the *Atelopus* in Panama, their country of origin. These experiments will allow us to utilize an ecological drone to test its effectiveness for golden frog population monitoring in the future.

A Sustainable Alternative: Chicken Waste as a Nutrient Source for Red Potatoes (*Solanum tuberosum*) in a Hydroponic System

Allie Houtz & David K Foster†

The purpose of this study was to determine the viability of growing potatoes hydroponically with chicken waste as an alternative, more sustainable nutrient source in substitute for fish waste in an aquaponics system. Hydroponics is a growing form of sustainable agriculture which utilizes a water-based method to deliver nutrients to plants. Hydroponics is a preferable alternative as it uses 90% less water than traditional agriculture and does not contribute to common agricultural issues such as land degradation. A popular form of hydroponics is aquaponics, which combines hydroponic technology with aquaculture. The fish waste in this system is the sole source of nutrients for the plants. The cost and difficulty of maintaining the fish in this system can be an

obstacle to farming communities, however. A proposed alternative to the use of aquaculture is the use of chicken waste as a nutrient source. Chicken waste is a much cheaper and more accessible resource for less developed farming communities. It is also not as high maintenance as aquaculture and contains a high content of essential nutrients such as nitrogen, potassium, and phosphorus. Potatoes are the crop of choice for this study due to their relatively high nutrient content, making them a popular staple crop in less developed countries. If chicken waste proves to be an effective nutrient source for staple crops such as potatoes in a hydroponic system, it may be a viable option for less developed farming communities who are in need of sustainable alternatives to traditional farming practices.

Analysis of the Impact of Water Treatment Plant Effluent on Antibiotic Resistance in Pennsylvania Stream Microbiomes

Christy J Sackett & Jeff S Erikson†

Though antibiotics have served as a powerful tool in humanity's fight against disease, the overuse of antibiotics has caused some bacteria to develop resistances to antibiotics, making these drugs less effective. One means by which bacteria may be exposed to antibiotics and develop resistance is through wastewater treatment plant effluent, which carries antibiotics from human wastes into local streams. We hypothesize that increased levels of antibiotics downstream of treatment plants have selected for increased antibiotic resistance in bacteria living in these downstream regions. As such, when both upstream and downstream samples of bacteria are exposed to the same antibiotic treatments, downstream samples should exhibit higher survival. To test our hypothesis, we sampled bacteria from Dogwood Run and Stony Run in Dillsburg, PA, selecting collection sites from both upstream and downstream of the local wastewater treatment plants. We cultured these bacteria in the lab, classified them based on colony morphology and Gram-stain results, and exposed them to antibiotics through the Kirby-Bauer and replica plating methods. When comparing all bacteria, there did not appear to be a clear trend of increased resistance in either the upstream or downstream samples. However, when comparing bacteria only within our classified groups, initial results showed 26 instances of increased downstream resistance and only 13 instances of increased upstream resistance. These results are promising for our future work in this area, and they may indicate that increased regulations on wastewater treatment plants are needed to combat further antibiotic resistance development.

Hydroponic Growth of Goldenseal (*Hydrastis canadensis*)

Regan M Wilton & David K Foster†

Goldenseal (*Hydrastis canadensis*) is a plant native to woodlands of east-central North America. Goldenseal roots contain antimicrobial compounds which promote respiratory and digestive health and the plant has been wild-harvested almost to the point of extinction. Plants are products of their environments and the concentration of medicinal compounds within a plant are affected by the conditions in which the plant was grown. Wild harvested plants are subject to the variations in their natural environment and it is almost impossible to accurately know the concentrations of desired medicinal compounds within plants that are wild-harvested. With all medicinal compounds there exists a therapeutic dosage in which the compounds are beneficial for the body. Outside of this window, the compounds are either ineffective or toxic. It is crucial

for supplements made from medicinal plants to contain known concentrations of the desired medicinal compounds in order to ensure the effectiveness and safety of the supplement. Due to these reasons it is crucial to develop a sustainable and controlled method of growing goldenseal, and aquaponic technology is a form of agriculture that serves as a hopeful solution to these concerns. Previously, goldenseal was aquaponically grown at Messiah University somewhat successfully, but many of the plants died prematurely of root rot. Other than the work completed at Messiah, there are no published studies that include growing goldenseal aquaponically. It is proposed that goldenseal be grown in an aquaponic system at various water levels to find a growing method that maximizes the plant's survival and productivity.

Biochemistry

Expression Vector Construction for a Heme C Attachment Assay

Spencer P Clements & Jesse Kleingardner†

The world is rapidly approaching a point where they need to find a source of sustainable clean energy. Something that could aid this problem is new efficient catalysts, one potential catalyst is Cobalt substituted Holocytochrome C. Holocytochrome c is a protein that is attached to a heme group at a CXXCH amino acid motif. These are found naturally occurring in the mitochondria and are part of the electron transport chain. These enzymes have the capability to catalyze the reduction of water into a diatomic hydrogen which provides a fuel source for energy production. Cytochrome *c* naturally has an iron heme cofactor, but this wild type does not have the same enzymatic capabilities as a cobalt substituted heme to catalyze the reduction of water. In order to synthesize a large quantity of the desired coenzyme, they must be created *in vitro* using *E.* coli expression vectors. The primary objective of my project was to create these expression vectors that would be capable of expressing large amounts of desired product by having the cytochrome *c* be expressed by cytochrome *c* heme lyase, the naturally existing enzyme responsible for holocytochrome c synthesis. These expression vectors can utilize mutations of both the cytochrome *c* and cytochrome heme lyase genes to express different mutants. These mutant products can then be quantitatively analyzed in different combinations to find what combination gives the highest enzymatic capability to combine cytochrome c and a cobalt substituted heme. Once the ideal combination is found, we will attempt to make a microbe and utilize the mutated proteins to effectively perform the process *in vivo.* Utilizing this could provide a catalyst that leads to efficient production of fuels from sustainable energy sources.

Developing a Reliable Computational Method to Determine the Allosteric Sites to Be Targeted in Finding Non-aggregating Allosteric Inhibitors of PTP1B

Sarah K Codd, Anne M Reeve†, Seth Kabonick, & Noah D Smith

Type 2 diabetes is a serious metabolic disease with a growing need for improved therapeutics. Traditional antidiabetic drugs can assist with the maintenance of blood glucose levels in diabetics but are not optimal treatments due to side effects. PTP1B is a validated therapeutic target for type 2 diabetes, as it acts as a negative regulator in the insulin signaling pathway. Inhibiting PTP1B may allow for increased insulin sensitivity, leading to glucose homeostasis and improved metabolism. In the past three allosteric sites were identified as targets for PTP1B

inhibition. This study analyzes the validity of these sites and the possibility of other allosteric sites through computational methods and enzymatic assays. A new allosteric site was identified based on the determination of the anti-diabetic drug Lobeglitazone's binding site and used to improve the computational method for testing possible inhibitors of PTP1B. Correlation of the computational method with known natural product PTP1B inhibitors revealed that the computational method is independent of kinetic properties. This study provided a new allosteric site to target and a computational model to eliminate non-inhibiting compounds in the search for type 2 diabetes therapeutics.

Genetic Code Engineering: Expression of Cytochrome C with a Methylated Histidine Ligand

Nathan E Cordell & Jesse Kleingardner†

Cytochrome c is a mitochondrial enzyme that is responsible for catalyzing many important chemical reactions in many different species of animals. My research was primarily concerned with the creation of a mutant variant to Horse Cytochrome c (HCc), with potentially different electrochemistry than the original. The target amino acid for the project was HCc's axial histidine, which I attempted to replace with an Unnatural Amino Acid (UAA) that is simply a methylated variant of histidine. At this point, I have successfully transformed BL21 cells with engineered plasmids to create cells with the potential to express the mutant HCc protein. Once the new protein is confirmed to have been created, its properties will be compared to the original to gauge the impact of the methylated, axial histidine on the electrochemistry of HCc.

Using Molecular Dynamics to Optimize Geometries of Metal Binding Sites

Kadie A Goodin & Jesse Kleingardner†

Molecular dynamics is a computational method that models a macromolecule in a solvent over time. Using the molecular dynamics software GROMACS to engineer metal-binding sites in proteins, precious time and energy can be saved by computational modeling of the metal-binding sites first. In this project, molecular dynamics was used to simulate metal binding sites with different combinations of ligands to obtain optimized geometries to ensure that the results of the simulations were realistic. Specifically, iron and zinc sites involving cysteine and histidine ligands were analyzed. Using GROMACS, these metal sites were subjected to simulated annealing, and the resulting bond angles and bond distances from the stable conformations were compared to the average values in the Protein Data Bank (PDB). By adjusting force constants, bond distances, and bond angles in the GROMACS force fields, two particular sets of parameters were identified for an iron site with four histidine ligands that produced realistic bond distances. However, when aligning these conformations to the metal binding site in Troponin C, the lowest RMSD value calculated was 0.72 which is not low enough to indicate good overlap, indicating that the specific combinations of amino acids in the model would not likely generate a good iron binding site in Troponin C. A lower RSMD value would indicate better alignment between the metal binding site backbone atoms and the protein backbone atoms. The bond angles will need to be optimized in addition to the bond distances to produce geometries that overlap favorably with proteins. This procedure can be applied to metal binding sites with different combinations of cysteine and histidine to determine what specific set of ligands will produce an iron binding site ligated by amino acids whose backbones overlay with the existing backbone structure of Troponin C. To increase the efficiency of engineering metal binding sites in proteins, only

binding sites with favorable backbone atom overlap would be produced and tested in the laboratory.

Synthesis of Novel Hydroxychalcone Derivatives as Potential Inhibitors of Protein Tyrosine Phosphatase 1B

Seth Kabonick, Sarah K Codd, Noah D Smith, & Anne M Reeve†

Protein phosphatases have been considered a potential target for drug-based therapy since their discovery. Mutations of phosphatases found in cell signaling pathways have been linked to type II diabetes, obesity, and certain types of cancer. One of these phosphatases, protein tyrosine phosphatase 1B (PTP1B), functions as a negative regulator of the insulin pathway. A gene knockout study in mice confirmed that mutations to this protein result in a dampened insulin sensitivity. Previous attempts at competitive inhibition through drug-based therapy have been unsuccessful due to the highly conserved active site across the phosphatase family. This study explores the plethora of natural products available as potential inhibitors for PTP1B in hopes of designing a molecular scaffold to generate site-specific inhibitors. Chalcones and stilbenes are two compound families that have previously exhibited inhibitory binding. Hydroxychalcone derivatives, specifically those in the para position, show promise as a drug scaffold for specific active site inhibitors. In addition, isoprenyl groups attached to branching aromatic rings have a significantly higher binding affinity to PTP1B and a lower IC50. Initial *in silico* results showed moderate to high binding affinities for a small hydroxychalcone library. Currently, enzyme activity levels in the presence of these hydroxychalcones are determined using absorbance assays. Duplicate assays are performed with detergent to account for non-specific inhibition caused by aggregation. Preliminary assays using commercially available hydroxychalcones display slight to moderate inhibition of PTP1B at the active site. In future studies, twelve hydroxychalcones will be synthesized and introduced into an absorbance assay to evaluate their potential as a scaffold for PTP1B specific inhibitors.

Mutagenesis of a Heme Attachment Enzyme for Expression of Metal-substituted Heme Proteins

Blake Rondon, Jesse Kleingardner†, & Kadie A Goodin

Heme is a molecule that plays a vital role in many biological processes. Because of its central iron atom, heme proteins are a class of metalloproteins, which are renowned for their ability to enable many of the daily processes such as oxygen transport to begin in an organism. Our research aims to substitute the central iron atom with a cobalt atom due to its unique chemistry that will allow us to expand the metalloprotein cofactors living systems employ to fine tune their catalytic properties. Modifying this molecule is best done using the metabolic pathways of *E. coli* which has an expression vector that we can manipulate to see if the mutants of the cytochrome c heme lyase enzyme, responsible for ligating heme to its attachment motif, would allow us to replace the central iron atom with a cobalt atom. Our experiments consist of raising *E. coli* colonies and extracting their DNA via MiniPrep so that we can induce mutations to the bacterial plasmid using several types of PCR based mutagenesis. We can then use MiniPrep to extract the newly transformed DNA and confirm that the bacterial cell successfully integrated the mutation into its plasmid with Sanger Sequencing. This part of the project was performed by Cornell Genomics Center. Out of the 4 variants created, only one, W97A, expressed the desired mutation that would allow for the possibility of heme substitution.

However, further tests will be performed on the additional 3 variants after their PCR products have been placed through a Dpn1 digest.

Site-directed Mutagenesis of the Fluorescent Calcium-sensing Troponin C Protein Complex to Bind Iron and Copper

Jenna Walter & Jesse Kleingardner†

Metal ion imbalances in the brain can lead to the formation of amyloid plaques, groups of misfolded proteins, characteristic of neurodegenerative diseases like Alzheimer's. The current method for detecting metal ions, the Patch-Clamp, requires excised tissue, expensive equipment, and skilled workers. This study seeks to develop a less labor-intensive technique by genetically encoding the binding site of a fluorescent calcium-sensing protein, troponin-C, to bind to iron and copper. The hypothesis of this research is that if enough mutations are introduced to the ten amino acids in the calcium-binding site of troponin-C, one mutant will be able to bind to iron and copper respectively. 1,024 possible mutant combinations were introduced to create a mutant library through a series of PCR reactions resulting in five mutated fragments. The five mutated fragments were then consecutively reconstructed through a series of COE-PCR reactions to rebuild the mutated troponin-C gene. Gel electrophoresis was used to verify the resulting fragment lengths after each reaction. Certain fragment combinations did not combine successfully despite altering COE-PCR parameters, while other combinations were successful. Gibbon Assembly was also used to reconstruct the troponin-C gene. In the future the mutant library will be verified through Sanger DNA sequencing and successfully mutated DNA will then be inserted into *E.coli* cells via transformation. The library will be screened based on fluorescence and successful sequences will be compared to fine-tune future mutation libraries.

Chemistry

Self-assembled Monolayers on ZnSe Surfaces

Jeffrey Gao, Bailey Rhodes, Niklas Hellgren†, & Alison R Noble†

Zinc selenide (ZnSe) has drawn particular interest as a possible supporting substrate for self-assembled monolayers (SAMs) due to its transparency in the infrared region and its functionality as a semiconductor. The naturally forming oxide layer on bulk ZnSe has been shown to support SAMs; however, the effect of modifications to this oxide layer through chemical means has not been well-established. In this study, the oxide layer and choice of adsorbate molecular head group are optimized towards the formation of high-quality SAMs. This is achieved by stripping the oxide later from optically polished ZnSe substrates using varying chemical surface treatments prior to SAM deposition. The resulting surfaces are characterized via contact angle analysis, Fourier transform infrared spectroscopy, and atomic force microscopy. While SAMs with thiolate head groups formed readily on both etched and un-etched substrates, we find that acid head groups show affinity only for certain etched surfaces. This can be reconciled with the surface compositions under various etching conditions, as determined via X-Ray photoelectron spectroscopy, to determine the surface compounds conducive to the formation of these SAMs.

Synthesis of Carbon Nitride Thin Films for Use in the Catalysis of the Oxygen Reduction Reaction

Daniel Vallette, Richard W Schaeffer†, & Niklas Hellgren†

One proposed solution to reduce global carbon dioxide emissions is to replace the traditional engine with one that does not produce harmful emissions. A hydrogen fuel cell meets this requirement and utilizes the oxygen reduction reaction (ORR) at the cathode of the cell. One major reason that they are not widely implemented is that the main catalyst used, platinum, is expensive and breaks down. Carbon nitride has been explored as a possible replacement for the catalyst of the ORR. Physical vapor deposition via DC magnetron sputtering was used to create thin films of CN_x for analysis by cyclic voltammetry, serving as a model ORR in the lab. It is necessary to deposit a metallic layer between substrate and CN_x for the thin films to be stable. However, upon heating the carbon nitride thin films become non-conductive and therefore cannot be measured by cyclic voltammetry. Future work will focus on solving the conductivity issues associated with heating in the deposition process as well as structural determinations of nitrogen content in CN_x to determine the optimal doping mode.

Computer & Information Science

Burkina's Promise

Joston W Chan, **Matthew S Coates**, & **Rebecca C Merendino**

We helped build a website application that is meant to store and track information concerning users, sponsors, and children for ease of looking up information. The Burkina's Promise organization started when several short-term mission trips to the country of Burkina Faso revealed a need for helping the children of pastors become educated. The need for the site itself arose when the Burkina's Promise organization outgrew its use of Microsoft Excel to handle all of its data and information. During our time developing the site, we added features such as a User Messaging feature, more visitor pages, and other features. Lastly, we tested the entire site and fixed various bugs such as getting the French translator API to work throughout the user pages and being able to delete children in the database on the frontend via a button.

Behavv

Bryce Doane, **Eddie Daniel**, **Matthew C Laven**, & **Gage L Sapp-Rahme**

Our application was built to aid special education teachers in the monitoring and tracking of students' behavior. Every day, these teachers are asked to record student's performance of specific tasks to track their progression over time. Teachers will then use this data to generate reports to share with other teachers, parents, or counselors of that student. Currently, teachers are using complex and unmanageable spreadsheets to record student's ratings on specific tasks within specific classes. Behavv serves to make reporting this data easier and more user-friendly by simplifying data entry and presenting clear visual representations of that information. Our application allows teachers to focus less on stressful spreadsheets and more on students' success.

Something WEAVER

Micah Johnson, Larry Mylin‡, & John Harms‡

ISeek

Nik m Mourelatos, **Sam J Gulinello**, Joseph M King, & Luke A Meads

The iSeek team has built an application to aid people that are visually impaired. If you are visually impaired and have misplaced something you own then you could spend an unwanted amount of time trying to find your object. iSeek intends to solve this problem. Through the use of your phone's camera, alongside a trained chatbot, helps users find misplaced or new objects in their surroundings. iSeek is built with the user in mind, all actions inside the app are voice-activated alongside the classic touch controls. We hope to have created an app that is unique to the market and that has real-world applications to help people live a more enjoyable, easy life.

Hit Pause

Will Newcomb, Tanner Stern, **Joshua Keong**, & Drew Weaver

We created an application that helps provide coping mechanisms for individuals struggling with anxiety. Everyone experiences stress and anxiety in different ways, so we sought after a solution, tailor made to the user, to help them find better ways that work for them to manage their stressors. By taking a few short surveys, our algorithm learns about the user's likes, dislikes, and past experiences with stress to offer handpicked suggestions generated from that data. Each suggestion comes with an excerpt on why it's effective for anxiety, and a playlist from Spotify to help calm the mind during the activity. The more they interact with the app by reviewing those suggestions, the better our algorithm gets at personalizing them for each user.

Development of a Graphical Advising Tool for Computing, Mathematics and Physics

Billy Park & Matthew J Farrar†

Effective degree planning, course scheduling, and advising are predicated upon an understandable and clear curricular hierarchy. Presently, there is no tool at Messiah University that allows students and advisors to simultaneously visualize course requirements, prerequisite structure, and course offerings in an intuitive format.

To this end, I built an application that lets the user—faculty or student—visualize these requirements for degrees in the Department of Computing, Mathematics and Physics. The output of this program is a tree diagram that provides "at a glance" mapping of course sequencing and availability.

HooDat

Eric J Weischedel, **Belosan Wubishet Jekale**, **Wesley Chong**, & Billy Park

We built a mobile app that quizzes you on peoples' names and faces in order to help users remember them. Humans are bad at remembering people's names. Despite really wanting to learn someone's name, many people struggle to recall it, which can lead to feelings of awkwardness. According to Charan Ranganath, the director of the Dynamic Memory Lab at UC Davis, people underestimate the work that needs to be done in order to commit somebody's name to memory: "People are often overconfident, and they underestimate how hard it will be later on." It is easy to focus on making a good impression, rather than learning a new name and

face. Ranganath asserts that one way to solve this problem is to test your name memory skills: "The act of actually testing yourself on the name will help you retain it better in the long term." Our application implements this solution by providing a way to create and take quizzes on people's names and faces. Users can group people into categories in order to associate people with certain characteristics, e.g., classmates or coworkers. Additionally, managers can create lists and share them with other users, making onboarding a new group of people a breeze.

Engineering

Prosthetic Knee for CURE Kenya

Samuel R Burgess, **Kay Laura Sindabizera**, Isaiah D Bryner, Nathan E Jaloszynski, Sarah N Kelchner, Josiah D Moyer, Carter D Urich, & Jamie R Williams†

Amputations, specifically lower limb amputations, are common in sub-Saharan Africa and across the broader global community largely due to infection and disease. All amputees struggle with the negative social stigma surrounding their missing limb and have difficulty getting from one place to another. Our project, The Prosthetic Knee Team, partners with the orthopedic workshop at the CURE International Hospital in Kijabe, Kenya to create a prosthetic knee design for a specific type of amputation known as a Knee Disarticulation (also called through-knee). Currently, the orthopedic workshop is only able to provide one very expensive prosthetic knee option for these patients. Often, these patients choose to undergo a second surgery, a trans-femoral amputation, because it is more affordable. The goal of our project is to provide the orthopedic workshop with a manufacturable prosthetic knee design that provides through-knee amputees with a cheaper prosthetic option and removes the need to have a second amputation above the knee. Throughout the past two semesters, our focus has been to modify and improve our current prosthetic design to best meet all of the specifications laid out by our partner. Those specifications include minimized thigh-lengthening, low weight, maximized stability and durability, and aesthetically pleasing. We also have performed simulated tests to determine the overall strength and lifespan of the design. Moving forward, our team is presently working on manufacturing the prosthetic knees at Messiah University's machine shop.

A Low-Cost, Portable Fluorescence Correlation Spectrometer for Disease Diagnosis

Nathan E Cordell, **Castine L Donoff**, **Jeffrey Gao**, Sam J Gulinello, Al W Mokris, Jessica E Paulus, Brittany Shirk, & Matthew J Farrar†

The Diagnostic for Viral Diseases Collaboratory team is developing a cost-effective technique for measuring HIV viral load (concentration) from a patient blood sample in resource-restricted regions. Our client is the Macha Research Trust in Zambia. Our method is based on fluorescence correlation spectroscopy – a method for obtaining particle sizes from their diffusion rates – and includes an engineered fluorescent protein probe, confocal optics, low-level light detection, and integrated electronics capable of digital signal processing analysis and providing a graphical user interface. The team is currently working to build and test the individual components of the system, moving towards system integration and a functional exploded prototype in Fall 2021.

Our recent focus has been on developing a system capable of correlating light signals from the optical assembly to display a viral load count. We have designed a modular detector circuit that converts incident photons into a digital signal, which is later analyzed to determine the viral load in a sample. We are also designing a user interface to receive data and display HIV load results in a graphical form using a Raspberry Pi and Arduino touchscreen. The success of the interface design connects the end user to the device and together with the light detector helps to move the team one step closer to final integration.

Cunningham Clubfoot Brace

Jacob R Cornwell, **Benjamin J Mellott**, Brittney D Fouse, Michelle Lo, Clint M Meekins, Sam J Rasinske, Leigha R Southall, Jordan M Witt, & Tim Howell†

Clubfoot is a musculoskeletal birth defect characterized by an inward twisting of an infant's feet. Currently, a series of casts are used to correct the clubfoot, and a boots-and-bar brace is used to maintain the correction. However, this method has concerns with compliance, comfort, and social stigma. Hope Walks and their clinic in Kijabe, Kenya are interested in implementing a new maintenance brace that addresses these concerns. Mr. Jerald Cunningham, CPO, designed and is utilizing a unilateral clubfoot maintenance brace called the Cunningham Clubfoot Brace. He asserts his brace reduces treatment time, lessens social stigma, and increases child mobility. However, to date, there is not enough published research on its biomechanics and patient success rates to confirm his findings.

The Cunningham Clubfoot Brace Collaboratory project seeks to validate the effectiveness of the Cunningham design through biomedical testing and increase brace availability through sustainable manufacturing. To do this, the team is measuring the biomechanical forces applied by the brace with multiple force sensor systems and an infant foot model. The team is assisting Mr. Cunningham in his plans to use injection molding to increase brace production by scanning and creating CAD files of the brace. The team is also completing a failure and reuse analysis of the Cunningham Brace for the clinic in Kijabe. Furthermore, the ongoing clinical study at CURE International's hospital in Kijabe, Kenya, and Dr. Emily Farrar's research paper will provide greater insight into the effectiveness of the Cunningham Brace. These collaborative efforts will allow for further understanding of the effectiveness of the Cunningham Brace and its acceptance as an alternative clubfoot maintenance brace.

Designing a Locally Manufacturable Wheelchair for Nepal

Harrison J Crosley, **Cade K Bender**, **Peter C Hopkins**, Ethan M Barnes, Riley Harro, Levi D Hauger, Joshua J Holley, & Timothy J Van Dyke†

Persons with disabilities in developing countries often lack the basic equipment needed to assist them in their daily lives. International Nepal Fellowship (INF) is a Christian medical organization located in Nepal that provides medical care and assistance to people with disabilities and other conditions. Because importing expensive wheelchairs involves a prolonged and unpredictable border process, INF has reached out to the Collaboratory to ask them to design

a wheelchair that can be manufactured in Nepal from locally available materials and which will withstand the challenges of operating in Nepal's rugged terrain. The Nepal Wheelchair team accepted this challenge and set out to design a wheelchair that can meet this need. The team began to develop a design and, in January 2020, the team traveled to Pokhara, Nepal to gain feedback from the staff at INF on the initial designs and to investigate what materials were available locally there. The team brought back some of these materials in order to construct a prototype. While there are a few remaining components yet to be prototyped, the team has completed most of its first prototype. This prototyping has motivated a number of design changes. Moving forward, the team plans to evaluate the prototype and make improvements to the design.

Adjustable Prone Trolley Design for People Suffering from Spinal Cord Injuries in Nepal

Dylan J Derstine, **Derek A Thrush**, Blake N Clemmer, Abby Miller, Jared T Pavlovich, & Timothy J Van Dyke†

For people suffering from spinal cord injuries, it is important to stay active. However, with spinal cord injuries, the use of a wheelchair isn't feasible. These patients require a prone trolley. A prone trolley is a horizontal pad with four wheels that a patient can maneuver and control while lying in a prone position. Our partner, International Nepal Fellowship (INF), deals directly with patients who suffer from spinal cord injuries on a daily basis. INF, a Christian, medical organization, manages a hospital in Pokhara, Nepal which specializes in treating patients with spinal cord injuries. The Nepal Prone Trolley Team's goal is to provide our partner with a sustainable prone trolley design and create the required manufacturing documentation to enable them to produce the prone trolleys in country at their Green Pastures hospital. The team began our work by researching what a prone trolley is, how it functions and what is currently available. During the research, the team discovered that there weren't many examples of a manually powered prone trolley or critical dimensions for ergonomics for manually powered trolleys. This drove the team to develop testing methods and preliminary designs specifically for INF. Various basic designs were considered, but, through communication with INF, a single design was chosen. Computer modeling of this design was used to decrease the overall weight of the trolley and simplify the frame. With most of the design finalized, the team is ready to begin prototyping next semester.

Land Development - Tree 4 Hope

Corey B Englehart, **Olivia R Allbee**, Caleb R Light, Jorge O Zambrano, & J Scott Heisey†

The Land Development Team has partnered with Tree 4 Hope and Hope Academy in Santa Lucía Milpas Altas, Guatemala to improve the outdoor facilities of the school. Jenn and David Hope-Tringali are the client/partner representatives of the school for this project. The goal of the project is to provide design and construction drawings for three main elements of the proposed land development: (1) a parking lot for buses and school vehicles that enter the site, (2) a single sports court that can accommodate basketball and soccer, and (3) a playground that is directed towards themes associated with STEAM (science, technology, engineering, art, and

math). The team has completed project drawings to allow construction by local personnel, or by student or church mission teams when travel is allowed to resume post-pandemic.

Panama Bridge Project

Mikayla R Eyster, **Crosby Harro**, **Zachary C Hartman**, **Drew W Moyer**, Jordan T Barner, Luke T Fetterman, Noah Ling, Noah W Thrush, & Brian D Swartz†

The Panama Bridge project has partnered with Rio Missions Panama to design a bridge for the village of La Gigi, Panama. The mountain community of La Gigi experiences heavy rainfall during the rainy seasons. A stream runs along the community, separating those in the village from their fields and other communities further up the mountain. While passable during dry seasons, the stream floods and becomes impassable after heavy rains. The residents are effectively cut off from their livelihoods, church, health services, and other communities during this time.

To accommodate this need, the Panama Bridge Team has spent the last two academic years designing an aluminum truss bridge, spanning 90 feet. The design includes a unique construction strategy to deal with challenging site constraints.

Gravity Fed Water System in Guatemala

Joseph Grant, **Ryan G Class**, & Thomas S Soerens†

In the village of Sipacapa, Guatemala, people lack direct access to safe and clean water. The team is partnering with Mennonite Central Committee (MCC) to work to design a gravity-fed water system to serve their needs. The system conveys water from an intake structure connecting three seeps to a collection box. The water then flows by gravity through approximately 3 km of pipe to several locations in the village. Hydraulic calculations were performed to select pipe size and pipe route. The team sized, designed, and located break-pressure tanks and storage tanks as well as tap stands. Materials, costs, and construction schedules were specified. The projected cost of the project is about $10,000. MCC will work with the existing community council for maintenance and oversight of the system. In addition to the technical design, a recommended church-led community water sanitation and hygiene (WASH) program was identified. MCC's partner organization Pastoral works with women's and community organizations in the villages and will cooperate in implementing the WASH program. The team will submit the design to our partner in May 2021.

Rapid Orthotics for Cure Kenya: Development of Safety Testing for 3D Printed Sockets

Gabi E Griffith, **Brandon J Weindorf**, Joey D Andrews, Rachel E Bruns, Elizabeth G Hargrove, Lauren N Seubert, Jarod A Snader, & Jamie R Williams†

The Rapid Orthotics for CURE Kenya team partners with CURE International Hospital in Kijabe, Kenya to implement a customizable 3D printing system to create orthotics and prosthetics. Within Kenya, amputees face stigmatization over disabilities and are often marginalized. The orthopedic workshop empowers amputees by supplying them with devices to overcome adversity. The overarching goal of the ROCK team is to develop a design that will allow the hospital to minimize costs, reduce manufacturing time, and ultimately allowing the hospital to serve more patients. The ROCK team has designed transtibial sockets, ankle-foot orthotics, and rigid hands. The current focus of the project is to evaluate the safety and performance of the transtibial sockets using the ISO 10328 Standard. ISO 10328 defines testing conditions for prosthetic sockets simulating different parts of the gait cycle for people of various masses. The standard also defines three main tests that simulate standing, falling, and walking for each testing condition. These are referred to as Static Proof Test, Static Ultimate Test, and Cyclic Test. Within this presentation, we discuss the design and manufacturing of hardware that allows the Material Testing System (MTS) available at Messiah University to interface with the transtibial sockets developed by the ROCK team. More specifically, we detail the intricacies of the design that allow the tests to be done in full accordance with the ISO 10328 Standard. Finally, we report on the preliminary results obtained from initial Static Proof tests and discuss plans for future Static Ultimate and Cyclic tests.

Sustainable Agriculture

Micah J Hess, **Brandon M Bickom**, Jacob Dean, Madalyn A Heckman, Miggy Matanguihan, Aleesa Wu, & Michelle L Lockwood†

The Sustainable Agriculture team is using a soil-free, closed-loop, agricultural technique called aquaponics to help lift communities out of malnutrition and poverty. Aquaponics techniques decrease the use of natural resources associated with growing crops. The Sustainable Agriculture team is partnered with Sheltering Wings, a non-profit based in Missouri that ministers with an orphanage and women's shelter in Yako, Burkina Faso and TransWorld Radio, a Christian radio broadcasting station located in Benin. The team currently has two best practice prototypes in operation at Messiah as well as a working prototype in Benin.

Throughout this year, our team has focused on reducing power consumption and costs for our clients. A modified ebb and flow prototype was created to suit TWR's resources and successfully installed in Benin. The development of the ebb and flow prototype reduced power consumption by eliminating the need for the air pump. We have also worked towards lowering the costs of water quality testing kits by introducing a Nutrient Film Technique with basil plants. Another focus of this year was improving the general operation of the prototypes. Biological problems were investigated and steps were taken to improve the quality of the prototypes. To support present and future clients, the year will conclude with the completion of manuals including a construction manual, an operations and maintenance manual, and a troubleshooting manual.

Fatigue Testing a Mechanized Percussion Well Drilling System for Water Access in Western Africa

Matthew R Higgs, **Tommy Denlinger**, **Robert H Donley**, Micah T Clark, Benjamin R Gates, & Philip M Tan†

The Mechanized Percussion Well Drilling (MPWD) Collaboratory project seeks to design a simple mechanized well drilling system for drilling shallow water wells in Western Africa. Our client, Open Door Development (ODD), seeks to make water accessible to all in the region, but has had difficulty drilling through hard soil layers. To combat this problem, the MPWD team has worked closely with Mr. Joseph Longenecker to develop a mechanized percussion well drilling rig that is capable of drilling through these harder layers. Currently, the MPWD team is seeking to provide recommendations to improve the lifetime of our client's new, fully mechanized rig design. This year, our team's work has been focused specifically on analyzing the lifetime of the rig's driveline chains and also on its frame. For the driveline chains, the team will be conducting fatigue testing on a model of the driveline system to determine which type of chain should be used on the rig. To determine the lifetime of the frame, the team will be performing a series of static, buckling, and fatigue finite element analyses on the rig's frame. The most recent accomplishments of the MPWD team have nearly proved that their design for the loading application will be feasible for use on the actual testing rig and that multiple studies of finite element analysis can be performed to simulate the different rig frame loading scenarios.

Energy Monitoring & Management System (EMMS)

Zachery D Holsinger, **Ben Weaver**, **David C Williams**, Bennett A Andrews, Seth Wilcox, & Tom Austin†

The Energy Monitoring and Management System (EMMS) is developing an electrical power meter to help make electricity more available in energy impoverished regions of the world. The meter fills a unique niche for energy tracking and regulation within micro-grid systems. The EMMS project has partners in Burkina Faso and Zimbabwe: Open Door Development (ODD), the Institut Missiologique du Sahel (IMS), and the Theological College of Zimbabwe (TCZ). Ties are also maintained on a regular basis with IEEE Smart Village for potential future widespread system implementation.

Recent work on the EMMS meter has been focused on resolving the last few remaining bugs, establishing a robust communication system, and developing a centralized server-based interface which aids with meter configuration and administration. The team has also begun several future developments which include datalogging and remote access features.

Fluency Assistance Device (FAD): Masker Impact & Development

Michael D Jenkins, **Corey Bean**, **Larry A Vega**, Jake T Finkbeiner, Chad M Long, & Harold R Underwood†

Around seventy million people internationally have a stutter, a form of a fluency disorder. Some fluency assistance devices are available to the public, but most are highly expensive or unreliable. The Fluency Assistive Device (FAD) team seeks to assist a niche community of these

individuals who currently rely on a device known originally as the Edinburgh Masker. To best reach this community, FAD is partnering with Dave Germeyer, who has invaluable experience repairing these masker devices for his clientele. To help with his efforts, FAD is developing two new redesigned versions of the masker to increase its portability, functionality, and cost-effectiveness. The first solution, known as the Analog Masker (Version 1.1), focuses on updating components and fixing flaws with the original. A prototype of the Analog Masker v1.1 has been developed, tested and is currently being revised based on the results. Revisions include updating the hardware and finalizing the power supply circuitry. The second solution, known as the Digital Masker (Version 1.0), will use a Bluetooth-enabled microcontroller to achieve masker functionality. Bluetooth audio output for the Digital Masker has been tested, and two algorithms have been created for the masking output. The supporting software for the Digital Masker is nearing completion. Development of the schematic and layout design for the hardware of the Digital Masker is also underway.

A Sustainable Mobility Solution for Persons Living with Disability in Burkina Faso

Faith N Kerlen, **Katie Bunch**, **Sam Fino**, Rachel Delate, Erin T Logsdon, Julia M Neborg, Rachel Rashford, Joey Sinsel, John Meyer†, & David T Vader†

The Sustainable Mobility project of the Collaboratory empowers people living with a disability in rural West Africa to pursue educational and work opportunities and more fully participate in family and community life. Our electric, 3-wheeled, off-road wheelchair has transformed the lives of dozens of clients through partnerships with the Center for the Advancement of the Handicapped in Mahadaga, Burkina Faso and the Center of Hope in Fada, Burkina Faso. Now, to reach more people in new locations and with more partners, Sustainable Mobility is working to reduce manufacturing time and cost, author image-driven fabrication guides to enable local fabricators to build trikes, create instructional trike assembly videos, and develop supply chains to bring parts and materials to build sites. We seek to put local fabricators to work building tricycles wherever they are needed.

Preparing For Extended Field Tests of the Intelligent Water System

Daniel J Labrie, **Evan Freed**, Josiah J McCarthy, & Randall K Fish†

The Intelligent Water System, which improves access to clean water by autonomously monitoring and reporting on the health of hand pumps in developing countries, has been under development for several years. This development has included short-term prototype field tests in several countries. The design has matured to the point that an extended field trial to demonstrate performance and reliability has been requested by our client. In light of this, the team analyzed the systems returned from our most recent prototype field test, implemented needed changes and has begun the manufacture of the five systems intended for the first extended field trial in Burkina Faso. This talk will focus on the design changes between our most recent prototype test and the extended test systems as well as the design and fabrication of an installation jig requested by our field installation partners.

Village Water Ozonation System

TJ J Malanga, **Benjamin K Burlew**, Grant Brubaker, Ruth C Galyen, Sam B Stone, Ray Knepper†‡, & Michelle L Lockwood†

The Village Water Ozonation System (VWOS) team's core mission statement is to provide economically sustainable and culturally sensitive drinking water solutions for communities, to empower communities with the ability to properly maintain their drinking water supply, and to transform people's lives by decreasing the occurrences of waterborne diseases.

Currently, the VWOS team is partnering with Friends in Action to design and implement two drinking water treatment systems for the community living on Rama Cay, an island in the Bluefields Lagoon on the eastern coastline of Nicaragua. The wells on the island are contaminated with E. coli and other bacteria and contain high levels of salt that cause the water to be unhealthy, distasteful, and corrosive to metal equipment in the system. The team hopes to design a system that will disinfect the water, remove salinity from the well water with a safe and efficient disposal of all byproducts, and decrease corrosion agents.

3D Mapping Jungle Airstrips with a Drone

Geoffrey D McClary, **Micah D Lehman**, **Kyle E Miller**, Pauline Deutcheu, & Dereck Plante†

Missionary aviation pilots often must land their planes on remote airstrips that might be unsafe due to runway obstructions such as encroaching vegetation or large objects that were unknowingly placed on the runway. The Falcon Unmanned Aerial Vehicle (UAV) team is partnered with Indigenous People's Technology and Education Center (ITEC) to develop an imaging system using a UAV to scan these airstrips to detect these obstructions. ITEC was founded by Steve Saint, the son of martyred missionary Nate Saint, to develop technologies to aid missionaries and indigenous peoples in their work. The Falcon UAV team focuses primarily on the use of automated 3D mapping and photogrammetry by drones to help identify obstructions to pilots landing on remote airstrips. Through our research and experimentation, the team is recommending the DJI Mavic Platinum Pro in combination with the Drone Deploy 3D mapping software.

SkinSafe: A Bacterial Skin Model for Studying Silicone Prosthetic Liners in Kenya

Hailey C Miller, **Esther C Seeland**, Keera L Dupler, & Philip M Tan†

The interface between an amputee's residual limb and prosthetic liner is at risk for high levels of bacterial growth which can lead to skin breakdown and in the worst cases, infection. This is particularly a concern in low-resource settings, such as that in Kijabe, Kenya, in which a lack of clean water can result in poor hygiene. It is believed that silicone prosthetic liners have a sealing effect that could heighten this issue. The SkinSafe team is working to conduct a prosthetic liner study to investigate these concerns. To accomplish this goal, the team needs a model to

accurately represent the interactions at the skin–liner interface. Traditional cell culture procedures incubate bacterial plates at a constant environmental temperature. However, the skin–liner interface is a dynamic environment, as the human body provides heat and water to the surface of the skin. Thus, the SkinSafe team has developed a bacterial skin model that captures the dynamic behavior of the skin–liner interface by incorporating a one-sided heat and water source that mimics the human body. This model will be used in future prosthetic liner studies as the team investigates silicone liner alternatives and developing minimum hygiene protocols for amputees in low-resource settings.

Landmine Neutralization: Air Excavation Unit

Evan Poust, Josh Card, Ethan R Cornwell, & Donald G Pratt†

The Landmine Neutralization presentation will show our team's progress towards designing and prototyping an air excavation unit. To accomplish this, the project team is working with the HALO Trust, the world's largest demining non-governmental organization. The HALO Trust works in many countries to remove the remnants of war, including improvised explosive devices (IEDs) and unexploded ordnance (UXOs). This project seeks to help deminers by providing a device that blows air at high velocity to clear dust and debris away from potential IEDs and UXOs in war-torn areas. The prototype is intended to be easily and quickly attached and detached from the custom excavator rakes that are used in the demining process. The current design is modular and consists of a hydraulic motor powering a fan from a backpack leaf blower, all of which is assembled within a steel frame which is attached to the rake. Due to circumstances beyond our control, our project will be closing at the end of this semester, which is sooner than anticipated. This means the goals of our project have been narrowed to having a functional prototype and relevant documentation that we can present to our client.

Hybrid Thermal Lance

Josiah Stitt, **Lucas C Honebrink**, **Frank G Honey**, **Emily L Keane**, & Donald G Pratt†

The Hybrid Thermal Lance (HTL) is a device used to burn through the outer casing of landmines to safely destroy the explosive inside. Designed for the team's client, HALO Trust, the HTL has proven to work well in destroying explosives, which has been shown by field trials conducted in a number of countries, including, but not limited to Afghanistan and the Republic of Georgia. The HTL works by igniting acrylic burn tubes, which both act as a fuel source and focus the flame on a specific location on unexploded ordnance. The system is controlled via a user-friendly, rugged control box that can run the HTL automatically and allow the device to burn for different lengths of time upon user command.

Trans World Radio - Culvert Design

Daniel O Thomas, **Darren J Heisey**, Warner C Hockenberry, Logan J Horst, Seth M Kline, Gabriel J Tiday, & J Scott Heisey†

Trans World Radio (TWR) is a mission organization focused on broadcasting the Gospel around the world. TWR now serves 190 countries by transmitting in 275 languages, using radio to deliver the message to as many people as possible. The organization's West Africa Transmitter Site in Benin currently has accessibility problems due to high streamflows and saturated ground conditions during the rainy season. The site also needs a secure perimeter to reduce trespassing and theft.

To address these issues, our client, Garth Kennedy, Director of the West Africa Transmitter Station, has asked the team to design two culverts, one at the upstream property boundary and one at the downstream boundary. Culverts are advantageous for this scenario because they can act as a bridge, while the pipe size can be restricted to inhibit trespassing. Once the culverts are built, the fence and perimeter road can be extended over them.

For both sides of the property, the team has designed a series of U-shaped, pre-cast concrete box culverts. The team calculated the design flows based on rainfall data and the topography of the site to determine the size and number of box sections. The team has also designed the culverts and the supporting concrete structures to bear the load of vehicles and the machinery on-site. TWR plans to construct the pre-cast culverts on site, and then build the supporting structures and install the culverts during their dry season.

Muscle Activated Prosthetic

Samuel F Whittle, **Sam Sparks**, **Eddie J Yesilonis**, Lindsay L Haseltine, Jaymie R Monday, Meghan L Sampson, & Tim Howell†

Due to the rapid growth of children and the cost of myoelectric technology, children are not always given the same opportunities to use myoelectric prosthetics as adults. The Muscle Activated Prosthetic (MAP) team is working on designing a myoelectric prosthetic that will cost under $1,000 as opposed to the $10,000-$20,000 cost of commercial myoelectric prosthetics, making it more affordable for our client. The basic mechanism by which this device operates is electrical signals emitted via muscle contractions that are detected and used to open/close the hand. Our product is built with EMG sensors, electrodes, a microprocessor, linear actuators, fishing line tendons, a battery and a specialized printed circuit board (PCB) to connect it all together. This semester, the team has assembled a fully functioning prototype that has been sent to the client and is currently working on making a second and final prototype. The next steps include incorporating user feedback and/or size changes as well as small improvements such as aesthetic changes and sanding surfaces. This final prototype will then be reviewed by our partner, Ability Prosthetics for their assessment and approval before shipping to our client and beginning the process of pivoting our team's overall direction and goal in the 2021-22 academic year.

Better Pumps: Reliable Handpump Infrastructure

Emma L Workman, **Caleb E Danehower**, **Darren Kulp**, **Jacob Valentine**, Matt J Caldwell, Andrea Hunsberger, Darin M Landis, Joshua L Maxson, Tony Beers†‡, Matthew Schwiebert†‡, & David T Vader†

Approximately 90 million people in Africa lack access to safe drinking water, despite having water infrastructure installed in their community. The India Mark II and the Afridev handpumps are among the most widely used handpumps in the world. Sadly, studies show that approximately 30% of these handpumps are non-operational due to failures of the bearings, seals, head flange, and other common components. The Better Pumps team of the Collaboratory provides engineering support for partners who are working to improve handpump sustainability. We partnered with Tony Beers and AlignedWorks to validate a bearing test methodology for the India Mark II handpump. By modifying the loading conditions in our handpump test machine, we were able to replicate failures observed by AlignedWorks in a field trial of their bearing design. We partnered with Matt Schwiebert and Living Water International to test new seal designs for the India Mark II and Afridev handpumps and to measure head flange deflections in the India Mark II handpump. Seal performance data collected by the team was used to validate a new design in advance of field trials by Living Water International. Head flange deflection data was collected for partner benchmarking of their computational analysis. Test methodologies and results are reported.

Exercise Science

The Role of CYP1A2 and ADORA2A in Individual Response to Caffeine Consumption Under Anaerobic Conditions

Kathryn J Armstrong, Michael Shin†, H. Scott Kieffer†, & Michael Sedhom

Caffeine is the most commonly used psychoactive drug in the world and has wide implications in medicine, athletics, and public health. Studies have shown that metabolism, clearance, reception, and response to caffeine vary significantly among individuals. The pharmacokinetics are primarily dictated by the cytochrome p450 enzyme CYP1A2 while the adenosine neuroreceptor ADORA2A heavily influences the drug's pharmacodynamics. Polymorphisms of the -163 A>C CYP1A2 and the 1976 T>C ADORA2A are thought to influence these interindividual responses; therefore, the purpose of this project is to determine the effect of caffeine consumption on anaerobic exercise. 12-15 female college athletes completed two maximal WAnT30 anaerobic bike tests on a Velotron cycle ergometer. The participants ingested a capsule of caffeine ($5mg \cdot kg^{-1}$ bodyweight) or a placebo capsule (maltodextrin) one hour prior to testing. The order of the bolus was randomized, counterbalanced, and administered in a double-blind manner. Peak power ($W \cdot kg^{-1}$), anaerobic capacity ($W \cdot kg^{-1}$), and total power output ($W \cdot kg^{-1}$) were recorded during each test. Buccal epithelial cells were collected using a 0.9% NaCl mouth rinse with DNA extraction conducted using proteinase k to lyse cells and collection using QiAmp Mini spin columns. Allelic discrimination was obtained using TaqMan® SNP Assay for CYP1A2 (rs762551) and ADORA2A (rs5751876) and a One-Step qPCR. Each sample was run in duplicate positive and negative quality controls. Each variable was analyzed using a factorial ANOVA with repeated measures ($p > 0.05$).

The Influence of Caffeine and the C957T (DRD2) Polymorphism on Measures of Unilateral and Bilateral Motor Tasks

Abigail Beveridge, Rachel Caldwell, H. Scott Kieffer†, & Michael Shin†

The effects of caffeine on task performance and attentional focus during movement have been suggested to be influenced by the interaction between the adenosine (ADORA2A) and dopamine (DRD2) receptors in the dopaminergic pathways and specific centers in the brain. Individual polymorphisms of 1976 T/C (ADORA2A) and C957T (DRD2) are thought to influence the varied response of caffeine during movement. **PURPOSE:** The purpose of this study is to determine the influence of caffeine, 1976 T>C ADORA2A polymorphism, and C957T (DRD2) polymorphism on measures of unilateral and bilateral motor tasks. **METHODS:** The Purdue Pegboard Test and the Minnesota Dexterity Test were used to assess fine motor abilities following both placebo or caffeine ingestion (5mg·kg-1 bodyweight). Buccal epithelial cells of each subject were retrieved by a 0.9% NaCl mouth rinse. To collect the DNA samples, the cells were lysed via proteinase k and isolated with QiAmp Mini spin columns. The polymorphisms were identified using a TaqMan® SNP Assay for ADORA2A (rs5751876) and DRD2 (rs1800497) and allelic discrimination via One-Step qPCR amplification. All samples were run in duplicate with negative and positive controls. **RESULTS/DISCUSSION:** The Results and Discussion will be presented at the Symposium.

Effects of Caffeine and CYP1A2, ADORA2A Polymorphisms on Exhaustive Anaerobic Performance

Adam D Cole, H. Scott Kieffer†, & Michael Shin†

Individual responses to caffeine are suggested to be genetically influenced by polymorphisms of the cytochrome P450 enzymes, specifically the -163 A>C CYP1A2, for metabolism in the liver and through the adenosine receptor, 1976 T>C ADORA2A, for sensitivity of specific target cells. Individuals with the AA variant are caffeine responders, while those with the AC/CC variants are caffeine non-responders. ADORA2A TT variants demonstrate an increased sensitivity to caffeine compared to TC/CC variants. **Purpose** To examine the effect of caffeine and CYP1A2 and ADORA2A polymorphisms on anaerobic power during exhaustive exercise. **Methods** Fifteen elite NCAA male athletes (age=20.1 yrs, weight=77.4 kg, height=176.7 cm) participated in a double-blind study. Subjects performed two separate 90-s Wingate Tests (WAnT90) separated by two to four days on a Velotron cycle ergometer, resistance=0.05 kg·BW(kg)$^{-1}$. Subjects ingested a bolus of caffeine, 5mg·kg^{-1}BW, or a placebo (maltodextrin) one hour prior to each trial that were administered in a randomized/counterbalanced design. Peak power (W·kg^{-1}), total power (W·kg^{-1}), and average power (W·kg^{-1}) were calculated for the 90-s and each 30-s interval. Buccal epithelial cells were collected using a mouth rinse, 0.9% NaCl, and DNA was extracted via spin columns and proteinase k. Allelic discriminations for CYP1A2 (rs762551) and ADORA2A (rs5751876) were procured via an assay and a One-Step qPCR amplification. Samples were run in duplicate, with positive and negative controls. The data was analyzed using a factorial ANOVA with repeated measures ($p > 0.05$) for each variable. **Results/Discussion** The results and the discussion will be presented at the Symposium.

The Effects of Static and Dynamic Stretching on Muscular Power in Dancers and Jump Athletes

Elizabeth R Vlieg, Sydney De Poto, Grace Brewster, H. Scott Kieffer†, & Abigail K Gibson

Flexibility and power output are two critical components of dance performance; however, recent research suggests that acute bouts of stretching may decrease muscular power. **PURPOSE**: The purpose of this study was to compare the effects of dynamic stretching (DS) and static stretching (SS) on muscular power in dancers. **METHODS**: 12 female, collegiate dance majors volunteered for this study. The subjects attended a familiarization session, gave informed consent, and were oriented to all testing procedures. Three different experimental sessions were conducted in randomized order for each stretching condition: DS, SS, and no stretching (NS), with a minimum of 48 hours between testing days. The sessions began with a warm-up (5 min walk) followed by a guided stretching protocol for each condition. Muscular power (peak torque, Watts) was obtained for the plantar/dorsiflexors with an isokinetic dynamometer (60o/s and 180o/s) and vertical power (W/kg BW) was obtained using both a squat (SJ) and countermovement jump (CMJ). One-way ANOVA with repeated measures with Tukey HSD post-hoc tests was conducted to determine significance ($p \leq 0.05$) for each variable. **RESULTS**: The results indicated that there were no significant differences for the power output of dancers after incorporating DS, SS, or NS as part of a warm-up. Isokinetic muscular power isolating the plantar flexors demonstrated no significant difference at 60o/s (DS, 57.1 $\pm$ 22.7 W; SS, 57.8 $\pm$28.4 W; NS, 62.7 $\pm$ 19.3 W, $p = 0.82$) or at 180o/s (DS, 28.3 $\pm$ 9.50 W; SS, 30.5 $\pm$ 14.7 W; NS, 32.2 $\pm$16.2 W, $p = 0.76$). In addition, measurements of power incorporating whole body power demonstrated no significant difference in jump height for the SJ (DS, 21.05 $\pm$ 3.36 cm; SS, 20.83 $\pm$3.55 cm; NS, 20.9 $\pm$ 3.14 cm, $p = 0.893$) or the CMJ (DS, 23.8 $\pm$ 3.9 cm; SS, 23.7 $\pm$ 5.7 cm; NS, 24.7 $\pm$6.8 cm, $p = 0.98$). **CONCLUSION**: Although some research suggests that varying forms of stretching may decrease muscular power, this study suggests that neither acute static stretching nor dynamic stretching will elicit a significant change in muscular power in dancers. The jump athlete data is still being collected at the time of abstract submission.

Mathematics

Applications of Options Greeks and the Black-Scholes Equation

Ryan B Althoff

Options Greeks are embedded in the definition of the Black-Scholes equation and have direct applications to the financial markets, specifically through the pricing of options contracts. This presentation endeavors to provide an overview of the Greeks by contextualizing increased retail investor participation and its collective influence on short-term price volatility for select equities. In conjunction with a consideration of the underlying theoretical mathematics, a practical example is presented to demonstrate contemporary relevance and to evaluate the implications for established hedge funds and other financial firms.

Staircase Tableaux

Kasey B Caras, **Maelyn Elder**, **Ian Parzyszek**, & **Morgan Zimmerman**

In this presentation, we will discuss our research on staircase tableaux, which are mathematical objects in the field of combinatorics that have applications in physics and biochemistry. Staircase tableaux are constructed in a similar way to Sudoku puzzles. Boxes are aligned in a staircase

shape and these boxes are filled with alphas and betas depending on a few simple rules. In our research, we determined the probability of staircase tableaux that have an alpha followed by a beta on the main diagonal as well as a beta followed by an alpha. These results are interesting because of the applications staircase tableaux have in other disciplines.

Is Your Professor Exaggerating? Investigating the Importance of Conditions in One-Sample Testing

Emily Decker

The conditions of a given hypothesis test should always be taken into account when deciding what test best fits the given data. But what happens when the conditions of normal, rank based, or sign based tests are not fully met? After an introduction to the nuances and power of the usual t, Wilcoxon sign rank, and Fisher sign test, this paper will analyze the efficiencies of these tests and analyze how each responds to violations of the conditions. Through the use of a significant number of iterations of SAS created data sets, it will be shown how frequently normal, symmetric, and non-symmetric data affect the theoretical outcomes of each test. These results will play an important role in understanding the impact conditional violations have on the conclusions and overall which test is best in each data setting.

Sabermetrics

Paul M Hansel

In 1971, the Society for Baseball Statistical Research (SABR) was founded. This society was the first to coin the term “sabermetrics” which refers to the empirical analysis of baseball. Prior to sabermetrics, decisions about players, teams, and coaches in baseball have always been made based upon a subjective interpretation. However, this society started a massive movement of applying an objective-based science such as mathematics to the subjective world of baseball. This combination of an objective science and a subjective sport makes sabermetrics an interesting part of mathematics to explore.

Fisher's and Neyman's Tests in Treatment Analysis

Sarah E Hartman

Fisher’s and Neyman’s tests are both randomization-based tests that can be used to detect effects of experimental treatments; however, they differ significantly, particularly in the construction of the hypotheses. As a result, each test has specific limitations: in certain cases, Fisher’s test can result in either Type I or Type II errors, and Neyman’s test relies on approximations of variance and normality. This project offers a comparison of these two tests and an analysis of their limitations through simulations.

Mathematics at Play: A Study of Game Theory Applications

Reece J Horne

The purpose of this presentation is to give an example of applied mathematics being used to enhance mathematics instruction. We show several examples of game theory applications in

mathematics where connections are made to the mathematical topics of saddle points on three-dimensional graphs and the use of matrices in representing payoff functions. The main examples explored are two player games where we explore crucial information about each scenario such as the Nash Equilibrium point. The discussion follows as to how these examples can be worked into a mathematics curriculum for enrichment. We conclude by evaluating how effective practices such as game theory applications encourage appreciation, engagement, and performance in instructional settings.

Sprouts & Brussels Sprouts

Joshua J Kantner

Since its conception in the early 18th Century, graph theory, or the mathematical study of graphs, has become a useful tool in solving and analyzing a variety of problems. One of these problems is the seemingly simple pen-and-paper game of Sprouts created by John Conway and Michael Paterson in 1967. Sprouts is an easy game to learn and play for the casual player, but it has proven to be a much more difficult game from a competitive point of view. In order to get a better understanding of how to win any game of Sprouts, the game's properties can be analyzed and studied mathematically with the help of graph theory. The results of such analyses can be helpful in determining each game's outcome as well as understanding new variants of the game when changing a few mechanics.

Deus Absconditus and Game Theory

Devan K Miller

Deus Absconditus is a Latin phrase used by Martin Luther and John Calvin in explaining the hiddenness, or unknowability of our God. In Isaiah 45, the prophet Isaiah writes "Truly you are a God who has been hiding himself, the God and Savior of Israel." Why is it that Christian faith accepts that God cannot be known to us? The study of Game Theory might have an answer. Game Theory is a fascinating extension of Mathematics that seeks to understand risk, analyze social interaction, and provide researchers with an understanding of why "players" behave the way they do. By researching Game Theory, I hope to support my Christian faith by providing an understanding of why our creator has chosen to be unknowable, and require belief without seeing.

Ordering Polynomial Rings

Abigail Mitchell

Polynomial rings are a foundational concept in understanding the characteristics of number systems. Through the examination of specific polynomial rings and their properties, it is possible to develop methods of ordering the polynomials within these rings. In this presentation, a broad introduction to polynomial rings will be provided in addition to an in-depth exploration of ordered polynomial rings and their properties. An understanding of ordered polynomial rings is useful for further development in both number theory and calculus.

Proof and Application of The Central Limit Theorem

Ian Parzyszek

The broad applications of the Central Limit Theorem cannot be understated. It allows one to approximate other distributions with that of a normal distribution, which is crucial in hypotheses testing and modern day statistical analysis. The properties of this theorem have been observed for hundreds of years but its actual discovery and proof did not happen until 1810. The elegance of this mathematical proof is shown in this paper, along with its applicability to modern day hypotheses testing using real world data.

The Tower of Hanoi and Recursive Sequences

Sunny Shao

The Tower of Hanoi is a famous puzzle invented by a French mathematician Edouard Lucas in 1883. The task of that is we are given a tower of eight disks, initially stacked in decreasing size on one of three pegs. The objective is to transfer the entire tower to one of the other pegs, moving only one disk at a time and never moving a large one onto a smaller. How do we solve this problem? This problem may not appear to be related to mathematics, but we can actually use a recursive sequence to solve it. Sequences are mathematical objects discussed in Calculus II and the Tower of Hanoi is just one example of how sequences can be applied in the real world.

The Black Scholes Model

Katie R Stottlemyer

In this presentation, I will discuss the Black-Scholes Model which is an important mathematical model in financial theory for pricing options. I will discuss the background of this model and an example of how it is used. I also will discuss some contradictions that have been found when using the model and how it is still being used.

Mathematical Approaches to Political Gerrymandering

Morgan Zimmerman

With the speculation that electoral maps are divided into boundaries that favor one election outcome over another, the Supreme Court has deemed political gerrymandering as unconstitutional. Various mathematical models have been proposed in an attempt to combat the lack of standardized parameters for detecting the extent to which this occurs. The efficiency gap metric is a simple formula introduced as an evaluation of how extensive districting has favored one party over another within a specific boundary. With the desire to produce a more statistical-heavy method, a hypothetical state is explored and various iterations of boundaries are illustrated in an attempt to find the most effective algorithm. It is concluded that Markov chains could prove useful in the area of redistricting.

Physics

Interaction of Dark Matter Dark Photon Particles with Plasma in the Universe

Nathan Branson & Abaz Kryemadhi†

Dark matter is hard to detect because it does not interact with normal matter with a few exceptions. A dark matter candidate of interest is the dark photon which could convert with some probability to a regular photon. The dark photon is not directly detectable due to small interaction. There is theoretical motivation however to increased conversion to regular photon when it is in the presence of a plasma. We have explored what dark photon masses and what plasma conditions create favorable conditions for its detection. Previous works look at the extreme cases of mass being much larger or smaller than the plasma frequency, while we look for plasma frequency at a similar magnitude to the dark photon mass. We use the earth's ionosphere as the medium for this project. The areas of focus are the regions of plasma frequency matching the dark photon mass where high conversions might occur due to resonance.

This is a computational research project, which uses many concepts from computer science in addition to physics. The analysis for this project is done using the python programming language.

Laser Sheet Imaging of *Arabidopsis thaliana* Roots

Dalton J Daugherty & Matthew J Farrar†

Plant root systems are inherently 3-dimensional structures. To effectively understand these root systems—such as the model organism *Arabidopsis Thaliana*— it is necessary to develop and employ imaging methods capable of generating 3-D data sets. One such method is laser sheet imaging of plant roots grown in transparent media, such as in hydroponic/aquaponic cultures or hydrogels. We have been working on a custom-built laser sheet system to achieve this end. This modality requires the coordination of smooth translation stage motion, camera exposure, and camera frame rate settings. Multiple graphical user interfaces (GUIs) written in custom Python software are necessary to guide the user through the imaging process. This software communicates with the stage, controls laser power, and triggers the camera in order to synchronize motion and imaging to get clear images with definite dimensions. This system can then be used to perform time-lapse imaging of plant root structures to analyze how environmental factors affect root structure and development.

Development of a Detector System for Dark Photon Dark Matter

Ryan J Thurber, Abaz Kryemadhi†, & Niklas Hellgren†

Dark Matter is assumed to exist because of the gravitational effects on stars as they move around galaxies. Efforts have been made to discover its properties in ways such as studying possible inelastic collisions in particles, but they have not yielded conclusive results. Dark photons have been motivated from theory as dark matter candidates. They can convert to regular photons at a small rate. Therefore, my study will strive to discover the regular photons emitted by the dark photons. This can be done by setting up an experimental area shielded from all light sources where the existence of a dark photon would be manifested by the appearance of regular photons in a completely dark area. To increase the chance of detection, a spherical mirror will be used to reflect the photons towards our detector, which increases our effective surface area. So far, I have worked with the photodetector and the data acquisition system in order to understand detector performance in a vacuum, which allows for photons of a shorter wavelength to last longer before they are absorbed, and how to determine the background noise from actual signals.

Development of a Detector for High Energy Gamma Ray Studies

Brandon J Weindorf, Aeowyn Kendall, Al W Mokris, Abaz Kryemadhi†, & Matthew J Farrar†

Despite the rapid progression of knowledge present in the field of particle physics, many mysteries still abound that have yet to be fully solved and understood; one such example is that of dark matter as there is relatively little known about it and many experiments today endeavor to detect it. This project focuses on the use of a relatively new technology, Silicon Photomultipliers (SiPMs), to detect high energy particles incident from space. It is currently hypothesized that dark matter serves as the source of high energy particles that are particularly more energetic than those incident from the sun and supernovae. The Silicon Photomultipliers are coupled with deionized water and scintillating materials to generate and absorb Cherenkov Radiation. While classic photomultipliers (PMTs) can solely be used for this experiment, they cost more and require significantly more power than that of SiPMs. While SiPMs are significantly smaller than PMTs, use of a large volume of water can increase the effective detection range of the SiPM. We detail the design, construction and overall development of multiple Cherenkov Radiation detectors. Moreover, we report on the performance of each design and discuss comparisons among each combination of models. Finally, we present the new data programming tool that has been developed to aid in the analysis and visualization of all collected data.

Acridine Orange as a Novel Tool for Identifying Partially Double-Stranded DNA

Eli Whitehead-Zimmers & Matthew J Farrar†

One of the distinguishing features of hepadnaviruses is the presence of partially double-stranded DNA. Hepatitis B is one of the well-known examples of this virus family, yet there remains a paucity of effective methods for probing this nucleic acid structure. This study investigates the use of the dye Acridine Orange (AO) as a possible probe for understanding these genomes. AO is of interest for identifying partially double-stranded DNA because of its unique spectral properties. Specifically, AO fluoresces red (~650 nm) if bonded to single-stranded DNA (ssDNA), or green (~525 nm) if bonded to double-stranded DNA (dsDNA) by intercalation. To test this system, confocal optics and single-photon counting modules were used to probe dsDNA or ssDNA at the single-molecule level. The ratios of green/red fluorescence by AO-stained DNA in varying concentrations of AO were assessed.

Poster Abstracts

Biology, Cellular & Molecular

Efficacy Analysis of Zebrafish, *Danio rerio*, as a Novel Demyelination Model Through Exposure to Cuprizone-infused Food

Halle L Becker & Jennifer K Ness-Myers†

The central nervous system demonstrates its functional reliance on glial cells in support of neurological function primarily through its use of oligodendrocytes and the myelin they produce. Damage to myelin is caused by different pathways, one exemplified in Multiple Sclerosis (MS), where the immune system incorrectly attacks healthy tissue of the CNS. Most research focusing

on these biological processes of demyelination has relied on rodent models. However, zebrafish, *Danio rerio*, are an ideal model for these goals because they are transparent and therefore are easy to monitor for demyelination and remyelination progression. In this study, the viability of zebrafish as a model for demyelination and remyelination research studies was evaluated. The initial steps were the development of cuprizone-infused food and the design of a cuprizone drug feeding protocol. Following treatment, myelin staining techniques were used to assess the level of myelin damage. The goal of this study is to develop a reliable new demyelination research model that will improve imaging of myelin damage and repair during demyelinating diseases.

Laboratory Culture of *Plasmodium falciparum* with Cryopreserved Human Erythrocytes

Michael L Bellerose, Mina Diep, Matthew T O'Neil, & Lawrence M Mylin†

Uninfected human blood free of anti-malarial medications is essential for the laboratory culture of *Plasmodium falciparum*, the causative agent of severe malaria, but is of limited availability in sub-Saharan Africa where research is conducted by the Macha Research Trust. Cryopreservation of erythrocytes obtained from uninfected donors in the US would provide an adequate supply. Our goal has been to identify methods of freezing that will preserve the capacity of RBCs to support asexual propagation and gametocyte formation by *P. falciparum* strain NF54 in serial asynchronous culture, as well as the shelf life of RBCs stored at -80ºC. We have investigated RBCs cryopreserved using two types of freezing solutions: 1) a mixture of hydroxy ethyl starch (HES) and polyvinyl alcohol (PVA); 2) glycerol supplemented with human serum. Our results reveal that blood frozen with hydroxy ethyl starch and polyvinyl alcohol failed to support asexual propagation of *P. falciparum* beyond three serial passages. Blood frozen using the glycerol & serum solution was able to support asexual propagation through at least nine serial passages, and later passages were capable of gametocyte generation. NF54 parasites maintained in serial culture using glycerol/serum-frozen remained unable to proliferate when serially passaged using HES/PVA-frozen blood, confirming the absence of mutations that may have adapted the parasite to long-term culture in frozen blood. These results will be discussed with respect to essential functions of the apicoplast, a parasite organelle required for erythrocyte invasion.

Quantifying In Vitro Matrix Deposition to Elucidate the Target of Proglumide-mediated Fibrotic Inhibition in Pancreatic Cancer

Silas Buckwalter & John F Harms†

Pancreatic cancer has a 5-year survival rate of 8%, and the average prognosis has not improved in recent history. Chemotherapy is the primary clinical approach but carries poor efficacy and offers little improvement in patient survival due to the highly fibrotic collagen-rich tumor microenvironment. Our lab has previously confirmed that the gastrin receptor antagonist, proglumide, significantly decreases pancreatic tumor fibrosis *in vivo*. *In vitro* efforts to elucidate the cellular target of proglumide have been unable to demonstrate a decrease in the mRNA expression of collagen (COL1A) or its post-translational processing enzymes, in either myofibroblast-like pancreatic stellate cells or cancer cells. We hypothesize that quantification of

deposited collagen matrix *in vitro* will better reflect the decreased fibrosis apparent *in vivo*. Utilizing the Sirius Red/Fast Green protein staining assay, collagen deposition was measured in cultured cells. To normalize collagen to potentially disparate cell proliferation rates, we confirmed that the alamarBlue cell quantification assay does not interfere with subsequent Sirius Red/Fast Green protein staining. Human pancreatic stellate cells (RLT-PSC), mouse pancreatic cancer cells (Panc02), and human pancreatic cancer cells (PANC-1) were stained 4 days after seeding. Significantly higher collagen was evident in Panc02 and PANC-1 wells compared to stellate cells ($p<0.05$). To test the impact of proglumide treatment, cells were treated with proglumide (200 μg/ml). Lines showed no change in normalized collagen deposition after 4 days compared to untreated controls. These preliminary data reflect the unchanged COL1A mRNA levels previously apparent in the stellate and cancer monocultures. Further inhibition studies will utilize co-cultures to address potential heterotypic intercellular communication between these cell types, characteristic of the tumor microenvironment.

Altered Heavy Metal Stress Response of Mutant *Arabidopsis thaliana* Plants in the Presence of Nickel

Annelyse M Cruzan, Michael Shin†, & Richard W Schaeffer†

While heavy metals such as zinc and nickel are necessary for several life-sustaining processes, elevated levels of these metals pose a threat to humans, plants, and other forms of life. Researchers have found that certain plants known as hyperaccumulators tolerate higher levels of heavy metals than non-hyperaccumulating plants. They can be planted in heavy metal contaminated soil to take up excess heavy metals, thus decontaminating the soil in a process known as phytoremediation. In this research project, we will examine the altered heavy metal stress response of two Columbia-line *Arabidopsis thaliana* mutants that we hypothesize are hyperaccumulators of heavy metal divalent cations. This hypothesis will be tested using nickel tolerance and accumulation assays. The tolerance assays will compare the root length, a key indicator of plant health, of the two mutant *A. thaliana* plants against the Columbia-line wildtype plant in varying concentrations of nickel. The accumulation assay will be used to quantify the concentration of heavy metals present in the plant tissue. These analyses will allow us to determine the impact of the mutations on the nickel heavy metal stress response of the plant.

Cryopreservation of Human Erythrocytes for Laboratory Propagation of *Plasmodium falciparum*

Mina Diep, Michael L Bellerose, Matthew T O'Neil, & Lawrence M Mylin†

The cryopreservation of fresh, uninfected human red blood cells (RBCs) is fundamental in culturing *Plasmodium falciparum*, to create better preventive measures in combatting malaria. We hypothesize that human blood cryopreserved with hydroxyethyl starch and polyvinyl alcohol (HES/PVA) or glycerol and serum will support asexual parasite growth and gametocyte formation in culture following prolonged freezing at -80°C of A+ or O+ blood. These freezing techniques utilized multiple conditions to evaluate each of the ability for the culture to propagate sexually and asexually with the NF54 parasite strain. The conditions tested through serial propagation: fresh, never frozen blood, which acted as the control; short-term cryopreserved

blood, which was stored at -80ºC for less than two weeks; and long-term frozen blood, which was stored at -80ºC from six months to one year. This shelf-life study found that HES/PVA cryopreservation technique supported the asexual propagation of *P. falciparum* through only two serial passages; parasites could not be rescued by subsequent addition of non-frozen blood. Blood frozen using the glycerol & serum solution was able to support NF54 propagation beyond a ninth dilution and re-plating (passage). Blood cryopreserved by either method supported the generation of mature gametocytes in the first passage. Additionally, our results suggest that the NF54 strain did not accumulate mutations that abnormally enhanced or sustained growth on cyropreserved blood.

Generation of Bacteriophage T4-specific Monoclonal Antibodies

Josiah Gonzalez, Ukamushu A Undieh, & Lawrence M Mylin†

Vaccines are an integral part of modern-day disease prevention. Healthcare professionals should be aware of the basic immunological foundations of how vaccines work. Students in past offerings of Messiah University's Microbiology course were able to carry out an exercise in which commercially obtained polyclonal neutralizing antisera (samples from simulated patients) were compared for ability to inhibit infection of *E. coli* B cells by bacteriophage T4. Unfortunately, the antiserum is no longer available for purchase. Therefore, we seek to produce similar antibodies using monoclonal technologies. Hybridoma clones capable of generating T4 neutralizing monoclonal antibodies will be produced from T4-immunized Balb/c mice. T4 virus stocks of known titer with reduced concentrations of endotoxin must be produced to vaccinate the mice. Our approach has been to reduce endotoxin concentrations in commercially obtained T4 phage stocks by mild organic extraction, followed by dialysis and concentration using Centricon centrifugal filtration units. titers of the concentrated stocks were determined by infection of *E. coli B* cells. The endotoxin levels in the virus concentrates were measured to confirm reduction relative to viral titer.

Generation of Engineered SV40 T Antigen-Expressing Immortal Murine Kidney Cells for Vaccination Against Pancreatic Cancer

Daniel L Guevin & Lawrence M Mylin†

That pancreatic cancer is unusually aggressive and deadly has prompted extensive research into its mechanisms and options for therapy. Our research seeks to develop a vaccine that will activate the tumor-bearing host immune system against a novel protein expressed by aggressive pancreatic tumors. Our approach is to engineer immortalized syngeneic cells to express a derivative of the Simian virus 40 Large Tumor antigen protein (SV40 T ag) that contains a 20 amino acid insertion corresponding to a unique sequence found in the CCK2i4svR pancreatic-cancer associated growth factor receptor. These cells will be used as a vaccine to cause the host to develop cellular or humoral immune responses against the cancer-associated CCK2i4svR epitope. Immortalized cell lines were produced by transfection of C57Bl/6 primary kidney cells with four plasmids, each encoding a different derivative of the SV40 T antigen with respect to presence and insertion site of the CCK2i4svR 20 codon sequence. Transfected cultures were

monitored for growth of dense foci which result from immortalization as a result of expression of the SV40 T ag. Foci were harvested, expanded and evaluated by indirect immunofluorescence using two SV40 T ag-specific monoclonal antibodies to confirm that they had been successfully transformed and expressed the SV40 T antigen protein derivative. SV40 T ag-positive lines were cryopreserved for characterization in subsequent immunization and tumor immunotherapy studies.

The Role of Muscarinic Acetylcholine Signaling in Oligodendrocyte Maturation

Brittany Hayden & Jennifer K Ness-Myers†

The myelin sheath is a protective insulating layer that covers neurons in the nervous system. Myelin supports the generation of rapid electrical signals to ensure efficient communication between neurons. Within the central nervous system, myelin is produced by specialized cells called oligodendrocytes. Unfortunately, in neurodegenerative diseases such as multiple sclerosis, the myelin sheath undergoes damage which leads to neuron degradation and disruption in neuron communication. In order to prevent and treat neurodegenerative diseases, current research seeks to further understand the mechanism underlying myelination and identify treatments that could promote remyelination of neurons. One pathway known to be involved in regulating myelination in oligodendrocytes is the muscarinic acetylcholine signaling pathway. Research suggests inhibiting specific receptors in this pathway promotes the generation of new myelin in humans. Our lab sought to further investigate the impact of several antagonists on oligodendrocyte precursor cells (OPCs) differentiation and remyelination. OPCs were isolated from neonatal rat brains, cultured, and treated with specific antagonists. Gene expression analysis was performed in order to assess oligodendrocyte maturation.

Inhibition of Muscarinic Receptor Subtypes and Effects on Oligodendrocyte Differentiation

Kylee M Kimbel & Jennifer K Ness-Myers†

Oligodendrocytes are the myelinating cells of the central nervous system, and these cells and their myelin sheaths are the autoimmune targets in multiple sclerosis (MS). Increasing the differentiation of OPCs to myelinating OLs is a promising method for treating MS. Recent clinical trials have revealed a positive effect of clemastine, an antihistamine/muscarinic antagonist, in stimulating myelin repair in patients with MS. This study is investigating the combinatorial effects of muscarinic receptor antagonists darifenacin (M1) and pirenzepine (M3) on the rate of oligodendrocyte (OL) maturation. Cultured oligodendrocyte progenitors were used to assess receptor subtype effects on myelin-specific gene expression and the rate of OL maturation during treatment for 4 days. Combination treatments were also studied in the larval zebrafish model. RNA was isolated and analyzed for changes in expression of myelin-specific genes MPZ and MAG. Several dosages of combinatorial treatment and time points were tested, but no significant changes in larval zebrafish gene expression were identified. However, muscarinic agonist, cevimeline, was shown to reduce the expression of myelin-specific genes MPZ and MAG, which supports the hypothesis of the involvement of the muscarinic pathway in

myelination. Muscarinic antagonists do not appear to enhance the vigorous myelination program of zebrafish larvae.

The Effect of Stress on Drug Seeking Behavior

Christine A Milbrath, Sara C Stambaugh, Isela M. Rodriguez, & Jennifer L Thomson†

Post-Traumatic Stress Disorder (PTSD) can develop after experiencing an intensely distressing event. The condition exhibits as a maladaptation of the fear response, causing frequent activation of the sympathetic nervous system (fight and flight behavior) in response to benign stimuli. Persistent sympathetic activity results in chronic stress and precipitates other physical and psychological conditions—severely impacting the individual's quality of life. Current treatments yield low recovery rates and poor prognosis, particularly when initiated long after the original trauma. New research explores opioids as a prospective preventative medication, as morphine seems to diminish PTSD acquisition if received during the critical period of memory consolidation. However, PTSD is often comorbid with increased rates of substance abuse, including opioid addiction. Our research examined if PTSD treatment with morphine increases drug seeking behavior using a rodent model. Rats underwent Stress-Enhanced Fear Learning (SEFL) to induce PTSD like symptoms. After SEFL was completed, the rats were given two injections of morphine in a unique "Context A" and two injections of saline in "Context B". The rats were then allowed to pass between the chambers, and the relative amounts of time spent in either Context A or Context B was measured to determine if the rodents exhibited a Conditioned Place Preference (CPP). If SEFL subjected rats exhibit CPP for Context A, this indicates a correlation between receiving morphine treatment for PTSD and increased drug seeking behavior.

Cryopreservation of Human Erythrocytes for *Plasmodium falciparum* Culture

Matthew T O'Neil, Mina Diep, Michael L Bellerose, & Lawrence M Mylin†

Malaria is caused by multiple species of the parasite *Plasmodium* and disproportionately affects people living in the developing world where effective control of or protection from the parasite is lacking. This study seeks to support ongoing research at the Macha Research Trust (MRT) which is located in the Southern Province of Zambia where the virulent species, *Plasmodia falciparum* is prevalent. Our goal is to support the capacity of the laboratory at MRT to culture (propagate and preserve) locally-isolated or laboratory strains of *Plasmodium*. Laboratory cultivation of *P. falciparum* requires fresh human blood. However, it is difficult to assure the steady supply of fresh, uninfected human blood needed to sustain culture experiments at MRT because blood from local residents cannot be used, and because many visiting scientists and physicians routinely take prophylactic anti-malarial drugs which can make their erythrocytes unable to support asexual propagation of or gametocyte generation by *P. falciparum* in culture. We are investigating methods that should allow for the cryopreservation of erythrocytes obtained in the US from uninfected individuals, with subsequent shipment to Zambia. We have cryopreserved preparations of fresh, leukocyte-depleted erythrocyte suspensions using minimal aqueous volumes of solutions containing the hydroxyethyl starch (HES) and polyvinyl alcohol (PVA), or

with glycerol-based solutions. This presentation will describe ongoing efforts to determine if cryopreserved RBCs can effectively support asexual propagation or gametocyte formation for the *P. falciparum* laboratory strain NF54, and how well this capacity is maintained following long-term storage at -80°C.

Utilizing a Plasma Membrane Enrichment Protocol to Optimize Western Blot Detection of the CCK2 and CCK2i4sv Receptors

Noah Scholl & John F Harms†

Pancreatic cancer is currently the fourth most deadly form of cancer in the United States. One factor implicated in pancreatic cancer growth is the hormone signaling pathway between gastrin and its receptor, CCK2R. In the early 2000's, it was also discovered that pancreatic cancer cells can contain a variant ($CCK2_{i4sv}R$) of the normal receptor protein. Crucially, this longer variant has been shown to be hyper-stimulated and to drive increased cancer growth. Measuring the relative abundance of these two receptors at the protein level can help us understand their role in pancreatic cancer and may represent prognostic value as a biomarker. However, while RNA detection and measurement have been reproducible, protein detection has been problematic. Using western blot analysis, we have been able to detect the receptors in cells expressing them at high levels; however, detection in wild type and stably-transfected lines more representative of physiological expression has been unclear. Thus, we hypothesize that low, natural abundance of the receptors requires enrichment for reliable quantification. Herein, we report our initial attempt to enrich for green fluorescent protein-tagged variants, CCK2R-GFP and $CCK2_{i4sv}R$-GFP, utilizing a membrane extraction protocol based on the non-ionic detergent, Triton X-114. Following enrichment, western analysis demonstrated a significant decrease in cytosolic protein in control cells transfected with untagged GFP. Efforts to verify retention of membrane proteins in the hydrophobic fraction, and subsequent specific detection of the CCK2R variants, are ongoing.

The Effect of Morphine on the Reconsolidation of Traumatic Fear Memories: A Pilot Study

Sara C Stambaugh, Christine A Milbrath, Isela M. Rodriguez, & Jennifer L Thomson†

Post-traumatic stress disorder (PTSD) is a chronic, debilitating disorder in which the heightened fear and arousal experienced during a traumatic event generalizes to nonthreatening stimuli. Previous research has suggested that administering morphine within 24-48 hours of a traumatic event inhibits memory consolidation and prevents future development of PTSD. The goal of this study was to block the reconsolidation of previously stored stress memories by administering morphine within 24-48 hours of conscious recall of the original stressor. A rat model of PTSD called stress-enhanced fear learning (SEFL) was used, which employed two distinct contexts, Context A and Context B. The severe stressor, which mimics trauma experienced by humans, occurred in Context A, during which 15 foot shocks were administered within a 90-minute period. Two reminder shocks were delivered in Context B. The first was followed by an injection of morphine or saline, and the second was delivered three weeks later without a subsequent pharmacological injection. The animals were video recorded for seven weeks in Context B to

assess for freezing behavior, a classic fear response in rats. We hypothesized that the morphine-treated rats would exhibit less freezing behavior following both reminder shocks, whereas the saline-treated rats would exhibit a heightened fear response.

Differential Efficacy of Antifibrotic and Immunotherapy on T-cell Infiltration in Murine Pancreatic Adenocarcinoma

Kally Tan, Lawrence M Mylin†, & John F Harms†

Pancreatic cancer is projected to become the third leading cause of cancer-related deaths in the United States and has a low 5-year relative survival of 10%. The poor response to treatment is attributed to the immunosuppressive and highly fibrotic nature of the pancreatic tumor microenvironment. Previous studies have demonstrated that proglumide treatment results in decreased fibrosis and may increase T-lymphocyte infiltration. Additionally, our lab has demonstrated that peptide immunization targeting the human $CCK2_{i4sv}$ receptor, a cancer-associated splice variant of CCK2R, evokes a potent T-cell response in mice. We hypothesized that combination therapy utilizing concomitant immunization and anti-fibrotic treatment will further enhance T-cell infiltration and decrease tumor burden. To address this, we first analyzed T-cell infiltration in a pilot group of orthotopic murine pancreatic tumors (Panc02) obtained from mice treated +/- proglumide. Tumors were analyzed by immunohistochemistry (n=3 per grp), and infiltrating CD-3+ T-cells were numerated in five random photomicrographs for each tumor. Tumors treated with proglumide exhibited increased T-cell infiltrate and significantly less fibrosis. To assess the efficacy of combination therapy, mice (C57Bl/6) were immunized weekly with $CCK2_{i4sv}$ and control peptides starting three weeks prior to orthotopic injection of syngeneic cancer cells (Panc02) engineered to express human $CCK2_{i4sv}$R. Both groups received oral proglumide following tumor establishment. Upon necropsy (≤6 wks), mice exhibited no significant difference in tumor burden. Immunohistochemical analysis showed the number of infiltrating CD-3+ T-cells per tissue area trended higher in these proglumide-treated mice compared to the previous untreated pilot controls. However, no difference in infiltration was observed in tumors from $CCK2_{i4sv}$R-immunized mice compared to control immunization. Our data confirm proglumide treatment may enhance immune access to the tumor; however, simultaneous analysis of the circulating lymphocyte response is necessary to thoroughly determine the efficacy of combined immunotherapy on T-cell infiltration.

Genetic Control of Heavy Metal Nutrient Uptake in *Arabidopsis thaliana*

Nathan Thorne, Richard W Schaeffer†, & Michael Shin†

Plant cells uptake metals from the soil by operating specific cellular pumps to transport metal nutrients. These pumps are crucial for maintaining the healthy concentration of metal elements. Metal Micronutrients, like zinc or nickel, are essential to plant growth and their uptake is controlled by the MTP1 transporter on the vacuoles in cells. Using *Arabidopsis Thaliana* as a model organism, the effect of mutations on the MTP1 gene can provide a window into MTP1 transporter function and metal nutrient uptake pathways. Two preliminary metal tolerance experiments were conducted on Columbia line wild-type seeds with zinc and nickel

contamination to learn what effects the metal ions were having on the plants' growth. Unfortunately, a problem found during a preliminary wild-type experiment highlighted problems somewhere in the experiment. After that result, the primary purpose became to troubleshooting the experimental method. Both experiments with zinc and nickel contamination yielded results that unexpectedly appeared to be hyper-concentrations of metal ions. Hyper-concentration pointed to a problem with the media, the metal ion solutions, or the seeds. The media was examined by chemical digestion and atomic absorption spectroscopy to quantify metal ion concentration. The analysis demonstrated the concentrations were not hyper-concentrated in the media meaning the problem was with the metal ion solutions. This semester began with a complete remake of metal ion stock solutions and growing a new batch of seeds that would hopefully fix the problems. Early experiments showed the same apparent hyper-concentration problem but with a noticeable consistency. The plants were reacting consistently to the presence of metal ions in a range between 20-60 uM with nickel contamination. This consistency is shown over multiple experiments and means an adjusted experiment range to can be used to test MTP1 mutant tolerance.

Titer Determination, Enrichment, and Endotoxin Quantitation in Bacteriophage T4 Stocks for Generation of Monoclonal Antibodies

Ukamushu A Undieh, Josiah Gonzalez, & Lawrence M Mylin†

Carolina Laboratory Supplies was the sole commercial supplier of polyclonal antisera specific to bacteriophage T4, an essential item used in the Phage Neutralization Lab in Microbiology courses at Messiah University. Carolina ceased production of this antiserum, necessitating discontinuation the Phage Neutralization Lab. the goal of this study is to produce monoclonal T4-neutralizing antiserum so that the Phage Neutralization Lab can be restored. The T4 suspensions used need to have confirmed titers and be depleted of lipopolysaccharide (endotoxin) which are lethal to mammals. Determining the titer of the bacteriophage suspension, purifying it, and verifying that the endotoxin levels are below lethal concentrations must be done before immunization of the mice. The successful generation of antibodies against bacteriophage T4 would enable the project to continue to the next stage, where monoclonal antiserum can be produced using hybridoma technology.

Biology, Organismal & Ecological

Analysis of the Impact of Water Treatment Plant Effluent on Antibiotic Resistance in Pennsylvania Stream Microbiomes

Christy J Sackett & Jeff S Erikson†

Though antibiotics have served as a powerful tool in humanity's fight against disease, the overuse of antibiotics has caused some bacteria to develop resistances to antibiotics, making these drugs less effective. One means by which bacteria may be exposed to antibiotics and

develop resistance is through wastewater treatment plant effluent, which carries antibiotics from human wastes into local streams. We hypothesize that increased levels of antibiotics downstream of treatment plants have selected for increased antibiotic resistance in bacteria living in these downstream regions. As such, when both upstream and downstream samples of bacteria are exposed to the same antibiotic treatments, downstream samples should exhibit higher survival. To test our hypothesis, we sampled bacteria from Dogwood Run and Stony Run in Dillsburg, PA, selecting collection sites from both upstream and downstream of the local wastewater treatment plants. We cultured these bacteria in the lab, classified them based on colony morphology and Gram-stain results, and exposed them to antibiotics through the Kirby-Bauer and replica plating methods. When comparing all bacteria, there did not appear to be a clear trend of increased resistance in either the upstream or downstream samples. However, when comparing bacteria only within our classified groups, initial results showed 26 instances of increased downstream resistance and only 13 instances of increased upstream resistance. These results are promising for our future work in this area, and they may indicate that increased regulations on wastewater treatment plants are needed to combat further antibiotic resistance development.

Hydroponic Growth of Goldenseal (*Hydrastis canadensis*)

Regan M Wilton & David K Foster†

Goldenseal (*Hydrastis canadensis*) is a plant native to woodlands of east-central North America. Goldenseal roots contain antimicrobial compounds which promote respiratory and digestive health and the plant has been wild-harvested almost to the point of extinction. Plants are products of their environments and the concentration of medicinal compounds within a plant are affected by the conditions in which the plant was grown. Wild harvested plants are subject to the variations in their natural environment and it is almost impossible to accurately know the concentrations of desired medicinal compounds within plants that are wild-harvested. With all medicinal compounds there exists a therapeutic dosage in which the compounds are beneficial for the body. Outside of this window, the compounds are either ineffective or toxic. It is crucial for supplements made from medicinal plants to contain known concentrations of the desired medicinal compounds in order to ensure the effectiveness and safety of the supplement. Due to these reasons it is crucial to develop a sustainable and controlled method of growing goldenseal, and aquaponic technology is a form of agriculture that serves as a hopeful solution to these concerns. Previously, goldenseal was aquaponically grown at Messiah University somewhat successfully, but many of the plants died prematurely of root rot. Other than the work completed at Messiah, there are no published studies that include growing goldenseal aquaponically. It is proposed that goldenseal be grown in an aquaponic system at various water levels to find a growing method that maximizes the plant's survival and productivity.

Biochemistry

Expression Vector Construction for a Heme C Attachment Assay

Spencer P Clements & Jesse Kleingardner†

The world is rapidly approaching a point where they need to find a source of sustainable clean energy. Something that could aid this problem is new efficient catalysts, one potential catalyst is Cobalt substituted Holocytochrome C. Holocytochrome c is a protein that is attached to a heme group at a CXXCH amino acid motif. These are found naturally occurring in the mitochondria and are part of the electron transport chain. These enzymes have the capability to catalyze the reduction of water into a diatomic hydrogen which provides a fuel source for energy production. Cytochrome *c* naturally has an iron heme cofactor, but this wild type does not have the same enzymatic capabilities as a cobalt substituted heme to catalyze the reduction of water. In order to synthesize a large quantity of the desired coenzyme, they must be created *in vitro* using *E.* coli expression vectors. The primary objective of my project was to create these expression vectors that would be capable of expressing large amounts of desired product by having the cytochrome *c* be expressed by cytochrome *c* heme lyase, the naturally existing enzyme responsible for holocytochrome c synthesis. These expression vectors can utilize mutations of both the cytochrome *c* and cytochrome heme lyase genes to express different mutants. These mutant products can then be quantitatively analyzed in different combinations to find what combination gives the highest enzymatic capability to combine cytochrome c and a cobalt substituted heme. Once the ideal combination is found, we will attempt to make a microbe and utilize the mutated proteins to effectively perform the process *in vivo.* Utilizing this could provide a catalyst that leads to efficient production of fuels from sustainable energy sources.

Developing a Reliable Computational Method to Determine the Allosteric Sites to Be Targeted in Finding Non-aggregating Allosteric Inhibitors of PTP1B

Sarah K Codd, Anne M Reeve†, Seth Kabonick, & Noah D Smith

Type 2 diabetes is a serious metabolic disease with a growing need for improved therapeutics. Traditional antidiabetic drugs can assist with the maintenance of blood glucose levels in diabetics but are not optimal treatments due to side effects. PTP1B is a validated therapeutic target for type 2 diabetes, as it acts as a negative regulator in the insulin signaling pathway. Inhibiting PTP1B may allow for increased insulin sensitivity, leading to glucose homeostasis and improved metabolism. In the past three allosteric sites were identified as targets for PTP1B inhibition. This study analyzes the validity of these sites and the possibility of other allosteric sites through computational methods and enzymatic assays. A new allosteric site was identified based on the determination of the anti-diabetic drug Lobeglitazone's binding site and used to improve the computational method for testing possible inhibitors of PTP1B. Correlation of the computational method with known natural product PTP1B inhibitors revealed that the computational method is independent of kinetic properties. This study provided a new allosteric site to target and a computational model to eliminate non-inhibiting compounds in the search for type 2 diabetes therapeutics.

Systematic Screening for Expression of an Engineered Heme C Protein in *E. coli*

Michael A Florio & Jesse Kleingardner†

Hemoproteins in the class of metalloproteins have been shown to be incredibly efficient catalysts of oxygen reduction reactions. These have also shown promise in engineering synthetic metalloproteins which could serve as more efficient catalysts than the commonly used inefficient expensive platinum fuel cells. A porphyrin structure with the ability to rapidly transfer four electrons to dioxygen producing water has been shown to possibly be the replacement of the platinum fuel cells. CHIP which is a protein homodimer has presented the ability to bind with the heme *c* attachment and is able to orient so more hemes can attach. Given that this CHIP heme is synthetic, finding vectors which could successfully produce this CHIP heme could prove helpful for the fuel cell research. This study attempted to express this protein in different E. coli genomes*, BL21, pLysY*, and with the addition of other genes such as *pEC86,* and *HemH.* Also, a *NrfA* and *PelB* signal sequence with the CHIP-gene. Most transformations were able to grow in the minigrowths but were unable to produce any successful heme expression to date.

Synthesis of Novel Hydroxychalcone Derivatives as Potential Inhibitors of Protein Tyrosine Phosphatase 1B

Seth Kabonick, Sarah K Codd, Noah D Smith, & Anne M Reeve†

Protein phosphatases have been considered a potential target for drug-based therapy since their discovery. Mutations of phosphatases found in cell signaling pathways have been linked to type II diabetes, obesity, and certain types of cancer. One of these phosphatases, protein tyrosine phosphatase 1B (PTP1B), functions as a negative regulator of the insulin pathway. A gene knockout study in mice confirmed that mutations to this protein result in a dampened insulin sensitivity. Previous attempts at competitive inhibition through drug-based therapy have been unsuccessful due to the highly conserved active site across the phosphatase family. This study explores the plethora of natural products available as potential inhibitors for PTP1B in hopes of designing a molecular scaffold to generate site-specific inhibitors. Chalcones and stilbenes are two compound families that have previously exhibited inhibitory binding. Hydroxychalcone derivatives, specifically those in the para position, show promise as a drug scaffold for specific active site inhibitors. In addition, isoprenyl groups attached to branching aromatic rings have a significantly higher binding affinity to PTP1B and a lower IC50. Initial *in silico* results showed moderate to high binding affinities for a small hydroxychalcone library. Currently, enzyme activity levels in the presence of these hydroxychalcones are determined using absorbance assays. Duplicate assays are performed with detergent to account for non-specific inhibition caused by aggregation. Preliminary assays using commercially available hydroxychalcones display slight to moderate inhibition of PTP1B at the active site. In future studies, twelve hydroxychalcones will be synthesized and introduced into an absorbance assay to evaluate their potential as a scaffold for PTP1B specific inhibitors.

In Vivo Synthesis of Zinc Horse Cytochrome C in *E. coli* Via Zn-supplemented Autoinducing Media

Courtney P Smith & Jesse Kleingardner†

Horse cytochrome *c* (hCc) is a heme protein that operates in essential metabolic processes, including the electron transport chain to generate energy. Substitution of hCc's heme with zinc can create Zn-hCc. Zn-hCc is a useful tool for studying protein folding and hCc's cellular

activity due to zinc's intrinsic fluorescence. However, Zn-hCc has only been artificially synthesized, and the process is complicated, expensive, and wasteful. This study attempted an *in vivo* method that induces *E. coli* BL21 cells to synthesize Zn-hCc by themselves, eliminating the complications of artificial synthesis. The cells were grown in autoinducing minimal media supplemented with minimal iron and excess zinc to induce zinc-takeup in the heme. In addition to the wild-type HCCS, *E. coli* with an HCCS E138A mutant were grown. Analysis via UV/Vis spectrophotometry showed that greater Zn to Fe concentrations up to 250 uM Zn/5 uM Fe correlated to increased incorporation of zinc into the heme and the possible production of Zn-cyt c. The HCCS E138A mutant showed increased incorporation of zinc into the protein and higher yields of hCc than the wild-type, with the highest yield in the 250 uM Zn/5 uM Fe growths. A purification of the 5 uM Fe/25o uM Zn growths was attempted to confirm the synthesis of Zn-hCc. However, the purification was unsuccessful due to a very small yield of hCc. Future research will conduct larger-scale growths and attempt the purification again. If successful, the purified protein will be analyzed with SDS-PAGE, UV/Vis spectrophotometry, and fluorescence spectroscopy to determine whether Zn-hCc was synthesized.

Chemistry

Synthesis of Stilbene Derivatives as Inhibitors of Protein Tyrosine Phosphatase 1B

Noah D Smith, Anne M Reeve†, Sarah K Codd, & Seth Kabonick

The insulin signaling pathway is a biochemical signaling path that regulates the level of glucose within the blood. Protein phosphatases are central enzymes in cell signaling pathways and several phosphatases are associated with the onset of conditions like diabetes, cancer, and obesity. Protein tyrosine phosphatase 1B (PTP1B) is a negative regulator of the insulin pathway that shows significant promise as a target for anti-diabetic therapy. Through a gene knockout study, mice deficient in PTP1B were found to be notably more sensitive to insulin and less prone to high-fat diet induced obesity. Past studies of competitive inhibitors of PTP1B have proven fruitless because the active site is highly conserved across phosphatase-type proteins. (*E*)-Stilbene compounds have been identified as a potential scaffold to design specific inhibitors of PTP1B through computational analysis. This study explores the synthesis of an (*E*)-Stilbene analog from vanillin starting material in order to design a consistent synthetic method for later (*E*)-stilbene derivatives. In the future, twelve stilbene class compounds will be synthesized with methoxy and hydroxy substitutions in varying positions to analyze trends in aggregation and enzyme kinetics. Subsequent analysis of synthetic analogs through enzyme assay will help to classify the compounds as useful or unviable for PTP1B inhibition.

Engineering

Energy Monitoring & Management System (EMMS)

Bennett A Andrews, **Seth Wilcox**, Zachery D Holsinger, Ben Weaver, David C Williams, & Tom Austin†

The Energy Monitoring and Management System (EMMS) is developing an electrical power meter to help make electricity more available in energy impoverished regions of the world. The meter fills a unique niche for energy tracking and regulation within micro-grid systems. The EMMS project has partners in Burkina Faso and Zimbabwe: Open Door Development (ODD), the Institut Missiologique du Sahel (IMS), and the Theological College of Zimbabwe (TCZ). Ties are also maintained on a regular basis with IEEE Smart Village for potential future widespread system implementation.

Recent work on the EMMS meter has been focused on resolving the last few remaining bugs, establishing a robust communication system, and developing a centralized server-based interface which aids with meter configuration and administration. The team has also begun several future developments which include datalogging and remote access features.

Rapid Orthotics for Cure Kenya: Mechanical Design and Modeling of 3D Printed Sockets

Joey D Andrews, **Rachel E Bruns**, **Lauren N Seubert**, **Jarod A Snader**, Gabi E Griffith, Elizabeth G Hargrove, Brandon J Weindorf, & Jamie R Williams†

Rapid Orthotics for Cure Kenya (ROCK) works with CURE, a non-profit orthopedic workshop in Kjabe, Kenya, to implement a 3D printing system for manufacturing custom prosthetics and orthotics. The goal is to reduce the production time and cost for the current transtibial sockets being manufactured in the orthotic clinic to give the patients a way to integrate into society and reduce stigma from their communities. The team has developed a transtibial socket for below-the-knee amputees produced by a 3D printing system that converts a scan of the residual limb to a model that takes a third of the time to print versus the current manufacturing method. The current focus of the team is to develop a rigorous testing procedure adhering to the requirements set by the ISO 10328 Standard, an internationally recognized testing method. In order to ensure the safety of the sockets, tests must be run demonstrating that the product can withstand the different forces experienced during the gait cycle. Due to the complex geometry of the applied forces outlined in the ISO 10328, the team has designed a novel testing rig that interfaces with the MTS machine at Messiah University to apply the necessary forces according to the geometry outlined in the standard. Additionally, computer-based simulations are being developed in SolidWorks, a 3D modeling software, to determine how the components will behave under certain loading conditions. This is done to ensure accordance with the 10328 Standard and will be critical in the future for developing necessary cyclic tests.

Designing a Locally Manufacturable Wheelchair for Nepal

Ethan M Barnes, **Levi D Hauger**, Cade K Bender, Harrison J Crosley, Riley Harro, Joshua J Holley, Peter C Hopkins, & Timothy J Van Dyke†

Persons with disabilities in developing countries often lack the basic equipment needed to assist them in their daily lives. International Nepal Fellowship (INF) is a Christian medical organization located in Nepal that provides medical care and assistance to people with

disabilities and other conditions. Currently, INF imports expensive wheelchairs that undergo a prolonged border process before being received. INF has reached out to the Collaboratory to design a wheelchair that can withstand the challenges of Nepal's terrain and can be manufactured from local materials. The Nepal Wheelchair team accepted this challenge and set out to design a wheelchair that can fulfill their needs. The team began by researching wheelchair models for inspiration and eventually settled on two preliminary designs. In January 2020, the team traveled to Pokhara, Nepal to gain feedback from the staff at INF and select a final design based on their comments. During this trip, the team acquired and brought back locally available materials and parts available in Nepal in order to construct a prototype. This year, prototyping has been completed for the following elements: the central frame, wheel lock, footrest, wheel mounting assemblies and armrest fixtures. As a result of knowledge gained during prototyping, some design changes to the wheelchair have been made. Moving forward, the team will test the overall prototype for durability and prepare a manufacturing manual for INF.

Village Water Ozonation System

Grant Brubaker, **Ruth C Galyen**, **Sam B Stone**, Benjamin K Burlew, TJ J Malanga, Ray Knepper†‡, & Michelle L Lockwood†

The Village Water Ozonation System (VWOS) team's core mission statement is to provide economically sustainable and culturally sensitive drinking water solutions for communities, to empower communities with the ability to properly maintain their drinking water supply, and to transform people's lives by decreasing the occurrences of waterborne diseases.

Currently, the VWOS team is partnering with Friends in Action to design and implement two drinking water treatment systems for the community living on Rama Cay, an island in the Bluefields Lagoon on the eastern coastline of Nicaragua. The wells on the island are contaminated with E. coli and other bacteria and contain high levels of salt that cause the water to be unhealthy, distasteful, and corrosive to metal equipment in the system. The team hopes to design a system that will disinfect the water, remove salinity from the well water with a safe and efficient disposal of all byproducts, and decrease corrosion agents.

VWOS is partnering with Forward Edge International for the third time (Nicaragua 2009 and Mexico 2016) to design water treatment systems for communities in Oaxaca, Mexico and Kijabe, Kenya. The system for Oaxaca is ready for implementation and awaits availability to travel. The system for Kijabe is in the initial stage of communicating with the client on specifics for the design.

Prosthetic Knee for CURE Kenya

Isaiah D Bryner, **Sarah N Kelchner**, **Carter D Urich**, Samuel R Burgess, Nathan E Jaloszynski, Josiah D Moyer, Kay Laura Sindabizera, & Jamie R Williams†

Lower limb amputations are common in sub-Saharan Africa due to the prevalence of disease and infection in communities that lack access to adequate healthcare. Our project, Prosthetic Knee,

partners with the CURE International Hospital in Kijabe, Kenya. In the region surrounding our client's facility, there is a large number of lower-extremity amputations due to various infections and diseases. While many of these amputees only require a through-knee amputation, the lack of an affordable through-knee prosthesis often forces patients to undergo a more invasive transfemoral amputation to enable them to use a cheaper above-knee prosthesis. The goal of our project is to design and manufacture a financially accessible and user-friendly prosthetic knee for knee-disarticulation patients that can be manufactured in Messiah University's machine shop (and ultimately, at the orthopedic facility in Kijabe).

This year, the team's work has included finalizing the prototype design, conducting finite element analysis in SOLIDWORKS to evaluate the knee's static and fatigue strength, and beginning the process of physical manufacturing. Additionally, a damping mechanism driven by a spring-loaded system has been developed and integrated into the design after completing motion analysis and SOLIDWORKS modeling. Our future goals include manufacturing a complete metal prototype of the knee, conducting physical strength and fatigue testing on the metal prototype, and continuing iterative prototyping of the damper in preparation for physical implementation.

Landmine Neutralization: Air Excavation Unit

Josh Card, **Ethan R Cornwell**, Evan Poust, & Donald G Pratt†

The Landmine Neutralization team's poster will present our team's progress towards designing and prototyping an air excavation unit. To accomplish this, the project team is working with the HALO Trust, the world's largest demining NGO. The HALO Trust works in many countries to remove the remnants of war, including improvised explosive devices (IEDs) and unexploded ordnance (UXOs). This project seeks to help deminers by providing a device that blows air at high velocity to clear dust and other debris away from potential IEDs and UXOs in war-torn areas to help in their effort of demining. We have focused on designing our prototype to operate reliably in harsh environments while fulfilling our client's specifications. Our client requested that we design our excavation unit to be easily installed onto their Volvo 220 backhoes and their custom-made backhoe attachments. The current design is modular and consists of a hydraulic motor powering a fan from a backpack leaf blower, all of which is assembled within a steel frame that attaches to the rake of the HALO excavators. Due to circumstances beyond our control, our project will be wrapping up at the end of this semester, which is sooner than anticipated. This means the goals of our project have been narrowed to having a functional prototype and relevant documentation that we can present to our client.

Fatigue Testing a Mechanized Percussion Well Drilling System for Water Access in Western Africa

Micah T Clark, Tommy Denlinger, Robert H Donley, Benjamin R Gates, Matthew R Higgs, & Philip M Tan†

The Mechanized Percussion Well Drilling (MPWD) Collaboratory project seeks to design a simple mechanized well drilling system for drilling shallow water wells in Western Africa. Our client, Open Door Development (ODD), seeks to make water accessible to all in the region, but

has had difficulty drilling through hard soil layers. To combat this problem, the MPWD team has worked closely with Mr. Joseph Longenecker to develop a mechanized percussion well drilling rig that is capable of drilling through these harder layers. Currently, the MPWD team is seeking to provide recommendations to improve the lifetime of our client's new, fully mechanized rig design. This year, our team's work has been focused specifically on analyzing the lifetime of the rig's driveline chains and also on its frame. For the driveline chains, the team will be conducting fatigue testing on a model of the driveline system to determine which type of chain should be used on the rig. To determine the lifetime of the frame, the team will be performing a series of static, buckling, and fatigue finite element analyses on the rig's frame. The most recent accomplishments of the MPWD team have nearly proved that their design for the loading application will be feasible for use on the actual testing rig and that multiple studies of finite element analysis can be performed to simulate the different rig frame loading scenarios.

Adjustable Prone Trolley Design for People Suffering from Spinal Cord Injuries in Nepal

Blake N Clemmer, **Abby Miller**, **Jared T Pavlovich**, Dylan J Derstine, Derek A Thrush, & Timothy J Van Dyke†

For people suffering from spinal cord injuries, it is important to stay active. However, with spinal cord injuries, the use of a wheelchair isn't feasible. These patients require a prone trolley. A prone trolley is a horizontal pad with four wheels that a patient can maneuver and control while lying in a prone position. Our partner, International Nepal Fellowship (INF), deals directly with patients who suffer from spinal cord injuries on a daily basis. INF, a Christian, medical organization, manages a hospital in Pokhara, Nepal which specializes in treating patients with spinal cord injuries. The Nepal Prone Trolley Team's goal is to provide our partner with a sustainable prone trolley design and create the required manufacturing documentation to enable them to produce the prone trolleys in country at their Green Pastures hospital. The team began our work by researching what a prone trolley is, how it functions and what is currently available. During the research, the team discovered that there weren't many examples of a manually powered prone trolley or critical dimensions for ergonomics for manually powered trolleys. This drove the team to develop testing methods and preliminary designs specifically for INF. Various basic designs were considered, but, through communication with INF, a single design was chosen. Computer modeling of this design was used to decrease the overall weight of the trolley and simplify the frame. With most of the design finalized, the team is ready to begin prototyping next semester.

Sustainable Agriculture

Jacob Dean, **Madalyn A Heckman**, Brandon M Bickom, Micah J Hess, Miggy Matanguihan, Aleesa Wu, & Michelle L Lockwood†

The Sustainable Agriculture team is dedicated to developing alternative and sustainable agricultural solutions to alleviate poverty in western Africa. The team is currently working with Sheltering Wings in Yako, Burkina Faso and Trans World Radio in Parakou, Benin. Both clients currently have a working aquaponics system, but the type of system varies between clients.

Sheltering Wings has a flood and drain system and Trans World Radio has an ebb and flow system.

Throughout this year, our team has focused on reducing power consumption and costs for our clients. The development of the ebb and flow prototype was a consequence of this mission as we reduced power consumption by one pump using a manual siphon. We have also worked towards lowering the costs of water quality testing kits by introducing a Nutrient Film Technique with basil plants. Lastly, we have strived to research best practice methods for fish food making and fish feeding in order to keep the biology of the prototypes healthy. To support present and future clients, the year will conclude with final deliverables for the ebb and flow prototype including an operations and maintenance manual, a construction manual, and a troubleshooting manual.

A Sustainable Mobility Solution for Persons Living with Disability in Burkina Faso

Rachel Delate, **Rachel Rashford**, **Joey Sinsel**, Katie Bunch, Sam Fino, Faith N Kerlen, Erin T Logsdon, Julia M Neborg, John Meyer†, & David T Vader†

The Sustainable Mobility project of the Collaboratory empowers people living with a disability in rural West Africa to pursue educational and work opportunities and more fully participate in family and community life. Our electric, 3-wheeled, off-road wheelchair has transformed the lives of dozens of clients through partnerships with the Center for the Advancement of the Handicapped in Mahadaga, Burkina Faso and the Center of Hope in Fada, Burkina Faso. Now, to reach more people in new locations and with more partners, Sustainable Mobility is working to reduce manufacturing time and cost, author image-driven fabrication guides to enable local fabricators to build trikes, create instructional trike assembly videos, and develop supply chains to bring parts and materials to build sites. We seek to put local fabricators to work building tricycles wherever they are needed.

Panama Bridge Project

Luke T Fetterman, **Jordan T Barner**, Mikayla R Eyster, Crosby Harro, Zachary C Hartman, Noah Ling, Drew W Moyer, Noah W Thrush, & Brian D Swartz†

The Panama Bridge project has partnered with Rio Missions Panama to design a bridge for the village of La Gigi, Panama. The mountain community of La Gigi experiences heavy rainfall during the rainy seasons. A stream runs along the community, restricting their access to schools, employment options, and other communities. While passable during dry seasons, the stream floods and becomes impassable after heavy rains. The residents are effectively cut off from their livelihoods, church, health services, and other communities during this time.

To accommodate this need, the Panama Bridge Team has spent the last two academic years designing a 90 foot aluminum truss bridge. The design includes a unique construction strategy to deal with challenging site constraints.

Fluency Assistance Device (FAD): Masker Upgrades

Jake T Finkbeiner, **Chad M Long**, Corey Bean, Michael D Jenkins, Larry A Vega, & Harold R Underwood†

Around seventy million people internationally have a stutter, a form of a fluency disorder. Some fluency assistance devices are available to the public, but most are highly expensive or unreliable. The Fluency Assistive Device (FAD) team seeks to assist a niche community of these individuals who currently rely on a device known originally as the Edinburgh Masker by partnering with Dave Germeyer. Utilizing his expertise in repairing the Edinburgh Masker, FAD is developing two new versions of the masker to increase its portability, functionality, and cost-effectiveness. The first is an update of the original called the Analog Masker (Version 1.1). A prototype of the Analog Masker v1.1 has been developed, tested and is currently being revised based on the results. Revisions include updating the layout of the board and finalizing the power supply circuitry. The second version, known as the Digital Masker (Version 1.0), will use a Bluetooth-enabled microcontroller to achieve masker functionality. Bluetooth audio output for the Digital Masker has been tested, and two algorithms have been created for the masking output. The supporting software for the Digital Masker is nearing completion. The schematic and the layout design have been started for future implementation of the hardware.

Design of a Transradial Myoelectric Prosthesis

Lindsay L Haseltine, **Jaymie R Monday**, **Meghan L Sampson**, Sam Sparks, Samuel F Whittle, Eddie J Yesilonis, & Tim Howell†

Due to the rapid growth of children and the high cost of myoelectric technology, children are not given the same opportunities to use myoelectric prosthetics as adults. The Muscle Activated Prosthesis (MAP) team is developing an affordable, transradial, myoelectric prosthetic for a thirteen-year-old girl. The MAP team is designing a myoelectric prosthetic that will cost under $1,000, over 90% less than custom myoelectric devices on the market. This device has an EMG sensor, a microprocessor, a printed circuit board (PCB), linear actuator motors, and a battery organized within a 3D-printed transradial prosthesis to open and close the hand grip when the EMG detects electrical signals via muscle contractions in the client's flexor carpi radialis. Currently, the team has fully assembled a prosthetic prototype and will obtain feedback from the partner, Ability Prosthetics, and the client to deliver a final prototype. This poster details the recent mechanical and electrical design optimizations, grip strength testing, and integration of mechanical and electrical components to build the current functioning prosthesis.

Trans World Radio - Culvert Design

Warner C Hockenberry, **Gabriel J Tiday**, Darren J Heisey, Logan J Horst, Seth M Kline, Daniel O Thomas, & J Scott Heisey†

Trans World Radio (TWR) is a mission organization focused on broadcasting the Gospel around the world. TWR now serves 190 countries by transmitting in 275 languages, using radio to

deliver the message to as many people as possible. The organization's West Africa Transmitter Site in Benin currently has accessibility problems due to high streamflows and saturated ground conditions during the rainy season. The site also needs a secure perimeter to reduce trespassing and theft.

To address these issues, our client, Garth Kennedy, Director of the West Africa Transmitter Station, has asked the team to design two culverts, one at the upstream property boundary and one at the downstream boundary. Culverts are advantageous for this scenario because they can act as a bridge, while the pipe size can be restricted to inhibit trespassing. Once the culverts are built, the fence and perimeter road can be extended over them.

For both sides of the property, the team has designed a series of U-shaped, pre-cast concrete box culverts. The team calculated the design flows based on rainfall data and the topography of the site to determine the size and number of box sections. The team has also designed the culverts and the supporting concrete structures to bear the load of vehicles and the machinery on site. TWR plans to construct the pre-cast culverts on-site, and then build the supporting structures and install the culverts during their dry season.

Better Pumps: Reliable Handpump Infrastructure

Andrea Hunsberger, **Joshua L Maxson**, Matt J Caldwell, Caleb E Danehower, Darren Kulp, Darin M Landis, Jacob Valentine, Emma L Workman, Tony Beers†‡, Matthew Schwiebert†‡, & David T Vader†

Approximately 90 million people in Africa lack access to safe drinking water, despite having water infrastructure installed in their community. The India Mark II and the Afridev handpumps are among the most widely used handpumps in the world. Sadly, studies show that approximately 30% of these handpumps are non-operational due to failures of the bearings, seals, head flange, and other common components. The Better Pumps team of the Collaboratory provides engineering support for partners who are working to improve handpump sustainability. We partnered with Tony Beers and AlignedWorks to validate a bearing test methodology for the India Mark II handpump. By modifying the loading conditions in our handpump test machine, we were able to replicate failures observed by AlignedWorks in a field trial of their bearing design. We partnered with Matt Schwiebert and Living Water International to test new seal designs for the India Mark II and Afridev handpumps and to measure head flange deflections in the India Mark II handpump. Seal performance data collected by the team was used to validate a new design in advance of field trials by Living Water International. Head flange deflection data was collected for partner benchmarking of their computational analysis. Test methodologies and results are reported.

Land Development - Tree 4 Hope

Caleb R Light, **Jorge O Zambrano**, Olivia R Allbee, Corey B Englehart, & J Scott Heisey†

The Land Development Team has partnered with Tree 4 Hope and Hope Academy in Santa Lucía Milpas Altas, Guatemala to improve the outdoor facilities of the school. Jenn and David

Hope-Tringali are the client/partner representatives of the school for this project. The goal of the project is to provide design and construction drawings for three main elements of the proposed land development: (1) a parking lot for buses and school vehicles that enter the site, (2) a single sports court that can accommodate basketball and soccer, and (3) a playground that is directed towards themes associated with STEAM (science, technology, engineering, art, and math). The team has completed project drawings to allow construction by local personnel, or by student or church mission teams when travel is allowed to resume post-pandemic.

Preparing For Extended Field Tests of the Intelligent Water System

Josiah J McCarthy, Daniel J Labrie, Evan Freed, & Randall K Fish†

The Intelligent Water System, which improves access to clean water by autonomously monitoring and reporting on the health of hand pumps in developing countries, has been under development for several years. This development has included short-term prototype field tests in several countries. The design has matured to the point that an extended field trial to demonstrate performance and reliability has been requested by our client. In light of this, the team has implemented design changes to address issues from our most recent prototype field test and begun manufacture of the first five systems intended for installation in Burkina Faso. This poster highlights the code changes enabling more accurate determination of the volume of water pumped and the simplified mounting of the system's Handle Motion Sensor.

A Low-Cost, Portable Fluorescence Correlation Spectrometer for Disease Diagnosis

Jessica E Paulus, **Al W Mokris**, **Brittany Shirk**, Nathan E Cordell, Castine L Donoff, Jeffrey Gao, Sam J Gulinello, & Matthew J Farrar†

The DVD team is developing a cost-effective technique for measuring HIV load in resource-restricted regions. Our client is Dr. Phil Thuma and the Macha Research Trust in Zambia. Our design is based on advanced fluorescence spectroscopy that utilizes a fluorescence protein probe, confocal optics, and low-cost, low-power electronics to assess viral load in a patient blood sample. Our timeline for a functional exploded prototype is Fall 2021.

Specifically, we are employing a method of spectroscopy that seeks to identify individual viruses in dilute samples by characteristic "bursts" in fluorescent and elastically scattered light. We have assembled a housing for a custom-designed detector, associated electronics, and signal processing hardware. One project goal is to integrate this modular design into a single printed circuit board. Communication between signal processing hardware and a software-based user interface implemented on a Raspberry Pi and touchscreen is achieved by the use of a Serial Peripheral Interface (SPI) protocol. The entire system is battery-powered. This system will allow for fast, effective viral load determinations in remote settings.

On-Campus Solar PV Lab: Component Selection is Only the Beginning

Noah A Rood, **Jonas E Kolb**, **Christian Pilawski**, **Garrison Shields-Seelig**, & Randall K Fish†

The work of the Solar PV Team is to design and install Solar PV systems which enable our clients to fulfill their mission in the presence of unreliable or non-existent electrical power. In order to experiment with different Solar PV configurations and train new members, the Solar PV team last year designed a Solar Lab to be installed in and next to Frey 70. This work paralleled the design/component selection typically performed prior to an installation site trip. This year, the team modeled the efforts typically done at the installation site by building and configuring the Solar Lab design. This poster will focus on the lessons learned about decisions that need to be made in the field to convert a Component Selection level design into a Functioning PV System.

Force Characterization and Manufacturing of a Dynamic Unilateral Clubfoot Brace

Leigha R Southall, **Michelle Lo**, **Clint M Meekins**, Jacob R Cornwell, Brittney D Fouse, Sam J Rasinske, Jordan M Witt, & Tim Howell†

Clubfoot is a musculoskeletal birth defect characterized by an inward twisting of an infant's feet. Currently, a series of casts are used to correct the clubfoot, and a boots-and-bar brace is used to maintain the correction. However, this method has concerns with compliance, comfort, and social stigma. Hope Walks and their clinic in Kijabe, Kenya are interested in implementing a new maintenance brace that addresses these concerns. Mr. Jerald Cunningham, CPO, designed and is utilizing a unilateral clubfoot maintenance brace called the Cunningham Clubfoot Brace. He asserts his brace reduces treatment time, lessens social stigma, and increases child mobility. However, to date, there is not enough published research on its biomechanics and patient success rates to confirm his findings.

The Cunningham Clubfoot Brace Collaboratory project seeks to validate the effectiveness of the Cunningham design through biomedical testing and increase brace availability through sustainable manufacturing. To do this, the team is measuring the biomechanical forces applied by the brace with multiple force sensor systems and an infant foot model. The team is assisting Mr. Cunningham in his plans to use injection molding to increase brace production by scanning and creating CAD files of the brace. The team is also completing a failure and reuse analysis of the Cunningham Brace for the clinic in Kijabe. Furthermore, the ongoing clinical study at CURE International's hospital in Kijabe, Kenya, and Dr. Emily Farrar's research paper will provide greater insight into the effectiveness of the Cunningham Brace. These collaborative efforts will allow for further understanding of the effectiveness of the Cunningham Brace and its acceptance as an alternative clubfoot maintenance brace.

A Low-Cost Egg Incubator to Provide Zambian Churches with Income and Food Security

Claudia M Tolley, **Jacob M Barton**, **Aaron Bashore**, **Matthew D Eells**, **Lydia Reber**, Shekinah Ellis, Brandon Koehnke, & Philip M Tan†

Partnering with Brethren in Christ (BIC) Church in Zambia, the Egg Incubator Team is seeking to help provide a source of income for the growing churches in Choma, Zambia. They will accomplish this by designing and building a high-quality, low-cost egg incubator fabricated from local parts and cheap internationally available parts for The Nahumba Mission, in Choma, Zambia. The team's design will provide the means for the Mission to hatch and sell chickens to provide both food security and a sustainable supplemental income. With the specifications to maintain temperature, humidity and constant ventilation, the team selected heating and humidity concepts for their incubator series. The team completed both mechanical and electrical designs for the setter and hatcher. In preparation for testing the incubator design with fertilized eggs, the team has also produced an incubation and hatching plan and achieved IACUC approval. Currently, the team is in the prototyping phase, while simultaneously monitoring the temperature and humidity in an existing incubator setter design. Once the team finishes their hatcher prototype and verifies that the temperature and humidity specifications are met, they will be ready to test designs using fertilized chicken eggs.

Exercise Science

The Role of CYP1A2 and ADORA2A in Individual Response to Caffeine Consumption Under Anaerobic Conditions

Kathryn J Armstrong, Michael Shin†, H. Scott Kieffer†, & Michael Sedhom

Caffeine is the most commonly used psychoactive drug in the world and has wide implications in medicine, athletics, and public health. Studies have shown that metabolism, clearance, reception, and response to caffeine vary significantly among individuals. The pharmacokinetics are primarily dictated by the cytochrome p450 enzyme CYP1A2 while the adenosine neuroreceptor ADORA2A heavily influences the drug's pharmacodynamics. Polymorphisms of the -163 A>C CYP1A2 and the 1976 T>C ADORA2A are thought to influence these interindividual responses; therefore, the purpose of this project is to determine the effect of caffeine consumption on anaerobic exercise. 12-15 female college athletes completed two maximal WAnT30 anaerobic bike tests on a Velotron cycle ergometer. The participants ingested a capsule of caffeine ($5mg \bullet kg^{-1}$ bodyweight) or a placebo capsule (maltodextrin) one hour prior to testing. The order of the bolus was randomized, counterbalanced, and administered in a double-blind manner. Peak power ($W \bullet kg^{-1}$), anaerobic capacity ($W \bullet kg^{-1}$), and total power output ($W \bullet kg^{-1}$) were recorded during each test. Buccal epithelial cells were collected using a 0.9% NaCl mouth rinse with DNA extraction conducted using proteinase k to lyse cells and collection using QiAmp Mini spin columns. Allelic discrimination was obtained using TaqMan® SNP Assay for CYP1A2 (rs762551) and ADORA2A (rs5751876) and a One-Step qPCR. Each sample was run in duplicate positive and negative quality controls. Each variable was analyzed using a factorial ANOVA with repeated measures ($p > 0.05$).

The Influence of Caffeine and the C957T (DRD2) Polymorphism on Measures of Unilateral and Bilateral Motor Tasks

Abigail Beveridge, Rachel Caldwell, H. Scott Kieffer†, & Michael Shin†

The effects of caffeine on task performance and attentional focus during movement have been suggested to be influenced by the interaction between the adenosine (ADORA2A) and dopamine (DRD2) receptors in the dopaminergic pathways and specific centers in the brain. Individual polymorphisms of 1976 T/C (ADORA2A) and C957T (DRD2) are thought to influence the varied response of caffeine during movement. **PURPOSE:** The purpose of this study is to determine the influence of caffeine, 1976 T>C ADORA2A polymorphism, and C957T (DRD2) polymorphism on measures of unilateral and bilateral motor tasks. **METHODS:** The Purdue Pegboard Test and the Minnesota Dexterity Test were used to assess fine motor abilities following both placebo or caffeine ingestion (5mg•kg-1 bodyweight). Buccal epithelial cells of each subject were retrieved by a 0.9% NaCl mouth rinse. To collect the DNA samples, the cells were lysed via proteinase k and isolated with QiAmp Mini spin columns. The polymorphisms were identified using a TaqMan® SNP Assay for ADORA2A (rs5751876) and DRD2 (rs1800497) and allelic discrimination via One-Step qPCR amplification. All samples were run in duplicate with negative and positive controls. **RESULTS/DISCUSSION:** The Results and Discussion will be presented at the Symposium.

Effects of Caffeine and CYP1A2, ADORA2A Polymorphisms on Exhaustive Anaerobic Performance

Adam D Cole, H. Scott Kieffer†, & Michael Shin†

Individual responses to caffeine are suggested to be genetically influenced by polymorphisms of the cytochrome P450 enzymes, specifically the -163 A>C CYP1A2, for metabolism in the liver and through the adenosine receptor, 1976 T>C ADORA2A, for sensitivity of specific target cells. Individuals with the AA variant are caffeine responders, while those with the AC/CC variants are caffeine non-responders. ADORA2A TT variants demonstrate an increased sensitivity to caffeine compared to TC/CC variants. **Purpose** To examine the effect of caffeine and CYP1A2 and ADORA2A polymorphisms on anaerobic power during exhaustive exercise. **Methods** Fifteen elite NCAA male athletes (age=20.1 yrs, weight=77.4 kg, height=176.7 cm) participated in a double-blind study. Subjects performed two separate 90-s Wingate Tests (WAnT90) separated by two to four days on a Velotron cycle ergometer, resistance=0.05 kg•BW(kg)$^{-1}$. Subjects ingested a bolus of caffeine, 5mg•kg^{-1}BW, or a placebo (maltodextrin) one hour prior to each trial that were administered in a randomized/counterbalanced design. Peak power (W•kg^{-1}), total power (W•kg^{-1}), and average power (W•kg^{-1}) were calculated for the 90-s and each 30-s interval. Buccal epithelial cells were collected using a mouth rinse, 0.9% NaCl, and DNA was extracted via spin columns and proteinase k. Allelic discriminations for CYP1A2 (rs762551) and ADORA2A (rs5751876) were procured via an assay and a One-Step qPCR amplification. Samples were run in duplicate, with positive and negative controls. The data was analyzed using a factorial ANOVA with repeated measures ($p > 0.05$) for each variable. **Results/Discussion** The results and the discussion will be presented at the Symposium.

The Effects of an Acute Bout of Controlled Breathing on Short-Term Heart Rate Variability

Sarah Roise Hartman & H. Scott Kieffer†

Heart rate variability (HRV) is a non-invasive measurement of the autonomic nervous system (ANS) which uses the frequency and time domains of an ECG to determine the magnitude of sympathetic and parasympathetic activity. Decreased HRV has been associated with several disease conditions whereas increased HRV indicates increased parasympathetic drive, which attenuates physiological and psychological stress conditions. The short-term HRV, 5-minute, average, denotes the temporal and dynamic functioning of the ANS and can be used to monitor acute changes in allosteric load. Increased parasympathetic activity secondary to acute bouts of controlled breathing holds conflicting results in current research. **Objective:** To determine the effect of an acute bout of controlled breathing on the time domain measures of HRV. **Methods:** Twenty-nine subjects (x= 19.4 $\pm$ 0.8 yrs) volunteered for the meditation activity. The subjects attended a familiarization session to give informed consent, to adjust to the lab setting, and to practice a guided breathing technique, 6 breaths/min. The subjects returned to the lab for a 15-minute experimental protocol during which, they sat quietly breathing normally for 5 minutes to establish a baseline, followed by 5 minutes of the guided breathing, and finished with 5 minutes of normal breathing. The ECG recorded continuously over the 15-minute protocol using a Polar H10 heart rate monitor. The raw data was downloaded into the commercially available Kubios HRV software program and the 5-minute average for heart rate (HR), low frequency (LF), high frequency (HF), peak HF/LF ratio, standard deviation of the normal-normal R-R (SDNN), and root mean square of the standard deviation (RMSSD) were calculated for baseline, guided breathing and recovery periods. **Results:** 5 minutes of guided breathing elicited significant differences in measures within the frequency domain (HF and LF) and the time domain (RMSSD and SDNN) heart rate variability. HF baseline was 52.0 $\pm$ 20.7 nu significantly decreased to 18.7 $\pm$ 10.1 nu following controlled breathing, and significantly increased to 37.0 $\pm$ 17.4 nu during the recovery. LF baseline was measured at 47.9 $\pm$ 20.8 nu, increased to 81.3 $\pm$ 10.1 nu during controlled breathing and returned to 63.0 $\pm$ 17.4 nu during recovery. The RMSSD baseline was 64.8 $\pm$ 36.3. It increased significantly to 79.5 $\pm$ 37.8 and returned to baseline measures 60.3 $\pm$ 35.4 following the breathing exercise. The SDNN baseline was 63.4 $\pm$ 32.2 which increased significantly to 114.9 $\pm$ 40.5 during controlled breathing, and then significantly decreased to 67.8 $\pm$ 31.2 during recovery. during recovery. **Discussion:** The frequency domains of HF (parasympathetic) and LF (sympathetic) appear to respond in the opposite direction from the anticipated change in parasympathetic activity; however, these variables are not reliable indicators of parasympathetic activity at paced breathing of 6 breaths/minute or slower. Within the time domain, RMSSD and SDNN have been identified as indicators of short-term parasympathetic activity, specifically vagal nerve activation. The RMSSD and SDNN results demonstrated that controlled breathing provided significant parasympathetic response during the controlled breathing technique; however, the acute increase in activity was not sustainable past the breathing intervention.

The Effects of a 2-week Meditation/Prayer Program on Heart Rate Variability

Hannah K Logan, **Rhianna Gonzales**, **Courtney Luckenbill**, **Spencer Cassel**, & H. Scott Kieffer†

Heart rate variability, the variation in time between heartbeats, is used to measure the autonomic balance between the sympathetic and parasympathetic nervous systems. It can be influenced by meditation and various breathing techniques. The purpose of this study was to evaluate the effects of a meditation and breathing training program on the heart rate variability of inexperienced meditators. Participants in the study were male and female college students, ranging in age from 18 to 22 years. They participated in a synchronous pre-test training session, during which baseline data was collected during spontaneous and controlled breathing. Over the following week, participants meditated independently using a guided breathing app. They then returned for a synchronous post-test training session, during which data was collected and compared to the pre-test baseline data. Results and data will be discussed at the Symposium.

The Effects of a 2-week Weighted Vest Training Intervention on Sprint Performance.

Joshua J Monturo, **Grant Myers**, **Jack Conrad**, H. Scott Kieffer†, & Eric Rawson†

The addition of resistance to a sprint interval training program could potentially increase different aspects of sprint performance. **PURPOSE:** To determine the effects of a two week, 10% resisted speed training program on power performance. **METHODS:** Six active males participated in a two-week resisted sprint interval training program that met three times a week (six total sessions). Each participant completed a pre and post-test that includes a 30 second Wingate test and a vertical jump test. The training regime began with three sprints on a Woodway Curve 3 treadmill and increased repetitions throughout each session. Each participant completed the training program with a weighted vest that contains 10% of their body weight. **RESULTS AND DISCUSSION:** The data and results will be discussed at the symposium.

The Effects of Static and Dynamic Stretching on Muscular Power in Dancers and Jump Athletes

Elizabeth R Vlieg, Sydney De Poto, Grace Brewster, H. Scott Kieffer†, & Abigail K Gibson

Flexibility and power output are two critical components of dance performance; however, recent research suggests that acute bouts of stretching may decrease muscular power. **PURPOSE**: The purpose of this study was to compare the effects of dynamic stretching (DS) and static stretching (SS) on muscular power in dancers. **METHODS**: 12 female, collegiate dance majors volunteered for this study. The subjects attended a familiarization session, gave informed consent, and were oriented to all testing procedures. Three different experimental sessions were conducted in randomized order for each stretching condition: DS, SS, and no stretching (NS), with a minimum of 48 hours between testing days. The sessions began with a warm-up (5 min walk) followed by a guided stretching protocol for each condition. Muscular power (peak torque, Watts) was obtained for the plantar/dorsiflexors with an isokinetic dynamometer (60o/s and 180o/s) and vertical power (W/kg BW) was obtained using both a squat (SJ) and countermovement jump (CMJ). One-way ANOVA with repeated measures with Tukey HSD post-hoc tests was conducted to determine significance ($p \leq 0.05$) for each variable. **RESULTS**: The results indicated that there were no significant differences for the power output of dancers after incorporating DS, SS, or NS as part of a warm-up. Isokinetic muscular power isolating the plantar flexors demonstrated no significant difference at 60o/s (DS,

57.1 ± 22.7 W; SS, 57.8 ±28.4 W; NS, 62.7 ± 19.3 W, p = 0.82) or at 180o/s (DS, 28.3 ± 9.50 W; SS, 30.5 ± 14.7 W; NS, 32.2 ±16.2 W, p = 0.76). In addition, measurements of power incorporating whole body power demonstrated no significant difference in jump height for the SJ (DS, 21.05 ± 3.36 cm; SS, 20.83 ±3.55 cm; NS, 20.9 ± 3.14 cm, p = 0.893) or the CMJ (DS, 23.8 ± 3.9 cm; SS, 23.7 ± 5.7 cm; NS, 24.7 ±6.8 cm, p = 0.98). **CONCLUSION**: Although some research suggests that varying forms of stretching may decrease muscular power, this study suggests that neither acute static stretching nor dynamic stretching will elicit a significant change in muscular power in dancers. The jump athlete data is still being collected at the time of abstract submission.

The Influence of Video Distraction During Exercise on Measures of Cardiometabolic Function and Perceived Exercise Enjoyment

Max Weaver, **Chloe A Milanesi**, **Laura Sauder**, **Michael Sedhom**, Melinda B Smith†, & H. Scott Kieffer†

As the presence of technology has increased, its incorporation into physical activity has become vital for making exercise more enjoyable and enticing. Various types of distraction have been used to test if, during exercise, a form of distraction may alter the VO2max, RPE, energy expenditure or heart rate values of a participant. **PURPOSE:** The purpose of this study is to investigate how the presence or absence of a predetermined 15-minute action clip during 15 minutes of biking affects RPE, heart rate, breathing rate, affect and performance. **METHODS:** Participants will come in for a familiarization session as well as two testing sessions. One will involve a 15-minute action clip and the other will involve no distraction. Pre-test, post-test, and every 5 minutes during the test the participants' heart rate, VO2, and breathing rate will be recorded as well as the participants' RPE and Affect using the Feeling Scale. **RESULTS AND DISCUSSION:** Results will be discussed at the Symposium.

Continuous and Interval Training Acutely Elevates Serum BDNF in Healthy College-age Males

Sam A Zercher, Grant Myers, H. Scott Kieffer†, Nathan T Romberger, & Jennifer L Thomson†

Brain-derived Neurotrophic factor or BDNF is a protein involved in the growth and maintenance of brain cells and overall brain function. Research has shown both moderate and high-intensity aerobic exercise improve brain health and function. This study sought to examine the relationship between serum BDNF levels and moderate-intensity continuous training (MICT) compared to high-intensity interval training (HIIT). Ten healthy males from Messiah University participated in this study and were taken through VO2 Max and peak power (W_{peak}) testing to establish a baseline to be used for the MICT and HIIT protocols. The HIIT protocol involved 60 seconds at 75% (W_{peak}) followed by 60 seconds at 40% (W_{peak}) and repeated 10 times. The MICT protocol involved 20 minutes at 60% (W_{peak}). Serum was collected before, immediately after, and 15 minutes post-exercise for both protocols. The serum was then tested for BDNF concentrations using the Quantikine enzyme-linked immunosorbent assay (ELISA) kit from R&D systems. The ELISA kit showed BDNF concentrations were significantly elevated immediately post-exercise compared to the pre-exercise levels. There were no significant differences between time points for the exercise protocols. This supported the hypothesis that

moderate and high-intensity aerobic exercise increases serum BDNF levels as well as supporting the American College of Sports Medicine statement about exercising at a range of intensities enhances overall health.

Nursing

Evidence-Based Recommendations to Improve Patient Outcomes Using Outpatient Cervical Ripening Methods.

Shayna Bashore, **Kiersten R Miller**, **Kaylee A Hollenbach**, KateLynne Siebert†‡, Karen Wagner†‡, Amanda Lawrence†‡, & Kate Bigler†‡

Background and significance of clinical problem/question: Cervical ripening for induction of labor (IOL) is commonly performed in the inpatient setting, but there is a growing push to perform this procedure in the outpatient setting, as this can improve patient satisfaction, the occurrence of adverse outcomes, and cost-effectiveness (Kruit et al., 2016).

PICO Question: In low risk pregnancies undergoing cervical ripening, do outpatient ripening balloons lead to lower CS rates and increased satisfaction compared to inpatient inductions utilizing the same method?

Methods of Literature Search: PubMed, CINAHL, and Cochrane databases were searched using the keywords, “balloon catheter,” and “induction.” Seventeen articles were found, 7 articles were critiqued, and 4 articles were selected for presentation. The dates of the published articles range from 2015-2020. The Johns Hopkins Nursing Evidence-Based Practice Model was applied to determine quality of evidence, which ranged from Level I-II, quality A-B.

Findings from EBP project: Evidence supports the use of outpatient IOL via cervical ripening balloon on the basis of reduction of cesarean section rates (Kruit et al., 2016; Policiano et al., 2017). Patient satisfaction was also increased when patients received outpatient IOL (Alfirevic et al., 2020; Beckman et al., 2020; Kruit et al., 2016).

Recommendations for Practice: Based on the literature, opportunities to provide outpatient inductions on a routine basis for low-risk women should be assessed and a protocol should be developed for incorporation of outpatient IOL into routine care.

Evidence-Based Recommendations to Prevent Delirium in Intensive Care Settings Using a Multi-Faceted Approach

Sarah M Chen, **Lauren Becker**, & **Skylar J Johnson**

Background and significance of clinical problem/question: Delirium occurs in 60-80% of Intensive Care Unit patients who are ventilated (Martinez, Donoso, Marquez, & Labarca, 2017).

Currently, there is no standard protocol used to prevent delirium in ICU patients. As a result, delirium commonly develops, leading to prolonged hospital stays, increased costs of care, and decreased patient satisfaction (Rivosecchi, Smithburger, Svec, Campbell, & Kane-Gill, 2015).

PICO Questions: In ICU patients, does the implementation of a multi-faceted approach improve the prevention of delirium?

Methods of Literature Search: The literature search was conducted using CINAHL and PubMed. The article dates range from 2015-2020. A total of 448 articles were found, and 6 articles were chosen that answer the PICO question. Based on the John Hopkins Nursing Evidence-Based Practice Model, the evidence ranged from a level II to level III with an A or B quality.

Findings from EBP Project: Evidence supports the use of a multi-faceted approach to prevent delirium. The multi-faceted intervention decreased delirium incidence from 38% to 24% (p = 0.02) and from 38% to 23% (p = 0.01) respectively (Martinez et al., 2017; Bounds et al., 2016). The odds of developing delirium were reduced by 78% (p = 0.001) and 60% (p < 0.002) using a multi-faceted approach (Smith & Grami, 2017; Pun et al., 2019). Similarly, mean reduction in the incidence of delirium was 24.7%, with a range of 9.7%-31.8% in a study using a multi-faceted approach (Rivosecchi et al., 2015).

Recommendations for practice: Based on the literature reviewed, the evidence supports the use of a multi-faceted approach to prevent delirium. The approach should be implemented in institutions after proper education of the staff to ensure successful application.

Evidence for a Delirium Bundle for PICU Patients

Payton Dierkes, Brittany J McCullough, Reagan Welch, Lauren E Nagy, & Micaiah J McClymont

Background and significance: A knowledge deficit exists in prevention, treatment, and recognition of pediatric delirium (Flaigle et al., 2016). Delirium is present in 30% of critically ill children (Flaigle et al., 2016). Pediatric patients with delirium had longer hospitalizations ($p < 0.001$) and days on mechanical ventilation ($p < 0.001$) (Ista et al. 2018; Simone et al., 2017).

PICO question: In PICU patients, what are the effects of implementing a delirium care bundle as compared to no delirium interventions on patient outcomes?

Methodology: The literature search included: Medline, PubMed, CINAHL, and AACN.org. A total of 128 articles were found ranging from 2013 – 2020 and 10 addressed the PICO question. Using John Hopkins Nursing Evidence Based Practice Model, there were 5 Level III and 5 Level V articles, of B quality.

Findings: Delirium risks include mechanical ventilation, sedation, and analgesic therapies (Flaigle et al., 2016; Holly et al., 2018). Symptoms include anxiety, poor eye contact, labile affect, and agitation (Holly et al., 2018; Ista et al., 2018). Delirium education improved delirium management confidence ($p<0.001$) (Simone et al., 2017). Prevention strategies include protocols

to promote alertness, mobilize early, and decrease sedation ($p< 0.001$) (Simone et al., 2017). Delirium screening every 8 hours promoted earlier diagnosis (Ista et al., 2018). Unidentified and undertreated delirium contributes to familial and financial burdens (Holly et al., 2018). Early recognition and prevention may be enhanced by routine use of valid screening tools (Flaigle et al., 2015, Holly et al., 2018, Malas et al., 2017, Mason et al., 2017, Smith et al., 2016).

Recommendations for practice: Delirium screening using psCAM-ICU or PD-scale is indicated for early recognition and intervention. Multi-site randomized controlled trials are needed to strengthen the evidence, explore prevention strategies and implementation barriers.

Ketamine Effectiveness in Sickle Cell Disease (SCD) Vaso-Occlusive Crisis (VOC)

Jordan Diethrich, **Amanda NN Rossi**, **Rachel Rupp**, & **Katie Carbaugh**

Background and significance of clinical problem/question: High dose opioid prescription is the common practice in the treatment regimen of vaso-occlusive pain crises (Applequist, 2017), which leads to an undesirable side effect profile. Thus, it is imperative to find better options of treatment that will protect patients from experiencing unnecessary effects and provide better pain control.

PICO Question: In patients experiencing vaso-occlusive crisis, does ketamine administration provide better pain control and a decreased side effect profile compared to opioid monotherapy?

Methods of Literature Search: The literature search included PubMed, CINAL, American Society of Hematology, MEDline, and UpToDate. The articles ranged from 2013-2020. Of the 37 total articles found, 13 were used to address the PICO question. The John Hopkins Nursing Evidence-Based Practice Model was used to rate the level and quality of the articles. The levels ranged from Level I to V with a quality range of A to B.

Findings from EBP project: Pain scores in patients without acute chest syndrome (ACS) were decreased with ketamine use compared to opioid use (Twafic 2014, $p=0.01$; Kilinc, 2018, $p<0.05$). Ketamine administration also resulted in a decreased need for opioid usage (Nobrega, 2018, $p<0.001$; Neri, 2014, $p=0.0038$; Tawfic, 2014, $p=0.007$; Hagedorn, 2019; Jennings, 2013; Gowhari, 2013). Side effect profiles were less severe with ketamine monotherapy (Palm, 2018; Hagedorn, 2019).

Early Childhood Oral Healthcare to Reduce Cardiovascular Disease

Joelle Hickey, **Shannah Hough**, **Kasie Pratesi**, **Rachel Smith**, & **Becky Slyder**

Background & Significance: Poor oral health is associated with increased incidence of cardiovascular disease (CVD). Oral health care interventions (OHC) improve serum cardiovascular markers including triglycerides, total cholesterol, and HbA1C (Lavigne & Forrest, 2020). Determining appropriate early childhood OHC may prevent CVD later in life.

PICO Question: In early childhood, what oral healthcare interventions reduce the risk of cardiovascular disease across the lifespan?

Methods of Literature Search: Literature search included Medline and CINAHL articles from 2013-2020. Ninety-one articles met the search criteria; 11 addressed the PICO question. Evidence included Level I, II, III, and V, with A or B quality using the John Hopkins Nursing Evidence-Based Practice Model.

Findings: Education about proper oral hygiene during the prenatal period ($p<0.05$), preschool years ($p<0.05$), and elementary years ($p<0.001$) is associated with decreased incidence of childhood caries (Blake et al., 2015; Burgette et al., 2017; Kuter & Uzel, 2020; Xiao et al., 2020). Topics of proper toothbrushing, flossing, and decreased sugar intake were found effective ($p<0.001$) (Blake et al., 2015). School dental screenings and professional flossing may also reduce caries' risk (Burgette et al., 2017; Salzer et al., 2017). Computerized reminders with older children encourage toothbrushing (Walker et al., 2015). Use of fluoride treatment modalities is also associated with better OHC outcomes ($p<0.001$) (Brecher & Lewis, 2018; Clarke & Stevens, 2019, Marinho et al., 2013). Physicians and dentists should work in collaboration to educate about other risk factors associated with CVD (Dietrich et al., 2017).

Recommendations for Practice: Multi-disciplinary OHC should begin during the prenatal period and continue into primary school using age-appropriate incentives. Education to reduce risk of CVD should include use of fluoride toothpaste, flossing, and decreased sugar intake. School-based dental screenings and professional flossing should be conducted.

Evidence-Based Recommendations for Adjunct Non-Pharmacological Peri-Operative Pain Management in Pediatric Patients

Olivia M Jimenez, **Faith Beattie**, & **Kimberly Thomas**

Background: Current standards of practice only focus on analgesic medication for pain, without considering nonpharmacologic interventions (Suresh, De Oliveira, & Suresh, 2015). Prolonged pain in children can result in adverse psychological effects such as increased anxiety, bedwetting, fear of pain, and eating disorders (Woragidpoonpo et al., 2013). It is crucial to determine effective pain management to provide patient-centered care.

PICO Question: In perioperative pediatric patients, which nonpharmacologic interventions with pharmacotherapy decrease pain and increase comfort compared to pharmacological therapy alone?

Methods of Literature Search: Literature was reviewed using PubMed and CINAHL with article dates ranging from 2013 to 2020. A total of 818 articles were found with 6 articles addressing the PICO question. According to the Johns Hopkins Nursing Evidence-Based Practice Model, evidence ranged from Level I to Level III with quality ratings of an A or B.

Findings: Evidence showed support for a wide variety of nonpharmacologic techniques. Audiobooks and music interventions (Suresh, De Oliveira Jr, & Suresh, 2015), quiet rooms (McKay, et. al., 2019), guided imagery (Vagnoli, et. al., 2019), and skin to skin contact (Lisanti, et. al, 2020) incorporated into post-operative care all showed a decrease in the child's pain scores. Parent-nurse-patient relationships and massages also correlated with pain relief (Woragidpoonpo, et. al., 2013). There was a discrepancy in parent and nurse expectations for pain control, suggesting a need for more family education in the pre-operative period (Harvey, & Kovalesky, 2018).

Recommendations: According to the reviewed literature, strong evidence of various nonpharmacological interventions including relaxation-guided imagery, music and audiobook therapy, and a quiet room, along with pharmacotherapy, decrease pain and increase comfort. These interventions should be implemented in pediatric patients undergoing surgery.

Key words: pediatrics, pain, nonpharmacologic, comfort, pre-operative, post-operative

End-of-Life Simulations & Nursing Students' Confidence

Jenny Jobson, Morgan Lieberman, Starr Nitschke, Alley Sell, & Grace E Davis

Background & Significance: Nurses spend more time with critically ill and end-of-life (EOL) patients than any other healthcare professionals (Ferrell et al., 2016). As the population ages, nurses need to be prepared to care for patients at the EOL. Unfortunately, undergraduate nursing students are beginning professional practice with little confidence in caring for patients at the EOL.

PICO Question: In undergraduate nursing students, what is the effect of end-of-life simulation on students' confidence in providing end-of-life care?

Literature Search Method: The evidence was obtained from CINAHL, Medline, and PubMed. Dates of publication ranged from 2014-2020, 38 total articles were retrieved with eight used. Based on the Johns Hopkins EBP model, evidence consisted of one Level I, quality B, four Level II quality B, and three Level V quality B.

Findings: The evidence reviewed supported the use of EOL simulation as having a positive impact on student confidence. Tamaki et al. (2019) found improved confidence among the EOL simulation group students for both, physical assessment ($p=0.00$) and psychological care ($p=0.00$). Using the Frommelt Attitudes toward Care of the Dying Scale (FATCOD), four studies reported significant increases in mean scores following simulations: Bryne et al. (2020) ($p=0.03$), Dame and Hoebeke (2016) ($p<0.001$), Kirkpatrick et al. (2020) ($p<0.001$), and Lewis (2016) ($p<0.001$). In literature reviews, Gillian et al. (2014) and Kirkpatrick et al. (2017) found that improved self-confidence and self-efficacy were outcomes of EOL simulations and Carman's (2014) case report indicated that EOL care simulation had an improved effect on students' confidence.

Recommendations: Based on the consistency of the evidence in support of EOL simulation, an undergraduate pilot project is recommended. However, experimental studies with larger sample sizes and identification of reliable and valid student confidence tools are needed to strengthen the evidence.

Mindfulness, an Intervention to Reduce Burnout Among Registered Nurses

Mikayla Lewin, **Katie Riggs**, & **Alexandria M Vecchio**

Background and significance of clinical problem/question: Nursing burnout is a phenomenon that affects individual nurses and nursing as a whole. Nursing burnout affects patient outcomes, patient satisfaction, nursing job satisfaction, and the longevity of a nursing career (Zhao et al., 2018).

PICO Question: For registered nurses, does the use of mindfulness reduce the future risk of burnout compared to no intervention?

Methods of Literature Search: The literature was reviewed using CINAHL and PubMed. The article dates ranged from 2013 to 2020. Of the 965 articles retrieved, six articles were used. Utilizing the Johns Hopkins Nursing Evidence-Based Practice Model, the levels of evidence ranged from level V to level I with a quality of A or B.

Findings from EBP project: Evidence supports mindfulness as a means to reduce nursing burnout. In various studies, implementing mindfulness resulted in stress decreases that were statistically significant ($p < .001$) (Duarte, 2016). Mindfulness also increases compassion satisfaction (95% CI, $p = .027$), decreases burnout (95% CI, $p = .003$), and decreases secondary trauma (95% CI, $p = .047$) (Hevezi, 2016). One quasi-experimental study showed a correlation among mindfulness and self-compassion and empathy as well as serenity and wellbeing ($p < .001$) (Penque, 2019; Van Der Riet, Levett-Jones, & Aquino-Russell, 2018). A separate literature review found that of various interventions used to reduce burnout, mindfulness was the most successful (Suleiman-Martos et al., 2020). A cross-sectional survey showed moderately reduced levels of burnout when mindfulness was utilized ($p < .05$) (Zhao et al., 2018).

Recommendations for practice: Based on the literature reviewed, mindfulness should be implemented as a pilot study in individual hospitals to reduce burnout among registered nurses. Measurement of mindfulness intervention implementation could provide additional evidence about its ability to reduce burnout.

Evidence-Based Recommendations to Reduce Delays in Care for Women Presenting with Chest Pain in the Emergency Department (ED)

Samantha A Mazzone & **Mary Underman**

Background and significance of clinical problem/question: Women have an increased mortality rate compared to men presenting to the ED with chest pain. 12-month mortality

following percutaneous coronary intervention for men versus women was 7% and 12% respectively (p= 0.035) (Dreyer et al., 2013). The use of a traditional triage tool in the ED often leaves room for subjectivity and could contribute to a delay in care.

PICO question: In women presenting to the Emergency Department with chest pain, does the use of a traditional triage index tool cause a delay in care as compared to men with the same complaint?

Methods of Literature Search: Comprehensive literature searches of MEDLINE and CINHAL were completed. Article dates ranged from 2010-2020. 539 articles were found, and 5 addressing the PICO question were selected. Based on the Johns Hopkins Nursing Evidence-Based Practice Model, all articles were Level III and Quality B.

Findings from EBP Project: Evidence supports that the average treatment time was faster for men presenting with chest pain compared to women ($p = 0.04$) (Kuhn et al., 2013). Proportionally more women were triaged incorrectly compared to men ($p < 0.001$) leading to significant delays in initial treatment of women (Kuhn et al., 2013). Specifically, women experienced significant delays in initial examination (Mnatzaganian et al., 2019), door to balloon time (Dreyer et al., 2013), and time to initial ECG (Zégre-Hemsey et al., 2011).

Recommendations: Findings suggest women experiencing chest pain who present to the ED are more likely to receive delayed care. Further research is needed to determine if an objective triage tool exists or if one should be developed to eliminate this disparity. Additional research may also identify why this disparity exists.

Evidence-Based Recommendations for Nurse Driven Communication in Adult Intensive Care Units and the Impact on Family Satisfaction

Bekah Warner, Emily Kirk, & Katarina L Sobolewski

Background and significance of clinical problem/question: Nurses are often responsible for facilitating communication with patient's family members. Due to continuous changes in patient status and care plans, this is often time-consuming and can result in inconsistent communication resulting in nurses spending less time on patient care and increased family dissatisfaction.

PICO Question: In the adult intensive care setting, does daily nurse driven communication with patient's family improve family satisfaction compared to less frequent communication?

Methods of Literature Search: The literature was searched using CINAHL and Medline. Article dates ranged from 2015-2020. Keywords included communication, intensive care unit, family satisfaction, and family centered care. A total of 357 articles were found and 5 articles that addressed the PICO question were utilized. Based on the John Hopkins Evidence-Based Practice Model, the evidence ranged from level II-level V, and quality ratings were level A to level C.

Findings: Evidence supports the idea that family involvement in rounds can increase family knowledge regarding the patient's care ($p<0.001$) and nurses felt that it supported patient advocacy (Allen, et al., 2016). Bell, et al. (2016) found that electronic communication via a portal improved family satisfaction and knowledge. Loeslie, et al. (2017) found that family knowledge and satisfaction increased with scheduled family meetings ($p<0.01$). The evidence consistently supports nurse driven communication improving family satisfaction and knowledge.

Recommendations for Practice: Based on the literature reviewed, there is not enough evidence to suggest a practice change and more research is needed. We recommend developing or finding a family satisfaction survey to assess communication strategies and performing a high quality randomized control trial with a consistent nurse education communication strategy based on that survey.

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Collaboratory/Engineering Department Project Review Panelists

Once a semester, each team in the Collaboratory takes part in a formal engineering design review in which faculty from the Department of Engineering and others, including professional engineers from industry, hear an in-depth report about the work that the team has been doing, are able to ask questions about the work and give their insights and suggestions based on their experience working as engineers. While these project reviews are a bit nerve-wracking for the students, they really help to make sure that projects are progressing along a path which will lead to eventual success of the project. Because of the time that the following individuals from outside the Department of Engineering have volunteered, these projects are much more successful than they could have been otherwise. Thank you!

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