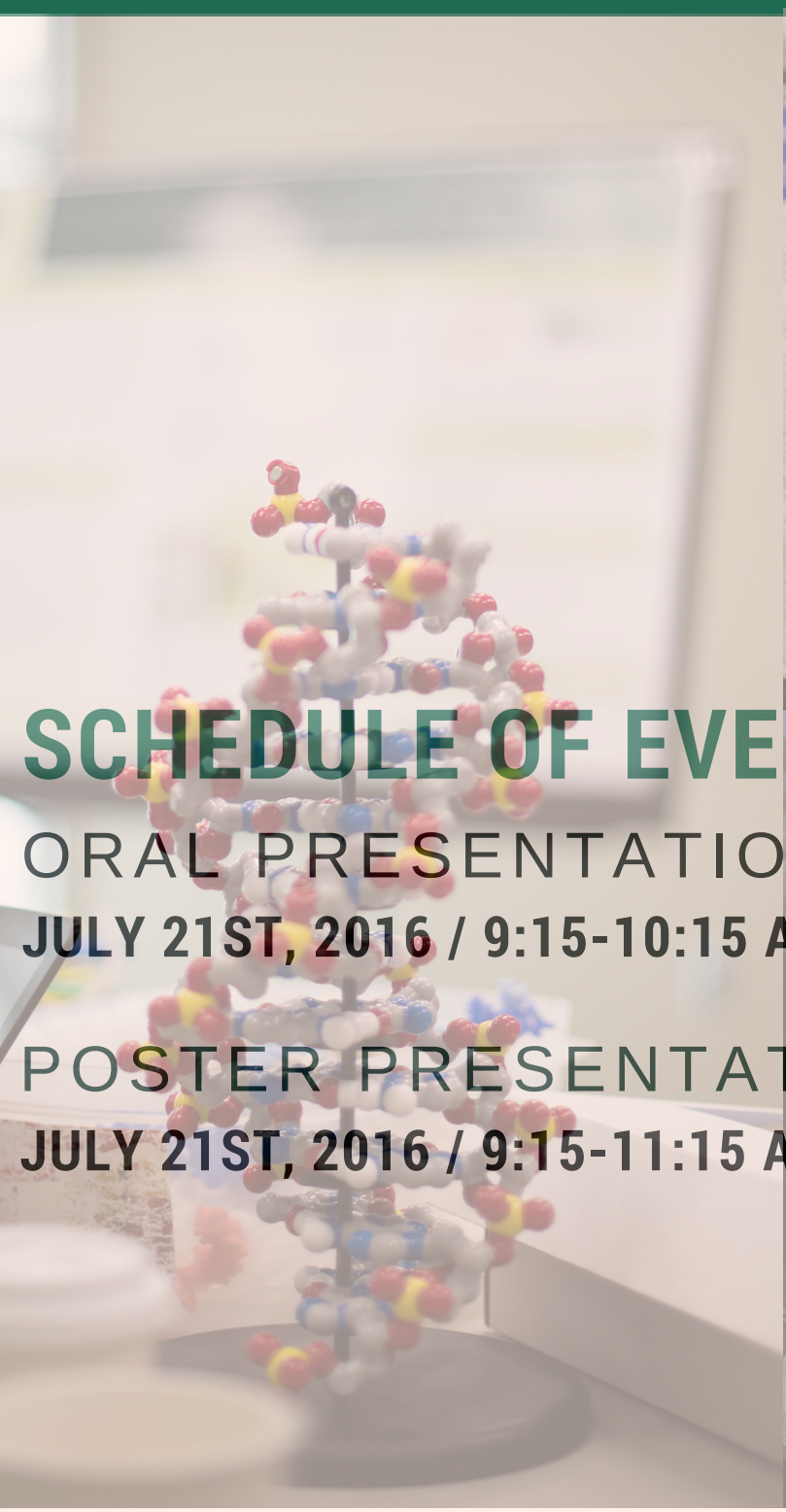
7-21-2016

2016 UAB Summer Research Expo

University of Alabama at Birmingham

UAB Summer EXPO 2016

AN EXPOSITION OF UNDERGRADUATE RESEARCH



SCHEDULE OF EVENTS

ORAL PRESENTATIONS

JULY 21ST, 2016 / 9:15-10:15 AM

POSTER PRESENTATIONS

JULY 21ST, 2016 / 9:15-11:15 AM

UAB Summer EXPO

An Exposition of Undergraduate Scholarship

Welcome

The University of Alabama at Birmingham and the Office of Service Learning and Undergraduate Research are proud to welcome you to the 2016 Summer UAB EXPO: An Exposition of Undergraduate Scholarship. This year's EXPO promises to be the largest to date, with over 100 student presentations and approximately 200 student participants, represented by all academic disciplines. We have observed a significant growth expressed by the undergraduate student research with their creative and innovative ideals that have been under-represented in the past. Therefore, we are excited to showcase a vast diversity of student achievements who have put in their hard work and effort. By working with faculty, graduate students, peers, or individually, these aspiring and highly motivated students are an inspiration to the entire university. Our faculty continually seeks to encourage undergraduate students in quality research, discovery and creative endeavors that will define their academic experience. We would like to give a special thanks to all faculty members who have helped assist in mentoring student presenters, as well as, a hearty congratulations to all our student participants for their contribution and their impressive body of work presented today.

Through collaborative efforts of the EXPO Volunteers, Undergraduate Research Ambassadors and Inquiro-Editorial Board, we would like to give a sincere thanks for their tireless efforts in planning and development.

We celebrate the established tradition of annually recognizing the research and creative accomplishments of our best and brightest undergraduate students.

Best Regards

Gareth Jones
Program Administrator for Service Learning and Undergraduate Research

Skyler Hendrix & Brennan Hickson
UAB EXPO 2016 Student Assistants

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Courtney Walker

Schedule of Events

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JULY 21ST, 2016

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Keynote Speaker

Dr. Despina Stavrinos is an Assistant Professor of Psychology and Director of the Translational Research for Injury Prevention (TRIP) Lab. As Director of the TRIP Lab, Dr. Stavrinos and her team are studying the cognitive aspects of transportation-related injury with a particular emphasis on the impact of distraction among at risk groups such as teens and young adults, older adults, and drivers with developmental disabilities. She has received funding from multiple federal agencies including the U.S. Department of Transportation, the Centers for Disease Control and Prevention, the National Institutes of Health, and the National Science Foundation. Dr. Stavrinos has received numerous prestigious awards including being selected as a USDOT Dwight D. Eisenhower Fellow, the UAB University Transportation Center Student of the Year, the Society for Public Health Education Injury Prevention Fellow and most recently the Routh Early Career Award in Pediatric Psychology. Dr. Stavrinos' applied behavioral background and expertise in unintentional injury and driving behavior has prepared her to serve as a research mentor to nearly a hundred of graduate, undergraduate and high school students creating highly relevant and engaging opportunities for students so they become active participants in the learning process.

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ARTS AND HUMANITIES

63-Aldridge Gardens Veteran Memorial Bust Project

Bryce Martinez

Aldridge Gardens, in association with the non-profit Vettes-4-Vets, has begun the construction of Veterans Memorial Arbor, an outdoor memorial for Alabama veterans. Professor of sculpture Stacey Holloway was contacted about the project; I was then put in touch with Mark Davis who is heading the project. A bust of Alabama veteran Thomas River's was wanted for the memorial. I applied for the President's Summer Research Scholarship (PSRS) with this project as a way to involve UAB in the community. Funding for the Veterans Memorial Arbor is primarily through donation and charity, and not wanting to take money away from the rest of the project I am very grateful to have the PSRS fund this project.

The project involves the sculpting and casting of a likeness to Thomas Rivers, the first Alabamian killed during the Iraq War. The bust, nearing completion, will be cast in bronze using the lost-wax casting process. Using 3D scanning and printing small scale replicas of the bust will be created for family members of Thomas as well as large donors to the project. Aside from the completion of this bust and replicas I am responsible for communications with the family of Thomas Rivers as well as departmental faculty whom I consult with on the project. Thomas' family has been involved to help ensure I capture the likeness of Thomas accurately. The family has also helped in providing photographs as well as allowing hands on access to Thomas' medals and uniform. Sculpting mold making and casting processes

107-You Are What You Eat: Kaneki's Struggle with Hybridity

Danielle Ivey

In Sui Ishida's Tokyo Ghoul, Ken Kaneki struggles with the acceptance of his hybridity, because of the connections and perceptions he has developed in the human and ghoul worlds. Tokyo Ghoul follows Kaneki's forced transformation into a half human, half ghoul, as well as his struggle with that transformation as he attempts to suppress his ghoul side in various ways, such as his attempt to use his eyepatch as a shield, hiding both his ghoul eye and serving as a physical representation of his refusal to accept that side of himself. In Stuart Hall's work Culture, Community, Nation, Hall identifies that individuals can be hybrids, being a part of multiple groups and sharing their ideas and traditions, while still being an entirely new being because they do not exactly fit in with each group that they are a part of. Kaneki's struggles are uniquely hybrid, as he is forced to find a way to live in both of the worlds that he is a part of simultaneously, and create his own path in the margins of each of these worlds. Kaneki develops his identity throughout the manga and eventually moves away from his efforts to only perform in the human world, as he is forced to accept his hybrid identity and progress as a member of both domains.

BIOLOGICAL AND LIFE SCIENCES

6-Hemopexin treatment attenuates the susceptibility to bacterial infection after acute lung injury

Henry Paiste, Adam Lam, Israr Ahmad, Nilam Vetal, Sadis Matalon and Saurabh Aggarwal

Exposure to toxic halogen gases, such as bromine (Br2) and chlorine poses an environmental and occupational hazard that result in acute lung injury (ALI) and death. Patients who survive initial insult develop respiratory infections with Pseudomonas aeruginosa. However, the mechanisms involved in the increased rate of infection are not clear. Recently, we demonstrated that Br2 exposure promotes hemolysis resulting in heme-dependent ALI. Elevated heme has been shown to cause fatal bacteremia, features that can be reproduced by the administration of the heme donor, hemin, in animals. Therefore, we hypothesized that treatment with the heme scavenging protein, hemopexin (Hx), would attenuate the bacterial infection after Br2 exposure. C57BL/6 mice were exposed to air or Br2 (600ppm) for 30 minutes. Thirty minutes later Br2 exposed mice received an intraperitoneal injection of Hx (4µg/g), while air exposed mice received hemin (50µmol/kg). Four days later, we challenged these mice to an intratracheal bolus (105 colony-forming units) of Pseudomonas aeruginosa. One day post infection; lungs were harvested and plated on an agar plate. Br2 exposed and hemin challenged mice were more susceptible to infection as indicated by numerous bacterial colonies on the agar plate which correlated with impaired bactericidal activity of neutrophils in these mice. Interestingly, Hx treatment reduced bacterial infection and lung inflammation (lower bronchoalveolar lavage fluid protein, total cell count, and neutrophils) and improved survival in Br2 exposed mice. Together, these data suggest that Hx may prove a useful adjuvant therapy to reduce the rate of bacterial infection in patients with ALI.

9-Feeding Study in Lethal Nf1 Knockout Mouse Model

Brennan Yoder, Ashley Turner M.S., and Robert Kesterson Ph.D.

Neurofibromatosis type 1 is one of the most common genetic disorders affecting 1:3,500 people worldwide. NF1 patients commonly suffer from benign tumors, neurofibromas, which develop across the nervous system. Other indicators of this disease are varied such as learning disorders, Lisch nodules in the eyes, and café au lait spots on the skin. The disorder begins with a mutation in the NF1 gene, which codes for a tumor regulator protein called neurofibromin, typically followed by a second-hit mutation or loss of heterozygosity in a particular cell population. The neurofibromin protein functions as a negative regulator of Ras signaling. Numerous mouse models have been generated to study and better understand the NF1 gene and disorder and explore therapies. With both alleles of the Nf1 gene removed, the mouse is embryonic lethal. However, the model and experiment of knocking out Nf1 systemically in an adult mouse is missing from the literature and from our understanding of this gene and protein. Therefore, a mouse model was generated with a tamoxifen-inducible CAGGCre-ER recombination system to assess other potential functions of this multi-domain protein. All adult CAGGCre-ER ; Nf1flox/flox mice die within 12 days following tamoxifen induction with weight loss and multiple tissues affected including skin, bone marrow and intestines. My project was to assess feeding and nutrient absorption efficiency following tamoxifen induction. This novel mouse model reveals other functions for the neurofibromin protein with multiple systems affected not originally affected in the NF1 disorder.

14-What Role Does NreB Play in the Stress Response to Hypochlorous Acid in Lactobacillus reuteri?

Jennifer Chavez, Poulami Basu Thakur and Michael J. Gray

Hypochlorous acid (HOCl), the active ingredient of household bleach, is one of many antimicrobial oxidants that play an important role in the innate immune system. Our interest focuses on how Lactobacillus reuteri, a Gram positive probiotic lactic acid bacterium with anti-inflammatory properties, detects HOCl and whether it is able to signal to the host to reduce inflammation in response to HOCl. Recent transcriptome data from L. reuteri exposed to HOCl demonstrated up-regulation of a gene homologous to NreB of Staphylococcus carnosus. NreB is a sensor protein involved in a two-component system that stimulates the expression of genes in nitrate respiration under anaerobic conditions. NreB regulates nitrate reductase, potentially producing nitrite and nitric oxide, which could contribute to HOCl defenses. It is possible that this is a novel mechanism by which L. reuteri senses inflammation and signals the host to stop producing toxic oxidants, thereby reducing inflammation. We aimed to construct a mutant of L. reuteri that lacks NreB using a CRISPR Cas-9 genome editing system. We plan to test the ability of this mutant to grow when exposed to oxidants, nitrate, and other stresses. To determine conservation of the mechanisms we characterized in L. reuteri across different bacteria, bioinformatic analyses were used to compare NreB and genes potentially controlled by NreB to similar genes in other organisms. By discovering the mechanism by which L. reuteri reduces inflammation we hope to understand the symbiotic relationship between such bacteria with the human intestinal tract.

15-Investigating the Mechanism of Positive Supercoil-Induced Cell Toxicity

Marina K. Triplett, Christine M. Wright, Mary-Ann Bjornsti

DNA topoisomerase I (Top1) is an enzyme that catalyzes the relaxation of DNA supercoils that form during DNA replication and transcription through a process of single-stranded cleavage, relaxation, and religation. Top1 is the target of the camptothecin (CPT) class of anticancer drugs. Previous studies suggest that CPT binding slows the Top1-dependent removal of positive supercoils in DNA. We hypothesize that cell toxicity results from the collision of the replication fork with positive supercoils ahead of the replication machinery. To study this relationship, cell toxicity and positive supercoil accumulation in yeast cells expressing human Top1 (hTop1) were observed in the absence and presence of CPT. Overexpression of hTop1 leads to the accumulation of positive supercoils, even in the absence of CPT. Self-poisoning hTop1 mutations at residues T718 and N722 stabilize Top1-DNA complexes when overexpressed in yeast cells. We have found that the T718A mutant, which reduces the rate of DNA religation, displays slight positive supercoil accumulation in the absence of CPT. In contrast, the N722H mutant, which increases the amount of Top1-DNA binding and the rate of DNA cleavage, exhibits a significant accumulation of positive supercoils in the absence of CPT. This difference between self-poisoning mutants suggests that these mutants contribute to cell toxicity through different mechanisms. By understanding toxicity in relation to Top1, it may be possible to increase the effectiveness of drugs derived from CPT or determine which compounds may be useful in combination therapy. This project was partially funded through the UAB President's Summer Research Scholarship Program.

16-Functionality and Mechanisms of the Chromo Shadow Domain in the Heterochromatin Protein 1 Family

Lanasia Jefferson, Heidi Glassie, Tabitha Hardy, Nicole C. Riddle

Chromatin is a complex of DNA and proteins which forms chromosomes in eukaryotic cells. There are two types of chromatin, heterochromatin and euchromatin. Heterochromatin is a highly condensed, late replicating form of chromatin, which contains sequences that are typically not expressed. Conversely, euchromatin is the form of chromatin which is less condense and has higher gene expression. Our research focuses on the function and maintenance of heterochromatin. The Heterochromatin Protein 1 (HP1) family is a family of proteins that plays an important role in heterochromatin formation. The HP1 proteins are comprised of three domains, a chromo domain which binds to H3K9me2, a hinge which binds DNA and RNA and chromatin, and a shadow domain which regulates protein-protein interactions. This versatile protein family is involved in gene silencing, DNA repair, and regulation of gene expression. The purpose of this project is to further characterize the function of the HP1 chromo shadow domains by introducing a single amino acid substitution within the domains of each of the three somatic HP1 proteins found in Drosophila melanogaster. Site-Directed Mutagenesis was used to introduce a single amino acid substitution within the chromo shadow domain of each HP1. We hypothesize that this change will affect the protein-protein interaction of the HP1 proteins. Once the constructs have been produced, we will introduce these transgenes into flies permitting further characterization of this essential protein family.

18-Chronic Co-treatment with Low Dose LPS and Tempol Preserves Afferent Arteriole Autoregulatory Behavior.

Estevan C Beltran, Justin P Van Beusecum, Shali Zhang, Anthony K Cook, Edward W Inscho.

Activation of toll–like receptor 4 (TLR4) with lipopolysaccharide (LPS) increases reactive oxygen species (ROS) causing oxidative stress. Tempol, a superoxide dismutase mimetic, scavenges endogenous ROS, thereby reducing oxidative stress. Afferent arteriole autoregulatory behavior is impaired following 7 days of LPS treatment. Accordingly, we hypothesized that chronic co-treatment with LPS and Tempol preserves autoregulatory behavior. Three treatment groups were used (n=4/group): LPS (0.1mg/kg/day), LPS+Tempol (2mmol/L), and saline (0.9% NaCl)+Tempol. Rats received osmotic minipumps (day 0) for infusion of LPS or saline, placed inside metabolic cages, and Tempol was added to the drinking water on day 1. Systolic blood pressures (SBP, tail cuff) were measured (days 0, 3, and 7) and collected urine was analyzed for electrolytes, and protein. Kidneys were harvested on day 7 for juxtamedullary nephron experiments. SBPs were similar across all three groups, averaging 140±4, 124±9, 127±7 mmHg, respectively on day 7. Urine output averaged 8±0, 8±1, 6±1mL/24Hrs., respectively. Urinary protein excretion averaged 9.2±0.3, 9.2±0.2, and 9.3±0.6mg/24Hrs., respectively, consistent with normal glomerular function. Autoregulatory behavior was assessed by increasing renal perfusion pressure from 65 to 170 mmHg (15 mmHg increments). Baseline diameters (100 mmHg) averaged 15.2±1.2, 14.0±1.0, and 15.0±1.5 microns, for saline (n=6), LPS (n=6) and LPS+Tempol (n=3), respectively. At 170 mmHg, arteriole diameter decreased by 25±1% ($P<0.05$) and 21±3% ($P<0.05$) in the saline and LPS + Tempol co-treated groups, respectively, signifying normal autoregulatory behavior whereas it did not change in the LPS group. Therefore, chronic co-treatment with LPS+Tempol preserves renal autoregulatory behavior, by scavenging endogenous ROS.

20-Cadmium from cigarette smoke increases the risk and severity of lower respiratory tract infection by decreasing the immune response of alveolar macrophages.

Ereny G. Gerges, Jennifer L. Larson-Casey, and A. Brent Carter

Cigarette smoking is associated with an increased risk of lower respiratory tract infection (LRTI). LRTIs are a prevalent infection that result in hospitalization; however, the molecular pathogenesis of cigarette smoke-induced LRTIs is unknown. Cigarette smoke contains more than 4500 chemical compounds, including carcinogens, toxins, oxidants, and metals. Cadmium (Cd) is one of the metals present in cigarette smoke. Cd specifically inhibits bacterial clearance in alveolar macrophages unlike other immune cells. The molecular mechanism by which Cd alters the immune response of alveolar macrophages is not known. One factor that is required for the immune response is ROS production by the NADPH oxidase. The small GTP-binding protein Rac2 is required for NADPH complex formation. We hypothesize that Cd from cigarette smoke impairs the host defense of alveolar macrophages via inhibition of the Rac2 GTPase. We tested our hypothesis by treating macrophages with different concentrations of cigarette smoke extract (CSE) or Cd to determine if there is a dose-dependent effect on Rac2. We isolated the cell fractions after treatment and found that localization of Rac2 in the cell membrane and ROS production by the NADPH oxidase are reduced in a dose-dependent manner. The mechanism of Rac2 inhibition is by impairing lipidation of the C-terminal cysteine. Overall, the results of these studies will allow us to characterize the molecular mechanism(s) by which cadmium in cigarette smoke inhibits host defense, thereby permitting the potential design of a clinically relevant therapeutic agent in subjects that smoke.

23-Determining Whether 14-3-3 θ Inhibition Plays a Role in the Regulation of α-syn Release and Toxicity Found in Parkinson's Disease

Maria O. Onatunde; Rachel Underwood; Talene Yacoubian, M.D., PhD

Parkinson's disease(PD) is one of the most prevalent neurodegenerative diseases. One of the pathological indicators of PD are Lewy bodies that form due to the misfolding and aggregation of alpha-synuclein (a-syn). Although the cause of this disease is still unknown, evidence supports that the protein a-syn plays a large role in this disease. Misfolded alpha-syn can be released by neurons and spread to other areas of the brain by infecting other neurons and further inducing further a-syn misfolding in a prion-like manner. Colocalized with a-syn in the Lewy bodies of Parkinson's disease is a family of seven proteins with chaperone-like functions called 14-3-3s. We have developed a paracrine culture model in which a-syn is overexpressed in cultured neuroblastoma cells (isyn cells) that releases excess, toxic a-syn to separately cultured neurons. Using difopein (pan inhibitor of 14-3-3) on the cells, we find that there is a decrease in a-syn release but an increase in toxicity. When the isoform 14-3-3theta is overexpressed in these isyn cells, we find that there is an increase in a-syn release, but a decrease in toxicity. Using the culture model which a-syn is over expressed, we used the shRNA lentivirus directed against 14-3-3theta. We successfully knocked down 14-3-3theta and we plan on using these 14-3-3θ KD isyn cells to analyze a-syn release by western blot and measure the corresponding toxicity. We will test if the absence of this isoform alone will alter release and toxicity or if the other six isoforms will compensate in its absence.

27-The examination of domoic acid content in oil dietary supplements, urine and kidney mouse samples using liquid chromatography–mass spectrometry.

Mohamad Noor Alsirafi, Catherine M Fuller, Phillip Darwin Bell.

Domoic acid (DA) is a naturally occurring neurotoxin produced by marine algae belonging to the genus Pseudo-nitzschia, and underlies the neurological disorder known as amnesic shellfish poisoning. Previous studies have shown that exposure to low levels of DA can also result in renal injury. This could have a significant impact on human health through ingestion of higher marine organisms. Oil supplements such as fish and krill oils are more likely to contain detectable levels of DA due to the fact that the supplements are derived from marine organisms' livers. This study examined five brands of oil dietary supplements that are commercially available (G.N.C Fish Oil, Rite Aid Triple Strength Krill Oil, Nature's Bounty Co-Q-10 with krill oil, Sundown Naturals Triple Strength Red Krill Oil, and Natrol Omega-3 Krill Oil). LC-MS was used to determine the amount of DA in these samples. Extraction methods were developed using hexane and water to maximize the percentage of recovered DA in the samples. The study found no detectable amounts of DA in the oil supplements, (limit of detection was 0.1 ng/mL). The study also examined 24h urine samples of six mice (3 males, and 3 females) treated with DA at different doses (0.05 mg/kg, 0.005 mg/kg, and control), after 7 days of exposure to DA. Our results show that female mice excrete lower amounts of DA over 24h than male mice. Further research is therefore needed to determine the physiological basis underlying the gender difference in these results.

28-Dissecting the TORC1 gene interaction network by perturbing different subunits

César J. Torres Gutiérrez, Sean M. Santos, John L. Hartman, IV

Target of Rapamycin Complex 1 (TORC1) is a kinase that controls cell growth. Regulation of TORC1 activity contributes to aging and the human orthologous complex mTORC1 often becomes dysregulated in cancer cells. Rapamycin (a TORC1 inhibitor) is an immunosuppressive agent used for patients with organ transplantation or cancer. In Saccharomyces cerevisiae, high Rapamycin concentrations create a nutrient starvation physiology resulting in decreased protein synthesis and entrance into a G0 state. TORC1 is composed of multiple subunits (Kog1/Lst8/Tco89/Tor1/Tor2). While the TORC1 complex has been extensively studied, we aim here to understand the full extent to which gene interactions vary between different members of the complex. Such gene interactions may reveal undiscovered roles in the TORC1 complex or different pathways that modulate TORC1 function. A tetracycline-regulated allele of KOG1 was introduced into the complete collection of yeast gene knockout and knockdown (YKO/KD) strains, and these ~6000 double mutant strains were grown in different concentrations of Doxycycline (0-5µg/ml) with an otherwise sub-growth-inhibitory concentration of Rapamycin (5nM). Kinetic parameters from growth curves were obtained by quantitative high throughput cell array phenotyping (Q-HTCP) method, and used to measure gene interactions. By analyzing the growth of the Tet-Off-Kog1 YKO/KD and comparing them to a distribution of references, we identified gene deletions buffering cell proliferation against loss of TORC1. These new data were compared to analogous data for Tor1 obtained previously. 155 gene deletions aggravated the loss of Kog1 and 196 aggravated the loss of Tor1 with an overlap of 34, suggesting gene interactions vary between subunits.

31-Selective targeting of histone methyltransferase EZH2 inhibits aggressive bladder cancer.

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Histone methyltransferase EZH2 is overexpressed in multiple aggressive cancers including prostate, breast and bladder cancer. Increased expression of EZH2 in cancer leads to the down regulation of tumor suppressor genes such as E-cadherin as well as multiple microRNAs. Thus, therapeutic targeting of EZH2 may serve as an effective strategy in treating aggressive cancer. Treating bladder cancer cells with the EZH2 inhibitor GSK126 reduced bladder cancer cell proliferation, and in vivo chicken embryo chorioallantoic membrane assays suggested reduced tumor growth upon EZH2 inhibitor treatment. Gene expression profiling data using RNA from EZH2 inhibitor treated cells identified multiple re-activated target genes. We further validated one of the re-activated targets SQSTM1 by performing qPCR using RNA from EZH2 inhibitor treated cells and controls. In addition, we confirmed the re-activation by performing immunoblots using specific antibodies. Our investigations suggest that treating bladder cancer cells with an EZH2 inhibitor blocks cancer cell proliferation and invasion. Furthermore, we showed that inhibition of EZH2 results in the re-activation of multiple genes including autophagy regulator SQSTM1. In summary, our study suggests that targeting EZH2 may be an effective therapeutic strategy in treating aggressive bladder cancer.

32-A Studying the functions of the HP1 protein family in Drosophila melanogaster

Anthony Foster

The Heterochromatin protein 1 (HP1) protein family represents proteins that support cellular processes including but not limited to DNA repair, gene silencing, and telomere maintenance. The HP1 family is highly conserved from Drosophila to humans, and within the genome of Drosophila are at least five HP1 paralogs that have different roles. HP1a is mainly associated with heterochromatin while HP1C is mainly associated with euchromatin. HP1B is associated with both heterochromatin and euchromatin. HP1D and HP1E are associated with female and male germline expressions respectively. The mechanisms by which these proteins function, however, are poorly understood. In order to improve our knowledge of the characteristics of HP1 proteins, we generated a double mutant line of Drosophila which combines a null mutant allele of Su(var)205 (HP1a) with a null mutant allele of HP1c. This double mutant reduces the expressions of HP1a and HP1c and provides focus on the functions of HP1B. To generate this stock, males carrying a mutation in Su(var)205 were mated with females carrying a mutation in HP1c; the reciprocal cross (switching male and female genotypes) was also arranged. The progeny from these crosses were screened for physical markers indicating that both the HP1a and HP1c mutant alleles were present to confirm the successful generation of a double mutant line. The double mutant line will be able to be utilized in various characterization studies – climbing assays, feeding assays, and lifespan assays – in order to better understand the functions of HP1b in the absence of other HP1 proteins.

33-Female rats acclimate more rapidly than males when challenged with a high salt diet: Possible role for skin sodium regulation

Elizabeth M. Daugherty, Eman Y. Gohar, Kelly A. Hyndman, Jermaine G. Johnston, Joshua S. Speed, Chunhua Jin and David M. Pollock

Men are more vulnerable to hypertension and cardiovascular disease than premenopausal women of the same age. Kidneys and skin work together to maintain sodium homeostasis. Females elicit a more rapid natriuretic response to a high salt (HS – 4% NaCl) diet. Vascular endothelial growth factor-C (VEGF-C) contributes to skin sodium storage by enhancing lymphangiogenesis and is generally up regulated on HS diets. We hypothesized that females mobilize their skin sodium storage during a HS diet due to an increase in VEGF-C. Male and female Sprague Dawley rats (16-18 weeks old) were implanted with telemeters, placed in metabolic cages and fed a normal salt (NS - 0.49% NaCl) diet for five days before being switched to a HS diet for five days. Blood pressure data, kidneys, skin and urine were collected. Western blots performed on kidney cortices after one day of HS revealed no significant differences between sexes on NS and HS in NCC, ENaCα, NHE3, and NKAα5 sodium transporters. Creatinine clearance increased in males and females on Day 1 HS (0.48 ± 0.07 vs. 0.86 ± 0.04; 0.38 ± 0.09 vs. 0.83 ± 0.18 ml/min/100g, $p<0.05$, respectively). Skin sodium in females only decreased when shifted to HS (99.46 ± 3.06 vs. 79.65 ± 5.69 μmol/g). Serum osmolality, urea and VEGF-C decreased on Day 1 HS in males only (urea: 0.42 ± 0.02 vs. 0.28 ± 0.01 mg/ml; VEGF-C: 214.93 ± 10.35 vs. 169.59 ± 11.50 pg/ml $p<0.05$). Collectively, these data suggest that female rats adapt more rapidly to increased dietary sodium intake and suggests important roles for urea and VEGF-C. Funded by AHA 15POST25090329 to EYG, P01 HL095499 to DMP, DK079337 to KH and UAB-UCSD O'Brien Center

35-Regulation of SIN3A and SIN3B expression in metastatic breast cancer cells by unique microRNA

Ananya Bandyopadhyay, Monica J. Lewis, Jianzhong Liu, Mick D. Edmonds, and Douglas R. Hurst

SIN3 chromatin remodeling complexes have been demonstrated to regulate breast cancer progression and metastasis. More recently, the Hurst lab has shown differential functions for the two paralogs of SIN3, SIN3A and SIN3B, with data supporting metastasis suppressive roles for SIN3A and metastasis promoting roles for SIN3B. Although SIN3A and SIN3B share a significant amount of sequence similarity, they are quite different within their 3' untranslated region (UTR) leading us to hypothesize that SIN3A and SIN3B expression is regulated by unique microRNA (miRNA). We utilized three publicly available databases (TargetScan.org, miRDB.org, and microRNA.org) to obtain putative miRNA that target SIN3A and SIN3B. We identified 9 miRNA that are predicted to target SIN3A, 5 miRNA for SIN3B, and 4 miRNA for both paralogs. Future studies will validate the direct binding of these putative miRNA to the 3' UTR of SIN3A or SIN3B using luciferase reporter assays and test the ability of each validated miRNA to lower the expression level of SIN3A or SIN3B in human cell lines. A more complete understanding of how the expression these genes is uniquely regulated is an important step towards comprehending their role in the process of breast cancer metastasis and may help elucidate possible targeting strategies for patients.

36-Evidence for altered circadian heart rhythms in response to elevated dietary Na+ in rats.

Isaac D. Campos, Joshua S. Speed, Malgorzata Kaztan, Jermaine E. Johnston, Kelly A. Hyndman, Martin E. Young, and David M. Pollock

Circadian rhythms in physiologic function are mediated by a group of transcription factors that oscillate over a 24-hour period, termed the molecular clock. Disruption of clock mechanisms promotes cardiovascular disease (CVD). High salt (HS) intake promotes endothelin-1 (ET-1) production by the vasculature and is a risk factor for CVD; however, it is unknown if HS affects circadian clock components in the heart. Therefore, we hypothesized that HS intake disrupts components of the molecular clock in the heart, via activation of ET-1 receptors. Control and endothelin type B receptor (ETB) deficient (ETB def) rats were placed on either HS or normal salt (NS) for two weeks and euthanized every 4 hours over a 24-hour period. RNA expression was assessed for clock genes (Bmal1, Cry1, Per1, Per2, DBP, CLOCK) by RT-PCR. Cosinor analysis was performed to determine mesor (daily average), amplitude (trough-to-peak), and phase (timing of peak) of gene expression oscillations. Our results indicate that HS intake suppresses both the mesor and amplitude of Per1, Per2, CLOCK, Cry1, DBP, and Cry2 expression. These effects occurred similarly in control and ETB def rats. HS intake increased heart ventricle weight (HVW) to a similar level in both genotypes. Our data indicate that HS intake disrupts the molecular clock and promotes hypertrophy in the heart independent of ETB receptor activation. We speculate that alteration in circadian components may contribute to CVD risk associated with high dietary Na+.

37-Maresin 1 Directs the Resolution of Inflammation Post-Myocardial Infarction

Roman Travis, Vasundhara Kain, Kevin A. Ingle, Priscilla Maddox, and Ganesh V. Halade

Introduction: Current inflammation inhibitory measures failed to improve healing post myocardial infarction (MI). Resolution of inflammation presents a unique avenue for clearing inflammation though the effects on heart failure pathology are unclear. Hypothesis: Tested whether Maresin-1 (MaR1) resolves post-MI inflammation thereby limiting pathological remodeling.

Methods: 8-12 week old male C57BL/6J mice underwent permanent coronary artery ligation surgery and were treated daily 3 hours post-MI with Maresin (10ng/25g) until day 1 (n=7) or day 5 (n=14). Mice with no treatment of MaR1 served as MI-controls, and mice with no-MI served as d0 naïve controls. Post-MI neutrophils, macrophages and collagen density were measured using histology. Left ventricle (LV) function was measured using echocardiography. Pro-resolving and pro-inflammatory immune cell markers were assessed using real-time PCR followed by immunoblotting.

Results: MaR1 treated mice displayed increased neutrophil clearance at day 5 post-MI. MaR1 treated mice displayed no change in macrophage density, however, mRNA levels of pro-inflammatory markers i.e CD10, TNFα, IL-6, and IL-1β were decreased at day 5 post-MI (all $p<0.05$). Mice treated with MaR1 displayed improved cardiac output and ejection fraction. Post-MI MaR1-treated mice displayed increased levels of pro-resolving markers YM-1 and Arg-1 at day 1 post-MI with decreased levels of the lipoxygenase (Alox-12,-15 and -5) at day 5 post-MI compared with MI-control group. However, post-MI fibrosis remained unaltered in MaR1 treated group compared with MI-control.

Conclusion: Maresin-1 increases pro-resolving markers in post-MI heart failure pathophysiology thereby protecting heart function.

38-Cell and Gene Therapies for Rescue of Cooley's Anemia Mouse Model

Shaidy Moronta, Jonathan Lockhart, Michael Berlett, Suean Fontenard, Shanrun Lui, and Thomas M. Ryan

β-thalassemia is a group of inherited blood disorders that result in defects in β-globin chain production. Cooley's anemia (CA), or β-thalassemia major, is the most severe form of the disease and occurs when an individual has mutations in both copies of the adult β-globin gene. Patients with CA fail to make adult hemoglobin, have ineffective erythropoiesis, suffer from severe anemia, and are transfusion dependent for life. Interestingly however, individuals with mutations that promote fetal hemoglobin expression in adulthood, termed 'hereditary persistence of fetal hemoglobin' (HPFH) mutations are protected from β-hemoglobinopathies like β-thalassemia and sickle cell disease. In light of this and recent advances in targeted nuclease technology, several genome editing approaches aimed at reactivating the silenced γ-globin gene are under active investigation. The transcription factor Bcl11a, which strongly represses γ-globin expression, has been targeted by the Ryan lab using CRISPR/Cas9 microinjection in embryos derived from a humanized mouse model of erythropoiesis. Cleavage by Cas9 and imperfect repair via Non-homologous end joining (NHEJ) can produce indel mutations in an erythroid specific enhancer element within the second intron of the Bcl11a gene. This mutation would reduce Bcl11a expression in erythroid cells and produce elevated levels of γ-globin expression.

Founder mice derived from these microinjected embryos displayed a mosaic mixture of mutations. Droplet digital PCR (ddPCR) was employed to quantify the overall efficiency of indel formation in the Bcl11a enhancer site in these founder mice. Quantifying the efficiency of Cas9 editing is important for movement into clinical applications and potential therapies for β-hemoglobinopathies.

39-Exosomes secreted by Human Broncho-Epithelial cells change in gene and miRNA expression after hyperoxia exposure

Abby Fyfe, Nelida Olave PhD, Brian Halloran MS, Namasivayam Ambalavanan MD

Introduction: Preterm infants often require respiratory support including supplemental oxygen, exposing lungs to higher than normal oxygen concentrations (hyperoxia). This oxidative stress can harm lung cells and lead to chronic lung disease called bronchopulmonary dysplasia (BPD). The mechanisms by which oxygen induces injury are little understood at the cellular level. Our laboratory has become interested in how lung cells communicate with each other via release of small particles called exosomes, and how changes in these exosomes may mediate some of the effects of oxygen toxicity.

Methods: Human broncho-epithelial cells (HBE) were exposed to both hyperoxia and room air. Exosomes were isolated from the media supernatants in which the cells are grown. The exosomes were counted and analyzed for vascular endothelial growth factor (VEGFA), basic fibroblast growth factor (FGF-2), and three microRNAs involved in lung injury (miR214, miR219, and miR876).

Results: Hyperoxia exposure decreased VEGFA and FGF-2, increased miR214, and did not change miR219 compared to room air controls. miR876 was not undetected in exosomes. The overall number and size of the exosomes increased with hyperoxia.

Discussion: The alterations in exosomes induced by hyperoxia may mediate some of the effects of oxygen toxicity. The decrease in VEGFA and FGF-2 as well as increases in miR214 may impair lung development. The lack of change in miR219 and the lack of miR876 indicate that these miRNA probably do not affect the development of BPD via exosomes.

40-Development of an Ift88 Conditional Knockout Allele in the Rat Using CRISPR/Cas9 Technology

Daniel La Rota1, Laura J. Lambert1, Anil K. Challa1, Bradley K. Yoder2, Robert A. Kesterson1

Primary cilia are present in vertebrate cell membranes and play a major role in extracellular reception. Intraflagellar Transport 88 (ift88) encodes a ciliary protein that is linked to polycystic kidney disease and situs inversus and is highly conserved in various model organisms. To further investigate the role of ift88 a conditional knockout allele is being generated using CRISPR/Cas9 technology in the rat to avoid embryonic lethality and allow induction of time and tissue specific loss of the protein. CRISPR guides were designed to target introns 3 and 6 to induce double strand breaks and incorporate repair templates containing loxp sites by homology-directed repair. Breeding flanking loxp allele bearing animals with Cre recombinase lines will excise exons 4-6 and yield a nonfunctional protein. Pronuclear microinjection of rat embryos with CRISPR/Cas9 reagents resulted in eight founder animals of 47 embryos transferred (17.0%). Tail genomic DNA was analyzed by PCR and PAGE for CRISPR activity. Samples showing single bands were sequenced directly and those with multiple bands, indicative of insertions and deletions (indels), were cloned before sequencing to discern the nature of the mutations. Nine indels were detected (56.3%) among which two specimens were positive for loxp insertions with one having the desired insertion at both targeted loci (18.8% knock-in efficiency). Long range PCR amplifying the entire 2.5 Kb region will determine the cis/trans status of the inserted loxp sites. Phenotyping rats after loss of this conditional knockout allele will allow further understanding of human ciliopathies.

41-Renal denervation reduces neuronal activity markers in the brain stem of normal rats but not salt-sensitive hypertensive rats on high salt diet

Amanda C. Feagans, Bryan K. Becker, David M. Pollock

Endothelin-1 (ET) modulates sympathetic nerve activity and sodium excretion, but the connection between sympathetic nerves and ET in controlling salt-sensitive hypertension is not understood. We hypothesized that renal denervation reduces markers of neuronal activity in autonomic control areas of the brain in a model of ET dysfunction and salt-sensitive hypertension. Endothelin B receptor deficient (ETB-def) and transgenic control (TG) rats underwent bilateral renal sympathetic denervation (Dnx) or sham operation and were fed a normal salt (0.49% NaCl) diet for 5 days followed by14 days of high salt (4.0% NaCl) diet. We measured norepinephrine and glutamate content via ELISA in the paraventricular nucleus (PVN) and the nucleus tractus solitarius (NTS). All data are expressed relative to TG sham levels. There was less norepinephrine in the NTS of TG Dnx (0.411 ± 0.148; p=0.047; n=4-6/group), but not in ETB-def sham or Dnx rats (0.992 ± 0.133 vs. 0.951 ± 0.121, respectively; p=0.995; n=7/group). Glutamate was similar across groups (1.00 ± 0.0874 TG sham vs 0.857 ± 0.124 TG Dnx; p=0.670; 0.748 ± 0.0658 ETB sham vs. 0.839 ± 0.0664 ETB dnx; p=0.845). Norepinephrine and glutamate in the PVN were not different between groups or treatments. We conclude that on a high salt diet, denervation did not affect markers of neuronal activity in central autonomic control centers in the ETB-def model of hypertension; however, denervation may reduce central autonomic activity in normal controls on high salt. Future studies will evaluate the effect of denervation on central autonomic control areas following normal salt diet.

42-Targeting DNA methylation at Bdnf exon IV using CRISPR

Alexis Pulliam, Anderson Butler

The understanding of memory loss in epileptic patients is still incomplete. Many patients suffer from traumatic loss of memory, and pharmaceutical treatments are often ineffective. To better understand epilepsy, we are studying the epigenetic changes associated with the disorder. Epigenetics is the study of the modification of gene expression. One of the most extensively studied mechanisms is DNA methylation. DNA methylation is the process of silencing genes by adding a methyl group directly to the DNA at specific sites on the nucleotide residues. In epileptic rodents, the Bdnf gene expression is increased. Previous studies used methyl supplements and treatments to manipulate DNA methylation on Bdnf on a global scale. DNA methylation can decrease the expression of the gene. However, no one has used a targeted method to increase DNA methylation. Therefore, the objective of the project is to target DNA methylation at the Bdnf exon IV using a CRISPR effect system. Plasmids were grown and the endonuclease restriction sites, BstXI and XhoI, cleaved the sequence creating a fragment with sticky ends. The new target specific sequence was ligated into the plasmid. This will be used in conjunction with the dcas9-DNMT3a to mediate site-specific methylation. In the cell, the Bdnf gene expression will decrease and methylation will increase, potentially impacting memory in rodent models.

44-The Effects of HIV-Tat and Morphine on M17 Neuroblastoma Cells

Felicia Peoples,Tonia Tse, Emily Haley, John J. Shacka

Introduction: Parkinsonism has been reported in HIV/AIDS, and alpha-synuclein (αsyn), involved in Parkinson's disease pathogenesis, is higher in postmortem HIV patient brains. Many HIV/AIDS patients take chronic opiates for pain. Recent evidence indicates combined treatment with morphine and the HIV protein Tat exacerbates neurotoxicity. Whether the opiate morphine +/- Tat regulates αsyn metabolism and autophagy lysosome pathway (which mediates αsyn clearance) markers in HIV/AIDS is unknown. Objective: Determine if morphine +/- Tat regulates cell viability, levels of endogenous αsyn/ALP markers, or clearance of over-expressed αsyn.

Methods: M17 human neuroblastoma cells were used that conditionally over-expresses human wild-type αsyn. Cells were treated with HIV-Tat (100nM) +/- morphine (0-500μM). Cell viability was measured via MTT assay. Western blot analysis assessed levels of endogenous αsyn and LC3-II (autophagosome marker) and p62 (ALP substrate), and clearance of over-expressed αsyn.

Results: Morphine induced cell death at ≥100μM; Tat did not affect viability in the presence or absence of 500nM morphine. Western blot analyses suggested reductions in endogenous αsyn, LC3-II and p62 with 500nM morphine ± 100nM Tat. 500nM morphine did not affect clearance of over-expressed αsyn.

Discussion: It appears that M17 cells are less sensitive to Tat + morphine than cultured neurons, though previous studies have indicated that glial cells are important for mediating Tat +morphine toxicity. Western blot results suggested that 500nM morphine + Tat induced ALP function. Ongoing studies are determining the effects of higher morphine concentrations on Tat-induced toxicity of M17 cells, and whether glial cells are important for eliciting these effects.

45-Novel preclinical rat model of late onset Parkinson's disease

Sidhanth Chandra, Tyler Maltbie, Hisham Abdelmotilib, Valentina Krendelchtchikova, Xianzhen Hu, Vedad Delic, Andrew West

Parkinson's disease (PD) is a debilitating movement disorder affecting millions worldwide. PD is characterized by the loss of dopaminergic neurons in the substantia nigra pars compacta (SNpc), leading to a dopamine deficiency in the striatum. Lewy bodies (LBs) are protease resistant, insoluble aggregates of alpha-synuclein protein (α-syn) and are the histological hallmark of PD. Accumulating evidence suggests that intermediate size α-syn oligomers, precursors of LBs, are able to seed abnormal aggregation of endogenous α-syn and are, therefore, thought to be the causative species of PD. Most of what is known about PD comes from studies of rare genetic mutations in the α-syn gene, over expression of α-syn using viral vectors, and studies in immortalized cell lines. These models do not fully recapitulate the pathology observed in late onset PD. Absence of a pre-clinical model of PD with face validity is likely the reason why to date most PD therapeutics have failed in clinical trials. Preformed fibrils (PFFs), generated from recombinant α-syn, are able to seed the recruitment of endogenous α-syn into aggregates without the need for over expression. The objective of this study was to create the most relevant preclinical model for late onset PD by injecting sonicated PFFs into SNpc of rat brains. We report caudo-rostral cell-to-cell transmission of abnormal α-syn aggregates, degeneration of dopaminergic neurons, and decrease in tyrosine hydroxylase positive cell innervation of the striatum. We also report LB inclusions localize to medium spiny neurons in the striatum.

46-Endothelial-derived endothelin-1 (ET-1) does not contribute to α1-adrenergic sensitivity in the aorta

Hannah Clark, Brandon Fox, Paramita Pati, Jennifer Pollock

Acute behavioral stress induces increased circulating ET-1 levels and elevated blood pressure in both humans and rodents. We have previously shown that endothelial-specific ET-1 knockout (VEETKO) mice do not exhibit an acute stress-induced increase in circulating ET-1 and exhibit a blunted blood pressure response to acute stress compared to control mice. Additionally, endothelial-derived ET-1 is implicated as a potent pro-inflammatory stimulus. We hypothesized that endothelial-derived ET-1: 1) mediates sensitivity to α1-adrenergic constriction in vascular tissue, and 2) promotes systemic inflammation. To test the first hypothesis, we conducted wire myography utilizing ex vivo aortic rings from VEETKO and control mice to generate cumulative concentration-response curves to the α1-adrenergic receptor agonist phenylephrine. In the aorta, no difference was observed in EC50 (-7.07±0.05 vs. -7.03±0.06 log[PE,M], p=0.57) or maximum constriction (90.2±0.6 vs. 88.3±2.0, %KCl, p=0.39), in control vs. VEETKO mice respectively, suggesting that endothelial-derived ET-1 does not contribute to the aortic sensitivity to α1-adrenergic activation. To test the second hypothesis, male mice were subjected to acute behavioral stress, and at 30 minutes post-stress, plasma cytokine and pro-inflammatory marker levels were examined using a 144 target protein array. E-Cadherin was significantly lower (p=0.03), and IGFBP2 was higher (p=0.01), in plasma of VEETKO compared to flox mice. No difference was observed in the other 142 markers examined. Together, these findings suggest that the aorta does not contribute to the blunted blood pressure response to acute stress in VEETKO mice and that endothelial-derived ET-1 does not considerably affect systemic inflammation after 30 minutes of behavioral stress.

47-Inhibition of HGF activation: A Novel Approach to Overcome Resistance to MET-Targeted Therapy in Lung Cancer

Shantasia Thomas, Benjamin Y. Owusu, Phanindra Venukadasula, Robert A. Galemmoand and Lidija Klampfer

Abberant HGF/MET signaling supports proliferation, invasion, metastasis and angiogenesis. Lung cancer cells with amplified MET are sensitive to MET kinase inhibitors. Our preliminary data demonstrated that hepatocyte growth factor (HGF) inhibits the responsiveness of the cells to the MET tyrosine kinase inhibitor (TKI), JNJ-38877605. Fibroblasts are the predominant source of HGF in the tumor microenvironment. HGF is secreted in an inactive form called pro-HGF. Proteolytic processing of the single-chain pro-HGF to the active (mature) heterodimeric form by one of the serine proteases, hepatocyte growth factor activator (HGFA), hepsin, or matriptase is required for HGF/MET signaling. We developed a novel inhibitor of HGF activation, SRI 31215, and tested its ability to overcome HGF-mediated resistance to JNJ-38877605 in lung cancer cells with amplified MET. The primary objective of this study was to show that concurrent inhibition of the ligand (HGF) and its receptor (MET) is required to inhibit proliferation of MET-amplified lung cancer cells. We showed that both recombinant HGF and fibroblasts inhibit the sensitivity of MET-amplified lung cancer cells, EBC-1 and H1993, to JNJ-38877605. Inhibition of HGF activation by SRI 31215 overcomes fibroblast-mediated resistance to JNJ-38877605. Conversely, A549 lung cancer cells which do not have amplified MET, did not respond to JNJ-38877605. Taken together, we have demonstrated that small-molecule inhibitors of HGF activation, such as SRI 31215, would greatly improve the outcome of lung cancer patients that display amplification of the MET receptor.

50-Investigating role of breast tumor kinase/protein tyrosine kinase 6 (Brk/PTK6) during zebrafish development using TALEN-generated knockout alleles.

Samantha Foster, Sarah Glover, Kiranam Chatti, Anil Kumar Challa, and Robert Kesterson

Breast tumor kinase/protein tyrosine kinase 6 (Brk/PTK6) was initially discovered in a screen of tyrosine kinases overexpressed in metastatic breast cancer tissue. It is currently known to be expressed in several cancer types, and cell culture studies indicate a pro-oncogenic role for PTK6. PTK6-null mice are viable, but show hyperproliferation of colonic villi and hyperactivation of AKT. Contrasting evidence supporting a pro-apoptotic role of PTK6 has also been reported. In cultured non-cancer cells, PTK6 appears to sensitize the cells to inducers of apoptosis. The contrasting functions of PTK6 are likely to be a result of cell-type specific genetic interactions, biochemical regulation and environmental effects during oncogenesis. In order to gain a comprehensive understanding of the developmental role of PTK6 genes and to potentially explain their contrasting effects on cell growth and differentiation in vertebrates, we generated several indel alleles of the two zebrafish genes ptk6a and 6b using TALENs. Our current findings suggest that homozygous mutant animals are viable presumably because these two genes complement each other, or because of maternal expression of the genes that can rescue the zygotic loss-of-function. Further characterization is underway to generate and analyze double mutants as well as maternal-zygotic mutant animals for each of these genes to identify any developmental roles.

51-Ultrastructural Study of the Oligodendrocytes in Postmortem Schizophrenia Substantia Nigra

Vidushi Sinha, Courtney K. Walker, Rosalinda C. Roberts

Schizophrenia (SZ) is a neurological disorder characterized by abnormal social behavior, often as a result of hallucinations, delusions, and reduced emotional expression. It affects about 1% of the population worldwide. Dysregulated dopamine levels have long been implicated in the disorder; the main locus of dopamine neurons is in the midbrain, specifically the substantia nigra/ventral tegmental area. The focus of this study, the substantia nigra (SN), is a basal ganglia nucleus that projects to the striatum; hyperactivity in the SN has been seen in many imaging studies (Yoon et al, 2013, 2014). Dysfunctional oligodendrocytes and resulting abnormal myelin production have also been implicated in changes in synaptic formation, which may lead to the cognitive dysfunction seen in SZ. The objective of this study was to determine the presence of morphological differences in oligodendrocytes between controls (n=8) and SZ patients (n=13). Postmortem SN was obtained from the Maryland Brain Collection and analyzed using electron microscopy. Cellular area, nuclear area, percentage heterochromatin/euchromatin, cytoplasmic regularity and density, and myelin inclusion, among other characteristics were recorded. Data for normal controls was compared to that of treatment responders, and treatment resistant/off drug patients. Many of the variables measured showed no significant difference, though there was in fact a significant reduction in myelin inclusions in SZ subjects ($p = .022$, 1.531 ± 0.119 NC vs. 1.682 ± 0.225 SZ). This study contains possible limitations, including small sample size, variable antipsychotic drugs used, and use of postmortem tissue. Knowledge of morphological regularity progresses us towards a comprehensive understanding of SZ.

54-Evaluating lipid changes in septic kidney injury using SWATH lipidomics coupled with MALDI tissue imaging

Kelly B. Walters, Landon Wilson, Bo Chen, Subhashini Bolisetty, Stephen Barnes, Anupam Agarwal, Janusz H. Kabarowski

In a previous study, we discovered two candidate lipids for early time-point biomarkers of AKI and demonstrated that mass spectrometry lipidomics coupled to imaging mass spectrometry can be used for the molecular characterization of kidney injury pathobiology. In this study, we have been testing different tissue preparation methods in order to increase lipid and metabolite imaging sensitivity, including using different matrices that may better ionize lipids, and washing tissue sections to replace ion adducts in order to simplify the spectra and enhance lipid signal intensity. We have also been able to significantly increase the speed of tissue section processing with recent modifications to the vacuum sublimation system that we developed for matrix application last year; this has helped us to move much closer to being able to provide “high throughput” imaging in which multiple sections from a single tissue/organ can be imaged in order to localize a lipid or metabolite and render a 3-D image of its expression throughout the organ. Using these modifications made this summer, we are now starting work to determine whether similar or distinct lipid changes are associated with the development of acute kidney injury during sepsis, a major cause of acute kidney injury clinically. This work has allowed us to solicit support from much bigger and more diverse groups of UAB investigators/researchers for our upcoming UAB HSF-GEF grant application that will fund our future work aimed at integrating our Imaging Mass Spectrometry system into the UAB Core research technology Infrastructure.

57-Osteoblast-derived Runx2 Regulates Bone Marrow Cell Populations

Courtney Swain, Justin Gibson, Yang Yang

Multiple myeloma (MM) is an incurable plasma cell malignancy that preferentially grows in the bone marrow (BM). MM frequently metastasizes from the primary bone site to new bone sites, resulting in a 5-year survival rate of only 43%. Within the BM, MM cells form complex interactions with resident BM cells (i.e. osteoblasts, osteoclasts, immune cells, etc.). MM cells remodel the extracellular matrix and secrete soluble molecules that favor further MM progression at both primary and secondary sites. Recent studies have demonstrated that in MM, osteoblast (OB) differentiation and bone formation are inhibited via the suppression of runt-related transcription factor (Runx2) in OBs. Importantly, we have shown that suppression of OB-Runx2 also occurs at distant sites prior to arrival of MM cells in these sites. OBs, under the control of Runx2, also interact with other components of the BM microenvironment. The goal of this study was to investigate the effect of OB-Runx2 suppression on immune cells in the BM microenvironment. To study the impact of OB-Runx2 suppression on BM microenvironment, we have developed an animal model in which Runx2 is specifically deleted in OBs (Runx2 OB-/-). Tissues obtained from wild type (Runx2 OB+/+) and homozygous (Runx2 OB-/-) C57Bl/KaLwRij mice were used in immunohistochemistry to observe different cell types within the BM microenvironment. Results demonstrated an increase in the population of cytotoxic T cells and a decrease in osteoblasts and macrophages in 5 week and 3 month-old Runx2 OB-/- mice, compared to Runx2 OB+/+ control mice . Further studies will aim at determining immune regulatory cells, such as T reg cells, MDSCs, and soluble molecules in the BM microenvironment, that are altered by OB-Runx2 suppression.

58-Purification Strategy of Active RNA Polymerase I from Stationary Phase Saccharomyces cerevisiae

Hannah Newman, David Schneider

There are three nuclear RNA polymerases expressed in Eukaryotes. RNA polymerase I is responsible for the synthesis of ribosomal RNA (rRNA) which is folded in the core of the ribosome. Ribosome synthesis is proportional to cell growth and proliferation. Due to this correlation, RNA polymerase I is emerging as a major cancer therapy and requires more characterization. Our lab characterizes RNA polymerase I function, using Saccharomyces cerevisiae as a model system. Currently, RNA polymerase I is isolated and purified from cells harvested during the log growth phase. This takes a large quantity of Saccharomyces cerevisiae to get a small amount of polymerase, which proves to not be cost effective or practical. The aim of this project is to create a purification strategy of RNA polymerase I using stationary phase cells to be able to recover larger quantities of active polymerase from smaller media quantities. This will be done using Fast Protein Liquid Chromatography (FPLC). We hypothesize that if RNA polymerase I can be isolated from the stationary phase, that it will be just as active from being purified from the log phase. Through this experiment, we were able to perform a purification that resulted in partial purification, where protein was lost in one of the intermediate steps. We believe that with a different nickel column, protein would not have been lost. In the future, we hope to redo this procedure, while adding a Mono Q column after the final nickel column to ensure we only isolate our polymerase

61-Targeted Transcriptional Activation of Brain-Derived Neurotrophic Factor Using CRISPR/dCas9 Tools

Roxanne Robidoux, Svitlana Bach, Faraz Sultan, Katherine Savell, Jeremy Day

Brain-derived neurotrophic factor (Bdnf) is a complex gene important for synaptic plasticity, neuronal differentiation, and memory formation. In addition, Bdnf dysregulation has been implicated in neurocognitive and neuropsychiatric disorders. The Bdnf gene is complex, with nine different 5' non-coding exons (each with a unique upstream promoter) and one 3' coding exon. While each exon is capable of producing a unique mRNA through differential splicing, all variants are translated into the same mature protein. However, although neuronal depolarization, synaptic potentiation, and memory formation have all been shown to induce expression of specific Bdnf transcript variants, the function of these variants remains poorly understood. Here, we describe modular CRISPR-based tools for selective activation of specific Bdnf transcripts in rodent model systems. We designed guide RNAs (gRNAs) targeting promoters for specific variants of the Bdnf gene, which enabled site-specific recruitment of a dCas9 protein fused with VP64 (a transcriptional activator) to variants of interest in primary neuronal cultures. Using RT-qPCR, we show that targeting a specific region in the Bdnf gene with dCas9-VP64 fusion protein is capable of upregulating Bdnf variants. Finally, we show successful transduction of Bdnf-targeting CRISPR constructs in the prefrontal cortex and hippocampus of adult animals using lentiviral vectors. To further understand the roles that different Bdnf variants play in neuronal function, future experiments will utilize single-molecule Fluorescence In Situ Hybridization (FISH) to localize different mRNAs within cultured neurons. Understanding Bdnf transcription using these CRISPR/dCas9 tools will allow for better approaches to study synaptic plasticity disorders by site-specific epigenome editing.

62-Morphological studies of inhibitory interneurons in the medial prefrontal cortex of Rett Syndrome mice

Rahul Gaini, Mary Phillips, Lucas Pozzo-Miller

Although dysfunction of the medial prefrontal cortex (mPFC) has been implicated in autism spectrum disorders (ASDs), the specific structural and functional properties of this brain region remain unclear. To better define the role of the mPFC in ASDs, we utilize Mecp2 knockout (KO) mice because they recapitulate several neurological symptoms presented by individuals with the ASD Rett syndrome. Previous data from our laboratory indicate that the mPFC network is hypoactive in Mecp2 KO mice, and we hypothesize that enhanced activity of inhibitory interneurons is responsible for this network consequence. To characterize inhibitory interneuron populations in symptomatic male Mecp2 KO mice, we performed immunohistochemistry for the Ca2+ binding protein calretinin (CR) and for the neuropeptide somatostatin (SST) as markers for interneuron subtypes. Brain sections were imaged by laser-scanning confocal microscopy and CR-positive neurons were further classified based on their morphology. The number of CR neurons was significantly larger in the Mecp2 KO mPFC compared to WT littermate controls; however, the ratio of CR morphology was not affected. The number of SST neurons was not different in the Mecp2 KO mPFC. As CR expression levels are enhanced by neuronal activity, one interpretation of these results is that CR interneurons are hyperactive in the mPFC of Mecp2 KO mice, which would contribute to network hypoactivity.

65-Bmal1 knockout mice lack diurnal rhythms in food and water intake that mirrors loss of circadian blood pressure rhythm and urinary sodium excretion

Guillermo Najarro, Daian Chen, Ijeoma Obi, Dingguo Zhang, David Pollock, Jennifer Pollock

Many forms of life have circadian rhythms to perform specific biological functions. Bmal1 is a transcription factor and central element of the molecular clock but its role in fluid and electrolyte regulation is unclear. We hypothesized that loss of Bmal1 would disrupt fluid and electrolyte balance in response to normal (0.4% NaCl) and high salt (4% NaCl) diets. To test this hypothesis, we utilized Bmal1 knockout (Bmal1KO) (n=6) and wildtype (WT) mice under both diets in metabolic cages and collected urine during active (lights-off) and inactive (lights-on) phases. Our results show that Bmal1KO mice have reduced water and food intakes on both diets compared to WT in the active phase. In addition, Bmal1KO exhibit a loss of diurnal intake of water and food, which corresponds to a loss of diurnal blood pressure (measured by telemetry) rhythm on both diets. On normal and high salt diet, WT had a diurnal pattern in urinary sodium excretion that is higher in the active phase and lower in the inactive phase (normal salt: 41±4μEq/12h vs. 21±5mEq/12h, P=0.007, respectively; high salt: 490±50μEq/12h vs. 100±20μEq/12h, P<0.0001, respectively). Whereas, Bmal1KO had similar sodium excretion in the active and inactive phases on both normal and high salt diets. Blood pressure followed a diurnal pattern in WT, but not Bmal1KO mice on both diets. We conclude that Bmal1 facilitates maintenance of a normal diurnal pattern in blood pressure control, food and water intake and sodium excretion.

70-Localizing the corticospinal and rubrospinal tracts within the porcine spinal cord

Blake T. Brown, Anna Victoria Leonard, Joshua York Mendez, Ross Dawkins, Betty Maki Pat, Candace L. Floyd

Introduction: Although spinal cord injuries (SCI) are a devastating clinical problem, there continues to be failure in translation of research. Rodents continue to be the main and most well established model for traumatic spinal cord injury research. However, motor tract anatomy differs between humans and rodents. Because of this, some research groups are turning to the pig as an intermediate model due to their extensive anatomical and physiological similarities to humans. However, the neural anatomy and localization of major spinal tracts of the pig spinal cord has not yet been described. The goal of this study is to localize the major motor tracts, the corticospinal tract (CST) and the rubrospinal tract (RST), within the pig spinal cord.

Methods: Mature female domestic pigs received injections of fluorescent dextran tracers into the primary motor cortex and the red nucleus using StealthStation® image guided navigation. 5-6 weeks post injections, the pigs were euthanized and their brain tissue and spinal cord were collected. The tissue was serially sectioned and examined using confocal microscopy to observe to location of the CST and RST.

Results: The results demonstrated that the CST in pigs is laterally located in the white matter, very similar to that of humans. However, the CST does not appear to descend past the cervical regions. The exact location of the RST cannot be determined at this time, but preliminary results also point to a lateral location within the white matter throughout the entire length of the neural axis.

Discussion: The location of the CST and RST in the porcine model are anatomically similar to humans. Further work needs to be done to pinpoint the exact location of the RST, however, these results point to the porcine model as a valuable pre-clinical tool to improve translation of promising SCI treatments.

71-Characterization of HP1b Overexpression strains in Drosophila melanogaster

Melissa L. Garcia, Andrew D. Thomas, Heidi M. Glassie, Tabitha M. Hardy, and Nicole C. Riddle

Chromatin structure plays a vital role in the health and development of all organisms through its contribution to the regulation of gene expression as well as its role in maintaining genome integrity. Heterochromatin, the highly condensed form of chromatin, is maintained partly by members of the Heterochromatin Protein 1 protein family (HP1). The HP1 family is highly conserved, with homologs in both humans and Drosophila. HP1B most closely resembles human HP1 proteins based on its conserved domains. Previous investigations by the Riddle Lab using HP1b null mutant flies have found that HP1B loss impacts aging and metabolism. Our lab has generated strains of Drosophila that carry an additional copy of the gene encoding HP1B. The goal of this work is to provide basic characterization of these novel Drosophila strains and to determine if the additional gene copy increases the amount of total HP1b mRNA and protein. Studies of HP1b null mutants indicate that modulating HP1B proteins levels can impact lower activity levels and life span. Therefore, we are using larval crawling and climbing assays to assess the activity levels of the flies. We plan to utilize starvation assays in the HP1B overexpression lines to determine stress resistance, which serves as a direct proxy for longevity. Finally, we plan to cross the upregulated lines of flies to our null mutant HP1b strains (HP1b16 and HP1b86) in an attempt to rescue the previously observed mutant phenotypes. These experiments will provide valuable insight into how upregulation of HP1B affects the organism's vital processes.

75-Densitometry analysis of vGLUT1, vGLUT2, and GAD67 in the substantia nigra in human postmortem tissue

Sam Mabry, Emma Bloom, Rosalinda Roberts

The substantia nigra (SN) is one of the largest dopaminergic nuclei in the brain and has dopaminergic projections to the striatum. Dopamine abnormalities have long been implicated in schizophrenia because antipsychotic drugs block the dopamine D2 receptor, and there is increased dopamine in the striatum. To date, the SN has been studied very infrequently in schizophrenia. In recent years, imaging studies have shown a hyperactivity of the SN in patients with schizophrenia, resulting in the increased striatal dopamine seen in patients with schizophrenia. In this study, immunohistochemistry was performed on postmortem human brain tissue (5 controls and 6 schizophrenia) with 3 different antibodies: vesicular glutamate transporters 1 and 2 (vGLUT1 and vGLUT2, respectively) and glutamic acid decarboxylase (GAD67), which will measure the amount of glutamatergic (vGLUT1 and vGLUT2) and GABAergic (GAD67) projections to the SN. We hypothesize that there will be an increase in glutamatergic (excitatory) projections and/or a decrease in GABAergic (inhibitory) projections in patients with schizophrenia when compared to normal controls (NC). Through densitometry analysis of the neuropil, we observed no significant differences between schizophrenia and NCs. While we observed no significant results, vGLUT2 (14.6% increase, $p=0.126$) and GAD67 (21.8% increase, $p=0.562$) were slightly increased in patients with schizophrenia. In contrast, schizophrenia subjects exhibited a slight decrease in vGLUT1 (39.3% decrease, $p=0.140$) versus normal controls. However, the current investigation ($n=11$) requires addition of more subjects to increase power before further interpretation. Therefore, despite the slight changes observed, future studies with larger cohorts are needed.

79-Effects of Ghrelin and Restricted Time Feeding on Circadian Clock Rhythms

Joshua Washington, Daniel Mount, Jennifer Davis, Siva Tekumalla, Hira Munir, Assata Pyatt, Karen Gamble

The suprachiasmatic nucleus (SCN), the primary pacemaker of the circadian clock, coordinates 24-hour oscillations throughout the brain and periphery that prepare the body for different changes. When these central and peripheral clocks desynchronize with one another and/or with the environment, negative health effects may ensue— increased adiposity and cognitive decline. When external synchronizing signals (e.g., light and food availability) conflict with each other, the SCN is primarily reset by light exposure, but other pacemakers within the brain (e.g., hippocampus) and periphery (e.g., liver) are synchronized by food intake. The hunger hormone ghrelin is released in a 24-h rhythm, anti-phase to food intake; however, it is not known how food intake, lighting conditions, and ghrelin receptor activation interact to synchronize central and peripheral clocks. Therefore, this project tested whether ghrelin receptor activation that is mis-timed with food intake will differentially entrain circadian clocks in the hippocampus, liver, and SCN. Using a computerized time feeder, two groups of mice (N = 10/group) were fed pellets containing a ghrelin agonist at the beginning of lights on or lights off. These groups were divided into groups that are fed on either a light-restricted or dark-restricted 12-h availability schedule. The peak expression of the clock gene, Period2 (Per2), was measured using a transgenic reporter mouse model that rhythmically produces PER2-Luciferase protein. Chronic bioluminescence monitoring of organotypic cultures of the SCN, hippocampus and liver was used to determine circadian clock phase.

80-Effects of EGCG, an Active Component of Green Tea, on Obesity-related Autophagy

Shelby Luikart, Guang Ren, Teayoun Kim, and Jeong-a Kim.

Epigallocatechin-3-gallate (EGCG), a major component polyphenol in green tea, has been associated with the reduction in the symptoms of Type II diabetes through ECGC-induced autophagy in endothelial cells.1,2,3 However, the dependence of autophagy on these metabolic effects remains inconclusive. The proposed experiments attempted to rectify this disparity by conducting various studies with diet-induced obese (DIO) mice with a deficiency in the endothelial specific autophagy gene 7 (ATG7), which is essential for macroautophagy.2 Four groups of mice (control with or without EGCG, and endothelial-specific ATG7 KO [End-ATG7-KO] with or without EGCG) were exposed to a treatment regimen for 8-12 weeks. All mice received a 45% high-fat (HF) diet, but only two groups (one in the control, one in the KO) received daily gavages of EGCG. After treatment, their body weights were measured and their blood glucose levels were evaluated via the glucose (GTT), insulin (ITT), and pyruvate tolerance tests (PTT). The presence of P62, a protein that targets cells for autophagy,4 was evaluated and quantified via Western blot. Overall EGCG showed similar effects on glucose levels for both wild type and ATG7 knockout mice as well as increased autophagy in iMAEC (Immortalized Mouse Aortic Endothelial Cell) lines. However, long-term treatment with EGCG had adverse effects on insulin tolerance and gluconeogenic activities in both WT and End-ATG7-KO mice. Thus, despite EGCG increasing autophagy, the physiological effects of EGCG on glucose homeostasis may be a detriment to the cell by affecting other metabolic pathways.

81-Schwann cells require the endosomal-sorting pathway for peripheral nerve myelination.

Tina Tian, Julie A. Wilson, and Scott M. Wilson

Disruption of the endosomal-sorting pathway is associated with several neurodegenerative diseases. The endosome serves as a sorting station for internalized cell membrane proteins such as receptor tyrosine kinases, and defects in the sorting of these receptors can affect both the specificity and duration of the receptor tyrosine kinase signaling. Unfortunately, very little is known about endosomal sorting in cells of the nervous system. One neural cell type that is sensitive to endosomal dysfunction is the myelin-producing Schwann cell. The inability of Schwann cells to myelinate axons results in motor and sensory deficits due to impaired propagation of action potentials along the axon. Charcot-Marie-Tooth Disease (CMT) is one of the most commonly inherited neurological disorders and is associated with motor and sensory dysfunction due to loss of peripheral nerve myelination. While many of the mutations linked to CMT are suggested to impair endosomal sorting, a direct examination of endosomal sorting in Schwann cells has not been conducted.

This study investigated the effects of disrupting a core component of the endosomal-sorting pathway in Schwann cells. The endosomal sorting complex required for transport (ESCRT) components function to sort endocytosed proteins through the endosomal compartment. To determine if endosomal sorting is required for peripheral nerve myelination, we disrupted the ESCRT component hepatocyte growth factor-regulated tyrosine kinase substrate (HGS) specifically in Schwann cells. While ESCRTs have traditionally been thought to be negative regulators of cell signaling, our data suggest that HGS is required for the maturation of Schwann cells to a myelinating state. To investigate if the expression of key myelin-inducing pathways is compromised in the HGS deficient mice, we isolated RNA from peripheral nerves and performed quantitative PCR. Our findings suggest that HGS is required for the suppression of several myelin gene repressors as well as for the induction of transcription factors that induce myelin gene expression. They also provide evidence that endosomal sorting is necessary for the developmental transition from a non-myelinating to a myelinating Schwann cell and that endosomal signaling is a positive regulator of myelin gene expression.

86-Immunohistochemical analysis of excitatory and inhibitory input to the substantia nigra in schizophrenia

Emma Bloom, Samuel Mabry, Rosalinda Roberts

Schizophrenia is a widespread psychiatric disorder in which dopamine abnormalities have been implicated. Schizophrenic patients have excessive dopamine in the striatum and antipsychotic drugs primarily block dopamine receptors. The focus of the present study was on the substantia nigra (SN), a basal ganglia nucleus that hosts one of the largest clusters of dopaminergic cells in the brain and projects to the striatum. Previous research has demonstrated hyperexcitability in the SN of schizophrenic patients, but the cause of this is unknown. We aimed to determine whether this abnormality is modulated by increased excitatory (glutamatergic) input and/or decreased inhibitory (GABAergic) input. Postmortem SN of schizophrenic patients (SZ, n=6) were compared to matched controls (NC, n=5). We used immunohistochemistry to localize the vesicular glutamate transporters, vGLUT1 and vGLUT2, and the GABA precursor, GAD67. Densitometry was performed to calculate the density of glutamatergic and GABAergic terminals in the SN of SZ and NC subjects. No significant differences were obtained for the density of vGLUT1 (NC: mean=18.3±5.5; SZ: mean=12.6±6.2), vGLUT2 (NC: mean=36.4±3.6; SZ: mean=41.8±6.4), or GAD67 (NC: mean=26.3±13.6; SZ: mean=33.2±19.7). VGLUT2 trended in the anticipated direction, so we aim to draw conclusive results with a larger sample size. This study contains possible limitations, including a small cohort, confounds arising from the use of postmortem tissue, and our subjects' variable use of antipsychotic drugs. Regardless, in better understanding the reason behind hyperactivity of dopaminergic neurons in the SN of schizophrenic patients, we will be better equipped to identify targets for new treatment mechanisms.

87-Analysis of protein expression to optimize the leading sequence in mitochondria

Ashli Moore, Kah Yong Goh, Huixian Hong, Patrick Ernst, LuFang Zhou

Mitochondrion is an important organelle in cell. Mitochondrial function directly or indirectly contributes to a range of pathological disorders such as cancer, cardiovascular diseases, and diabetes. Optogenetics, a new technique which combines utilization of light and genetics to control cellular activities in living tissues recently emerged. In particular, channelrhodopsin-2 (ChR2), a gene that is responsive to light, has been widely used to control cellular functions in the neuroscience field. In this project, we would like to design and compare ChR2 gene that is targeted to mitochondria with different leading sequences. HeLa cells were plated onto different glass coverslips and transfected with four different plasmids, namely ABCB-ChR2-eYFP, ABCB-YFP (control), Mito-ChR2-eYFP, and Mito-eYFP (control). After 48 hours, the cells were stained with MitoView633 mitochondrial dye and imaged using a confocal microscope. The results showed that ChR2 are better localized to mitochondria in cells transfected with ABCB-ChR2-eYFP than Mito-ChR2-eYFP. Taken together, ABCB is a better leading sequence to localize ChR2 to mitochondria.

92-Rous Sarcoma Virus (RSV) Interacts Electrostatically with Plasma Membrane Phosphatidylinositols

Gunnar Eastep

Retroviral assembly is dependent on proper localization of viral components to the plasma membrane (PM). The PM localization of the retroviral Gag structural protein is mediated by Gag N-terminal domain, matrix protein (MA), interactions with the PM. Successful HIV-1 assembly depends on the N-terminal myristoylation of MA and its interaction with PI(4,5)P2, present as a signaling phospholipid in the inner leaflet of the PM. Depletion of PI(4,5)P2 leads to poor localization of HIV-1 Gag to the PM and impairs HIV-1 replication. Rous sarcoma virus (RSV) lacks the myristoyl group in MA and its PM binding is thought to be purely electrostatic. There are, however, conflicting reports whether RSV Gag localization and binding to the PM depends on PI(4,5)P2. We sought to study the binding of RSV MA to PI(4,5)P2 and other phosphoinositides (PIs) to help clarify the discrepancy and determine the structural basis for the putative interaction. To characterize these interactions, we use NMR spectroscopy and other biophysical methods. We found that RSV MA binds PIs and we demonstrate a direct relationship between the charge of the PIs and the strength of the binding, supporting an electrostatic model of RSV MA binding to PM. Furthermore, we examine the binding of several mutants of RSV MA to various PIs to determine the importance of several lysine residues that compose a basic membrane-binding patch on the surface of the RSV MA. Finally, by utilizing nanodiscs we study how RSV MA interaction with a lipid bilayer depends on its overall lipid composition.

96-Effect of Tumor Microenvironments on Glioblastoma Growth

Adetokunbo Ayokanmbi, Nathaniel H. Boyd and Anita Hjelmeland

Glioblastoma (GBM) is a primary malignant and deadly brain tumor due to genetic and epigenetic differences between tumor cells. Chromodomain-helicase-DNA-binding protein 7 (CHD7) was identified as an epigenetic factor consistently repressed by acidic stress that binds to SMAD transcription factors which are downstream mediators for Transforming Growth Factor (TGF) signaling, regulating self-renewal, cell proliferation, differentiation, and migration. These new findings suggest that CHD7 may be able to control TGF to regulate GBM growth and differentiation. The BTICs and 293T cells infected with the shRNAs in the pGIPZ vector were imaged before and after puromycin selection. Brain tumor initiating cells (BTICs) and human kidney cells transfected with different shRNAs in the pGIPZ lentiviral vector were subjected to real-time quantitative PCR and western blot. The results demonstrate the expression of CHD7 decreased in the shRNAs chosen for potential testing. These results suggest that shRNA containing the pGIPZ vector targeted the CHD7 and decreased the mRNA and protein expression of CHD7.

100-The Role of miR-214, miR-219, & miR-876 in Bronchopulmonary Dysplasia

Melanie Veizaga Zurita, Nelida Olave, Brian Halloran, Gabriel Rezonzew, Charitharth Vivek Lal, Namasivayam Ambalavanan

Introduction: Bronchopulmonary dysplasia (BPD) is the chronic lung disease of prematurity characterized by inadequate alveolar septation and lung inflammation. MicroRNAs (miRs) are small non-coding RNAs that down regulate the gene expression of target mRNAs. The role of epigenetic regulation of alveolar septation by miRs is not well studied. We hypothesized that the expression of miR-214, miR-219, miR-876 and their corresponding predicted target genes (Cox-2, PDGFRα, MCL-1, RBBP6) would be dysregulated with hyperoxia exposure, in vitro.

Methods: Human bronchial epithelial cells were grown via cell cultures and exposure to normoxia (21% O2) and hyperoxia (85% O2) conditions for both 12 and 24 hours. After total RNA was isolated, miRNA and mRNA analysis was performed via reverse transcription and qPCR.

Results: miR-214 and miR-219 both increased at 24 hrs under hyperoxia (fold change= 1.7 and 1.3 respectively) while miR-876 decreased in expression (0.6 fold change). All the mRNAs (Cox-2, PDGFRα, MCL-1, RBBP6) showed an increase in expression at 24 hr hyperoxia with Cox-2 having the most drastic increase. For RBBP6, there was great variability in data at 12 hr hyperoxia with no clear outlier.

Discussion: An inverse miR-mRNA relationship has been observed between miR-214 and Cox-2 as well as miR-219 and PDGFRα in previous studies. Hence there is a possibility that our result are due to a random error, and require further testing. Alternatively, these miRNA-mRNA regulation may be specific to non-epithelial cells, which we haven't tested. The down regulation of miR 876 with a corresponding up regulation of MCL-1 with hyperoxia exposure, may explain the lung inflammation seen in BPD. RBBP6 expression under hyperoxia requires further testing due to the high variability of data seen.

101-Central projections of intrinsically photosensitive retinal ganglion cells in the Macaque monkey

Treffalls, J. A., Chang, K. Q., & Gamlin, P. D.

The hypothalamic suprachiasmatic nucleus (SCN) is the master clock responsible for controlling circadian rhythms in mammals. These rhythmic cycles generated by the SCN are entrained to the environmental light/dark cycle via signals from intrinsically photosensitive retinal ganglion cells (ipRGCs) expressing the photopigment melanopsin. Orexin is a neuropeptide that regulates arousal, wakefulness, and appetite. Orexingeric cells in the posterior hypothalamus receive input from the circadian system. It has been reported that orexin is present in melanopsin-expressing ipRGCs. However, most knowledge of these cells has been gained from studies on nocturnal rodents, whose visual systems are distantly related to those of humans and other diurnal primates. Injection of cholera toxin B (CtB), an anterograde axonal tracer, was used to visualize retinal projections in the brain, and anti-orexin antibodies were used to determine if these projections also contained orexin. One male macaque monkey, aged 20 years, received bilateral intra-vitreal injections of 100 µl of 0.5% CtB conjugated with either Alexa Fluor 488 (right eye) or biotin (left eye). After a survival time of 6 days, the animal was deeply anesthetized, perfused, and the brain and eyes were both removed. Immunohistochemical staining of revealed penetration of CtB into the retinal projections of the brain, as well as the presence of orexin in the retina. This is the first study to observe orexin in the retina of nonhuman primates, supporting the hypothesis that orexinergic neurons in the SCN may be retinal projections of ipRGCs.

102-Alterations in dendritic spine density and morphology associated with tau pathology along Alzheimer's progression

Benjamin Boros, Erik Gentry, Betsy Birchall, Jeremy Herskowitz

Cognitive decline is a clinical hallmark of progression from healthy brain aging to Alzheimer's disease (AD) while accumulation of amyloid-β (Aβ) and tau-derived neurofibrillary tangles are pathological hallmarks of AD. There is strong evidence that cognitive impairment in AD is due to Aβ and tau's negative impact on synaptic plasticity. Development of AD pathology begins years prior to clinical symptom onset, and during this time, we hypothesize that accumulation of Aβ and tau negatively impacts synaptic plasticity. Dendritic spines are the post-synapse of excitatory neuron connections, and the number as well as the shape of spines highly influences cognition. Past work revealed that dendritic spine density is reduced in AD patients compared to age-matched controls; however these studies neglected alterations in spine shape as well as changes across disease progression. To address these gaps, I investigated spines change from early to late stages of AD across two brains regions. Human tissues were impregnated by Golgi method and imaged in 3D using brightfield microscopy. To analyze spines, I developed an innovative, novel method of digital image reconstruction using Neurolucida360. My efforts revealed that loss of spine density and increased spine neck extent/head diameter directly correlates with neurofibrillary tau pathology in AD. My results strengthen the hypothesis that detrimental changes in spine density and morphology are progressive in AD and that synaptic changes correlate to AD pathology. These findings are likely to fuel new experimental models of AD to examine how neurofibrillary tau pathology is linked to dendritic spine abnormalities in AD.

103-Development of a Mass Spectrometry Assay for measurement of the marine biotoxin domoic acid.

Mohamad Noor Alsirafi, Catherine M Fuller

Domoic acid (DA) is a naturally occurring neurotoxin produced by marine algae belonging to the genus Pseudo-nitzschia, and underlies the neurological disorder known as amnesic shellfish poisoning. Previous studies have shown that exposure to low levels of DA can also result in renal injury. This could have a significant impact on human health through ingestion of higher marine organisms. Oil supplements such as fish and krill oils might contain detectable levels of DA due to the fact that the supplements are derived from marine organisms. This study examined five brands of oil dietary supplements that are commercially available (GNC Fish Oil, Rite Aid Triple Strength Krill Oil, Nature's Bounty Co-Q-10 with krill oil, Sundown Naturals Triple Strength Red Krill Oil, and Natrol Omega-3 Krill Oil). LC-MS was used to determine the amount of DA in these samples. Extraction methods were developed using hexane and water to maximize the percentage of recovered DA in the samples. Our studies found no detectable amounts of DA in the oil supplements, (limit of detection was 0.1 ng/mL). Other studies were performed in mice. Recently we have found that female mice are more susceptible to DA renal injury compared to male mice. We examined 24h urine samples from six mice (3 males, and 3 females) treated with DA at two doses (0.05 mg/kg, 0.005 mg/kg per 24h via implanted osmotic minipumps, and saline as control). After 7 days of exposure to DA the results indicate that female mice excrete lower amounts of DA over 24h compared to male mice. These result suggest that the greater toxicity to DA in female mice maybe related to a reduced ability to excrete DA and therefore a greater effective renal concentration of this renal toxin.

105- Immunohistochemical analysis of neural activity-induced Tet localization

Richard Trieu

Changes to the expression of memory-related genes are a necessary process for long-term consolidation of information in the brain. These changes are regulated by epigenetic modifications that involve DNA methylation, a process whereby methyl groups are added to DNA. It is the presence or absence of these methyl-groups within and around memory-related genes which determine the extent they are transcribed. Studies have shown that DNA methylation patterns in the nervous system are dynamic. DNA demethylation is mediated by a family of methylcytosine dioxygenase enzymes named TET1, TET2, and TET3. However, how these enzymes regulate demethylation, particularly in the nervous system, is not well understood. In a recent study using non-neuronal cells, TET subcellular localization patterns were shown to be altered under certain cellular conditions, suggesting that changes in TET localization might act as a mechanism to control their DNA demethylation activity (Arioka, 2012). We hypothesize that excitatory stimulation of neurons might alter TET localization thus DNA methylation patterns. We have successfully cloned mouse TET1, 2 and 3 and shown that in neuronal cells, TET2 is localized primarily to the nucleus. To test our hypothesis, we are cloning mouse TET1, 2 and 3 and expressing them in primary hippocampal neuronal cultures. This will be followed by artificial stimulation using KCl, immunostaining for the TET enzymes and quantitative analysis of their subcellular localization. We hope to elucidate the location of TET 1 and 3 and their location upon stimulation in primary hippocampal neuronal cultures.

8-Conducting Teacher Research to Determine the Merit of Engaging Instructional Practices

Victoria Stanley, Kaitlyn McAvoy, Mariah Wilkins

Teacher research is a valuable process within the classroom, and it can be utilized across many subjects and areas. Teacher research was conducted by a cohort of three pre-service teachers to determine the merits and benefits of various instructional practices while serving as teacher assistants. During regular curriculum, lessons, and ongoing assessments in a summer enrichment program, these teacher researchers gathered data through ongoing observations, conferencing, and questioning. Students were observed during center time specifically in the dramatic play area to assess their use of mathematics within this center and it was concluded that the students discussed both numerical and geometric mathematics, and utilized them to problem solve while playing. Another pre-service teacher incorporated conferencing to encourage student self-reflection throughout the writing process. This enabled the students to reflect upon their own work to see the positive aspects of their writing, as well as the areas which needed improvement. The ability for students to make connections was also researched by a pre-service teacher and concluded a read aloud does increase the probability that students will make connections with a text. The pre-service teacher incorporated questioning into the read aloud, which created opportunities for the students to make personal connections. The key findings from the teacher research within different content areas indicated that these instructional practices are advantageous and valuable within the classroom, and can be utilized to create engaging student learning opportunities.

83-Emergent Bilinugal Pre-Kindergarteners' Oral Language Development

Katie Watkins

The purpose of this research is improved instructional practices in helping emergent bilingual students increase their oral language skills as they discuss with a teacher and classmates in a structured group format. Providing opportunities for students to learn language in a safe environment is critical to their future success. This research also proposes the utilization of a child's home language as a valuable asset to developing proficiency in English. Incoming kindergartens were observed for this qualitative study in an urban school setting during a 5-week enrichment program.

The methods that were used focused on wordless picture books and group discussions in the students' native language, Spanish, and English. Anecdotal notes in observational journals were kept on each student with notations regarding student-teacher conversations, growth in oral language, and student-student interactions. From these methods, findings were made that informed instruction and showed oral language growth in the students.

Through this process, patterns emerged within the students' oral language. By the end of the program, the daily group discussions, repeated shared viewings, and bilingual conversations about wordless picture books led to increased opportunities for talking about what was being read resulting in growth of all students' oral language development.

The above findings support current research on wordless picture books and bilingual children. These books are a valuable resource for generating discussions and the use of bilingual conversations support linguistic transfer of knowledge from one language to another to bolster understanding of subject matter.

104-Collaboration in Pre-Service Teacher Research to Guide Best Practices

J. Woods, A. Kizer, K. Scott, K. Watkins

The School of Education teacher candidates planned and implemented qualitative studies to answer questions of teaching methods. This research is significant to the pre-service teachers' continued growth of their pedagogical knowledge as preparation for their future classrooms. The pre-service teacher researchers' goal is to bridge the gap between structured university instruction and the realities and challenges of the real-world classroom.

This research was accomplished through collaboration between UAB, an urban school in the Birmingham area, and pre-service teacher candidates. Data was collected in classrooms of rising Kindergarten through 2nd grade students. Each pre-service teacher worked with 8-10 students for 5 weeks collecting data through self-designed curriculum. The teacher candidates will use the conclusions to further shape their teaching philosophies.

Each teacher candidate identified research questions that they focused on during the 2016 summer program. Inquiries range from autonomy and how it relates to student academic achievement, wordless picture books and how it relates to the support of Emergent Bilingual students, student-generated rules and how it promotes autonomy, and the effects of making personal connections on reading comprehension.

Results and conclusions benefited each researcher and will be listed individually. The teacher candidates plan to further their research when given the opportunity and also use qualitative study as a basis for new research and best practice.

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ENGINEERING

11-Experimental Validation of Fiber Orientation Modeling for Different Charge Placement Techniques

Victor Torres, Benjamin Willis, Mohamed Selim

Composites are used in today's world everywhere with examples being apparent in weapons, vehicles, and prosthetic limbs. The manufacturing of stronger and more durable composite products has been sought for by many companies yet is a challenging and time consuming process. The most common composites utilized are fiber reinforced. Depending on fiber orientation, the composite can be stronger in one location and weaker in another. This research focuses on determining the fiber orientation in composites that undergo compression molding. The focus involves computer simulation and mechanical testing of fiber orientation in Polypropylene Glass Fiber material that was compressed into a plate. The goal of this project is to study the effect of various charge placement techniques within the large press on the orientation of the fibers within the molded plate. The purpose is to eliminate threats of warpage, increase tensile strength, and decrease cost. The compression of this material is then simulated in Moldex3D, a commercial simulation software, which predicts fiber orientation. The data from the original experiment is recorded and inputted into the simulation software with all parameters mimicking what actually occurred. The mechanical test involves the plates, which are cut into 120 samples, undergoing a Flexure test (ASTM-D79003) to determine the flexural modulus. This data will verify whether what has been simulated in Moldex3D is actually what has occurred in this experiment. With this verification, charge placement can be manipulated in further experimentation to control fiber orientation in various products through the utilization of Moldex3D fiber orientation software.

17-Alternative replacement for Birmingham Alabama concrete storm drains

Julian A. Dover1,Benjamin Geiger-Willis2, and Selvum Pillay

The goal of the research is to use 100% recycled materials to make into a fiber reinforced polymer in the hopes to one-day replace the concrete storm drains in the Birmingham Alabama area, using material that is easy to attain and cheap. The composite Materials that was selected was High Density Polyethylene Recyclate and Kohler Waste made up of Glass fibers and polyester. Making four Test samples to see which perform the best under stress using Regrind, 10%, 20% and 30% of Kohler Waste mixed with the (HDPE) Regrind into a 10lb mix. The plan is to tests the strength of the composites to see if they can withstand bending and physical Impacts. Using the American Society for testing material (ASTM) D256-10 IZOD test, which is a pendulum raised to a fixed height used to determining the impact resistance of materials and (ASTM) D790-15 Flexural test which provides the Modulus of elasticity stress-strain of the material. The idea is to put each of the samples under the IZOD and flexure test to see which Composite has the overall best performance. The (HDPE) regrind alone is a good alternative and performed the best to replace storm drains but the goal was to add Regrind with another material that also can function well in an environment with frequent exposure to water so, Kohler Waste was added to the mix but since it has a high melting point some of the composites samples did not mix well with the (HDPE) Regrind.

19-Deposition Rate Calibration and Thickness Profile of Thin Films by Pulsed Laser Deposition

Kaleb Burrage, Sumner Harris, Eric Remington, Renato Camata

Thin films have played a paramount role in shaping modern science and technology. Breakthroughs in the study of the physics of materials and in semiconductor devices such as transistors, light emitting diodes, charge-coupled device cameras, and touch computer screens have all benefitted from widespread applications of thin films. Ensuring the successful production of quality films is just as paramount. Controlling the thickness and the thickness variation of the film is essential in virtually all applications. Thin films can crack or rupture due to a variety of reasons including defects, stress, and lattice mismatch with the substrate. Cracks can drastically reduce the performance of a film, or even render it useless. In this study, zinc selenide (ZnSe) thin films were deposited onto silicon substrates by Pulsed Laser Deposition (PLD) with varying laser fluence of the focused beam of a KrF excimer laser (248 nm). Other PLD parameters were kept constant (deposition pressure: 10^-5 Torr, laser angle of incidence: , laser spot size: 0.9 cm^2). The thickness and thickness variation of the films were measured by interference effects using UV-Vis-NIR spectroscopy. The study shows that a fluence of 3.2 J/cm^2 produced films with an essentially flat thickness profile (less than 10% variation) within a radius of 3 mm from the center of the ablation plume. Higher fluences of 5.9 J/cm^2 and 7.2 J/cm^2 produced significantly more peaked profiles with thickness variation of approximately 15%. These results suggest that there is an optimal fluence for deposition of ZnSe thin films of uniform thickness.

22-Analysis of Distributed Generation with Renewable Energy Conversion in ASHRAE Climate Zone 3A

Christy Green

A distributed generation (DG) system produces power at the point of use. DG systems can increase energy system reliability, reduce peak power requirements, and improve energy infrastructure resilience. This is done by integrating multi-generation with renewable energy conversion devices. Overall utilization factors of more than 95% have been reported in technical literature. In this study, commercial energy modeling software was used to model a DG system using renewable energy sources and energy storage for a typical residential building in ASHRAE climate zone 3A. 3D graphical modeling software was used to generate the model for the building based on Building America House Simulation Protocols. The 3D geometry was then imported to TRNSYS software, which was used to model the building's energy consumption and the dynamic behavior of the building energy equipment. Standard weather data from four cities across climate zone 3A was used. The energy sources considered in this analysis included a photovoltaic array and wind turbine. The results of these simulations were used to determine the most efficient and reliable DG system for four locations across climate zone 3A. This report shows that geographic location has a significant impact on photovoltaic and wind energy conversion, as well as the energy profile of residential buildings. This computer simulation may be used to assess the performance of renewable energy technologies and energy storage systems for any given climate, making the implementation of renewable energy technology more practical for homeowners.

74-A Biomimetic Cardiac Tissue Model (BCTM) to determine effects of cellular orientation on structural remodeling.

James Ansell, Palaniappan Sethu

Myocardial infarctions present one of the leading causes of death in America. The adverse effects of infarcted/damaged cardiac tissue results in the decline of patients' health due, in part, to the hypertrophic responses of surrounding tissues as the hearts tries accommodating to a decrease of cardiac output. Cardiac patches, consisting of cardiac tissue grown in vitro have been suggested as a novel treatment modality for infarcted patients. It has been shown that cardiomyocytes tend to orient themselves along primary axis of strain. However, if cardiac patch technology is employed, these cells may end up oriented in a matrix that forces these cells to be aligned against these primary strain profiles. This study evaluated the effects of orientation of H9C2 rat cardiomyoblastic cells using a novel biomimetic cardiac tissue model (BCTM). Cells cultured within the BCTM in an elliptical cell culture chamber experience stretch predominantly along the short axis of the ellipse. By patterning cells either in line with the primary direction of stretch or perpendicular to the primary direction of stretch, we sought to determine the effects of stretch on structural remodeling. Overall, we measured cell thickness, cell length and ratio of cell thickness to cell length to quantify the type of hypertrophy. Our results suggest that cells aligned parallel with the primary direction of stretch have thicker morphologies than cells aligned perpendicular to the direction of primary stretch.

76-Organ On A Chip: Modeling the Alveolar-Capillary Interface

David T. Lee, Thomas A. Haglund, Palaniappan Sethu

The use of cell culture and animal models in biomedical research is prevalent, especially in the development of pharmaceuticals. However, these models present inherent limitations: static cell culture models may not replicate the environment observed in vivo, and animal models are generally expensive and poorly screen ineffective treatments prior to clinical trials. Recent studies have replicated various systems in the body, such as the alveolar-capillary interface where gas exchange occurs. Previous works have developed small scale devices that mimic this functional region, separating lung epithelial cells from microvascular endothelial cells using a porous membrane of polydimethylsiloxane (PDMS). To improve upon previous designs, we constructed a device with a layer of fibrin gel, rather than porous PDMS, upon which a monolayer of cells were grown. Fibrin, a naturally occurring protein involved in blood clotting, has been utilized extensively as a cell scaffold because it more accurately mimics the extracellular matrix. Once cells have been cultured onto the fibrin membrane, the device will be placed in a closed loop with a steady flow of media with controlled amounts of pressure and shear stresses on the endothelial side and air on the epithelial side, mimicking the gas exchange architecture seen in vivo. This device will be further characterized by permeability assays using fluorescently labelled dextran to ensure that permeability of this device matches what is observed in the body.

85-Assessing the Toxicity of Carbon Nanomaterials in the Body

Matt Medders, Daneesh Simien

Carbon nanomaterials are emerging as an unrivaled class of materials providing new and intriguing potential applications for the industrial and medical fields due to their mechanical, electrical, and optical properties. Single-walled and multi-walled carbon nanotubes, fullerenes, and graphene all possess these unique characteristics that bring them to the forefront of the research industry; however, current research on the toxicity of these materials does not lead to a definitive conclusion about the adverse effects of exposure to society. This review seeks to compile data on the above classes of nanomaterials based on size, length, species, functionalization, and delivery route to determine the effect these properties have on toxicity in the body. Research on mice was collected from multiple sources to assess the degree of toxicity in six preliminary areas: nervous and digestive systems, along with brain, muscle, lung, and liver tissues. Our literature findings to date indicate that functionalization is a key characteristic to inhibiting toxicity, and that irrespective of delivery route – whether instillation, inhalation, or injection, of un-functionalized nanomaterials – resulting toxicity was found in the area under study in mice. Furthermore, many sources suggested additional research is needed to identify how size, length, and species all contribute to toxicity in the body due to conflicting results based on these parameters. We seek to however isolate characteristics that delineate the specificity of each type of nanomaterial used in these studies to characterize commonalities to understand how to safer handle and limit risk of exposure in the use of these valuable materials.

91-Size, Morphology, and Loading Efficiency of Microbubbles Fabricated using Double Emulsion and Coaxial Electrospray

Makena Dettmann, Karim I. Budhwani, Vinoy Thomas

More than 8 million lives are claimed each year by cancer alone and experts estimate that over the next 5 years, the lifetime probability of getting cancer will raise to 1 in 3 for females and 1 in 2 for males. Chemotherapy, the predominant cancer treatment modality ahead of surgery, radiation, and others suffers from elimination in renal and hepatic systems causing reduced bioavailability and increased toxicity causing harmful side effects. Recently, drug-encapsulating ultrasound-activated microbubbles have captured researchers' interest as a method for targeted drug delivery capable of improving efficacy of the drug while simultaneously reducing toxicity to benign tissue. Narrow size distribution, spherical morphology, and a high loading efficiency are all important parts of a good delivery system. Double emulsion, the most common method for microbubble fabrication suffers from low loading efficiencies and a wide size distribution. Coaxial electrospray, a more emergent technique, was developed to address these issues. Here we present an analysis of size, morphology, and loading efficiencies in MBs fabricated using double emulsion and coaxial electrospray techniques. Microbubbles from same set of raw materials were fabricated using both techniques in parallel for the analysis and characterized using scanning electron microscopy (SEM), fluorescent microscopy, and image analysis software. Microbubbles produced via coaxial electrospray displayed higher loading efficiency and a narrower size distribution while double emulsion microbubbles showed better morphology. These results indicate that the coaxial electrospray technique shows great promise as a fabrication method.

3-In Vitro Wear of 3 Glass-ionomer Based Dental Restorative Materials

SJ. Kwon, N. Lawson, P. Beck, JO. Burgess

The aim of this study is to measure and compare the in vitro wear of 3 glass-ionomer restorative materials using the Alabama wear tester against natural enamel for 100,000 cycles (9 months). Methods: Eight flat rectangular specimens were prepared for each material, using a flexible elastomeric mold. The material was placed in one increment (excluding the composite resin which was placed in 2mm increments). A clear matrix was placed over the specimens and a glass slide was placed on top of the specimens. For the Equia Forte Fil and Equia Fil with coatings groups, a layer of the coating was applied. The resin composite and the coatings were light cured using an Elipar S10 (3M ESPE) for 20 seconds. After fabrication the specimens were stored in distilled water at 37ºC in an incubator for 24hours. The specimens were mounted in brass holders using self-curing acrylic material. The specimens were not polished to preserve the coating and more closely replicate the clinical situation.

Conclusion: All glass ionomer materials demonstrated significantly more wear than the reference resin composite

The use of a coating on the glass ionomer materials did not improve its wear resistance

Equia Forte Fil and Equia Fil (with and without a coating) demonstrated better wear resistance than Chemfil Rock.

5-Assessing Undergraduate Students' Knowledge with an Interest in Health Professions of Medical Genetics and Genetic Counseling

Brooke Gresham, R. Lynn Holt

Genetics in medicine and genetic counseling as a profession are growing quickly with the new era of personalized medicine and the completion of the Human Genome Project in 2000. President Obama's Precision Medicine Initiative displays how the scope of genetics is expanding in medicine. As a result, there will be an increased demand for genetic health professionals such as genetic counselors. Undergraduates with an interest in healthcare may not have an accurate understanding of this field of study. The purpose of this project was to assess the knowledge and awareness of undergraduate students at UAB about the fields of medical genetics and genetic counseling. Gathering this data was completed through an IRB approved survey administered to students within the School of Health Professions and the School of Public Health at UAB. The results of this study revealed that the majority of students have little familiarity with genetic counseling and have not completed UAB genetics based courses. This implies that there is a potential gap in the current undergraduate education about genetics and genetic counseling. This information may be useful in improving the education and awareness of undergraduate students with an interest in health professions. Due to the lack of research regarding undergraduate students and their knowledge of genetics, it is difficult to compare the results to other studies. I hope the findings of this survey will serve as a catalyst for further research with a larger sample population in the future.

10-Perceived Stress, Sleep Disturbances, Fatigue and Blood Pressure in Children with Sickle Cell Disease

Barley, C., Bryant, A., & Soistmann, H. SJ. Kwon, N. Lawson, P. Beck, JO. Burgess

Background: Sickle Cell Disease (SCD) is a chronic autosomal recessive disorder in 1 in 400 African Americans, and causes red blood cells to take on an abnormal sickled shape. This abnormal shape often times causes red blood cells to clump together decreasing blood flow and oxygenation. The purpose of this study is to examine the relationships between perceived stress and sleep disturbances, and their influence on blood pressure and fatigue in patients diagnosed with SCD. This study is important because although there is some literature on these factors in adults, there is limited research in children with SCD.

Methods/procedure/approach: Anthropometric measurements were taken of each child and included: height, weight, waist circumference, and blood pressure. Two salivary samples were also obtained from each child, one in the morning and one in the afternoon. The parents and the children completed questionnaires related to the study variables, and a sleep diary.

Results/conclusions: It is hypothesized that perceived stress and sleep disturbances are positively correlated with increased fatigue and blood pressure in children with sickle cell disease.

Implications: Stress, fatigue, sleep disturbances, and blood pressure have an inverse relationship with health. As stress, fatigue, and blood pressure increase, overall health decreases. In order to improve the health and quality of life in children with SCD, interventions to address blood pressure, stress, fatigue, and sleep disturbances must be implemented. Knowledge of factors that influence blood pressure will inform these interventions.

21-Onsite Recruiters Increase Participation in Adjunct Cancer Therapy Trial

Catherine Black, Ashley Evatt, Heather Lipscomb, Jodee Robinson, Barbara Gower, PhD

Background: Limited information exists related to the recruitment of a vulnerable female population in the clinical setting. The purpose of this study is to explore the effect of face-to-face recruitment of an adjunct ketogenic diet therapy trial in women diagnosed with a gynecological cancer.

Methods: Eligible participants were approached while attending a regular scheduled clinic visit at a large cancer center in the Southeast. Providers obtained preliminary approval for initial recruitment discussion by onsite research staff. Individuals who expressed interest in obtaining additional information were provided study-related information in written and oral form. Those who were interested in participation were referred to project coordinator for possible study enrollment. Non-identified demographic and clinical information was collected from the medical records. Analysis was conducted via descriptive statistics, frequency distributions, Kruskal-Wallis, and t-tests, as appropriate.

Results: No demographic or clinical related differences were noted between those who enrolled in the study and those who declined participation. On-site staff presence increased recruitment by 310% (p=0.001) and increased total study enrollment by 62%. Prior to involvement of onsite clinical recruiters, the ketogenic diet study had enrolled 13 participants over an 8 month period. In 6 weeks, the on-site recruiters increased the recruitment numbers by 10.

Implications: The physical presence of research staff significantly increases recruitment in a clinical trial in a vulnerable, middle-aged, female population.

25-Perceived Stress, Sleep Disturbances, Fatigue and Blood Pressure in Children with Sickle Cell Disease

Michelle Sonnenberger, Carmen De Miguel, and Jennifer S. Pollock

Akita mice develop diabetes due to a mutation in the Ins2 gene that causes incorrect folding of insulin. Recently, an Akita mouse on the FVB background (FVB/NJ-Ins2+/C96Y) was created. We aimed to determine if these mice are sensitive to the development of diabetic nephropathy (DN), since Akita on other genetic backgrounds are resistant to DN. Urinary markers of kidney damage were measured in Akita and FVB control mice (n= 4-12/group). Akita mice showed increased excretion of renal damage markers: protein (FVB vs. Akita: 0.77 ± 0.14 vs. 21.81 ± 3.55 mg/day; p=0.002), albumin (0.05 ± 0.02 vs. 6.26 ± 1.66 mg/day; p=0.04), KIM-1 (0.55 ± 0.19 vs. 8.57 ± 2.75 ng/day; p=0.02), NGAL (26.17 ± 4.25 vs. 52.39 ± 0.62 ng/day; p=0.01), and nephrin (1.72 ± 0.41 vs. 43.92 ± 9.23 ng/day; p=0.002), indicating that FVB/NJ-Ins2+/C96Y mice present diabetic glomerular and tubular damage. Inflammation is a hallmark of DN, thus we assessed renal inflammatory markers to begin to understand the mechanism(s) of renal damage. Renal infiltration of macrophages and T lymphocytes was not different between genotypes. Akita mice demonstrated decreased expression of markers involved in inflammasome pathway compared to FVB in the outer medulla: Cd40lg (4 fold decrease; p=0.04, n=2-3/group), Chuk and Pea15a (both 2 fold decrease; p=0.02), Panx1 (2 fold decrease; p=0.05), and Mok (4 fold decrease; p=0.02). However, in cortex only a 3 fold decrease of inflammasome Nlrp3 was detected in Akita mice (p=0.04). In conclusion, FVB/NJ-Ins2+/C96Y mice present many DN characteristics.

26-Defining the Role of BAF45a in Chromatin Remodeling of Osteoblasts

Joshua Lee, Nylah Caves, Tanner Godfrey, Mohammad Hassan

Bone mass loss disorders, such as osteoporosis, affects over 53 million people in the United States (NIH Osteoporosis Overview). This issues a calling to develop novel and effective therapeutic approaches to treat this growing issue. Chromatin remodeling by the BAF complex plays a central role in the regulation of the differentiation of many cell types, including those that are involved in skeletogenesis. We have identified BAF45a, a component of the BAF complex, to play a major role in osteoblast differentiation. The purpose of this study is to define the role of BAF45a in osteoblast differentiation by studying osteoblasts' gene expression profile during differentiation. Two main transcription factors that are involved in osteoblast differentiation that will be used in this study are RUNX2 and SP7 (Osterix). Real time PCR and Western Blotting will be utilized in order to monitor these factors. By observing the levels of these factors and comparing them with the levels of BAF45a, some major conclusions could be drawn. The following marker genes will be used in order to track specific stages of cellular differentiation: COL1 (Collagen Type 1) as an early marker, ALP (Alkaline Phosphatase) as a marker of mature osteoblasts, and BGLAP (Osteocalcin) as a late marker of mature osteoblasts. A glycolysis enzyme, GAPDH, will be used as an internal control. The results showed that BAF45a exhibited a similar expression pattern to RUNX2. Since RUNX2 is the master transcription factor, this gives strong evidence that the BAF45a is crucial in the process of osteoblast differentiation.

52-Assessing Pannexin 1 Channel Function in Mouse Cardiomyocytes

Andrei M. Tuluca

Pannexin 1 (Panx1) is a ubiquitously expressed large pore conductance membrane channel that allows for the passage of small molecules like ATP. A role for Panx1 has been described during cell apoptosis, seizures, tumorigenesis, melanomas, migraines and HIV-1. In addition, our lab has preliminary data suggesting that Panx1 facilitates Ca2+ dependent focal ectopic arrhythmias. In order to assess Panx1 channel function in the heart it was necessary to isolate single mouse cardiomyocytes suitable for different assays. We chose to measure Panx1 channel function via patch clamp technique, which requires stable membranes, and via ATP release from cell batches, which requires high cell yield. Cardiac tissues were separated and digested into individual cardiomyocytes with the use of a perfusion system that delivered 35-degree bubble free solution via a roller pump. Multiple collagenase "Cocktails" supplemented with protease to optimize for (1) high yield and (2) membrane stability were tested. We also explored if Panx1 channel internalization would hamper functional measurements by measuring the expression of the receptor responsible for internalization, P2X7, using qPCR. We identified cardiomyocyte isolation conditions that were suitable for patch clamping, receiving a 6 out of 10 score for patchability. Moreover, we identified conditions that produced cardiomyocyte yields > 30%. Taken together, even though time constraints prevented us to attempt Panx1 function measurements, cardiomyocyte isolation procedures are now in place for each assay. Our data also suggests that P2X7 is likely present on cardiomyocytes and the possibility of internalization during the isolation procedure requires future considerations.

59-Hospital Utilization and Patient Centered Medical Home's

Rosa Lee Aderhold

The Patient Centered Medical Home (PCMH) is an innovative way of delivering healthcare to ensure that patients receive care when needed and the care is delivered in a form that the patients understand. This study addresses the effects PCMH's have on hospital utilization. According to a study done by James Adams, "The United States healthcare system overuses emergency department services, resulting in 38 billion dollars wasted annually in unnecessary emergency department visits." The PCMH aims to address the patient's needs while lowering the countries annual healthcare cost. This study focuses on individuals suffering from depression and who are from a lower socioeconomic class. This study found that those living in a suburban area had a p-value of 1.28 compared to an urban area of .54. This indicates the need among the urban homeless and housed poor for the need of a PCMH. This study also found an extremely high comorbidity rate with a p-value of 2.29. This supports the theory that those utilizing the PCMH have more than one illness present. The need for a primary care physician increases significantly when comorbidities are present. By implementing PCMH's comorbidity rates could decrease substantially because individuals would finally seek the appropriate type of healthcare services rather than seeking help from an emergency room. A major advantage the PCMH offers compared to an emergency department is a lasting relationship with a physician. By forming this relationship individuals better understand their diagnosis and are more informed of their medications.

66-The Impact of Community Orientation and Ambulatory Care Sensitive Hospitalization Rates

Shelby Wallace, Larry Hearld, PhD, and Jessica H. Williams, PhD, MPH

Purpose: Community orientation leads to better preventative care, and ambulatory care sensitive hospitalizations ACSHs are indicators of poor access to timely and efficient ambulatory and preventive care in populations.

Introduction Community orientation leads to better preventative care, and ambulatory care sensitive hospitalizations ACSHs are indicators of poor access to timely and efficient ambulatory and preventive care in populations.

Methods: Descriptive statistics and linear regression analysis were used to identify the relationship between community orientation and ACSH rates in white and black patients in a nationwide sample. A sample of data was selected across six different states from the 2012 Healthcare Cost and Utilization Projects State (HCUPS) Inpatient Database, the American Hospital Association (AHA) Annual Survey, the Centers for Medicare and Medicaid Services (CMS) Case Mix File, and the Area Resource File (ARF).

Results: The level of community orientation was negatively associated with the white ACSH rate for both white and black patients ($b = -0.001$, $p < 0.000$).

Conclusion: Results confirmed that increased community orientation leads to lower levels of ACSH rates. However, other factors such as the type of ownership and the unadjusted Case Mix Index that had a larger impact on ACSH rates for white and black patients. Further research is needed to understand other health system factors that might impact ACHS rates, particularly across different races.

72-Cost and Clinical Implications of Assaying CK-MB and Troponins Together: A Retrospective Look

Krista Doss

Despite current clinical practice guidelines identifying cardiac troponin I (cTnI) as the prevailing diagnostic tool for detecting myocardial necrosis, many clinicians continue to add creatine kinase (CK-MB) to their troponin order. While some find this to be an advantage of detecting cardiac damage, others claim this leads to over diagnosis, over treatment, and inappropriate test utilization that can all lead to an increase in medical costs. A retrospective review of physician practice patterns was performed to examine cTnI and CK-MB test ordering at the University of Alabama at Birmingham Hospital from January to May of 2015 and 2016. The study goal was to determine how many times CK-MB and troponins were assayed alone and in parallel, identify hospital units not in compliance with current clinical practice guidelines, and calculate the cost of the inappropriate parallel test utilization. From 2015 to 2016 CK-MB orders have decreased 35%, cTn have increased 102%, and parallel orders have decreased 36%. The majority of orders came from the emergency department, neonatal intensive care unit, and the cardiac care unit. The hospital spent $11,315.00 in 2015 and $7,186.00 in 2016 performing the additional redundant tests. Therefore, while the parallel ordering of CK-MB and cTnI is trending down, a significant number of physicians are still ordering these tests together. Interestingly, the inappropriate test utilizations are largely clustered within five hospital units. A large portion of the extra test ordering are not clinically indicated and are resulting in unnecessary costs.

90-Barriers and facilitators of antihypertensive medication adherence in older African Americans with HIV

Ashouri, S. A., Etherton, L. G., Hicks, L. J., & Gakumo, C. A.

Background: Previous research has extensively addressed antiretroviral therapy adherence in people living with HIV. However, less is known about adherence to antihypertensive medications. Nonadherence to antihypertensive medications can lead to increased cardiovascular complications. This is especially true for African Americans (AA) as they are less likely to achieve hypertension (HTN) control and more likely to be nonadherent to their medications. The purpose of this study is to explore barriers and facilitators to antihypertensive medication adherence in older African American people living with HIV (PLWH).

Methods: Qualitative descriptive design utilizing 5 semi-structured focus group sessions (N=36) with participants recruited from a local HIV clinic in the Birmingham metropolitan area. To participate, the following inclusion criteria must have been met: AA, age >50, HIV diagnosis >1 year, currently prescribed HTN medication. Data analysis was conducted using NVivo to code themes and SPSS for demographics. Themes were sorted and interpreted by 4 research team members.

Results: Identified barriers to medication adherence are: forgetfulness, perceived unimportance, and side effects. Identified facilitators to antihypertensive medication adherence include: routine/reminders/preparation, motivation & accountability, blister packs & medication mailing.

Implications: More studies are needed to identify the challenges faced by older AA with HIV regarding antihypertensive medication adherence. Future studies should focus on patient centered approaches to improve adherence in disparate populations.

97-Three Dimensional Cell Culture Model for Drug Efficacy Testing

Steven Roberts, Steven M. Rowe, Jennifer Guimbellot, Zhongyu Liu, Stephen Mackay

Cystic fibrosis (CF) is a chronic disorder generated by mutations in the cystic fibrosis transmembrane conductance regulator (CFTR) channel. CFTR has over 2,000 mutations causing abnormalities in many organs, predominately in the lungs. Recent studies have developed CFTR modulators, which increase the activity of CFTR. However, these drugs are not applicable to all CF mutations. Our research is directed towards optimizing ex vivo assays for responses to different drugs, which will advance CF therapy toward a more individualized approach when applied in a clinical setting. The study is designed to develop 3-D organoid models of human nasal epithelium (HNE). Two sphere formations were created through cell culturing, “apical membrane out” (AMO) and “apical membrane in” (AMI). AMO spheres shrink in response to CFTR activation, and AMI spheres swell in response. Both models were sectioned for H&E staining allowing cross sectional analysis and imaging. 3-D models were also fixed and immunofluorescence (IF) was performed. Photomicrographs were taken of spheres displaying their morphology in relation to that of a human nasal epithelium. We obtained AMO H&E stained spheres that are ciliated and AMI spheres with mucus formation, displaying differentiation. We are currently developing IF microscopy protocol to observe other proteins that indicate differentiation. Future directions would be to continue adjusting culturing conditions to optimize differentiation, to evaluate other markers of differentiation, and to test drugs to stratify the response of the spheres and determine which is most discriminating to judge effectiveness.

106-Barriers to Research Participation in Nicklaus Children’s Hospital Service Area

Beatriz Maciel, Yenni Cedillo, Jose Fernandez Ph.D.

Inclusion of minors in clinical research has been a long-existing challenge in the scientific community that oftentimes results in children being prescribed medication that has not been tested in their age group. Understanding the reasons preventing minors from participating in clinical research can aid in the recruitment of minors to research studies and allow for improvement in evidenced-based medicine for the pediatric population.

The purpose of this study is to identify the reasons guardians choose to allow their children to participate or not participate in clinical research. A phone questionnaire developed under the guidance of the Nicklaus Children’s Hospital in Miami was delivered to parents of children under 18 in Miami-Dade, Broward and Palm Beach counties asking about their willingness to allow their child to participate in clinical research. The results from 1,048 stratified randomized surveys were assessed. Using descriptive analysis, we were able to identify the percentage of respondents that were “Willing”, “Somewhat Open”, and “Not Willing” to allow their child to participate in clinical research and characterize the motives behind their choice. Of the 1,048 respondents surveyed, 10.5% of respondents were “Willing”, 34.8% of respondents were “Somewhat open”, and 54.6% of the respondents were “Not willing” to allow their child to participate in clinical research.

Determining the factors that influence the responder’s view of their children participation in research allows us to focus on ways to improve recruitment of children into clinical trials and ultimately lead to better research on diseases that affect children differently than adults.

3-Diets, Electrolytes, and Antiacid Food and Customer Service Prep

Lee,Walker, Santiago, et ., al 1997-2016

This Word and Poster Presentation can prepare many consumers and business marketers with Food and Customer Service Prep items to assist with diets and food nutrition chemical dissolves in order for patients to feel free from sickness and diseases.

7-Temperature-sensitive PVCL based polymersomes for sensing and drug delivery

Graham Dupont,1 Fei Liu,1 Veronika Kozlovskaya,1 Eugenia Kharlampieva

Temperature-responsive polymeric systems have attracted extraordinary interests in sensing and targeting drug delivery. In particular, nanostructured polymersomes with appealing thermo-responsiveness have been intensively studied due to the notable loading capacity and exceptional stability. We report on novel types of triblock copolymer polymersomes with temperature-dependent permeability within physiological temperature range of 37-42 ºC for controlled delivery of anticancer drug, doxorubicin. Incorporating with the properties of biocompatibility and biodegradability, reported polymersomes are also demonstrated with superior stability and facile procedures in regulating size and thermal responsiveness. The temperature responsive triblock copolymer poly (methylvinyl-caprolactam-co-vinylcaprolactam)-b-poly(dimethylsiloxane)65-poly(methylvinyl-caprolactam-co-vinylcaprolactam) (P(VCLm-MVCLn)-PDMS65-P(VCLm-MVCLn)) with m/n=0, 1, 10, 15, 19 and 29 are synthesized by controlled RAFT polymerization. Synthesized polymers are assembled into polymersomes at room temperature with the diameter decreased from 500 nm to 40 nm as the hydrophilic PVCL-MVC chain length increases. In addition, we also demonstrated the first example of finely controlling permeability within physiological temperature range by adjusting hydrophobicity of P (VCL-MVC) block as the polymersomes' corona. Reversible gradual shrinkage and swelling behaviors result in sustainable doxorubicin release from synthesized polymersomes in heating and cooling cycles. Both concentration and time dependent cytotoxicity of doxorubicin-loaded polymersomes to human alveolar adenocarcinoma cells (A549 cells) evidenced an extensive application potential as a novel type of stimuli-responsive nano-carriers in cancer therapy.

13-Ordering of Energy Levels in the Six Vertex Model

Sam Wunderly

We studied the relationship between the six vertex model and the 3 coloring problem that Temperley and Lieb found. My poster will verify this relationship through Matlab script. We have considered the subspaces spanned by all basis vectors with a particular value of magnetization. The transfer matrix is block diagonal, consistent with this decomposition. We plotted the eigenvalues for each block on the vertical, and the magnetization on the horizontal. We used Matlab to generate the spectrum for n= 5 up to n= 13. The results verify Lieb's conjecture. Sutherland's claim is more subtle. In the future we would like to explore the possibility of rigorously proving Lieb's conjecture.

29-Temperature Dependence of Spectroscopic Properties and Electrical properties of Cr(Fe)(Al):ZnSe Laser Active Materials

Rick Wakins, Ozarfar Gafarov, Vladimir Fedorov, Sergey Mirov

There is a high demand for high power, broadly tunable mid-IR lasers for many practical applications in medicine, material processing, environmental monitoring, industrial process control, and defense. Transition Metal (TM) doped II-VI semiconductors are proving to be potential candidates in developing these lasers in the 1.9-6 μm spectral range. Temperature influence on spectroscopic characteristics is crucial for many aspects of novel laser development including output noise, single frequency oscillation, and thermal bistability. We report on the spectroscopic characterization of chromium and iron doped ZnSe gain media at temperatures ranging from 77K to 389K. Heating of Cr:ZnSe resulted in the absorption peak shifting to a shorter wavelength from 1.806 μm at 77K to 1.753 μm at 389K. It also resulted in broadening of the absorption band from λ=260 nm to λ=373 nm and a decrease of the absorption cross section by 69%. Similar characterization was done for Fe:ZnSe laser material. The cooling of the Fe:ZnSe crystal from room temperature to 77K resulted in a 32% decrease of the absorption coefficient at 2.94 μm which is usually used as a pump source. We also studied the absorption of the electrical free-carriers in n-type Al:ZnSe crystals in visible and mid-IR spectral ranges. Diffusion of Al into ZnSe samples was achieved by annealing at 1000°C during 4 days in Al vapors. It was demonstrated that free-carriers absorption of Al:ZnSe samples with resistivity of 100-150 ohm per cm resulted in an increase of the absorption coefficient at 2.4 μm up to 2.5 cm^-1.

30-Magnetic Transitionsin Samarium at High Pressures and Low Temperatures Ordering of Energy Levels in the Six Vertex Model

Craig Johnson, Georgiy Tsoi, and Yogesh Vohra

The magnetic transitions in rare-earth metal Samarium have been studied up to 40 GPa and 10 K in a diamond anvil cell. The rare-earth metal Samarium is known to undergo structural phase transitions from Samarium phase to double hexagonal close packed phase at 1 GPa to face centered cubic phase at 14 GPa with increasing pressure. We employ designer diamond technology to measure electrical resistance changes at magnetic transitions under high pressures and low temperatures. The diamond anvil cell was cooled to 10 K using a Cryomech cooler and the pressure was measured by ruby fluorescence technique. Electrical resistance was measured at high pressures and low temperatures by the use of the four probe technique. We track the two known magnetic phase transitions temperatures (Neel temperatures) in Samarium to high pressures observed through electrical transport. A merging of Neel transition temperatures was found within the double hexagonal close packed phase at 12.2 GPa. The magnetic transition temperatures show a correlation with the underlying structural phase transitions between the hexagonal close packed layered structures. The magnetic transition persists up to the highest pressure of 40 GPa achieved in the present experiments. The present study provides information on the changes in the magnetic ordering temperatures and electronic structures induced by high pressures.

34-Photoluminescence Spectroscopy of GaAs Quantum Wells and Monolayer MoS2

Matt Stiles, Biplob Barman, David Hilton

High-quality semiconductors are essential for consistent results in the research and development of computer chip technology. Even miniscule amounts of unintentional impurities significantly reduce the mobility of electrons in semiconductor samples. Photoluminescence spectroscopy was used to detect defects in microscopic layers of gallium arsenide (GaAs) and molybdenum disulfide (MoS2). Each material was excited with a 447 nm laser, and the resulting light was captured in an optical spectrometer. The two samples of GaAs emitted a narrow range of near infrared frequencies with low-temperature linewidths under 10 nm, indicating high quality. The MoS2 sample displayed a much broader spectrum of visible and infrared frequencies with a linewidth of roughly 250 nm, which is insufficient for research applications. The GaAs will be transferred to a high magnetic field laboratory in the fall for further study.

43-Study of Defects in Strontium Titanate Using Electron Paramagnetic Resonance Spectroscopy

Brianna Kenney, Subash Paudel, Jamiyanaa Dashdorj, Mary Ellen Zvanut, Violet Poole, Matthew McCluskey

Strontium titanate is a material of considerable interest for electronic applications. A recent study revealed that strontium titanate (STO) annealed in strontium oxide (SrO) powder exhibits large persistent photoconductivity (PPC) after exposed to sub-bandgap light of 2.9 eV or higher. To better understand this phenomenon a titanium dioxide (TiO2) annealing treatment was applied to STO substrate to see if this property would be exhibited under altered conditions. Using electron paramagnetic resonance (EPR) spectroscopy, we were able to detect Fe3+ and Cr3+ defects in as-received STO samples. After annealing the samples, Fe3+ defects were not detected in both the SrO and TiO2 annealed; moreover, the signal intensity of Cr3+ defect decreased by no more than 20% for both annealed samples. To further study the samples we applied light illumination also referred to as photo-EPR by using LEDs and laser diodes that ranged from 1550 to 397 nm. Similar behavior was seen in both the SrO and TiO2 annealed samples in which the Cr3+ signal intensity reduced by at least two orders of magnitude; however, the TiO2 annealed sample did not exhibit this “giant” PPC. These results imply that Cr3+ is not responsible for this novel property. Ongoing studies are necessary to better understand what defect is responsible for this significant change in the electronic properties of STO.

55-Ordering of Energy Levels in the Six Vertex Model

Sam Wunderly

Temperley and Lieb found the relationship between the six vertex model and the 3 coloring problem. We are taking the six vertex model and entering a mock six vertex model into matlab. We then studied the eigenvectors and eigenvalues of our six vertex problem. The six vertex model can also be seen in real life when looking at the square ice molecule. My poster will go into greater detail of that. We used Matlab to generate the spectrum for n = 5 up to n = 13. The results verify Lieb’s conjecture. Lieb's conjecture was that the six vertex transfer matrix commutes with Heisenberg's quantum spin chain. Later, Sutherland explained this by introducing an auxiliary spin to construct the transfer matrix. Our matlab script also gave us graphs in order to visualize the eigenvalues. In the future we would like to explore the possibility of rigorously proving Lieb's conjecture.

56-Showing the Flag: Modeling Alien Megastructures

Aidan Farr

It has been proposed that placing a structure in orbit around a star would serve to indicate the presence of intelligent life by altering the light flux of the star in a manner distinct from natural phenomena. Interest in modeling this form of alien megastructure was jump-started by abnormal flux readings from the star KIC8462852 in October 2015. A Mathematica script is presented which allows the user to become familiar with the flux profiles of various flag designs as viewed from another star system, and to evaluate whether an observed flux profile could reasonably result from the presence of a flag. This process is applied to KIC8462852, furthering the consensus that the observed deviations are not likely to be the result of a flag. Some factors that would need to be considered in the process of building a flag of our own are also discussed, including the possibility of using a flag to transmit a binary-encoded message.

60-Analysis of Herbal Supplements Claiming Male Enhancement

Rebecca Smith, Dr. Elizabeth Gardner

This research project involves the analysis of supplements advertised for enhancing sexual performance. The supplements were purchased from a popular online market. Twenty-three samples were purchased and analyzed by gas chromatography mass spectroscopy (GC-MS) to determine if the pills or capsules contain any undeclared prescription drugs. If the products contain prescription drugs, they are being sold illegally. The supplements were extracted in chloroform and analyzed by gas chromatography mass spectroscopy (GC-MS). Initial results indicate that some of the supplements do contain the prescription drugs Tadalafil (Cialis) and Sildenfil (Viagra). Thiosildenafil, an analog of Sildenafil, was also detected in two of the supplements. Other substances found in the supplements include osthole, caffeine, and hexadecanoic acid. Future wok will include confirming the presence of these and other undeclared ingredients.

67-Assay Development for the Investigation of Viral Protein Function

Hayes, Kadeem O., Brady, Pamlea N., Chan. Matthew C., Johnson, Margaret A.

The severely pathogenic viruses, Severe Acute Respiratory Syndrome (SARS) and Middle East Respiratory Syndrome (MERS), which caused epidemics in 2003 and 2012, respectively, are members of a large family of viruses known as coronaviruses (CoVs). Viruses in this family are responsible for moderate to severe respiratory infections in humans and animals, and to date, there is no established treatment.1 Specifically, this work involves novel proteins found within bat coronaviruses. These domains in a central region of the nonstructural protein 3 are essential for viral replication and are predicted to be novel nucleic acid-binding proteins. However, these proteins have no well-characterized biochemical function. They are of particular interest due to their detectable sequence similarity to the SARS-unique region of the human SARS virus. The overall goal of the work presented is to develop an assay which will allow for a more in depth understanding of the function of a protein by the identification of binding partners. This knowledge can lend insight to potential methods of treatment of those infected by the virus. Development of the assay first involved the successful recombinant expression and purification of the proteins of interest under optimized conditions. Once pure protein samples were obtained, the thermal shift assay was chosen for functional analysis due to its ease of implementation in 96-well format. Proteins and ligands were identified for the assay and an assay format was developed. Preliminary results from the thermal shift assay will be reported.

68-Analysis of Boron Atom Incorporation in Boron-Doped Nanostructured Diamond Films Using X-Ray Photoelectron Spectroscopy

David Goodloe; Paul Baker, Ph.D

It is well established that boron doping of nanostructured diamond (NSD) films increases conductivity. While natural or synthetic diamond is electrically insulating, boron-doped diamond or NSD films are p-type semiconductors with numerous applications because of diamond's other advantageous material properties. However, the location and mechanism of boron atom incorporation in doped diamond films has not been discovered, and optimization of boron-doped diamond applications is thus limited. In seeking an explanation for doping activity, NSD films were chemical vapor deposited onto Ti-6Al-4V substrates. Samples were prepared using gas flow-rates of 500 sccm H2, 88 sccm CH4, and 8.8 sccm N2, and diborane (B2H6) gas flow-rates between 0.1 and 0.3 sccm. The project targeted nitrogen as the mechanism of boron incorporation, and four conditions were tested: low diborane with nitrogen, high diborane with nitrogen, low diborane without nitrogen, and high diborane without nitrogen. X-Ray Photoelectron Spectroscopy (XPS) surface analysis was utilized to detect specific binding energy chemical fingerprints, with high-resolution focus on the C1s and B1s regions. Curve fitting analysis of high resolution XPS spectra revealed relative intensities of various chemical bonds that indicated a strong correlation between boron and nitrogen incorporation in the form of boron-nitride (BN). Without nitrogen, boron did not bind substitutionally with carbon, indicating that nitrogen acts as a vehicle for boron incorporation into the diamond lattice.

69-Study of defects in Strontium Titanate Using Electron Paramagnetic Resonance Spectroscopy

Brianna Kenney, Subash Paudel, Jamiyanaa Dashdorj, Mary Ellen Zvenut, Violet Poole, Matthew McCluskey

Strontium titanate is a material of considerable interest for electronic applications. A recent study revealed that strontium titanate (STO) annealed in strontium oxide (SrO) powder exhibits large persistent photoconductivity (PPC) after exposed to sub-bandgap light of 2.9 eV or higher. To better understand this phenomenon a titanium dioxide (TiO2) annealing treatment was applied to STO substrate to see if this property would be exhibited under altered conditions. Using electron paramagnetic resonance (EPR) spectroscopy, we were able to detect Fe3+ and Cr3+ defects in as-received STO samples. After annealing the samples, Fe3+ defects were not detected in both the SrO and TiO2 annealed; moreover, the signal intensity of Cr3+ defect decreased by no more than 20% for both annealed samples. To further study the samples we applied light illumination also referred to as photo-EPR by using LEDs and laser diodes that ranged from 1550 to 397 nm. Similar behavior was seen in both the SrO and TiO2 annealed samples in which the Cr3+ signal intensity reduced by at least two orders of magnitude; however, the TiO2 annealed sample did not exhibit this "giant" PPC. These results imply that Cr3+ may not be the causing this "giant" PPC. Ongoing studies are necessary to better understand what defect is responsible for this significant change in the electronic properties of STO.

73-Effects of Low Rank Factor Matrices in Tensor Decomposition

Carmeliza Navasca, Elizabeth Liddle

When using the Alternating Least-Squares to find the tensor Singular Value Decomposition, the rank of the factor matrices composing the tensor can affect the speed of convergence. Decomposing tensors created from low rank matrices can lead to periods of slow convergence called swamps. We test if the existence of a swamp depends on whether the matrix has columns of zeros, collinear columns, or columns that are linear combinations of each other.

77-Electronic Structure of the Mid-Infrared Laser Iron-doped Zinc Selenide

Evan Garrison

Iron-doped Zinc Selenide (Fe:ZnSe) is a doped insulator laser with frequencies in the mid-infrared that shows much promise for room-temperature and above applications. With the ultimate goal in mind being to understand how diffusion of Fe in ZnSe works in order to maximize the diffusion rate, this study seeks to obtain a correct crystal and electronic structure that agrees with experiment. Density-Functional Theory, Pseudopotential Plane-Wave theory, and periodic boundary conditions are used to solve for total energies of ZnSe and Fe:ZnSe. First an accurate cutoff energy for the wave function is found. After this, the wave function obtained from an accurate energy calculation is used to optimize crystal structures, compute electronic density of states and band structure, and plot electron orbitals for both Fe:ZnSe and ZnSe to compare the doped and non-doped crystal.

78-Risk Perceptions of Ovarian Cancer Among College-Age Women

Ashley Crowe

There is a high demand for high power, broadly tunable mid-IR lasers for many practical applications in medicine, material processing, environmental monitoring, industrial process control, and defense. Transition Metal (TM) doped II-VI semiconductors are proving to be potential candidates in developing these lasers in the 1.9-6 μm spectral range. Temperature influence on spectroscopic characteristics is crucial for many aspects of novel laser development including output noise, single frequency oscillation, and thermal bistability. We report on the spectroscopic characterization of chromium and iron doped ZnSe gain media at temperatures ranging from 77K to 389K. Heating of Cr:ZnSe resulted in the absorption peak shifting to a shorter wavelength from 1.806 μm at 77K to 1.753 μm at 389K. It also resulted in broadening of the absorption band from λ=260 nm to λ=373 nm and a decrease of the absorption cross section by 69%. Similar characterization was done for Fe:ZnSe laser material. The cooling of the Fe:ZnSe crystal from room temperature to 77K resulted in a 32% decrease of the absorption coefficient at 2.94 μm which is usually used as a pump source. We also studied the absorption of the electrical free-carriers in n-type Al:ZnSe crystals in visible and mid-IR spectral ranges. Diffusion of Al into ZnSe samples was achieved by annealing at 1000°C during 4 days in Al vapors. It was demonstrated that free-carriers absorption of Al:ZnSe samples with resistivity of 100-150 ohm per cm resulted in an increase of the absorption coefficient at 2.4 μm up to 2.5 cm^-1.

88-Theranostic Polymer Agents for Ultrasound Therapy and Controlled Drug Release

Jacob Norman

Theranostic agents provide innovative methods for treatment of disease, by combining multi-modal imaging with targeted drug release. These highly targeted treatments offer less severe side-effects and for more efficient drug usage. Ultrasound is the cheapest and easiest imaging modality, and serves an ideal method for visualization and treatment. Currently available imaging agents do not give long-term contrast, and have difficulty with drug-loading. In the described research, Tannic acid–poly(N-vinylpyrrolidone) (TA-PVPON) multilayer capsules are used to observe the effect of ultrasound therapy on both ultrasound contrast imaging and modulated drug release. These systems are uniquely able to modulate binding energy via adjustments in polymer chemistry. Ultrasound is currently a clinical standard for both ultrasound imaging and therapy. By utilizing various frequencies and amplitudes of applied ultrasound, along with variations in capsule thickness and surface morphology, the specific impact of ultrasound on the theranostic agents is observed. Imaging is done using a clinical ultrasound device (SONIX ultrasound) and analysis is preformed aby utilizing custom region-of-interest based Matlab software. UV-vis data was collected following ultrasound therapy to verify drug release. We have shown that ultrasound contrast is greatly enhanced by ultrasound therapy, with variations based on capsule morphology and ultrasound intensity. In some cases, ultrasound contrast was observed increased about 800%. Thus, this work clearly demonstrates the ability to modulate ultrasound contrast and drug release of TA-PVPON theranostic agents via ultrasound therapy. This work has far reaching implications for the future of targeted-drug delivery.

94-Synthesis of Molecularly Imprinted Polyitaconic Acid

Kate Davis, Robert Collins, Aaron Catledge

More than 1 million Americans get a knee or hip replacement each year. A major detriment of joint implants is that the surgical sight and tissue surrounding them can become infected. One approach that may help diminish infection rates involves use of implants coated with molecularly imprinted polymers (MIPs) that can act to reduce biofilm formation through sequestration of bacteria quorum sensing molecules. As a first step in this approach, we molecularly imprinted polyitaconic acid with fluorescein and evaluated imprinting capability relative to the non-imprinted control. Our attempt to use ammonium persulfate (APS) as a fast-acting free radical initiator led to an unstable polymer when exposed to air. Instead, we settled on use of 1,1’-azobis(cyclohexanecarbonitrile) (ACC). Our attempts to use the solvent, N,N-dimethylformamide (DMF) were unsuccessful because its excessive fluorescein quenching prevented adequate fluorescence at low binding concentrations. Instead, we found that methanol could be used more effectively. The polymer was imprinted with a 1x10^-5 M fluorescein solution and then rebound with fluorescein solutions with concentrations of 1x10^-7 M, 1x10^-8 M, 1x10^-9 M, and 1x10^-10 M. The observed quenching of fluorescence with increasing fluorescein concentration was in agreement with the Stern-Volmer equation. However, contrary to expectation, the non-imprinted control polymer yielded a higher Stern-Volmer quenching constant than the molecularly imprinted polymer, resulting in a low imprinting factor. Explanations as to why this might have happened are discussed.

SERVICE LEARNING

89-STH 250 Women in Science Project: Ways to Spread the Power of Knowledge to Young Women

Kristin Deneen

Though women have reached many achievements, it can still be difficult for them to prove that they can be competitive in the sciences. Many young girls can feel intimidated by the scientific community and my not have a chance to be exposed to female leaders in the science fields. Therefore, this project was conceived with two main goals in mind—to make the public more aware of the science going on while spotlighting some of UABs amazing researchers, and inspire young women to continue to pursue a future in the sciences. The Women in Science videos were designed to give information and inspiration girls while being easily accessible and give the community at large more information about the many branches of science that are being researched here at UAB.

SOCIAL AND BEHAVIORAL SCIENCES

4-Sex Differences in the Emotional Response to Warning and Safety Cues

Danielle R. Hurst, Nathaniel G. Harnett, Muriah D. Wheelock, Kimberly H. Wood, Sylvie Mrug, David C. Knight

Emotional disorders (e.g., anxiety and depression) are prevalent in today's population, and are more common in females than in males. Some studies suggest that males and females may differ in emotional regulation, which is an important aspect of emotional disorders. Prior research has used Pavlovian fear conditioning during functional magnetic resonance imaging (fMRI) to study the neural and behavioral components of the emotional response. However, limited research has assessed sex differences in Pavlovian fear conditioning. Determining sex differences in Pavlovian fear conditioning may enhance our understanding of sex differences in emotional disorders. Therefore, the current study investigated sex differences in behavioral and neural responses during Pavlovian fear conditioning. Participants (n = 110) were presented two tones (conditioned stimuli; CS). One tone served as a warning cue (CS+) and was paired with a loud white noise (unconditioned stimulus; UCS). The other tone served as a safety cue, signaling the omission of the UCS (i.e., CS-). Neural and behavioral conditioned responses were assessed via the skin conductance response (SCR) and fMRI signal change. SCR data showed a significant interaction between stimulus type and sex such that the effect of conditioned stimulus type depended on the participant's sex. Further, female participants demonstrated greater activity in the dorsomedial and dorsolateral prefrontal cortex during the CS+ compared to male participants. These brain regions may support emotion regulation processes. The current study demonstrates sex differences in the neural and emotional response, and suggests there are sex differences in the associative fear learning process.

48-An Increase in Fatigue Symptoms Affects Activity Levels the Following Day in Patients with Chronic Fatigue Syndrome

Cooper Bailey

Chronic Fatigue Syndrome (CFS) plagues over one million people, primarily women, in the United States. CFS manifests itself in many different ways; however, the most common symptom is severe fatigue. Pain, headache, and concentration and memory issues also commonly accompany a CFS diagnosis. Previous research has shown that having a higher body mass index (BMI) has a weak positive association with increased levels of CFS symptoms. It is commonly assumed that the individuals' increased weight is a contributing factor to the increase of severity of symptoms. However, it is possible that the debilitating fatigue symptoms of CFS can lead to the increased BMI by way of facilitating a decrease in the levels of activity the person has throughout the day. As part of this study, participants with a diagnosis of CFS completed a questionnaire at night every day for the forty day duration of the study period. The daily measure assessed daily fatigue levels and various levels of physical activity and exertion before the participant went to sleep. Each participant also underwent a body composition exam using a Tanita Body Composition Analyzer. It was found that there was a significant negative association between the level of fatigue on one day and the level of activity on the following day by using a time-lapse model assessment. In particular, if the participant reported a high fatigue rating, they then reported a lower activity rating the following day. We concluded that higher levels of fatigue are associated with lower levels of activity, which could be a factor in increased BMI.

53-One Thing Leads to Another: Untangling the Relationship between Depression and Pain in Outpatients with Serious Illness

Osisami, Oladele; Bagcivan, G., PhD; Ejem, D., PhD; Mancarella, G. A., MPH; Dionne-Odom, J. N., PhD, RN; Kvale, E., MD, MPH; Tucker, R., MD; Bakitas, M., DNSc, CRNP

Depression and pain are commonly experienced by patients with serious illness, such as cancer and heart disease. Depression and pain co-occur in patients 30-50% of the time resulting in the exacerbation of each symptom, warranting equal attention to develop an effective treatment plan. We conducted a secondary analysis of the UAB Supportive Care and Survivorship Clinic dataset of outpatients with serious illnesses treated from 2012-2016. The aim was to determine the correlation between depression (scaled 0-27, measured by the PHQ-9) and pain (scaled 0-10, measured by the Brief Pain Inventory [BPI]). We hypothesized that depression severity would be associated with pain intensity. The sample (n=120) was mean age 51.27 years, 58.3% female, 24.2% black/African-American, 74.2% white/Caucasian and 52.5% married. We found positive correlations between mean depression total scores (12.43±6.21) and "least" (mean=4.07±2.86, r=.347, p=.001), "worst" (mean=7.48±2.45, r=.285, p=.012), & "pain now" (mean=4.89±3.27, r=.312, p=.002) pain scores. One-way ANOVA and post hoc tests demonstrated that the "least" (p=.009) and "pain now" (p=.027) pain scores were significantly higher in the patients with severe depression than those with mild depression. These results suggest that outpatients with severe depression report higher "least" and "pain now" pain scores than those with mild depression. This finding is consistent with other literature describing a close relationship between pain and depression but adds specificity regarding the relationship of depression severity and pain intensity. Research and practice implications include considering depression severity and pain intensity assessment for effective management of their co-occurrence in patients with serious illness. v

82-Baclofen and Opioid Interactions: Reward and Analgesia

Remy Y Meir, Stacie K Totsch, and Dr. Robert E Sorge

Opioids remain the primary treatment for moderate to severe pain, but this class of drug has significant negative side effects that include addiction and abuse. Therefore a drug that can reduce the negative effects of opioids, while preserving pain relief, would be highly advantageous. Classes of receptors, known as GABA receptors, that have two main subgroups, GABAa and GABAb receptors, are involved in inhibitory functions. Opioids are drugs that produce an analgesic, rewarding effect by diminishing the inhibitory effect of GABAb receptors on the release of dopamine. Whereas, the drug baclofen works as a GABAb agonist, activating the receptors in order to regulate the amount of dopamine being released by dopaminergic cells. The activation of GABAb receptors is involved in the direct inhibition of dopamine release. The anti-spastic drug baclofen is a GABAb receptor agonist and may interact with opioids. It has been shown that the administration of baclofen prior to the administration of an opioid, such as morphine or fentanyl, shows synergy in regards to analgesic effect in the hot plate test. Currently, conditioned place preference testing is being performed in order to observe whether the two drugs also interact synergistically in this test of drug reinforcement. We demonstrate that the interaction between baclofen and opioid analgesics can be both synergistic and additive in different contexts. These findings could lead to safer administration of prescription opioids when mixed with baclofen to achieve high analgesic and low rewarding effects.

95-Factors Affecting Age of Diagnosis of Autism Spectrum Disorder: Mother's Education Level

Caleb Brasher, Maria I. Hopkins, Ph.D., Sarah A. Koch, B.A.

Autism spectrum disorder (ASD) is a group of developmental disabilities that can cause significant social, communication and behavioral challenges. When caught early enough, behavioral therapy can be extremely effective in reducing the severity of the effects of ASD, as the brain is far more adaptive during the early stages of development. Not receiving therapy until later stages of development, in turn, means that many of the symptoms will not be nearly as effected by treatment, making it hard for the individual with ASD to relate to the world around them. All in all, early detection of ASD is crucial to ensure that children with ASD are the least socially inhibited as possible.

This study examined diagnostic age and factors associated with diagnostic age for 49 children with autism spectrum disorder. The study examined several demographic factors that research has indicated influence the age at which children are evaluated for ASDs. Primary variables of interest included sex, race/ethnicity, urbanicity, and maternal education. The results of this study indicate that higher maternal education was associated with earlier ASD diagnosis ($F(3, 45) = 8.297$, $p < .01$).

Although diagnosis of ASD is possible in children as young as 14 months of age, most children in the study had not been diagnosed until after 4 years of age. Understanding factors associated with delay in diagnosis may inform public awareness campaigns, screening guidelines, and professional education programming with the aim of lowering the age at which autism is detected. This would result in earlier access to intervention for children with autism.

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98-Sensitivity to Pain in Autism Spectrum Disorders

Victoria C. Seghatol-Eslami & Rajesh K. Kana

One of the diagnostic features of autism spectrum disorders (ASD), according to the DSM-V, is hypo- or hypersensitivity to sensory input such as pain. Unusual sensory sensitivity in ASD has a significant impact on the behavioral symptoms of this population. While there is growing interest in studying pain sensitivity in ASD, the number of empirical studies of pain in ASD is significantly less compared to other functions in this disorder. Therefore, the primary goal of this study was to review the literature on pain sensitivity in ASD in order to arrive at a consensus on its role in the behavioral profile of ASD. Twenty-three studies (17 experimental and 6 case studies) were included in the current review after a literature search on Google scholar and PubMed with the search criteria “pain and autism” and “pain in ASD.” Five case studies indicated pain hyposensitivity and insensitivity among individuals with ASD; the other indicated pain hypersensitivity. Five experimental studies indicated no difference between ASD and TD (typically developing) individuals, one reported hyposensitivity in ASD, and six showed hypersensitivity. The remaining five experimental studies investigated other aspects of pain that cannot be suitably categorized into any of the three categories listed above. Although the findings reported are largely inconsistent, it is clear from experimental studies that individuals with ASD do experience pain, in contrast to the common belief. It may be useful for future research to examine the neurobiology of pain as well as pain sensitivity as a function of autism symptom severity.

99-Interpersonal and Intrapersonal Mechanisms Play Independent Mediating Roles in the Effect of Internalized HIV Stigma on ART Adherence

Victoria C. Seghatol-Eslami, Heather Dark, Whitney Smith, Janet M. Turan, and Bulent Turan

Adherence to antiretroviral therapy (ART) is crucial for people living with HIV (PLWH) to achieve optimal health outcomes. Internalized HIV stigma occurs when PLWH accept negative beliefs about themselves because of their HIV infection, and predicts sub-optimal ART adherence. However, mechanisms through which internalized HIV stigma affects ART adherence remain elusive. We examined the independent mediating roles of two factors in the association between internalized HIV stigma and sub-optimal ART adherence: Concerns about being seen while taking HIV medication (an interpersonal mechanism) and HIV treatment self-efficacy (confidence to adhere to ART—an intrapersonal mechanism).

Using validated measures, 180 PLWH (65 women, 115 men) in Birmingham, AL self-reported internalized HIV stigma, concerns about being seen while taking HIV medication, HIV treatment self-efficacy, and ART adherence. We calculated bias-corrected 95% confidence intervals (CIs) for indirect effects to test the hypothesis that concerns about being seen while taking HIV medication and self-efficacy simultaneously mediate in a parallel fashion the relationship between internalized stigma and ART adherence

Concerns about being seen while taking HIV medication and lack of treatment self-efficacy were parallel mediating mechanisms in the stigma – adherence association. That is, internalized HIV stigma predicted increased concerns about being seen while taking HIV medication and lower self-efficacy, and both mediators predicted sub-optimal ART adherence (in paths that are independent of each other).

Adherence interventions may need to target interpersonal as well as intrapersonal mechanisms, since both types of mechanisms appear to operate simultaneously and independently of one another.

WORKS IN PROGRESS

12-Effects of Botanical Microglia Modulators in Gulf War Illness: A Novel Screening Approach

Rebecca L. Massey

Gulf War Illness (GWI), is a chronic multisymptom illness of unknown etiology that affects approximately 250,000 veterans of the 1990-1991 Persian Gulf War. There are currently no objective diagnostic tests or targeted treatments for pain, fatigue, and other debilitating symptoms associated with this illness. We hypothesize that GWI involves abnormal sensitization of the immune system, resulting in low-level neuroinflammation. This study aims to both help patients who are suffering with the symptoms of GWI and to give insight into possible mechanisms of disease and potential targets for future treatments. In order to achieve these aims, we will be testing the effects of nine anti-inflammatory botanical compounds in 40 male veterans with GWI. We use a novel, blinded study design in which each participant trials three separate botanicals, as well as placebo. Participants will also complete a symptom severity report twice a day and have blood drawn at 4 time points for inflammatory markers. A botanical compound will be considered successful if the symptom severity reduction meets both statistical and clinical criteria. As an exploratory endpoint, blood will be tested for levels of inflammatory markers to see if there are mechanistic differences in symptom reduction associated with different treatments. This unique design allows us to efficiently screen a large number of potential therapeutic agents in a small group of individuals, and daily outcome assessments provide superior statistical power to detect effects. We propose this design as a powerful way to explore disease mechanisms and prioritize possible treatment options for further study.

24-Differences in Prevalence of Childhood Obesity among Hispanic/Latino Subgroups from the National Health Interview Survey, 2008-2014

Calvin Colvin, Lucia Juarez, and Bertha Hidalgo

Objective: The prevalence of obesity in Hispanic/Latino children is higher compared to other racial/ethnic groups in the United States. The objective of this study is compare prevalence of childhood obesity across racial/ethnic groups and within the Hispanic/Latino group. Methods: Data from the National Health Interview Survey (NHIS) years 2008-2014 (N=27913) were used for this study. Obesity status for the children was defined as being at or above the 95th percentile on the Center for Disease Control's BMI-for-age Charts. Logistic regression models included obesity status as the dependent variable and race/ethnicity as the independent variable. We controlled for age, sex, NHIS survey year and clustering weights. Mexicans, Mexican-Americans, Puerto Ricans, Central and South Americans were included in the analysis along with non-Hispanic Blacks, Whites, and an 'Other' category. Analyses were stratified by sex.

Results: Mexican-American males had the greatest prevalence of obesity for most years included in the study. Non-Hispanic blacks and foreign-born Mexicans had increased odds of obesity (OR(CI)= 0.85 (0.74, 0.98) and OR(CI)= 0.84 (0.72, 0.99), respectively) compared to Mexican-Americans. Whites and individuals in the 'Other' category had the lowest odds of obesity in this sample (OR(CI)= 0.49 (0.43, 0.56) and OR(CI)= 0.59 (0.49, 0.69), respectively). These associations remained statistically significant after stratifying by gender among males; the effect was attenuated among females and no longer statistically significant.

Conclusions: Preliminary results show differences in the prevalence of childhood obesity between Hispanic/Latino background groups and suggest the need for a more complex categorization of race/ethnicity in obesity research.

49-Osseointegration of Photofunctionalized Titanium Dental Implants

Aileena Nelson

Serious complications can arise after dental implantation, a common procedure. Although a variety of factors can contribute to implant failure, implant loss is generally attributed to the failure of osseointegration in the healing process, where the bone does not adhere entirely to the surface of the implant. When complete osseointegration does not occur, the resulting spaces between the bone and the implant invite microbial habitation, leading to the inflammation of the surrounding tissues and loss of the surrounding bone. Osseointegration depends on a variety of factors, including surface topography and chemistry. Past literature has shown that increasing the surface roughness of implants increases osseointegration, as surface area increases and more cells attach. Various methods are used to alter the surface topography of an implant, including resorbable blast-texturing (RBT), and laser-etching. Another method used to alter the surface of titanium implants is photofunctionalization, an ultraviolet (UV) light treatment. By removing the accumulated hydrocarbons on the surface of titanium and creating a hydrophilic surface, cell attachment is increased leading to greater osseointegration. In this study, the osteogenic potential of resorbable-blast textured and laser-etched discs with and without photofunctionalization was examined. Titanium discs without surface alteration (called machined discs) were used as a control. Osteoblast-like MG-63 cells were cultured on the photofunctionalized and untreated resorbable blast-textured, laser-etched, and machined discs. Cell proliferation was examined on days 1, 5, 7, 12, and 14. The data showed cell proliferation was far greater on the photofunctionalized discs. In the future, this experiment will be replicated further

63-Inhibition of Arterial Intimal Hyperplasia via Implementation of 3D Printing to Fabricate a Geometrically Enhanced Nanofibrous Biomimetic Vascular Graft

Katie Herron, Vinoy Thomas

The failure of most tissue engineered vascular grafts is due to anastomotic intimal hyperplasia (AIH). The development of successful scaffolds requires that both the mechanical and physical properties of the scaffold match the properties of native vessels. Mechanical stress can lead to tearing and other damage on the vascular graft which can initiate intimal thickening and potential failure of the vascular graft. In this study, the development of a novel mandrel consisting of a 3D printed cylinder and a wire that attaches to the cylinder to be used as a collecting substrate for electrospun nanofibers. The fibrous membranes will be dual-reinforced through use of repetitive, geometrically idealized grooves as well as aligned fibers. Three different mandrels were designed with different groove pitch sizes (1mm, 2mm, and 3mm), and the mechanics of mechanically enhanced electrospun scaffolds were simulated with a pressure of 120 mmHg which is the systolic pressure exerted by a healthy heart. This study found that as the pitch size of the mandrels decreased, both the displacement and maximum principle strain decreased.

84-Mechanobiology of Scleral and Lamina Cribrosa Cells in Fibrous Matrices

Aaron B. Stuber, Joel Berry, Jacqueline Nikles, David Nikles, Rafael Grytz

Ribosome profiling experiments are used to discover small open reading frames (smORFs), previously presumed to be noncoding pieces of DNA. Mackowiak (2015) reported a smORF database for zebrafish. Based on smORFs that were highly conserved across species and sufficiently long enough to indicate expressivity, one gene was selected. A sequence annotated as a smORF was identified to encode for a 91 amino acid peptide. Upon closer inspection, this sequence was found to be part of the HID1b gene and not a smORF. Upstream sequences predicted to be potential enhancers of this misannotated smORF were found to be part of the USH1Gb gene. The HID-1b gene codes for a protein involved in intracellular Golgi trafficking and is conserved across species; however, very little is known about this gene (Wang et al. 2011). Similarly, the neighboring USH1Gb gene has been connected to the development of Usher Syndrome, which causes partial blindness and complete deafness. This gene has been shown in zebrafish to have a similar function affecting the fish's hearing and balance (Phillips et al. 2015). The purpose of this experiment was to assess the effectiveness of designed CRISPR/sgRNAs and to observe genotypic and phenotypic changes resulting from HID-1b and USH1Gb knockout embryos. A cloning-free method was used to generate CRISPR/sgRNA templates, and in vitro transcribed sgRNA was injected with Cas9 protein into embryos. Analyzing the injected embryos provides insight into the effectiveness of CRISPR nucleases, the extent of genotypic and phenotypic variation, and the function of HID-1b and USH1Gb in zebrafish.

93-The Possible Anti-Obesity and Anti-Nociceptive Potential of CB2 Receptor Agonist JWH-015 in Diet Induced Obese Mice: Work in Progress

Aaron R Landis and Dr. Robert E Sorge

The rates of chronic pain and obesity are rising in the American population and there is good reason to believe the two conditions are linked. The endocannabinoid system includes CB1 and CB2 receptors and primarily affects mood, pain, and appetite. Previous research in animals demonstrated that, although administration of CB1 receptor agonists caused weight loss, they also increased depression and other changes in mood. JWH-015 is a selective CB2 receptor agonist and may play an immunomodulatory role. This project is meant to discover the potential anti-obesity effects of JWH-015 in mice fed a standard American diet (SAD), a high-carbohydrate fat diet meant to reflect the average diet of obese Americans. To date, we have seen sex differences in weight gain and glucose tolerance during SAD consumption, but no sex or diet effects on mechanical or thermal sensitivity. Following 8 weeks on the SAD diet, mice will be subdivided and given osmotic minipumps with JWH-015 or control treatment for 2 weeks. It is expected that the chronic administration of JWH-015 will induce significant weight loss in the SAD-fed mice, but not the regular diet mice. In addition, following implantation of the minipumps, we will be looking at the CB2 receptor agonist and its effect on CFA-induced inflammation and hypersensitivity. If JWH-015 is found to have anti-obesity and anti-nociceptive properties, it would have major applications due to the rises in obesity and chronic pain-related illnesses in the American population.