UNIVERSITY OF
SOUTH DAKOTA
SANFORD SCHOOL OF MEDICINE

Annual Medical Student Research Forum 2018

Friday, July 20th
9:00 AM to 1:00 PM

Room 111
Lee Medical Building

Sanford School of Medicine
The University of South Dakota
414 East Clark Street
Vermillion, SD

Program and Abstracts

Program Schedule

9:00-9:15	Opening Remarks on Summer Research Program, T-35 Program, and Independent Research Program.
9:15-9:30	Katerina DeHaan-Influenza Viruses in Ferrets: Super-Infections and Immunity. (Victor Huber, PhD)
9:30-09:45	Andre Hafner-Veratridine, a Potential Plant Derived Anti-Cancer Compound in Human Colorectal Cancer. (Khosrow Rezvani, MD, PhD)
9:45-10:00	Alex Hanson-UBXN2A Promotes Proteasome-Dependent Degradation of Mortalin-2 Oncoprotein in Human Colon Cancer cells. (Khosrow Rezvani, MD, PhD)
10:00-10:15	Kayla Knutson-MARCKS Overexpression Does Not Rescue Behavioral Parameters or Brain Histopathology in Mouse Model of Alzheimer's Disease. (Jill Weimer, PhD)
10:15-10:30	Break
10:30-10:45	Gavin Nelson-Collateral Coronary Arteries May Be Protective of STEMI in Patient with Multi-Vessel Occlusions. (Adam Stys, PhD)
10:45-11:00	Casey Reihe-Necessity of Hypothalamic Paraventricular Nucleus for Venous Responses to Activation of the Cardiac Sympathic Afferent Reflex. (Douglas Martin, PhD)
11:00-11:15	Jackson Shriver-An Uptake Assay to Demonstrate the Role of Fc Mediated Phagocytosis in Influenza Immunity. (Victor Huber, PhD)
11:15-11:30	Lauren Van Hove-Activation of Cannabinoid (CB2) Receptors and Reactivity of Cerebral Arterioles. (William Mayhan, PhD)
11:30-11:45	Janice Baumberger Gerlach-A Qualitative Study of the Experiences of the South Dakota Healthcare Providers with the Hutterite Patients.
11:45-12:00	Scholarship Pathways Presentation
12:00-1:00	Lunch with Advisors.

Abstracts

Influenza Viruses in Ferrets: Super-infection and Immunity

Katerina N. DeHaan and Victor Huber

Division of Basic Biomedical sciences, Sanford Medical School, USD

Secondary bacterial superinfection continues to be the leading cause of death during influenza virus outbreaks. During the 2009 H1N1 pandemic, 27% of influenza A virus fatalities were associated with laboratory-confirmed *Streptococcus pyogenes*, also known as group A streptococcus (GAS). In addition to influenza A, there is also a strong association between influenza B viruses and severe GAS pneumonia. As influenza's interaction with the host creates an avenue of susceptibility to invasive bacterial infections, the mechanism by which this occurs remains poorly understood.

Ferrets are naturally susceptible to human influenza viruses and they have been used to study both virus transmission and vaccine efficacy. In addition, the domestic ferret's anatomical and physiological structure, as well as its' response to infection, closely resembles that of humans, making this animal ideal for the study of infection and transmission of disease. In this study, we aim to create a ferret secondary bacterial infection model to investigate the immune response both preceding and succeeding influenza B and GAS infection, using recently developed tools to study T cell responses by flow cytometry.

Identifying Human Islet circRNAs Associated with β Cell Loss in a Humanized Mouse Model

Connor Fullerton and Zhiguang Guo

Introduction: Circular RNA (circRNA) is a novel type of RNA that, unlike linear RNA, forms a covalently closed continuous loop, some of which are highly represented in the eukaryotic transcriptome and often show tissue/developmental-stage-specific expression. circRNA regulates insulin transcription and secretion in human islet B cells. We hypothesize dying or dead human B cells release a panel of islet-specific circRNAs into the circulation, and that the levels of circRNA in circulation after inducing B cell death are associated with B cell death. We are interested in use our humanized mouse model with adoptive lymphocyte transfer (ALT) to identify novel human β cell-specific circRNAs in human islet grafts related to early β cell loss.

Methods: NOD.scid mice were injected with STZ to induce diabetes, and blood glucose was >500mg/dL for 50 ± 6 days prior to human islet transplantation. Mice were transplanted with 3,000 islet equivalents (IEQs) each from human islet donors with purity >80% and viability >90%. Mice were normoglycemic (blood glucose <200 mg/dL) for at least 2 weeks before undergoing adoptive lymphocyte transfer, receiving lymphocytes ($1x10^6$) from diabetic NOD mice. Control mice received Phosphate buffer saline (PBS). Nonfasting blood glucose levels were measured daily during the first week, then twice a week. Blood and islet graft were collected immediately once blood glucose reached >200mg/dL. BiP and TUNEL staining were performed to confirm endoplasmic reticular (ER) stress and apoptosis, respectively, in islet grafts. Human circRNA Microarray (ArrayStar) was used to measure circRNA targets in isolated human islets and islet grafts. qRT-PCR was used to validate Microarray results.

Results: BiP and TUNEL staining successfully confirmed higher levels of ER stress and apoptosis, respectively, in ALT-treated grafts when compared to PBS-treated grafts from the same donor. circRNA expression profiling was successfully identified by Human circRNA Microarray (ArrayStar) and validated by qRT-PCR. Differential expression of circRNA between isolated human islets, PBS-treated human islet grafts, and ALT-treated human islet grafts is currently being elucidated.

Conclusions: circRNA expression profiling and circRNA differential expression in humanized mouse models will allow for study of B cell death's relation to up/down regulation of novel human B cell-specific circRNAs in human islet grafts. Future aims of the project include developing novel biomarkers of early B cell loss, as well as developing innovative approaches for the prevention and treatment of Type 1 Diabetes (T1D). We believe that characterizing changes in circRNA expression from transplanted human islets with or without inducing beta cell loss from the same donor will advance our understanding about the role of circRNA as a modulator of B cell death.

Li Y, Zheng Q, Bao C, Li S, Guo W, Zhao J, Chen D, Gu J, He X, Huang S: Circular RNA is enriched and stable in exosomes: a promising biomarker for cancer diagnosis, Cell Res 2015, 25:981-984
Memczak S, Jens M, Elefsinioti A, Torti F, Krueger J, Rybak A, Maier L, Mackowiak SD, Gregersen LH, Munschauer M, Loewer A, Ziebold U, Landthaler M, Kocks C, le Noble F, Rajewsky N: Circular RNAs are a large class of animal RNAs with regulatory potency, Nature 2013, 495:333-338
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Veratridine, a potential plant derived anti-cancer compound in human colorectal cancer.

Andre Hafner and Khosrow Rezvani

Division of Basic Biomedical Sciences, Sanford School of Medicine, USD

Anticancer agents derived from natural alkaloids are very promising and have already served as a rich reservoir for drug discovery. This study focuses on Veratridine (VTD), a natural steroid-derived alkaloid found in Liliaceae plants that has prospective anticancer properties. VTD transcriptionally increases a ubiquitin-like molecule UBXN2A which binds and inhibits mortalin-2 oncoprotein. Here, we aimed to establish VTD as an inducer of UBXN2A in an *in vivo* mouse model and examine its effects on tumor progression. Intraperitoneal injections of VTD over four weeks resulted in markedly increased levels of UBXN2A in colon tissues and slowed tumor progression in a UBXN2A dependent manner. Due to the potential neurotoxic effects of VTD, a subsequent aim was to ensure the biological viability of two VTD-conjugates that were modified to improve their pharmacokinetic profile: PLE-VT and PEG-PLE-VT. Flow cytometry analysis of colon cancer cells treated with one of the two conjugated forms of VTD revealed a significant increase in UBXN2A protein levels. More importantly, both PLE-VT and PEG-PLE-VT induced apoptosis in treated cells indicating that both compounds maintain the anti-cancer properties of VTD previously described by our group. Cumulatively, our results strongly support the anti-cancer effects of VTD and advance the objective of its potential use as a target therapy with acceptable safety profiles and favorable clinical outcomes in patients with colorectal cancer.

UBXN2A Promotes Proteasome-Dependent Degradation of Mortalin-2 Oncoprotein in Human Colon Cancer Cells.

Alex D. Hanson and Khosrow Rezvani

Division of Basic Biomedical sciences, Sanford Medical School, USD

Colorectal cancer (CRC) is the third leading cause of cancer-related deaths in the United States. Despite advances in treatment of primary tumors, one third of CRC patients will die from metastatic (disseminated) disease. Understanding the mechanisms of metastatic programs are critical steps in the development of more effective therapies for advanced CRC.
Mortalin-2 (mot-2) regulates protein folding processes in mitochondria in normal cells. However, in malignant cells, mot-2 relocates from the mitochondria to the cytoplasm where it binds to p53 tumor suppressor proteins preventing promoting growth and migration of tumor cells. The UBXN2A protein has been shown to occupy the binding domain of mot-2 and reactivate p53. We hypothesized that UBXN2A binds to mot-2 to recruit CHIP E3 ubiquitin ligase for ubiquitination and proteasomal degradation of mot-2 proteins. We showed that silencing CHIP and UBXN2A decreases ubiquitination of mot-2. In addition, overexpression of UBXN2A in silenced UBXN2A cells increased ubiquitination of mot-2. Finally, we found UBXN2A decreases the half-life of mot-2 protein while the silencing of UBXN2A increases half-life of mot-2 proteins in colon cancer cells. Our studies will address the central hypothesis that UBXN2A-CHIP complex reduces migratory, invasive, and metastatic properties of CRC by negatively regulating mot-2 oncoprotein.

MARCKS overexpression does not rescue behavioral parameters or brain histopathology in a mouse model of Alzheimer's Disease.

Kayla Knutson[1], Jon J. Brudvig[1,2], Samantha Davis[1], Brandon Meyerink[1], Rachel Laufman[1], Tyler Johnson[1], Deborah Stumpo[3], Perry Blackshear[3], & Jill Weimer[1]

[1]Pediatrics and Rare Diseases Group, Sanford Research, Sioux Falls, SD 57104 USA; [2]Basic Biomedical Sciences, University of South Dakota Sanford School of Medicine, Vermillion, SD 57069 USA; [3]Signal Transduction Laboratory, National Institute of Environmental Health Science, Research Triangle Park, NC 27709, USA

Appropriate levels of synaptic activity are vital for neuronal health, and loss and dysfunction of synapses has been shown to precede neurodegeneration in several disorders including Alzheimer's Disease (AD). To treat AD, many strategies aim to rescue synaptic pathology by reducing β-amyloid (Aβ) plaques. Our study aimed to stabilize synapses and prevent dendritic spine loss and neurodegeneration by modulating myristoylated alanine-rich C kinase substrate (MARCKS). In addition to its developmental role in cortical lamination and axon guidance, MARCKS interacts with dendritic spine machinery involved in AD and regulates preclinical synapse pathology in AD. Using the 5XFAD mouse model of AD, we overexpressed MARCKS in excitatory forebrain neurons starting at postnatal day 30 using a doxycycline controlled tet-off system. Behavioral parameters were tested at 2, 4, 6, 8, and 10 months of age, but MARCKS overexpression showed no improvement in AD phenotypes in either the Morris Water Maze or Rotarod tests. This was followed by histopathological analysis of AD phenotypes in the cortex and subiculum of the brain including gliosis and amyloidopathy. MARCKS overexpression partially rescued microglia and astrocyte reactivity but did not ameliorate Aβ plaque formation. Further work will include analysis of synapse loss, neurodegeneration, and dendritic spine characteristics.

Collateral Coronary Arteries May Be Protective of STEMI in Patient with Multi-Vessel Occlusions.

Gavin Nelson, Udit Bhatnagar and Adam Stys

Sanford Cardiovascular Institute

A 54-year-old male with a history of cardiovascular disease presented to the emergency room with chest pain. The patient was diagnosed with non-ST-segment elevation myocardial infarction (NSTEMI). He underwent a coronary angiogram and primary percutaneous coronary intervention (PCI) following admission. His angiogram showed severe multi-vessel coronary artery disease (CAD) along with extensive collateral circulation. We hypothesize collateral circulation helped in preventing acute transmural ischemia despite a large area of circulation from the affected artery, therefore, protecting this patient from ST-segment elevation myocardial infarction (STEMI).

Necessity of Hypothalamic Paraventricular Nucleus for Venous Responses to Activation of the Cardiac Sympathetic Afferent Reflex

Casey Reihe and Douglas Martin

Division of Basic Biomedical sciences, Sanford Medical School, USD

To date, 46% of hypertensive patients remain hypertensive despite available treatments, highlighting the need to advance understanding of the mechanisms controlling blood pressure. We targeted two understudied mechanisms, the venous system and the cardiac sympathetic afferent reflex (CSAR). This study tested the hypothesis that that activation of the CSAR would increase venous tone, and that neurotransmission in the PVN would be required for this response.

Experiments were performed on anesthetized male Sprague Dawley rats instrumented with femoral arterial and venous catheters for recording of arterial and venous pressures. A right atrial balloon catheter inserted via the jugular vein allowed estimation of venous tone. The rats were fitted with a catheter placed in the pericardial space and with injectors placed in the PVN for injection of drugs. Hemodynamic responses were recorded during pericardial injection of vehicle (saline) and bradykinin (CSAR stimulant) before and after PVN injection of ω-conotoxin (synaptic inhibitor).

CSAR activation increased arterial pressure (19±2.0 mm Hg). However, there was no effect on venous tone. These responses were abolished after PVN conotoxin injection. We conclude that neurotransmission in the PVN is required for the blood pressure and heart rate responses to CSAR activation.

An Uptake Assay to Demonstrate the Role of Fc Mediated Phagocytosis in Influenza Immunity

Jackson Shriver, Grigoriy Sereda and Victor Huber

Division of Basic Biomedical sciences, Sanford Medical School, USD

Vaccination against influenza can significantly minimize the severity of an influenza virus infection, but these vaccines are not 100% effective at preventing and eliminating infections. The typical target of vaccine-induced immunity is the hemagglutinin (HA) protein expressed by the virus. Additionally, interest has been made toward developing a vaccine against the Matrix-2 (M2) surface protein of influenza as it is highly conserved among influenza A viruses. In addition, to being a component of the virus itself, the M2 protein is also highly expressed on the surface of infected cells and antibodies against the M2 protein may enhance antibody dependent cellular cytotoxicity through natural killer cells within the immune system. Antibodies protect the host through neutralization, initiation of complement, or by interacting with host immune cells. Antibodies that enhance cellular response are critical for maintaining anti-influenza immunity as neutralization requires a constant high level of antibodies which may wane between vaccination and infection. It is known that Fc receptors on host cells are required for effective clearance of influenza viruses, but the exact mechanism of action remains undefined. Recombinant influenza protein was used as a model to isolate antibody protein interaction and to determine the effector function of antibodies on macrophage cells.

Activation of Cannabinoid (CB2) Receptors and Reactivity of Cerebral Arterioles

Lauren L. Van Hove, Denise M. Arrick, and William G. Mayhan

Division of Basic Biomedical sciences, Sanford Medical School, USD

Activation of cannabinoid (CB2) receptors has been shown to be neuroprotective, but no studies have examined whether this neuroprotection includes cerebral arterioles and whether activation of CB2 receptors can rescue cerebrovascular dysfunction during type 1 diabetes (T1D). Our goal was to test the hypothesis that administration of a CB2 agonist (JWH-133) would improve impaired dilation of cerebral arterioles during T1D. In vivo diameter of cerebral arterioles in nondiabetic and diabetic rats was measured in response to an eNOS-dependent agonist (adenosine 5'-diphosphate; ADP), an nNOS-dependent agonist (N-methyl-D-aspartate; NMDA), and an NOS-independent agonist (nitroglycerin) before and following JWH-133 (1 mg/kg IP). Dilation of arterioles to ADP and NMDA was greater in nondiabetic than in diabetic rats. Treatment with JWH-133 increased responses of cerebral arterioles to ADP and NMDA in both nondiabetic and diabetic rats. Responses of cerebral arterioles to nitroglycerin were similar between nondiabetic and diabetic rats and was not altered by JWH-133. It appears that activation of CB2 receptors can influence vascular function of cerebral arterioles during physiologic and pathophysiologic states. We speculate that treatment with CB2 receptor agonists may have potential therapeutic benefits for the treatment of cerebral vascular disease via a mechanism that can increase cerebral blood flow.

A qualitative study of the experiences of South Dakota healthcare providers with their Hutterite patients

Janice Baumberger Gerlach, Susan Anderson, Barbara Yutrzenka and Tianna Smith

Division of Basic Biomedical sciences, Sanford Medical School, USD

The primary researcher previously conducted qualitative research with Hutterite patients and found that cultural expectations affected their experiences with mainstream healthcare providers. It is unclear to what extent the providers are aware of these cultural expectations and whether these expectations affect their interactions with Hutterite patients. The present study employed the phenomenological method of qualitative research to explore this question. Eight providers participated in 30-minute interviews. The grand tour question of this study was, "What kinds of interactions have you had with Hutterite patients, and how are these interactions different from those of the general population?" The interviews were then transcribed and reviewed by trained personnel. The data analysis yielded three major themes: communication, considerations for medical service, and intervention implementation, with numerous subthemes. The Hutterite culture has presented unique experiences for healthcare providers in South Dakota that differ meaningfully from the general population. Experiences of providers with this culture have been overwhelmingly positive. By integrating the colony into medical care and increasing cultural knowledge, providers seem to have improved care for this population. This study is consistent with findings in the previous study conducted by the primary researcher describing Hutterite patients' perspectives of healthcare.

Posters

Benjamin Arbeiter: Identifying retained surgical needles using radiography.

Emma Bye: Medical student attitudes about optimal timing of interprofessional education in the medical school.

Kelly McKinght: Breaking bad news: Equipping future physicians with compassionate and empathetic communication skills.

Michael Frost: Medical Students keeping the heart of the community beating, CPR in the Rapid City area schools.

Daniel Parrott: Copper nanoparticle anti-microbial film: A solution for reducing hospital-acquired infections.

Research Programs and Opportunities for SSOM Students

The USD-SSOM Medical Student Research Program (MSRP) strives to create and support opportunities for medical students in the areas of research, service, and education. Three of our programs are described below. In addition, we can assist medical students who are not enrolled in these programs by helping find mentors and funding small projects and travel to conferences.
Contact Laura Rumohr, Program Assistant, Medical Student Research Committee (msrp@usd.edu).

The Medical Student Summer Research Program is an 8-10-week research experience for students as well as students newly accepted to the medical school. Students receive a $5,000 stipend and the hosting lab receives up to $2,000 for supplies. To apply, students choose a faculty member as a research mentor and submit an application to the Medical Student Research Committee.
For more information, please contact Laura Rumohr, Program Assistant, Medical Student Research Committee (msrp@usd.edu).

The Scholarship Pathways Program is an elective opportunity developed to enrich the medical school experience by promoting rigorous independent scholarship and scholarly excellence as well as produce leaders in medical education, research and service. The program spans all four years and develops critical thinking and independent learning skills.
For more information, please contact Candace N. Zeigler, M.D.,FACP (Candace.Zeigler@usd.edu), 605-357-1572, 1400 W. 22nd Street, Sioux Falls, SD 57105.

NIH T-35 Children's Health Innovative Research Program (CHIRP) at the Sanford School of Medicine and Sanford Research is focused on providing research training in clinical biomedical sciences to medical students. The ultimate goal of the CHIRP is to help meet the future needs of health-related research by contributing to the development of physicians who will be well-prepared to use evidence-based medicine in practice and contribute to translational research. This research program is self-paced, allowing participants to work closely with their mentors to complete 320 hours of research time during pillars 1 & 2 of their medical school curriculum. All participants receive a generous stipend. Additionally, participants are required to participate in a responsible conduct in research training program, meet routinely with their mentoring team and are encouraged to present their research at regional/national research conferences. Applications are now open for Year 2 of this competitive program. For more information and to apply, visit the Medical Student Research webpage.

All of these programs can be found at the Medical Student Research Website:
www.usd.edu/medicine/basic-biomedical-sciences/research/medical-student-research-program