MARSHALL UNIVERSITY
JOAN C. EDWARDS SCHOOL OF MEDICINE
PRESENTS

THE 33RD ANNUAL MARSHALL UNIVERSITY HEALTH SCIENCE RESEARCH DAY

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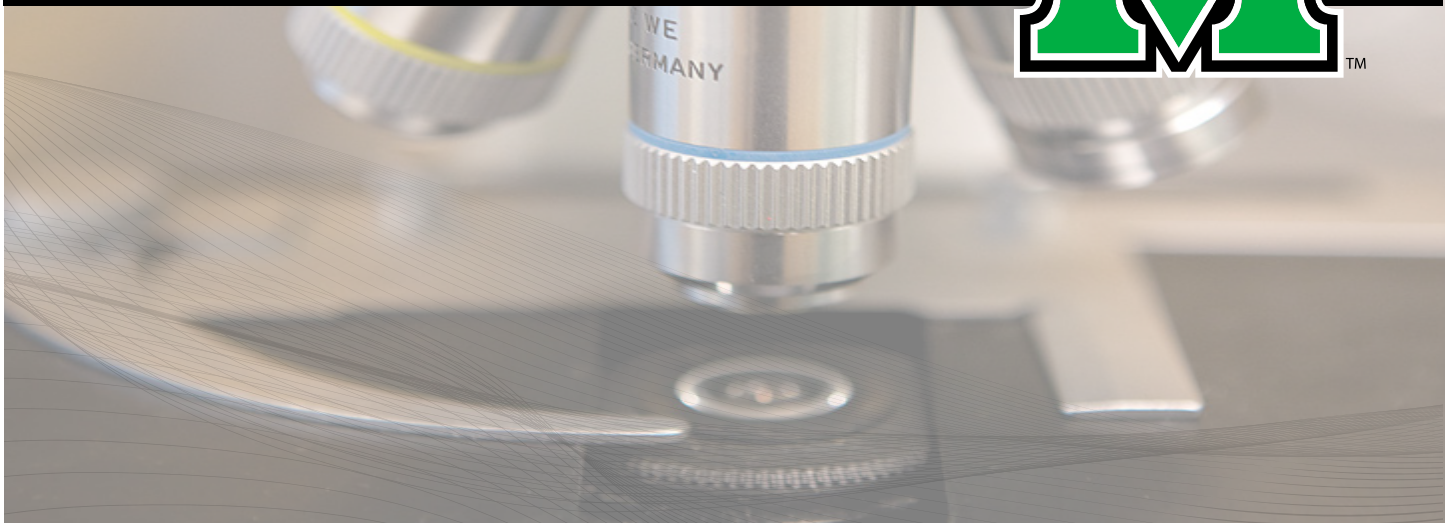
OCTOBER 29, 2021



2021 RESEARCH DAY

The conference consists of a series of oral and poster presentations by Marshall University students, residents and fellows. Please scan the QR code to view the complete syllabus and compilation of abstracts.

Specific learning objectives will be presented with each oral presentation.
Participant Q&A are encouraged throughout all sessions.
No relevant speaker conflicts are indicated as confirmed by disclosure.

This event is supported annually by educational grants from the following endowments:

- Dr. Albert C. Esposito Memorial
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Office of Research & Graduate Education

Robert C. Byrd Biotechnology Science Center
1700 Third Avenue
Huntington, WV 25755
jcesom.marshall.edu/research

Title: Updates from the National Institute of General Medical Sciences

JON R. LORSCH, PHD
Director, National Institute of General Medical Sciences

Jon R. Lorsch, Ph.D., became the director of the National Institute of General Medical Sciences (NIGMS) in August 2013.

In this position, Dr. Lorsch oversees the Institute's $2.9 billion budget, which supports basic research that increases understanding of biological processes and lays the foundation for advances in disease diagnosis, treatment, and prevention.

NIGMS supports more than 3,000 investigators and 5,000 research grants–around 11% of the total number of research grants funded by NIH as a whole. Additionally, NIGMS supports around 26% of the NRSA trainees who receive assistance from NIH.

Dr. Lorsch came to NIGMS from the Johns Hopkins University School of Medicine, where he was a professor in the Department of Biophysics and Biophysical Chemistry. He joined the Johns Hopkins faculty in 1999 and became a full professor in 2009.

A leader in RNA biology, Dr. Lorsch studies the initiation of translation, a major step in controlling how genes are expressed. When this process goes awry, viral infection, neurodegenerative diseases and cancer can result. To dissect the mechanics of translation initiation, Dr. Lorsch and collaborators developed a yeast-based system and a wide variety of biochemical and biophysical methods. The work also has led to efforts to control translation initiation through chemical reagents, such as drugs. Dr. Lorsch continues this research as a tenured investigator in the NIH's Eunice Kennedy Shriver National Institute of Child Health and Human Development.

NIGMS supported Dr. Lorsch's research from 2000-2013. He also received grants from the NIH's National Institute of Diabetes and Digestive and Kidney Diseases and National Institute of Mental Health, as well as from other funding organizations.

Dr. Lorsch is as passionate about education as he is about research. During his tenure at Johns Hopkins, he worked to reform the curricula for graduate and medical education, spearheaded the development of the Center for Innovation in Graduate Biomedical Education, and launched a program offering summer research experiences to local high school students, many from groups that are underrepresented in the biomedical sciences. In addition, he advised dozens of undergraduate and graduate students and postdoctoral fellows.

Dr. Lorsch received a B.A. in chemistry from Swarthmore College in 1990 and a Ph.D. in biochemistry from Harvard University in 1995, where he worked in the laboratory of Jack Szostak, PhD. He conducted postdoctoral research at Stanford University in the laboratory of Daniel Herschlag, PhD.

Dr. Lorsch is the author of more than 80 peer-reviewed research articles, book chapters, and other papers. He has also been the editor of six volumes of Methods in Enzymology and has been a reviewer for numerous scientific journals. He is the author on two awarded U.S. patents. His honors include six teaching awards from Johns Hopkins.

Dr. Lorsch's other activities have included membership on the American Society for Biochemistry and Molecular Biology's mentoring committee, the RNA Society's board of directors and NIH review committees.

Since joining NIH, he has taken on several leadership roles, including serving on the NIH Scientific Data Council, Administrative Data Council and Extramural Activities Working Group, which he co-chairs.

SCHEDULE OF EVENTS

7 a.m. Registration opens for morning and afternoon participants. Registration will remain open throughout the day.

7:45 a.m. **Welcome & Opening Remarks**

Uma Sundaram, MD
Vice Dean and Research Day Chair

Joseph I. Shapiro, MD
Vice President & Dean, Joan C. Edwards School of Medicine

8 a.m. to Noon **Poster Presentations Session 1: Basic Sciences**
See page 5.

8 a.m. **Oral Presentations Session 1**
Session Chair: Nancy Norton, MD

Time	Name	Abstract Title
8 a.m.	Christopher Walker	Adolescent binge ethanol exposure results in long-term changes in astrocyte morphology, the disruption astrocyte-synaptic colocalization, and the impairment of neuronal-astrocyte communication
8:15 a.m.	Mary Piscura	S426/S430 phosphorylation accounts for Beta-arrestin 2-mediated desensitization for cannabinoid sensitivity and tolerance in mutant mice
8:30 a.m.	Caleb Clark	Design of a Switching Molecular Beacon for the Detection of the Circular RNA circHOMER1_a in Cultured Neurons
8:45 a.m.	Skylar Cooper	Sex-specific effects of e-cigarette flavorants on addiction-related behavior and nicotinic receptor density in a rodent model of vapor self-administration
9 a.m.	Caroline Putnam	Loss of IRGM1 expression in the pathogenesis of myelodysplastic syndromes phenotypes in mice.
9:15 a.m.	Christian Harris	The intersection of alcohol, glia and behavior: Can astrocytes influence anxiety-like and reward-motivated behavior following adolescent ethanol exposure?

9:30 a.m. **Break**

9:45 a.m. **Introduction of Keynote Speaker**

Uma Sundaram, MD

Avinandan Mukherjee, PhD
Interim Provost & Senior Vice President for Academic Affairs, Marshall University

10 a.m. **Keynote Address – "Updates from the National Institute of General Medical Sciences"**
Jon R. Lorsch, PhD
Director, National Institute of General Medical Sciences

11 a.m. **Announcements**

Uma Sundaram, MD

Jerome A. Gilbert, PhD
President, Marshall University

11:15 a.m. **Oral Presentations Session 2**
Session Chair: Beverly Delidow, PhD

Time	Name	Abstract Title
11:15 a.m.	Jesse Lewis	The COVID-19 Effect: Severity and Type of Parent Reported Stress in Pediatric Patients with Attention Deficit Hyperactivity Disorder During 2020 compared to 2018
11:30 a.m.	Baylor Blickenstaf	Onodera's prognostic nutritional index a valuable measure in predicting complications after TKA
11:45 a.m.	Caroline Briggs	The effect of elevated blood pressure and stage one hypertension on perinatal outcomes
12:00 noon.	Lauren Hanna	Impact of SARS-CoV-2 Pandemic on Pediatric Viral Respiratory Infections

12:15 to 1:15 p.m. **Box Lunch**

12 to 3 p.m. **Poster Presentations Session 2: Clinical Science**
See page 7.

1:15 p.m. **Oral Presentations Session 3**
Session Chair: Richard Egleton, PhD

Time	Name	Abstract Title
1:15 p.m.	Shanmuga Sundaram	Intestinal bile acid absorption is upregulated in a diet induced rat model of Obesity associated dyslipidemia
1:30 p.m.	Jinju Wang	Compromised endothelial progenitor cell exosomal communication with endothelial cells in hypertensionischemia conditions
1:45 p.m.	Oliver Li	Na+/K+ ATPase alpha 1 subunit participates in platelet signaling and is a potential anti-thrombotic target
2:30 p.m.	Niraj Nepal	Nutrient-induced incretin hormone secretion by enteroendocrine cells is altered in obesity
2 :15p.m.	Jessica Wellman	Upregulated expression of phosphorylated N-Myc Downstream Regulated 1 (NDRG1) in clear cell renal cell carcinoma
2:30 p.m.	Ashley Cox	E-Cigarette Flavoring Agents Alter Mitochondrial Function and Autophagy Pathways in Human Kidney HK-2 Cells

2:45 p.m. **Oral Presentations Session 4**
Session Chair: James Denvir, PhD

Time	Name	Abstract Title
2:45 p.m.	Mariah Morris	Quasi-experimental evaluation of liposomal bupivacaine (Exparel®) on inpatient opioid consumption in patient undergoing bariatric surgery
3:00 p.m.	Ella Boggs	Regional Analysis of Cervical Spine Fractures in Patients 65 years and Older
3:15 p.m.	Lauren Morilla	Methamphetamines and the Effects on the Heart
3:30 p.m.	Darren Hylton	The effects of adolescent intermittent binge ethanol exposure on hippocampal astrocyte morphology and synaptic colocalization
3:45 p.m.	Jonathan Lash	What Is the Incidence of in Hospital Mortality After Hip Fracture? Does Treatment Options Matter?

3 to 4 p.m. **Poster Presentations Session 3: Case Reports**
See page 9.

4:30 p.m. **Presentation of Winners**

5 p.m. **Adjournment**

Evaluation Form

Scan the QR code to complete the 2021 Research Day evaluation form.
Your input will assist in the planning of future events.

SCHEDULE OF EVENTS

Poster Presentations Session 1 • Basic Sciences

8 a.m. to Noon

No.	Name/Poster Title
1	**Vijaya Sundaram** Adipose Derived Secretome Mediates the Regulation of SGLT1 in Rat Small Intestinal Epithelial Cells
2	**Renat Roytenberg** Thymidine Phosphorylase could be a novel therapeutic target for treating COVID-19
3	**Jared Youther** Effect of Cidec gene polymorphisms on obesity susceptibility in mice
4	**Holly Cyphert** The role of metabolic dysfunction in SARS-CoV-2 progression
5	**Harshal Sawant** The profile of extracellular vesicles in intracerebral hemorrhage patients
6	**Cora Miracle** The role of obese adipose derived secretome in colon cancer pathogenesis
7	**Heba Boustany** Influence of diet fat content on ethanol metabolism and PPAR-α-regulated lipid metabolism
8	**Jentre Hyde** Diverging effects of adolescent intermittent ethanol exposure on astrocyte morphology and synaptic proximity in prefrontal cortex subregions
9	**Hasitha Chavva** Sex-Dependent Changes in Vascular Function of Adult Rats Following Prenatal Exposure to Methamphetamine
10	**LaTaijah Crawford** Antinociceptive Effects of Decursinol on Cisplatin-Induced Chronic Neuropathic Pain
11	**Justin Merritt** The Synthetic Capsaicin-analog Arvanil sensitizes Cisplatin-Resistant Human Lung Cancer Cells to the pro-apoptotic activity of Irinotecan
12	**Cassaundra White** High-fat diet alters body composition and serum cytokines without affecting body mass
13	**Lauren Brooke Clower** Transformative Role of Endometriotic Milieu in Ovarian Cancer
14	**Morgan Brooke Elmore & Zach Mitchell** Sex-specific effects of green apple e-cigarette flavor on α4- and α6-containing nicotinic receptors of dentate gyrus and habenula neurons in a mouse model

15	**Pradeep Rajan** The α-1Na/K-atpase signalosome rescinded epigenetic changes in the progression of NASH-associated liver carcinogenesis
16	**Courtney Lulek** Role of Strain in Mediating Sex-Differences in Acute Cannabinoid Response and Tolerance
17	**Fatih Koc** Remodeling of Mitochondrial Energy Metabolism in Clear Cell Renal Cell Carcinoma
18	**Kristiana Sklioutovskaya-Lopez** Expression of cerebellar genes related to inflammation, insulin resistance, glial differentiation, and development in a mouse model of obesity
19	**Udoh Utibe Abasi Sunday** Tumor-Suppressor Role of the Caveolar α1-Na/K-ATPase Signalosome in NASH related Hepatocellular Carcinoma
20	**John Pickstone** Nephrotoxic Potential of Three Dichlorobenzene Isomers in Isolated Kidney Cells from Female Fischer 344 Rats
21	**Lady Kesler** Novel Protocol for Dual Immunofluorescence of Growth Plate Cartilage
22	**Autumn Pennington** Sex, Age, and Exercise Effects on Bone Density and Muscle Mass in Rats
23	**Meredith Kesler** Western Diet Accelerates Initiation and Progression of Myelodysplastic Syndrome in Mice
24	**Ashley Dague** Antimicrobial activities of secondary metabolites from model bryophites
25	**Travis Stevens** microRNAs as Potential Biomarkers for Drug-Induced Liver Damage
26	**Irina Kukharskaya** Impact of HSP90 inhibition on epigenetic drift in hematopoietic stem cells
27	**Lauren Morilla** Methamphetamines and the Effects on the Heart
28	**Levi Nolan and Joshua Keefer** The Association of the α1-Na/K-ATPase signalosome with Chaperone Mediated Autophagy in NASH related HCC
29	**Samuel Tetteh-Quarshie** The Effect of Continuous Sugar-Sweetened Beverage Consumption on Weight in Mice and Adipokine Expression in Preadipocyte Cells
30	**Anisha Valluri** Proteomic analysis provides evidence for metabolic reprogramming in clear cell renal cell carcinoma (ccRCC)

31	**Cecilia Sierra-Bakhshi** Salmonella Infection in Diabetic Mice
32	**Kailey Stuart** Tissue-specific reduction of Na/K-ATPase in mice uncovers a new mechanism of regulation of sodium balance and systemic blood pressure by the renal proximal tubule.
33	**Jared Mattingly** The Promise of Exosome-Based Therapy for Central Nervous System Diseases
34	**Sasha Zill** Sensing force dynamics is common to control of walking in humans, insects and robots

Poster Presentations Session 2 • Clinical Science

Noon to 3 p.m.

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35	**Abigail Samson & Mackenzie Bergeron** The effect of umbilical cord essential and toxic elements, thyroid levels, and Vitamin D on childhood development
36	**Adeoluwa Adeluola** Low dose Actinomycin D Predominantly Activates p53-p21 Pathway in Aerodigestive Tract Cancers
37	**Alec McCann** Synovial fluid absolute neutrophil count a promising marker for diagnosing periprosthetic joint infection
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39	**Andrew Ferguson** Assessment of Elements Affecting Nutritional Status in Elderly West Virginians
40	**Zainab Saeed** Parental Attitudes on Epinephrine Use during COVID-19 Pandemic
41	**Blass Morrone** Identifying Factors that Predict Positive Testing during a Pandemic in a Division I Sports Medicine Program
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43	**Danielle Roth** Use of a web-based sex education tool to improve reproductive health knowledge
44	**Dylan Smith** Operative Rib Fixation at a Rural Trauma Center: A Single Institution Retrospective Review
45	**Erin Shaver** Effects of Increased Opioid Dosage for Geriatric Hip Fracture Patients in an Inpatient Rehabilitation Setting

46	**Gavin Hayes** Effect of exogenous testosterone use on factors contributing to increased intraocular pressure and open angle Glaucoma
47	**Graham Sutherland** Retrospective Report of Antibiotic Resistance in Laboratory-Confirmed Bloodstream Infections in a Peruvian childrens' hospital
48	**Jaineet Chhabra** Perception of Virtual Interviews in Vascular Surgery Fellowship Application
49	**Jessica Tall** Early Detection for Women's Cancer: Disparities and Prevention in Vulnerable Women
50	**Jodi Plumley** Flu Vaccine Administration before and after COVID-19
51	**Karagan Mulhall** Improving LARC Use in Women with Substance Use Disorder
52	**Kassandra Flores** Adverse Outcomes of Intra-Articular and Peri-Tendon Corticosteroid Injections
53	**Kelly Melvin** COVID-19 Impact on Hospital Presentation of Adolescent Suicide Ideation and Attempts
54	**Kennedy Snavely** A descriptive study assessing temporal changes in infective endocarditis in Huntington, West Virginia
55	**Lauren Clower** Improving Pneumococcal Vaccination Rates Among Rheumatoid Arthritis Patients of the Rural Appalachian Population in an Academic Rheumatology Clinic
56	**Matthew Nudelman** Duration of Mother's Own Milk Feeding in Infants Post NICU Discharge and Maternal Perception of Reasons for Discontinuation
57	**Melissa Nehls** Complications and Hospital Admissions Among Pregnant Women with Substance Abuse
58	**Mohamed Tashani** Transitioning To Unfractionated Heparin In Treatment of NSTEMI patients on Direct Oral Anti-Xa Inhibitors
59	**Muhammad Jafary** Increased risk of type 2 diabetes in Appalachian patients with bipolar disorder after exposure to antipsychotic medications: a Cox Proportional hazard method
60	**Nana Bosomtwe** Neonatal Abstinence Syndrome (NAS) treated inborns at Cabell Huntington Hospital (CHH) in the SARSCoV2 pandemic era

61	**Rodrigo Aguilar** Sudden Cardiac Death in Heart Failure with preserved Ejection as compared to Heart Failure with reduced Ejection Fraction: A Nationwide Analysis
62	**Rodrigo Aguilar** In-hospital mortality of transcatheter versus surgical mitral valve replacement: A Nationwide Analysis
63	**Ron Carico Jr.** Evaluation of a pharmacy technician-based medication prior authorization program
64	**Ryan Vaught** Understanding biomarker expression in association with graded recoil analysis following shooting
65	**Seth Bergeron** Assessing Factors that Impact COVID Testing Rates in a Regional Testing Center
66	**Shadi Bashai** Impact of Setting Chapter Quizzes Benchmarks in a Capstone course on Exam Performance
67	**Shane Taylor** Risk Factors for Arthrofibrosis and Primary and Revision Total Knee Arthroplasty: A Multicenter Study
68	**Shelby Wellman** Improving LARC Use in Women with Substance Use Disorder
69	**Willie Kimler** Impacting Patient Perception of Medical Care and Communication in a Patient Centered Medical Home through an Informative Brochure
70	**Thomas McIntosh** Weight Loss in Community-living older adults during the Covid-19 pandemic
71	**Tristan Burgess** Coronary Artery Disease Status Modulates Non-Coding RNA Expression in Epicardial Fat
72	**Ty Bayliss** The Case for Using Composition Tomography to evaluate Perirectal Necrotizing Fasciitis: Is It Really Necessary?
73	**Vishavpreet Singh** The Utility of Leukocyte Esterase Test In Diagnosing Culture Negative Periprosthetic Joint Infections
74	**Wade Smith** Serum ESR and CRP are not reliable markers for screening/diagnosing PJI

Poster Presentations Session 3 • Case Reports

3 to 4 p.m.

List of Presenters' Abstracts
Oral Presentations

No relevant conflicts of interest as supported by disclosure.

List of Presenters' Abstracts
Poster Presentations

No relevant conflicts of interest as supported by disclosure.

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Adolescent binge ethanol exposure results in long-term changes in astrocyte morphology, the disruption astrocyte-synaptic colocalization, and the impairment of neuronal-astrocyte communication

Authors: C.D. Walker, H. Sexton, B.J. Henderson, M-L Risher

Department & Institution: Joan C. Edwards School of Medicine Marshall University, Biomedical Research, Huntington, WV 25701. Hershel 'Woody' Williams Veterans Affairs Medical Center, Huntington, WV 25704.

Background: Alcohol consumption, particularly binge drinking, is highly prevalent among adolescents, increasing the risk of long-term cognitive impairment. Animal models have shown neuronal structure and function disruption due to repeated binge-level ethanol (EtOH) exposure. However, the underlying mechanisms that elicit these changes are not well understood. Furthermore, how astrocytes contribute to EtOH-induced neuronal dysfunction is unknown. Astrocytes have complex morphologies with extensive perisynaptic astrocyte processes (PAPs) that ensheathe pre- and post-synaptic terminals and play an essential role in synaptic maintenance and signal transmission. Here we investigate the consequences of adolescent binge EtOH exposure (AIE) on astrocyte morphology, PAP-synaptic proximity, and neuronal-to-astrocyte communication in the form of astrocyte Ca2+ activity.

Study Hypothesis: AIE induces changes in astrocyte morphology that disrupt the PAP-synaptic colocalization, subsequently impairing astrocyte responsivity to neuronal activation.

Methods: Male Sprague Dawley rats received hippocampal intracranial injections of an astrocyte-specific adenoassociated virus expressing green fluorescent protein (GFP) or an astrocyte-specific Ca2+ indicator (GCaMP6f). Beginning post-natal day 30, animals received intermittent 5g/kg EtOH or water gavage over 16 days, followed by a 26-day forced abstinance period. PND72 hippocampal slices underwent immunohistochemistry (IHC, astrocyte morphology) or electrophysiology (neuronal-astrocyte responsivity). IHC was performed on GFP+ slices using the postsynaptic density marker PSD95. GFP+ astrocytes in the CA1 hippocampus were imaged using confocal microscopy and reconstructed using IMARIS-Bitplane. Astrocyte Ca2+ activity was recorded from GCaMP6f+ CA1 hippocampal astrocytes following Schaffer Collateral stimulation.

Results: Our data demonstrate that AIE results in a protracted decrease in astrocyte volume and PAP-synaptic colocalization that correlates with decreased astrocyte responsivity to neuronal activation as demonstrated by a reduction in astrocyte Ca2+ activity.

Conclusion: These findings contribute to our understanding of how adolescent EtOH disrupts neuronal-astrocyte communication, driving neuronal dysfunction that persists into adulthood.

S426/S430 phosphorylation accounts for Beta-arrestin 2-mediated desensitization for cannabinoid sensitivity and tolerance in mutant mice

Authors: Mary Piscura, Angela Henderson-Redmond, Diana Sepulveda, and Daniel Morgan

Department & Institution: Department of Biomedical Sciences, Joan C. Edwards School of Medicine, Huntington, WV

Desensitization is mediated via phosphorylation of residues S426 and S430 by a G protein-coupled receptor kinase (GRK). Following GRK-mediated phosphorylation at these residues, beta-arrestin 2 (bArr2) is recruited to facilitate desensitization of the receptor. Previous work demonstrated that tolerance for cannabinoid drugs was reduced in S426A/S430A mutant mice and in bArr2-KO mice. Since there are other phosphorylatable serine and threonine residues in the C-terminal tail of CB1R, it was not clear whether bArr2 recruitment to S426 and S430 accounts for all bArr2-mediated desensitization.

To address this question, we assessed cannabinoid acute response and daily tolerance in S426A/S430A x bArr2-KO double mutant mice. Both S426A/S430A and bArr2-KO single mutant mice display increased sensitivity and delayed tolerance to the antinociceptive effects of delta-9-tetrahydrocannabinol (Δ9-THC) and the synthetic cannabinoid, CP55,940. Double mutant mice did not differ from the S426A/S430A single mutant

model in respect to antinociception, suggesting that the effect of complete bArr2 deletion did not enhance the effect of the S426A/S430A point mutants. However, the hypothermic effects of acute Δ9-THC dosing were increased in male double mutant mice, relative to S426A/S430A and bArr2 single mutant mice.

These results indicate that phosphorylation of S426 and S430 are likely responsible for bArr2-mediated desensitization, with the exception of hypothermia induced by acute Δ9-THC dosing. Therefore, these results also suggest cannabinoid-induced hypothermia is dependent on the agonist (Δ9-THC or CP55,940) and duration of exposure (acute sensitivity vs tolerance).

Design of a Switching Molecular Beacon for the Detection of the Circular RNA circHOMER1_a in Cultured Neurons

Authors: Caleb Clark, Nadja Spitzer, David Neff, Michael Norton

Department & Institution: Caleb Clark: Joan C. Edwards School of Medicine, Nadja Spitzer: Department of Biology, Marshall University, David Neff and Michael Norton: Department of Chemistry, Marshall University

Background: HOMER1 is a well-known scaffolding protein that exists in two isoforms labeled HOMER1a and HOMER1b/c. The gene encoding for HOMER-1 produces mRNA transcripts that can undergo alternative splicing to produce a circular RNA by the name of circHOMER1_a, which has been shown to be the circular RNA most highly correlated with the development of Alzheimer's Disease.

Study Hypothesis: The Molecular and Biological Imaging Center at Marshall University hosts a microscope equipped to image samples concurrently via Atomic Force Microscopy (AFM) and Fluorescence Microscopy. Within this study, a molecular beacon intended to hybridize only to the circular RNA circ_HOMER1_a, was designed for in vivo imaging via AFM and Florescence Microscopy, demonstrating the potential of molecular beacons as a tool for single molecule detection in vivo.

Methods: Multiple versions of a molecular beacon were designed and ordered from Integrated DNA Technologies (IDT). Fluorescence spectroscopy was performed to analyze the capabilities of the molecular beacon under ideal circumstances, to allow optimization before in vivo testing. Fluorescence spectroscopy was performed so that molecular beacon designs could be tested for sensitivity and specificity to target DNA.

Results: The first version of the molecular beacon design did not provide adequate switching of fluorescent signal to be utilized as an intracellular probe. The molecular beacon sequence was then modified, and a subsequent version was shown to be specific to the target sequence and sensitive enough for target detection.

Conclusion: Version 2 of the molecular beacon design has demonstrated properties during extracellular testing that make it a promising intracellular probe for circHOMER1_a. At this time, the B35 cell line is being cultured so that the molecular beacon can be transfected for in vivo testing.

Sex-specific effects of e-cigarette flavorants on addiction-related behavior and nicotinic receptor density in a rodent model of vapor self-administration

Authors: Skylar Y. Cooper and Brandon J. Henderson

Department & Institution: Department of Biomedical Research, Joan C. Edwards School of Medicine, Marshall University

Background: Vaping remains prevalent among adolescents (4 million) and adults (10 million), with >70% of users preferring flavored products. This remains a concern considering the 15,000+ unique flavor options available. Based on previous findings that e-cigarette flavors are not simply additive, but instead, promote changes in neurobiology and neurophysiology, our goal is to identify green apple (GA) flavorant-specific effects on intracellular nAChR trafficking and assembly, midbrain neuron firing, and vaping-related behaviors.

Study Hypothesis: We hypothesize that popular GA flavored e-cigarettes (without nicotine) elicit changes in nAChR density and VTA dopamine neuron firing, and thus, enhance mesolimbic dopamine release that drives addiction-related behavior.

Methods: We utilized vapor self-administration assays with mice (genetically modified to contain fluorescent nAChRs) to study how GA flavorants (hexyl acetate, ethyl acetate, and methylbutyl acetate, and the combination of the three at a 3:1:1 ratio, respectively (GA-mix)) alter

vaping-related behaviors. Mice were trained to self-administer on fixed-ratio 1 (FR1) and FR3 schedules to measure acquisition and reinforcement-related behaviors. An additional cohort of mice were subjected to 10-day passive exposure of GA-mix or PGVG-control, and brains were extracted to determine nAChR density changes using confocal microscopy or to measure neuronal firing and dopamine release through electrophysiology and electrochemistry. Statistical differences were determined using one- and two-way ANOVAs with post hoc Tukey means comparisons.

Results: Male mice exhibited addiction-related behaviors to particular flavorants while female mice exhibited addiction-related behavior to all GA flavorants. We also observed that GA-mix vapor exposure induced nAChR upregulation in a region-specific manner dependent upon sex. GA-mix enhanced VTA dopamine neuron firing and subsequent DA release in the nucleus accumbens.

Conclusion: These GA-induced, and sex-dependent, behavioral and cellular changes display the importance of investigations into sex-dependent effects of e-cigarette flavors on vaping-related behaviors.

Loss of IRGM1 expression in the pathogenesis of myelodysplastic syndromes phenotypes in mice.

Authors: Caroline M. Putnam, Meredith B.A. Kesler, Lahari Kondeti, Melinda E. Varney

Department & Institution: Pharmaceutical Sciences and Research, Marshall University School of Pharmacy

Background: IRGM is a p47 immunity related GTPase. Its genetic location is on human chromosome 5q33.1. Deletions in this area of chromosome 5q are the most common cytogenetic aberrations in myelodysplastic syndromes (MDS). While IRGM and its mouse ortholog, Irgm1 have been studied for protective functions in hematopoietic stem and progenitor cells, its loss has not been studied as a potential contributor to del5q MDS.

Study Hypothesis: We hypothesize that Irgm1 loss contributes to overactive innate immune signaling and inflammation in hematopoietic stem and progenitor cells and that this loss ultimately leads to MDS-like phenotypes in mice. Furthermore, we hypothesize that current mouse models of MDS will be enhanced by inclusion of Irgm1 loss

Methods: First, we treated wild type and Irgm1 deficient bone marrow cells with empty vehicle or lipopolysaccharide (LPS) to determine if alterations in gene expression associated with innate immune signaling and inflammation occur. Additionally, we have treated wild type and Irgm1 deficient mice with LPS in vivo to determine if mice exhibit alterations in hematopoietic stem and progenitor cell frequency, spleen size, or blood composition which often occurs in MDS. Lastly, we have evaluated Irgm1 expression in a del5q MDS model to determine if inclusion of Irgm1 loss will more closely mimic human disease.

Results: We have found that in the presence of LPS, Irgm loss contributes to altered innate immune signaling and inflammation in the bone marrow of mice. Furthermore, we have determined that Irgm expression is upregulated in current del5q MDS mouse models.

Conclusion: Further studies will investigate a novel mouse model of del5q MDS in which combined loss of Irgm and two additional chromosome 5q immune regulation genes, Tifab and miR-146a, contribute to MDS pathogenesis.

The intersection of alcohol, glia and behavior: Can astrocytes influence anxiety-like and reward-motivated behavior following adolescent ethanol exposure?

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Background: Early onset binge drinking increases the likelihood of developing an alcohol use disorder later in life. However, the underlying cellular mechanisms that drive these changes are not well understood. Recent work has implicated neuroimmune glial processes in the development of alcohol-induced changes in brain function; however, the relationship between

neuroimmune activation and the development of sensitivity to addictive substances remains poorly understood. Previous work in our laboratory and others have demonstrated that microglia and astrocytes display stereotypical reactive phenotypes in response to repeated ethanol exposure. Of particular interest are interleukin 1 alpha (IL-1α), tumor necrosis factor alpha (TNF-α), and complement component 1q (C1q); proteins involved in the induction of astrocyte reactivity. Here we investigated the role of this pathway in reward-sensitivity and anxiety-like behavior following repeated adolescent EtOH exposure.

Study Hypothesis: Exposure to alcohol induces increased anxiety and morphometric changes in astrocytes that are dependent on astrocyte activation via TNFα, IL1α, and C1q.

Methods: Postnatal day (PND) 30 male and female C57BL/6J mice and triple knockout mice (3KO, ~ TNFα, C1q, IL1α) received ethanol (EtOH, 5.0 g/kg i.g.) or water intermittently over 16 days. Following a 7-day washout period mice underwent a battery of behavioral tests consisting of open field, light-dark box, and conditioned place preference (CPP) to test locomotor activity, anxiety, and reward sensitivity for nicotine, respectively.

Results: 3KO+EtOH female mice demonstrated less anxiety than C57BL/6J+EtOH females. Male 3KO+EtOH mice showed greater preference for nicotine compared to C57BL/6J+EtOH mice, whereas female 3KO's had greater preference for nicotine irrespective of water/EtOH exposure when compared to female C57BL/6J mice.

Conclusion: These data show unique genotype differences in the sex-dependent regulation of anxiety-like behavior and reward-sensitivity following binge EtOH exposure. Future work will investigate region-specific cellular and molecular changes which may further the current understanding of the role of neuroimmune glial cells in addiction.

The COVID-19 Effect: Severity and Type of Parent Reported Stress in Pediatric Patients with Attention Deficit Hyperactivity Disorder (ADHD) During 2020 When Compared to 2018

Authors: Jesse Lewis, Katina Nicoloudakis, James Lewis, Deborah Preston

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Background: Increased parental stress levels have been found to be higher in families whose children have ADHD and are directly related to the severity and frequency of child behavioral problems. The adverse effect of environmental factors on the severity and types of stress for these parents has not been studied.

Study Hypothesis: To determine if the level and etiology of stress reported by parent of pediatric patients with ADHD during the 2020 COVID-19 pandemic is elevated when compared to similar parent obtained data in 2018

Methods: During 2018 and 2020, two groups of parents of children diagnosed and treated by one behavioral pediatrician for ADHD according to current American Academy of Pediatrics (AAP) guidelines completed the Parenting Stress Index-Short Form (PSI-4-SF). The test measures total parental stress (TPS) and the 3 subdomains of parental distress (PD), parent-child dysfunctional interaction (PCDI), and difficult child (DC). In addition, both food and housing insecurity (FI,HI) were evaluated in the 2020 population using AAP approved screening tools.

Results: Both the mean scores and corresponding percentiles of COVID-19 group (n=46) TPS and the 2 subdomains of stress associated primarily with the parental distress (PD) and the parent and child dysfunctional interaction (PCDI) were statistically significantly higher according to the Wilcoxon-MannWhitney two sample t-test (p-value <0.05) when compared to the 2018 group (n=157). There was no statistically significant differences, however, in both the mean scores and corresponding percentiles of the difficult child CD) domain (Table). Both FI and HI were found in 8.7% of the COVID-19 group.

Conclusion: External factors, specifically the COVID-19 pandemic, can significantly increase TPS including both the subdomains of PD and PCDI, while interestingly sparing child-based stress. Environmental concerns, including FI and HI, should also be considered in assessment and management of children with ADHD.

Onodera's prognostic nutritional index a valuable measure in predicting complications after TKA

Authors: Baylor Blickenstaf, Alisina Shahi, Matthew Bullock, Ali Oliashirazi

Department & Institution: Marshall Orthopaedics

Background: The best marker for assessing nutritional status prior to total knee arthroplasty (TKA) remains unknown.

Study Hypothesis: The purpose of this study was to investigate the utility of Onodera's prognostic nutritional index (OPNI) in predicting early complications following TKA, and to determine the threshold above which the risk of complications increase significantly.

Methods: This prospective multi-center study evaluated primary TKAs. The OPNI was measured in patients within 14 days of surgery. Complications were assessed for 12 weeks from surgery and included prosthetic joint infection (PJI), wound complications, re-admission, and re-operation. The Youden's index was used to determine the cut-off for OPNI and albumin. Multiple regression model was also performed using the Charlson comorbidity index to compare the outcomes using OPNI and albumin levels as independent variables.

Results: Overall, 1325 patients (562 males, 763 females) were included in the study. OPNI cutoff score of 45.1 was determined as the optimal threshold associated with complications. Patients with lower OPNI (<45.1) were 9.8 times more likely to develop PJI compared to patients with higher OPNI (p=0.001). Re-admission and re-operation rates were 4.6 and 4.2 times higher in patients with OPNI below the threshold (p = 0.017 and p = 0.005, respectively). These complications remained statistically significant in multiple regression analysis. Unlike OPNI, albumin failed to show a significant association with complications (cutoff: 38.2 g/L).

Conclusion: OPNI is a valid and an excellent predictor of complications following TKA. It better reflects the nutritional status, has greater predictive power for complications, and can determine whether the body is in anabolic or catabolic status. Based on these findings, we recommend screening of all patients undergoing TKA using OPNI and for those who have a score lower than 45.1 the risk of surgery should be carefully weighed against its benefit and consider nutritional optimization.

The effect of elevated blood pressure and stage one hypertension on perinatal outcomes

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Background: There is literature in the obstetric population that suggests any degree of hypertension can lead to adverse maternal and fetal outcomes.

Study Hypothesis: We hypothesized that pregnant patients with elevated blood pressure have increased risk of preeclampsia compared to patients with no hypertension.

Methods: A retrospective chart review was performed on patients who received obstetric care with the Marshall University OB/GYN department and delivered at Cabell Huntington Hospital. Delivery outcomes were obtained for 218 deliveries from March 30, 2020 to September 30, 2020 in patients who did not meet obstetric criteria for chronic hypertension at the initial prenatal visit in the first trimester. Patients were divided into three groups: normotensive with blood pressure <120 systolic or <80 diastolic, elevated blood pressure of 120-129 systolic and <80 diastolic, or stage 1 hypertension of 130-139 systolic or 80-89 diastolic. Maternal demographics and pregnancy outcomes were compared.

Results: There were 75 normotensive patients (group 1), 60 patients with elevated blood pressure (group 2), and 83 patients with stage 1 hypertension (group 3). There was no difference in patient age, mode of delivery, or 5-minute Apgar. Birth weight (p=<.01) and gestational age at delivery (p=<.01) were significantly lower with increasing blood pressure. There was also a difference in BMI (p=<.01) among groups, with elevated blood pressures associated with higher BMI. The incidence of preeclampsia was positively correlated with hypertensive category, 15% for group 1, 20% for group 2, 25% for group 3 (p=<.01).

Conclusion: Elevated blood pressure in the first trimester increases the risk of preeclampsia and early delivery in Appalachian gravidas. Pregnant patients who do not meet obstetric criteria for chronic hypertension but have elevated blood pressure are at risk for adverse outcomes. Strategies directed at individuals with elevated blood pressures are needed to improve both maternal and fetal outcomes.

Impact of SARS-CoV-2 Pandemic on Pediatric Viral Respiratory Infections

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Background: Each year, pediatric viral respiratory infections (VRIs) have significant effects on the healthcare system, resulting in hundreds of pediatric deaths and accounting for a high proportion of hospital visits. Social distancing guidelines and mask mandates implemented at the start of the SARS-CoV-2 pandemic were put in place to reduce the spread of the virus. Marshall pediatricians have noticed that these guidelines being left in place during standard respiratory season have also caused a noticeable reduction from previous seasons in common pediatric viral respiratory infections that had yet to be quantified.

Study Hypothesis: We hypothesize that SARS-CoV-2 social distancing guidelines and mask mandates resulted in a quantifiable reduction of common pediatric VRIs during the 2020-2021 respiratory season.

Methods: Data was obtained on all nasopharyngeal viral polymerase chain reaction (PCR) tests administered to pediatric patients who were seen at Cabell Huntington Hospital. These tests include 7 major viruses with several subtypes. Additionally, Sars-COV-2 specific nasopharyngeal test results were obtained. Patient population included those birth to 18 years of age. Test results were used to analyze the prevalence of pediatric upper respiratory viruses during months with typical peak prevalence (October 1st to March 31st) in the past 5 years (2016-2021).

Results: Most viral cases displayed consistent positivity between 2016-2020, with the 2019-2020 season having peak total cases for nearly all viruses. The 2020-2021 season, on the other hand, saw a marked reduction in nearly all viral cases. Most notable was the reduction in RSV cases, with 399 cases in the 2019-2020 season, and 1 in the 2020-2021 season.

Conclusion: Social distancing and mask mandates have shown a significant reduction in pediatric VRIs during the most recent respiratory season. Further investigation into current VRI rates with lifted mandates would prove even more impactful and could influence future public health decisions for our area children.

Intestinal bile acid absorption is upregulated in a diet induced rat model of obesity associated dyslipidemia

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Department & Institution: Clinical and Translational Sciences

Background: Altered lipid homeostasis leading to dyslipidemia is central to the pathogenesis of obesity-related morbidities. Bile acids that aid in dietary lipid absorption in the intestine are known to be altered in obesity. Studies conducted in a monogenic rat model (Zucker rat) of obesity and in obese humans demonstrated that apical sodium-dependent bile acid cotransporter (ASBT; SLC10A2), the sole intestinal mediator of bile acid absorption was significantly increased in ileal villus cells, thus establishing itself as a potential participant in the pathogenesis of obesity associated dyslipidemia. However, how ASBT may be regulated in a rat model of diet induced obesity (DIO) is unknown.

Study Hypothesis: ASBT mediated bile acid absorption is uniquely regulated in rat DIO.

Methods: Sprague Dawley (SD) rats fed with high fat diet (HFD; 60% calories from fat) for 6 wks. served as DIO model with normal chow fed animals as controls. Na-dependent 3H-taurocholate uptake was performed to determine ASBT activity.

Results: HFD rats showed increased plasma levels of cholesterol and triglycerides (cholesterol 61 ± 4 mg/dl in control and 86 6 in HFD; n=3, $p<0.05$. Triglycerides 94±1.5 mg/dl in control and 147±26 in HFD; n=4, $p<0.05$) confirming dyslipidemia. ASBT activity was increased in intact villus cells from HFD animals (8.8±3.5 nmol/mg pro/min in normal and 22.7±3 in DIO; n=3, $p<0.05$). Na/K-ATPase activity that affects

ASBT function at the intact cell level, was found to be reduced (22.3±0.4 nmol/mg pro/min in normal and 14±0.4 in DIO; n=4, $p<0.0001$). ASBT in BBMV from DIO was significantly increased compared to that from normal (54.9±15 nmol/mg pro/min in normal and 167±8.8 in DIO; n=4, $p<0.05$). Western blot studies demonstrated that ASBT protein expression was significantly increased in whole cell lysate and BBM of villus cells from HFD rats.

Conclusion: Na-bile acid co-transport is stimulated in DIO likely due to increased expression of ASBT.

Compromised endothelial progenitor cell exosomal communication with endothelial cells in hypertensionischemia conditions

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Background: We have previously demonstrated that EPC-exosomes (EPC-EXs) can protect endothelial cells (ECs) against hypoxia injury in vitro. What's more, we found that extracellular vesicles released from starved EPCs have opposite functions to those released in inflammation conditions, suggesting that EPC-EXsmediated communication is functional and modulable by factors that affect the status of their parent cells. Given clinical studies showing the function of EPCs is declined in patients with hypertension, we speculate the function of EPC-EXs is altered in hypertension-ischemia conditions.

Study Hypothesis: The EPC-EX mediated communication with ECs is impaired in hypertension-ischemia conditions.

Methods: EPC-EXs were prepared from the bone marrow EPCs of wild type (WT) and hypertensive renin transgene (R+) mice and were denoted as WT-EPC-EXs and R-EPC-EXs, respectively. To mimic hypertensionischemia injury, ECs were treated with angiotensin II plus hypoxia and reoxygenation. To determine the function of EPC-EXs, ECs were co-cultured with EXs for 24hrs. EX uptake efficiency, cellular viability and function were assessed. To determine the EX uptake route, several pathway inhibitors were applied in the co-culture system.

Results: 1) Caveolae-dependent endocytosis is a major route in mediating EPC-EX internalization by ECs; 2) The incorporation efficiency of EPC-EXs from R+ mice by ECs was decreased. 3) Angiotensin II plus hypoxia reoxygenation-injured ECs displayed decreased cell viability, increased cell apoptosis and compromised angiogenic ability. 4) R-EPC-EXs displayed the impaired capability of rescuing ECs and improving their angiogenic ability on ECs as compared to that of WT-EPC-EXs did.

Conclusion: Our data have suggested that EPC-EXs mediated communication of EPC/EC is compromised in R+ mice hypertension-ischemia condition. These findings suggest that impairment of EPC exosomal communication might contribute to the exaggerated cerebral ischemia injury in hypertension-associated ischemic stroke.

Na+/K+ ATPase alpha 1 subunit participates in platelet signaling and is a potential anti-thrombotic target

Authors: Oliver Li1,2, Rodrigo Aguilar3, Renat Roytenberg2, Autumn DeHart2, Gretel Toloza-Alvarez2, Mark Hill2, Hong Yue2, Ellen Thompson3, Sandrine Pierre1,2, Jiang Liu1,2, Wei Li2

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Background: Thrombosis is a major cause of myocardial infarction and ischemic stroke. Sodium/potassium ATPase (NKA) is composited by alpha and beta subunits and plays an important role in maintaining the sodium and potassium gradient across the cell membrane. NKA also has signaling functions, and its α1 subunit (ATP1A1) reportedly binds to and inhibits the activity of Src, a tyrosine kinase, which plays a critical role during platelet activation.

Study Hypothesis: ATP1A1 participates in platelet signaling activation and is an anti-platelet and anti-thrombotic target.

Methods: Wildtype and ATP1A1 heterozygous mice aged 10-14 weeks were used. A FeCl3-induced carotid artery injury thrombosis model in combination with intravital microscopy was used for in vivo thrombosis study.

Platelet aggregation and Cellix flow chamber assays were used to evaluate in vitro platelet function. Western blot, co-IP, and blue native PAGE assays were used for the characterization of protein expressions and protein complex formations.

Results: ATP1A1 heterozygosity dramatically reduced its expression on platelets and inhibited in vivo thrombosis in male but not female mice. ATP1A1 heterozygosity did not affect initial platelet adhesion/aggregation on injured vessel walls and collagen-coated surfaces. However, it significantly delayed second wave platelet activation in vivo and inhibited ADP-induced platelet aggregation in vitro. ATP1A1 heterozygosity did not affect platelet intracellular sodium concentration, suggesting that the observed anti-thrombotic phenotype is not due to the altered activity of NKA. Intraperitoneal injection of the NKA inhibitor Ouabain (100 ng/g of body weight) for 24 hours significantly inhibited thrombosis in mice. ATP1A1 heterozygosity showed reduced ADP-induced AKT activation in platelets. ATP1A1 forms a complex with the ADP receptor P2Y12, and pretreatment of human platelets with Ouabain inhibited ADP-stimulated platelet aggregation in a dose-dependent manner.

Conclusion: ATP1A1 participates in platelet ADP signaling, which is essential for ADP-induced platelet activation. Targeting ATP1A1 could be a novel strategy for antiplatelet and antithrombotic therapy.

Nutrient-Induced Incretin Hormone Secretion by Enteroendocrine Cells is Altered in Obesity

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Background: Enteroendocrine cells (EECs) representing ~1% of the entire gut epithelial cells, constitute the largest endocrine organ of the body. In response to food intake, EECs release hormones, specifically the incretins glucagon-like peptide 1 (GLP1) and glucose-dependent insulinotropic peptide (GIP), that regulate energy balance and glucose homeostasis via their varied effects including stimulation of insulin secretion, regulation of appetite, gut motility and gastric emptying. EEC differentiation from intestinal stem cells is controlled by sequential expression of the transcription factors MATH1, Ngn3 and NeuroD1. In the small intestine, sensing luminal nutrients by transporter proteins expressed in EECs (such as SGLT1, PepT1) play critical role in triggering incretin secretion. Recent studies have shown decreased EEC and impaired incretin secretion in obesity. However, the mechanisms underlying dysregulation of EECs in obesity are not known.

Study Hypothesis: Dysregulated EEC differentiation and function alter incretin hormone secretion in obesity

Methods: mRNA and protein levels of EEC differentiation factors, the EEC marker chromogranin A, and GLP1 in the intestinal mucosa of lean versus obese Zucker rats, and control versus high fat diet-induced obese rats were measured by QRT-PCR and immunoblotting/immunofluorescence, respectively. Glucose and peptide-induced GLP1 and intracellular cAMP in vitro in mouse enteroendocrine cell line STC-1 was measured by ELISA.

Results: mRNA levels of MATH1, Ngn3 and NeuroD1, mRNA and protein levels of chromogranin A, and immunostaining of GLP1 were significantly decreased in the intestinal mucosa of obese Zucker rats and HFD-induced obese rats compared to the respective controls. In STC1 cells, glucose-induced GLP1 secretion was significantly reduced in response to inhibition of SGLT1, suggesting the role of SGLT1mediated glucose uptake by EECs in triggering GLP1 secretion.

Conclusion: Altered EEC function in obesity could involve mechanisms dysregulating EEC differentiation and impairing activity of nutrient transporters in EEC

Upregulated expression of phosphorylated N-Myc Downstream Regulated 1 (NDRG1) in clear cell renal cell carcinoma

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Background: Clear cell renal cell carcinoma (ccRCC) is the most prevalent type of kidney cancer. The mortality and incidence rates of ccRCC in Appalachia are higher than

non-Appalachia. This study aimed to determine signaling proteins that are differentially expressed in ccRCC relative to pair matched normal renal tissue from donors of central Appalachia.

Study Hypothesis: We hypothesize that Mechanistic Target Of Rapamycin Kinase (mTOR) signaling will be higher in renal tumors than pair-matched normal renal tissue.

Methods: Reverse phase protein arrays were used to identify statistically significantly differentially expressed proteins and phosphoproteins in renal tumors compared with pair-matched normal renal tissue. Western blot analysis was used to measure the levels of total NDRG1, phospho-NDRG1 (on threonine 346 and Serine 330), total SGK1 and phospho-SGK1 (serine 78), total GSK3 beta and phospho-GSK3 beta (serine 9) in total protein extracts isolated from renal tumors and normal renal tissue. The ChemiDoc MP Imaging System (image lab 4.0) was used to quantify band density and acquire Western blot images (Bio-Rad Laboratories, Hercules, CA, USA).

Results: The results showed statistically significant differentially expressed proteins in the mTOR pathway in renal tumors compared with normal renal tissue. The protein array results showed a statistically significant increase (by 3.3-fold) in the levels of phosphorylated NDRG1 (on Threonine 346) in renal tumors relative to normal renal tissue. Western blot studies showed upregulation of total NDRG1 and phospho-NDRG1 on Thr346 and Ser330 in renal tumors compared with normal renal tissue. The phosphorylation and thus activity of SGK1, the kinase that phosphorylates NDRG1, was also higher in renal tumors.

Conclusion: Prior reports show NDRG1 is a potent tumor suppressor of ccRCC. We therefore propose that targeting the hyperphosphorylation of NDRG1 would regulate its tumor suppressive activity in ccRCC and thus offering a potentially new therapeutic approach.

E-Cigarette Flavoring Agents Alter Mitochondrial Function and Autophagy Pathways in Human Kidney HK2 Cells

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Department & Institution: Ashley Cox, Kathleen C. Brown, Monica A. Valentovic. Department of Biomedical Sciences, Toxicology Research Cluster, Joan C. Edwards School of Medicine, Huntington, WV.

Background: Use of flavoring agents in e-cigarette liquids has become increasingly popular, but they are not tightly regulated which leaves gaps in knowledge regarding their safety. The kidney poses a potential site of action due to its filtering capacity.

Study Hypothesis: Cinnamaldehyde mediates renal cytotoxicity through mitochondrial and/or apoptotic pathways.

Methods: All studies were conducted using human non-cancerous renal proximal tubular cells (HK-2). HK-2 cells were plated, equilibrated for 48h, and treated with 0 (DMSO) or 5-100 uM cinnamaldehyde for 24 or 48h. Viability was assessed using MTT leakage and trypan blue exclusion using a Cell Countess. Western analysis probed for oxidative stress and autophagy markers (4-hydroxynonenal [4-HNE], LC3B-I, LC3B-II, PINK1, and PARKIN). Mitochondrial function was assessed as changes in Oxygen Consumption Rate (OCR) using a MitoStress Test kit and Seahorse analyzer. Results were obtained from at least 4 independent experiments using different cell passages. Differences between groups were analyzed using One Way ANOVA followed by post hoc Tukey test at a 95% confidence interval.

Results: MTT conversion to formazan was comparable between control and 0-20 uM groups ($p>0.05$).

Cinnamaldehyde was cytotoxic relative to control at 25-100 uM based on MTT assay ($p<0.05$). Cell Countess trypan blue exclusion assay showed no significant difference in cell membrane integrity at all concentrations tested when compared to controls. Expression of mitophagy-related protein LC3B-II increased significantly ($p<0.05$) at 100 uM cinnamaldehyde. Changes in PINK and PARKIN expression remained statistically insignificant. Seahorse assay showed significant decrease in mitochondrial maximal respiration rate and spare respiratory capacity at 20 uM cinnamaldehyde.

Conclusion: Cinnamaldehyde was cytotoxic to HK-2 cells, induced mitochondrial dysfunction, and may be activating the autophagic pathway. Additional studies are needed to explore the cytotoxicity mechanisms. (Supported by NIH Grant P20GM103434, A.C, supported by WV NASA Graduate Research Fellowship).

Quasi-experimental evaluation of liposomal bupivacaine (Exparel®) on inpatient opioid consumption in patients undergoing bariatric surgery

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Background: Excessive opioid consumption increases the risk of addiction and adverse drug events and should be limited. Liposomal bupivacaine (LB), which has been associated with reduced opioid consumption in other surgeries, was included for perioperative multimodal pain control for patients undergoing bariatric surgery at our institution. The effect of LB on inpatient opioid consumption has not been evaluated in patients who have undergone bariatric surgery.

Study Hypothesis: Evaluate opioid consumption in patients undergoing laparoscopic gastrectomy before and after implementation of LB as part of perioperative, multimodal pain control bundle.

Methods: A retrospective, pre/post quasi-experimental study was conducted to identify patients (n=483) who had undergone laparoscopic gastrectomy between 03/1/2017 and 06/30/2019. Intervention took place 03/07/2018, with no-LB (n=198) before and LB (n=285) after this time. Patients (n=362) were propensity score-matched (PS) 1:1 with inpatient morphine milligram equivalents (MME), ketorolac, intravenous acetaminophen, and baseline demographics compared. Multivariate linear regression was used to assess impact of variables on opioid consumption.

Results: Compared to no-LB, LB was associated with fewer median MME in the total (100.0 vs 173.8 mg, $p<0.001$) and PS (100.0 vs 170.0 mg, $p<0.001$) populations. In the multivariate analysis, LB significantly reduced MME (-94.84; 95% CI, -114.10 - -75.58, $p<0.001$).

Conclusion: LB given in the perioperative setting was associated with a significant decrease in post-operative, inpatient opioid consumption. These findings should be confirmed in a placebo controlled randomized trial. It is unknown if these findings extend to the consumption of opioids in the immediate, post-discharge setting.

Regional Analysis of Cervical Spine Fractures in Patients 65 years and Older

Authors: Ella Boggs, David Denning

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Background: Cervical spine fractures are a common occurrence among elderly populations. Mechanisms of acquiring cervical spine fractures in these patients can occur following a traumatic incident, mechanical falling, decreasing bone density, precipitating health conditions, or motor vehicle accident. As communities experience aging demographics, there is a transition for increase in specialized geriatric injuries.

Study Hypothesis: The purpose of this study was to analyze geriatric patients at a tertiary trauma center with cervical spine fractures and to bring awareness to its complex nature as an injury and for treatment, as well as outcome of survival based on individual ambulatory and discharge encounter progression.

Methods: Between 2016-2018, 75 geriatric cervical spine fractures were managed at St. Mary's Regional Medical Center a subsidiary of Mountain Health Network in Huntington, WV. This retrospective chart review included all patients 65 years and older with cervical spine fractures that had presented to the Emergency Department and analyzed the ambulatory visit characteristics, hospital stay, and discharge course up to 9 months post-fracture.

Results: Among geriatric patients analyzed for the study the average age was 81, C1-C2 vertebral fractures were most common, occurring frequently due to mechanical falls from standing height. 92% of patients had 2-11 comorbidities upon presentation. Hospital stays averaged 1-2 weeks, 65% of patients were unable to be discharged directly home following treatment, 29% died due to their cervical spine fractures alone, 37% had complications during their course of treatment, and 25% of spinal fractures were a repeat injury for the patient.

Conclusion: Cervical spine fractures in patients 65 years and older prove to be a serious injury with increasing incidence due to a growing elderly demographic. These injuries are multi-faceted in nature and prove to be a challenge for providers to treat, utilizing extensive hospital and healthcare resources, while still maintaining a poor prognosis overall.

Methamphetamines and the Effects on the Heart

Authors: Lauren Morilla

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Background: The chordae tendinea are an essential part of the heart and papillary muscles due to how it prevents mitral valve prolapse and regurgitation. The chordae tendinea originate from the fibrous heads of the papillary muscle and insert to the leaflet. The chordae anchor the atrioventricular valve to the wall of the ventricle which prevent the backflow of blood by stopping valve leaflets from inverting. Ruptured chordae affect the atrioventricular heart valves and is one of the primary causes of valve regurgitation. The use of methamphetamines causes an increase in catecholamine toxicity, which, causes alteration to the myocardium and remodeling/structural damage through electrical remodeling. The effects of methamphetamines on chordae tendinea and the heart is not yet fully understood.

Study Hypothesis: It was hypothesized that damage would occur on the chordae tendinea.

Methods: Rat hearts were obtained from a previous study of Dr. Hambuchen (School of Pharmacy). Rats were administrated 1mg/bw kg of methamphetamine, 3 times a day for 4 days. Then, rats were euthanized by opening the chest cavity to induce pneumothorax under isoflurane-induced anesthesia and stored in a -80o C freezer for further analyses. In this study the hearts were examined one at a time. The frozen hearts were defrosted in a cold phosphate buffer solution (in the refrigerator) overnight, then, carefully opened to expose the chordae tendinea. The opened heart was then placed in a buffer solution in a petri dish and observed under a stereomicroscope. The number and degree of chordae tendinea rupture and/or damage were recorded, and photographs were taken for each heart.

Results: A significant lack of chordae tendinea, along with significant thinning and ruptured chordae, was found.

Conclusion: The data collected supports the clinical observation that methamphetamine use is dangerous to the central nervous system but also to the cardiac function by changing the heart's structure.

The effects of adolescent intermittent binge ethanol exposure on hippocampal astrocyte morphology and synaptic colocalization

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Background: Binge drinking is highly prevalent among today's youth and emerging adults. Early alcohol use is associated with increased risk of alcohol dependency later in life. While progress has been made in understanding the consequences of binge drinking on neuronal and subsequent cognitive function, little is known about the role of astrocytes. Astrocytes ensheathe synapses and are critical in synapse formation, maturation, and neuronal transmission.

Study Hypothesis: Here we investigate the acute and chronic effects of adolescent intermittent binge ethanol (EtOH) exposure on astrocyte morphology and colocalization with synapses in the developing hippocampus.

Methods: Male Sprague Dawley rats received intracranial astrocyte-specific adeno-associated virus directly into the dorsal hippocampus (dHipp). Animals received intermittent EtOH (5g/kg, i.g.) 10 times over 16 days beginning PND30. Tissue was processed 24 hours after the 10th dose (PND 46) or after a 26-day forced abstinence period (PND 70). Immunohistochemistry was performed using post-synaptic marker PSD95. Imaging was performed using confocal microscopy and 3D astrocyte reconstructions were rendered using IMARIS (bitplane).

Results: Following intermittent binge EtOH exposure, there was a substantial decrease in dHipp astrocytes-synaptic colocalization after the 10th dose, despite no change in astrocyte volume. In adulthood, there was a significant decrease in hippocampal astrocyte volume as well as a decrease in astrocyte-synaptic colocalization in EtOH animals when compared to the age-matched controls.

Conclusion: Our results reveal that adolescent binge EtOH exposure results in changes in dHipp astrocyte

morphology that persist into adulthood. This is coupled with a protracted decrease in astrocyte-synaptic colocalization that isn't observed immediately following adolescent binge EtOH exposure.

What Is the Incidence of in Hospital Mortality After Hip Fracture? Does Treatment Options Matter?

Authors: Jonathan Lash, Alisina Shahi, Matthew Bullock, Ali Oliashirazi

Department & Institution: Marshall Orthopaedics

Background: Hip fractures are a common cause of disability, with high rates of morbidity and mortality. Hip fracture treatment includes hemiarthroplasty (HA), total hip arthroplasty (THA), and internal fixation.

Study Hypothesis: This study investigates the in-hospital mortality and complication rates in patients with hip fractures after surgical intervention.

Methods: The Nationwide Inpatient Sample from 2000-2015 was queried for hip fractures to identify and determine the incidence of in-hospital mortality and complications: myocardial infarction, stroke, length of stay(LOS), and disposition for hip fracture patients after internal fixation compared with HA, and THA. Multivariate logistic regression were used to compare these variables between the surgical interventions.

Results: The in-hospital mortality rate of hip fracture patients declined 0.04%; a 29% relative decrease, but hip fractures were associated with a greater incidence of in-hospital mortality (OR=1.63,95%CI:1.59-1.66). Internal fixation had lower mortality rates than HA (OR=0.53,95%CI:0.50-0.56) and THA

(OR=0.86,95%CI:0.77-0.96) and fewer in-hospital complication. Patients undergoing internal fixation had shorter LOS (5.77 days) than HA (6.39) and THA (6.08) while more likely to be discharged to home than HA (OR=3.99, 95%CI: 3.89-4.10) and THA (OR=1.58,95%CI:1.51-1.65). THA had lower in-hospital mortality than HA (OR=0.61, 95%CI: 0.55-0.68), and fewer in-hospital complications. Hip fractures had higher in-hospital mortality rates than cholecystectomies (OR=2.84,95%CI:2.78-2.89), and prostatectomies (OR=9.29,95%CI:8.68-9.95).

Conclusion: Based on these results, it appears in-hospital mortality of hip fracture patients is decreasing. Patients undergoing internal fixation had lower rates in-hospital complications, in-hospital mortality, a shorter LOS, and were more likely to be discharged home than patients undergoing HA or THA. Surgeons must be cognizant of the high in-hospital mortality potential of hip fractures. We encourage the orthopaedic community to optimize modifiable risk factors prior to surgery and select the surgical plan considering patients' risk factors.

Adipose Derived Secretome Mediates the Regulation of SGLT1 in Rat Small Intestinal Epithelial Cells

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Background: The most common complication of obesity is diabetes, resulting from altered glucose homeostasis. The most important step in this homeostasis is intestinal epithelial cell's absorption of glucose via the Naglucose co-transporter (SGLT-1). In obese Zucker rats (OZR), SGLT-1 is stimulated secondary to increased affinity for glucose. While it is known adipose derived secretome (ADS) has been shown to affect many physiological functions in obesity, how ADS may affect SGLT-1 and glucose homeostasis during obesity is not yet known.

Study Hypothesis: Hypothesis: ADS may regulate SGLT-1 in obesity-associated diabetes.

Aims: Determine the effects of ADS on SGLT-1 in intestinal epithelial cells.

Methods: Rat small intestinal epithelial cells (IEC-18 cells) grown to confluence in 24-transwell-plates and were treated on day 4 with either serum-free DMEM as control, mouse fibroblast conditioned media (FCM), or ADS. 3H-OMG uptake was performed for SGLT-1 activity. Na-K-ATPase activity was determined by measuring inorganic phosphate released. Western blot and RTPCR were performed using rat specific SGLT-1 antibodies and primers, respectively.

Results: ADS stimulated SGLT-1 in IEC-18 cells, while FCM did not. NA/K-ATPase was diminished in ADS treated IEC-18 cells. Kinetic studies revealed the mechanism of SGLT-1 stimulation was secondary to enhanced co-transporter affinity (1/Km) for glucose without change in the number of BBM co-transporters. Westernblot studies revealed no changes in SGLT-1 protein between ADS and DMEM-treated IEC-18 cells.

Conclusion: The in-vitro stimulation of SGLT-1 by ADS is similar to that seen in-vivo in obese Zucker rats. Thus, adipose derived secretome likely mediates the altered glucose absorption via SGLT-1 which may be important in the development obesity-associated diabetes.

Thymidine Phosphorylase could be a novel therapeutic target for treating COVID-19

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Background: COVID-19, the viral respiratory illness responsible for the current pandemic, is marked by a dysregulated hyperinflammatory state and thrombotic events, for which no comprehensive treatment has been established. The mechanism which mediates systemic thrombotic events in COVID-19 is not yet known. Thymidine phosphorylase (TYMP) plays an important role in platelet activation and thrombosis.

Study Hypothesis: TYMP mediates COVID-19-associated inflammation and thrombosis.

Methods: By co-transfecting SARS-CoV-2 spike protein (SP) and human ACE2 into Cos-7 cells, we examined how SP regulates ACE2 and TYMP expression. By using data provided by the Massachusetts General Hospital Emergency Department COVID-19 Cohort with Olink Proteomics, we analyzed the correlation between plasma TYMP and the severity of COVID-19.

Results: Overexpression of SP or its receptor-binding domain (RBD) leads to ACE2 shedding. Overexpression of SP also increases TYMP expression. In comparison to COVID-19-negative patients, plasma TYMP in COVID-19 patients is significantly increased in a severity-dependent manner. The increase of plasma TYMP is observed earlier than the increase of C-reactive protein, a predictive factor for inflammation and future risk of cardiovascular events. The increase of TYMP is positively associated with D-dimer, lactate dehydrogenase, and pulmonary symptoms as well. Receiver Operating Characteristic (ROC) analysis based on TYMP plasma levels on Day 0 (area-under-curve value 0.8721) suggests that TYMP is a very sensitive and specific marker in diagnosing severe COVID-19. TYMP is highly expressed in mouse asthmatic

lungs, human type II alveolar epithelial cells, and bronchial epithelium, all which mediate SARSCoV-2 entry.

Conclusion: TYMP is positively correlated with COVID-19-associated thrombotic events, inflammation, and organ damage. TYMP could be an acuity marker for COVID-19 diagnosis. Targeting TYMP could be a novel effective medicine for COVID-19 and/or its sequalae.

Effect of Cidec gene polymorphisms on obesity susceptibility in mice

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Background: A quantitative trait locus, tabw2a, was linked to obesity in TALLYHO/Jng (TH) mice, a polygenic model of obesity and type 2 diabetes. Using whole genome sequencing data of TH mice, we identified the gene "cell death-inducing DFFA-like effector c" (Cidec) as a candidate gene for tabw2a. Cidec is a lipid dropletassociated protein and is known to promote triglyceride accumulation in adipocytes. There was one nucleotide substitution in the coding sequence of Cidec in TH mice, 136 C>A, compared to C57BL/6J (B6) reference sequence. This results in an amino acid substitution R46S (Arginine to Serine). For functional evaluation of the Cidec R46S polymorphism in vivo, we have generated Cidec R46S knock-in mice where the S46 is exchanged for the R46 at the Cidec gene on a B6 background.

Study Hypothesis: The aim of this study was to characterize the obesity and energy metabolism in Cidec R46S knock-in mice.

Methods: Cidec R46S knock-in and B6 mice were weaned onto chow and high fat (HF) diets at 4 weeks of age (wk) and maintained. At 15-22 wk, energy expenditure, food intake, and locomotor activity were measured using an eight-chamber Comprehensive Laboratory Animal Monitoring System. At 20 wk, body composition including fat and lean mass was measured using EchoMRI-100 whole body composition analyzer.

Results: On chow, no genotype effects were shown on adiposity between knock-in and B6 mice for both males and females. However, on HF diets, for both sexes, knock-in homozygous mice had significantly larger body fat mass than B6, although their body weights were comparable. Food intake, locomotor activity, and respiratory parameters were not significantly different between knock-in and B6 mice for both sexes.

Conclusion: We conclude that the Cidec gene is the most likely candidate for the obesity effect of tabw2a, and that Cidec S46 variant contributes to the obesity susceptibility.

The role of metabolic dysfunction in SARS-CoV-2 progression

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Background: SARS-CoV-2 is a virus that mainly infects the respiratory tract and is responsible for more than 4 million deaths worldwide. The mortality of the virus displays heterogenicity with some populations more susceptible to severe illness and death including those of increased age, male, and poor metabolic health (diabetic and obese). We evaluated the role of diabetes and obesity in the progression of SARS-CoV-2 in hopes of elucidating the possible mechanisms(s) responsible for increased mortality using a diet induced obese (DIO) K18-hACE2-mouse model.

Study Hypothesis: We hypothesize that metabolically dysfunctional-infected and male mice will have an increase mortality rate in comparison to normal-weighted glucose controlled mice. We postulate that molecular signatures expressed during obesity will be further exacerbated by SAR-CoV-2 infection including genes responsible for normal glucose homeostasis.

Methods: K18-hACE-2 mice were placed under a high-fat/carbohydrate diet to recapitulate obesity and disproportional glucose clearance for 8 weeks (DIO group). Following induction, mice were infected with SARS-CoV-2 alpha variant and monitored over a week

period. Daily health checks were performed and animals were assigned clinical scores based on their overall health. Animal blood and tissues were harvested post-experiment and evaluated for changes in cytokine levels, RNA changes, and protein levels.

Results: DIO infected K18-hACE2-mice were more susceptible to mortality compared with control animals, correlating with human data. In addition, gender differences were observed, both at the physiological and molecular levels. FGF21, a modulator of glucose metabolism, was altered in DIO-infected mice along with other factors necessary for glucose and lipid partitioning.

Conclusion: This study revealed molecular changes that could be responsible for an increase in mortality in subjects with metabolic dysfunction. Understanding these molecular undertones is crucial in understanding SARSCoV-2 infection and illustrates potential targets for combating severity of the infection.

The profile of extracellular vesicles in intracerebral hemorrhage patients

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Background: Intracerebral hemorrhage (ICH) is one of the leading life-threatening types of strokes with high mortality. A prominent feature of ICH is neuroinflammation involving neutrophils and macrophages. Microvesicles (MV) and exosomes (EX) released from various cells are used as biomarkers for different diseases.

Study Hypothesis: The level of neutrophil or microglia MV/EX could be correlated with the ICH prognosis.

Methods: MV and EX were isolated from the plasma of ICH patients (n=39, 22/17, M/F) by using the serial centrifuge methods. Nanoparticle tracking analysis (NTA, NS300) was used to determine the type and concentration of neutrophil and macrophage-released MVs/EXs. Specific antibodies, CD66b, and P2RY12 were used for neutrophil and microglia, respectively.

Results: A predictive relationship between both hospital length of stay (R2=0.83) and ICU length of stay (R2=0.88) was found with MVs and EXs and patient data (including LDL, ICH volume, etc,). Further predictive multiple linear regression relationship was seen between MV and EX concentrations and MSRV3 (R2=0.46) and MSRV5 (R2=0.51), however, some issues existed with nonconstant error variance in these models. Further, slight multiple linear regression relationships were seen between MV and EX concentrations and both hospital length of stay (R2=0.18) and ICU length of stay (R2=0.26). LDL and ADM_SBP were found to have a slight predictive relationship with MV Neutrophils (R2=0.31) and age, race, htn, and the ICH location were found to be predictive of MV concentration (R2=0.46).

Conclusion: This study found predictive relationships between patient outcomes and MVs and EXs. When combined with generally collected patient data (LDL, etc.), measurements of MVs and EXs are strongly predictive of overall patient outcome. Further, larger studies should investigate these effects.

The role of obese adipose derived secretome in colon cancer pathogenesis

Authors: Cora Miracle, Chelsea Thompson, Travis Salisbury

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Background: Colorectal cancer (CRC) is the third most common cancer and second leading cause of death in within the United States. The survival rate of colon cancer has been on the rise due to screening, however the incidence rate has slowly climbed. Along with the rise in colon cancer is the rise in BMI. Obese individuals have a 30% higher chance of developing colon cancer. Obesity is known to cause; increased leptin as well as increases in PI3K/AKT pathway[1] They also are known to have increased plasma levels of the essential amino acid Leucine, a known stimulator of the mTOR pathway. These changes may be linked to the substances secreted by adipose cells termed adipose derived secretome (ADS). While the signaling factors in ADS have not been fully elucidated, obese ADS has been shown to increase mTOR

signaling in breast cancer promoting proliferation. While this is shown in breast cancer, the effects of obesity and adipose tissue is not known in colon cancer.

Study Hypothesis: Obese ADS as well as leptin treated cells will stimulate the mTOR pathway at a higher level compared to lean ADS.

Methods: CaCo2 were cultured in EMEM media with 20% FBS + P/S. Visceral fat from lean and obese mice were taken and cultured. The media of these culture cells was then taken and added to CaCo2 for 24 hours. Leptin was added to CaCo2 for 15 min. Protein analysis was performed via Western blot analysis.

Results: We have examined the various effects of ADS and leptin on colon cancer cells as well as the cell origin of colon cancer. We found that obese ADS and leptin stimulated CaCo2 colon cancer cells exhibit higher mTOR pathway activation as read by levels of phospho-S6 as compared to controls.

Conclusion: Obese ADS and increased leptin levels affect the mTOR signaling pathway.

Influence of diet fat content on ethanol metabolism and PPAR-α-regulated lipid metabolism

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Background: Alcohol-related Liver Disease (ALD) is one of the most prevalent causes of chronic liver disease. ALD presents as alcoholic fatty liver or steatosis and can progress to steatohepatitis and more severely, cirrhosis. This can be partially attributed to ethanol's ability to disrupt fatty acid β-oxidation (FAO), which is regulated by peroxisome proliferator-activated receptor α (PPAR-α).

Study Hypothesis: In this study, the effects of a high-fat diet, low-fat diet, and PPAR-α agonist, WY-14,643, on ethanolinduced steatosis were explored.

Methods: Mice were fed the Lieber-DeCarli liquid ethanol diet modified to high-fat (40% energy from fat) or low-fat diets (12.5% energy from fat). WY-14,643 was added at 10 mg/L. Mice were fed for 21 days to induce alcoholic fatty liver, then sacrificed following overnight fast.

Results: Results showed a serum triglyceride (TG) elevation following ethanol feeding, which was reduced by WY14,643. No differences in serum TG were seen between the low and high-fat diets. Liver TG levels did not show reduction by WY-14,643, but lipid-metabolism-related enzymes acyl-CoA oxidase 1 (ACOX1), catalase, and liver fatty acid binding protein (L-FABP) were induced by WY-14,643 more in the high-fat than low-fat diet. Serum ethanol clearance was higher in low-fat than high-fat diet, which is possibly attributed to the higher expression of alcohol dehydrogenase, a major ethanol metabolism enzyme, in the low-fat diet than the high-fat diet. Hepatomegaly due to peroxisomal proliferation was induced by WY14,643 to a higher extent in the low-fat diet than in the high-fat diet.

Conclusion: These results suggest that fat content in the diet has an influence on ethanol metabolism and WY-14,643induced lipid metabolism related enzymes and peroxisome proliferation.

Diverging effects of adolescent intermittent ethanol exposure on astrocyte morphology and synaptic proximity in prefrontal cortex subregions

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Background: Excessive alcohol consumption is prevalent among adolescents. Animal models show that adolescent intermittent ethanol exposure (AIE) disrupts neuronal function, increasing the risk of persistent cognitive impairment. However, the role of non-neuronal cells (astrocytes) in these processes remains unclear. Astrocytes have extensive perisynaptic astrocyte processes (PAPs) that ensheathe synaptic terminals and play an essential role in synaptic maintenance and signal transmission.

Study Hypothesis: We have previously shown that

astrocyte maturation continues throughout adolescence into early adulthood in the prefrontal cortex leading us to hypothesize that AIE induces changes in astrocyte morphology and PAP-synaptic proximity, that may contribute to neuronal dysfunction.

Methods: Male Sprague Dawley rats received intercranial injections of an astrocyte-specific green fluorescent protein (GFP) virus in the medial prefrontal cortex (mPFC), anterior cingulate cortex (ACC), or orbitofrontal cortex (OFC). Beginning on PND30, animals received intermittent EtOH or water (5g/kg i.g.) over 16 days. Tissue was collected on PND72. Immunohistochemistry was performed using the post-synaptic density marker PSD95. Astrocyte imaging, reconstruction, and co-localization with PSD95 was conducted using confocal imaging and IMARIS-Bitplane.

Results: There was a decrease in PAP-synaptic colocalization in ACC following AIE with no change in astrocyte volume. There was a decrease in PAP-synaptic colocalization in the ventral OFC following AIE with an increase in astrocyte volume. There were no changes in any measure in the mPFC or lateral OFC.

Conclusion: Our results reveal that AIE results in protracted changes in astrocyte morphology and PAP-synaptic decoupling in a region-specific manner within the prefrontal cortex.

Sex-Dependent Changes in Vascular Function of Adult Rats Following Prenatal Exposure to Methamphetamine

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Background: Prenatal exposure to cocaine or nicotine leads to vascular changes such as endothelial dysfunction and potentiation of agonist-induced vasoconstriction in adult offspring. However, the impact of prenatal exposure to methamphetamine on the adult vasculature has not been well characterized.

Study Hypothesis: The objective of the present study was to examine the hypothesis that prenatal methamphetamine exposure alters agonist-induced changes in vascular tone in adult offspring.

Methods: Pregnant rats received daily injections of saline or methamphetamine (5mg/kg, s.c.) throughout gestation. Responses to phenylephrine, angiotensin-II, serotonin, acetylcholine, and sodium nitroprusside were measured in aortic rings isolated from adult (5 months old) offspring.

Results: Acetylcholine-induced relaxation was attenuated in aortas from adult male rats (but not females) that had been prenatally exposed to methamphetamine. This methamphetamine-induced effect was dependent on the presence of intact perivascular adipose tissue (PVAT). Prenatal methamphetamine had no impact on nitroprusside-induced relaxation regardless of whether PVAT was present. Angiotensin II-induced contraction was significantly potentiated in male (but not female) aortas following prenatal methamphetamine exposure. This effect was abolished by L-NAME. Prenatal methamphetamine had no effect on phenylephrine or serotonin-induced contractile responses in either male or female aortas.

Conclusion: These findings suggest that prenatal exposure to methamphetamine leads to PVAT dysfunction, disruption of NO signaling, and potentiation of angiotensin II-induced contraction of the aorta in a sex-dependent manner.

Antinociceptive Effects of Decursinol on Cisplatin-Induced Chronic Neuropathic Pain

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Background: Presently, about 20.4% of adults suffer

from chronic pain. Despite increased susceptibility to tolerance and dependency, prescription opioids are commonly used for the management of chronic pain. Chemotherapy induced peripheral neuropathy (CINP) is a debilitating side effect of chemotherapy. Besides opioids, few treatment options are available for managing CINP. Therefore, there is a need to identify non-opioid alternatives to manage chronic pain. The Korean Angelica gigas Nakai (AGN) root has been utilized in Korean herbal medicine due to reported neuroprotective, anti-inflammatory, antioxidant, anti-cancer and analgesic properties. One of the major pyranocoumarin compounds purified from the AGN root is decursinol. Previous studies, though limited, suggest that decursinol possesses acute analgesic properties; however, the effects of decursinol on chronic pain and potential for tolerance development remain unknown.

Study Hypothesis: Therefore, the goal of this study was to assess both the antinociceptive effects and the potential for tolerance development to once-daily treatment of 50 mg/kg decursinol using models of both acute thermal (hot plate and tail-flick) and chronic (CINP) pain.

Methods: Mice were treated with 50 mg/kg of decursinol or vehicle (IP) and assessed 30 minutes later for antinociception on the hotplate and tail-flick or for reversal of mechanical allodynia via von Frey in a model of CINP once-daily for 14 consecutive days.

Results: Decursinol dose-dependently reversed antinociception and mechanical allodynia. Repeated daily treatments with 50 mg/kg of decursinol induced antinociceptive responses on both hot plate and tail flick along with full reversal of mechanical allodynia in a model of CINP. Likewise, in both pain models, there was evidence of tolerance to the antinociceptive and anti-allodynic effects of decursinol but, the rates of tolerance development varied.

Conclusion: Future directions include elucidating the mechanism of action for the antinociceptive and anti-allodynic effects and tolerance development of decursinol in both acute and chronic pain.

The Synthetic Capsaicin-analog Arvanil sensitizes Cisplatin-Resistant Human Lung Cancer Cells to the pro-apoptotic activity of Irinotecan

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Background: Cisplatin-based combination therapy is the standard of care for the treatment of human lung cancer. Initially, cisplatin shows excellent therapeutic response, but the tumor inevitably relapses and is resistant to cisplatin. These patients are said to have platinum-refractory lung tumors. Patients with platinum resistant/refractory disease have limited options, as the only standard chemotherapy with an FDAapproved drug, irinotecan, has an objective response rate of approximately 3% and little or no survival benefit.

Study Hypothesis: Agents which improve the therapeutic response (of human lung cancers) towards irinotecan may be useful for the treatment of cisplatin-resistant human lung cancer. Our published data show that the nutritional compound capsaicin sensitized human small cell lung cancer (SCLC) cells towards camptothecin-induced apoptosis. Arvanil is a synthetic non-pungent capsaicin-analog which displays enhanced growthsuppressive activity in human SCLC. The present study aims to investigate the combinatorial apoptotic activity of arvanil and irinotecan in cisplatin-resistant lung cancer.

Methods: Caspase-3 activity assays measuring apoptotic activity in H69-CPR and PC9-CDDP human cisplatinresistant SCLC cells and Chou-Talalay isobologram analysis.

Results: Caspase-3 activity assays reveal that the combination of irinotecan and arvanil displayed greater apoptotic activity in H69-CPR human cisplatin-resistant SCLC cells than the drugs used alone. These experiments were repeated in a second cisplatin-resistant lung

cancer cell line PC9-CDDP and similar results were obtained. Chou-Talalay isobologram analysis showed the interaction between irinotecan and arvanil is synergistic in H69-CPR and PC9-CDDP cells.

Conclusion: The co-administration of arvanil and irinotecan suppressed the growth of H69-CPR tumors (xenografted in SCID mice) better than the individual drugs. Taken together, our findings pave the way for the discovery of novel combination therapies for the therapy of cisplatin-resistant human lung cancer.

High-fat diet alters body composition and serum cytokines without affecting body mass

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Background: Accelerated bone growth, a hallmark of juvenile obesity, can lead to irreversible skeletal damage. Paradoxically, obese children have low to normal levels of the growth promoting hormone insulin-like growth factor-1. We previously found that a high-fat diet increases bone growth and alters serum cytokines in young mice before significant changes in body mass were evident.

Study Hypothesis: Here we tested the hypothesis that mice on a high-fat diet exhibit changes in body composition along with higher levels of inflammatory cytokines before they develop overt obesity.

Methods: Male 3-week-old C57BL/6 mice were put on high-fat or control diet at weanling age. Serum cytokines were measured by ELISA. Skinfold thickness in the rump was measured using a dial micrometer to estimate body fat. Statistical significance ($p<0.05$) was determined in SPSS.

Results: After 2 weeks, tibial elongation rate was over 10% higher in mice on a high-fat diet with no difference in body mass. The cytokines TNF-α and IL-6 were both decreased in the high-fat diet group, while VEGF increased. Rump skinfold was nearly 7% greater in the high-fat diet group, indicating a higher proportion of body fat.

Conclusion: Our results support the hypothesis that a high-fat diet alters body composition and inflammatory cytokines before overt signs of obesity. The most robust change was a 1.5-fold decrease in IL-6, which has a role in energy metabolism indicating that the changes in body composition that we observed might play a role in altering serum cytokines. Reduced inflammatory cytokines could serve as a useful biomarker to provide a timely and effective treatment to mitigate long-term skeletal damage before it might otherwise be recognized.

Transformative Role of Endometriotic Milieu in Ovarian Cancer

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Background: Ovarian cancer is the 4th largest cause of cancer death in women. Approximately 10-15% of women of childbearing age suffer from endometriosis. Endometriosis is defined by the growth and presence of endometrial tissue (lesions) outside of the uterus. The women with endometriosis also have an increased presence of peritoneal fluid (PF) that comprises of inflammatory cells, growth factors, cytokines/ chemokines, etc. Epidemiological studies have shown that >3% of women with endometriosis develop ovarian cancer (low-grade serous or endometrioid types).

Study Hypothesis: Our hypothesis is that the PF from women with endometriosis induces transformative changes in the ovarian cells, leading to ovarian cancer development.

Methods: PF from women with and without endometriosis was collected after IRB approval and patient consent. IOSE (normal ovarian epithelial cells) and TOV-21G cells (human clear cell carcinoma cell line) were treated with various volumes of PF (no endometriosis or endometriosis) for 48 or 96 hours and proliferation was measured. Expression levels of epigenetic regulators and FOXP3 an inflammatory tumor suppressor, were determined. A Human Cancer Inflammation and Immunity Crosstalk RT2 Profiler PCR array was used to

measure changes in cancer related genes in treated cells.

Results: Results showed increased growth of TOV-21G cells treated with PF from women with endometriosis versus without endometriosis compared to IOSE cells. Endo PF treatment induced EZH2, H3k27me3 and FoxP3. The RT2 PCR array of TOV-21G cells treated with endo PF showed upregulation of various inflammatory genes (TLRs, Myd88 etc).

Conclusion: These studies show that PF from women with endometriosis can both proliferate and transform ovarian cells.

Sex-specific effects of green apple e-cigarette flavor on α4- and α6-containing nicotinic receptors of dentate gyrus and habenula neurons in a mouse model

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Background: Despite a decrease in e-cigarette use during the onset of the COVID-19 pandemic, vaping remains high at roughly 4 million and 10 million adolescent and adult users, respectively. E-cigarette flavors have been shown to alter the neurobiology of midbrain neurons, a region critical for drug dependence and rewardrelated behaviors. Following these findings, we aimed to identify the effect e-cigarette flavors have on other important addiction-related brain regions, including the hippocampus and habenula.

Study Hypothesis: We hypothesize that popular green apple-flavored e-cigarettes (without nicotine) enhance α4-containing (α4*) nAChRs in the hippocampus, and α4* and α6* nAChRs in the habenula, following exposure to green apple flavorants.

Methods: We utilized non-contingent vapor exposure for 10-days with male and female mice (genetically modified to contain fluorescent nAChRs) to study how a green apple (GA) flavorant mixture (hexyl acetate, ethyl acetate, and methylbutyl acetate flavorants at a 3:1:1 ratio) alters nAChR density in the hippocampus and habenula, compared to PGVG control-exposed mice. Brains were extracted following the 10-day protocol, sliced using a cryostat, and imaged through confocal microscopy. Statistical differences were determined using one- and two-way ANOVAs with post hoc Tukey means comparisons.

Results: We observed nAChR changes in a sex-specific manner. GA-exposed females exhibited an increase in nAChR density in both the hippocampus and habenula, compared to PGVG-control and air-control mice. However, GA-exposed males exhibited a non-significant increase in nAChR density.

Conclusion: Given the prominent role of the hippocampus in learning and motivational signals involving drug dependence and the reward-mediating, aversive habenular pathway involved in dependence and withdrawal, these changes in neurobiology highlight an interesting finding that, similar to nicotine, flavorsalone may impact addiction-related circuitry to promote increased e-cigarette use.

The A-1Na/K-Atpase Signalosome Rescinded Epigenetic Changes in the Progression of Nash-Associated Liver Carcinogenesis

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Background: Hepatocellular carcinoma (HCC) is one of the most aggressive human cancers and is characterized by an acquisition of multiple abnormal phenotypes driven by genetic and epigenetic alterations. The Epigenetic changes associated with NASH-associated Hepatocellular carcinoma (HCC) include DNA methylation and histone modifications

Study Hypothesis: Most of the existing clinical and experimental reports provide only a snapshot of abnormal histone modifications in HCC rather than their dynamic changes. Hence, the present study hypothesised that the histone acetylation/methylation disturbances mediated by the α-1Na/K-ATPase signalosome may help to elucidate the significance of these changes in the development of HCC

Methods: In vitro studies were done using two human HCC cell lines, complemented with our in vivo murine models of NASH and NASH-HCC. Experimental groups were exposed to pNaKtide, a selective α-1 subunit signalling inhibitor. Acetylated/tri methylated H3K9 in cell lysates and liver homogenates were measured by ELISA, and their expression by confocal microscopy on immuno-stained livers. Significant differences among groups were established at $p<0.05$ using ANOVA/t-test

Results: Epigenetic changes in cell lines and liver malignancies correlated with morphological changes in NASH progression to malignancy. Acetylated and tri-methylated H3K9 were significantly increased in untreated cells and livers when compared with pNaKtide treated cells and animals ($p<0.05$). Quantitative analysis of H3acetylK9 and H3tri-methyl K9 proteins on confocal images from cells and livers confirmed our findings

Conclusion: Normalization of the α-1Na/K-ATPase signalosome in human HCC cell lines and murine NASH-HCC results epigenomic reprogramming

Role of Strain in Mediating Sex-Differences in Acute Cannabinoid Response and Tolerance

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Department & Institution: Department of Biomedical Sciences, Joan C. Edwards School of Medicine, Huntington, WV

Background: Cannabinoids have been increasingly used to alleviate chronic pain; however, tolerance to the antinociceptive effects of cannabinoids, including delta-9-tetrahydrocannabinol (Δ9-THC), may limit their therapeutic utility. Likewise, with more women than men now using medical cannabis for pain relief, it is imperative that we understand how sex may influence cannabinoid-mediated antinociception and subsequent tolerance. While studies in rats have consistently found female rats to be more sensitive to the acute antinociceptive effects of cannabinoids compared to male rats, work in our lab consistently finds the opposite finding that male mice are more sensitive to the acute antinociceptive effects of both Δ9-THC and CP55,940 compared to female littermates. Studies in our lab have consistently utilized mice on a C57BL6/J (B6) background.

Study Hypothesis: Therefore, the purpose of the present study is to examine whether our observed sex-differences in Δ9THC-induced antinociception and tolerance are consistent across multiple mouse strains or are strain dependent.

Methods: Male and female B6 and DBA mice were first assessed for differences in acute Δ9-THC-induced antinociception using the tail-flick assay across a range of doses of (0-100 mg/kg). After a significant washout period, these mice were subsequently assessed for sex-differences in antinociceptive tolerance development to 30 mg/kg Δ9-THC following once-daily treatment for seven consecutive days.

Results: Consistent with our previous findings, male B6 mice were more sensitive to the acute antinociceptive effects of Δ9-THC than female B6 mice. Male and female DBA, however, mice did not differ in their antinociceptive response to Δ9-THC, suggesting that sex-differences in cannabinoid-induced antinociception in mice is likely strain-specific.

Conclusion: These studies highlight the therapeutic potential of Δ9-THC in pain management and underscore the importance of considering sex, when evaluating their clinical utility.

Remodeling of Mitochondrial Energy Metabolism in Clear Cell Renal Cell Carcinoma

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Background: Clear cell renal cell carcinoma (ccRCC) is one

of the most common renal cell carcinomas. The defining morphological hallmark of ccRCC is the accumulation of glycogen and lipid droplets in the cytoplasm of the cells due to the reprogramming of glucose and fatty acid metabolism and oxidative phosphorylation (OXPHOS). Energy metabolism by OXPHOS is supported by both nuclear and mitochondrial-encoded genes. Alterations in the expression of energy metabolism are well documented in ccRCC; however, its correlation to mitochondrial biogenesis is limited

Study Hypothesis: We proposed that the remodeling of mitochondrial energy metabolism regulated by mitochondrial biogenesis also contributes to the progression of ccRCC.

Methods: To test our hypothesis, immunoblotting analyses of ccRCC biopsies and their matched normal tissues were performed to investigate the role of mitochondrial energy metabolism and biogenesis in the remodeling of OXPHOS complexes. To support our findings on the key factors involved in energy metabolism and biogenesis further, genomics and proteomics data mining analyses of publicly available ccRCC databases at the Cancer Genome Atlas (TCGA) were also performed.

Results: We discovered that the expressions of several key metabolic enzymes and factors involved in mitochondrial biogenesis were significantly reduced in ccRCC biopsies. Expression of epithelialmesenchymal transition markers, vimentin, and E-cadherin, were also correlated to energy metabolism in tumor biopsies.

Conclusion: Based on the evidence provided in our studies and publicly available ccRCC data, we propose that the change in mitochondrial biogenesis is part of the remodeling of mitochondrial energy metabolism and pathogenesis of ccRCC.

Expression of cerebellar genes related to inflammation, insulin resistance, glial differentiation, and development in a mouse model of obesity and type 2 diabetes

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Background: A substantial body of evidence demonstrates that obesity and type 2 diabetes (T2D) are linked to altered neurobehaviors. Obesity- and T2D-related behavioral alterations are postulated to result from impaired insulin signaling and increased oxidative stress and inflammation. Preliminary findings from our lab revealed that TALLYHO/Jng (TH) mice, which serve as a polygenetic model for obesity and T2D, demonstrate abnormal motor behaviors compared to normal C57BL/6J (B6) mice.

Study Hypothesis: Because the cerebellum is recognized as an important brain structure for balance and motor control, we assessed cerebellar mRNA levels of genes related to inflammation, insulin resistance, glial differentiation, and development in TH and B6 mice.

Methods: To assess the effect of diet on cerebellar mRNA expression, male and female TH and B6 mice were weaned onto chow and high fat (HF) diets at 4 weeks of age and maintained on these diets throughout the study. Mice were then euthanized at 5 and 20 weeks of age, and total RNA was isolated from their cerebellums. Real-time RT-PCR for Cox2, IL6, IL1b, Mcp1, Tnfa, Ins2, Irs1, Irs2, Glut4, Lrp1b, Bdnf, ApoE, Igf1, and Gfap was then conducted in SYBR Green PCR core reagents. Data were analyzed using a twoway ANOVA, with main factors of strain (TH vs B6) and diet (HF vs chow), followed by multiple comparison posttests with Bonferroni corrections, using GraphPad Prism 8 software.

Results: We found that the expression of Gfap and Igf1 were significantly reduced in TH compared to B6 mice, and Gfap expression appeared to be further reduced in mice on the HF diet.

Conclusion: Because GFAP is a major astrocyte protein and IGF-1 is a major regulator of lifelong neural development and plasticity, these changes in gene expression may contribute to differences in cerebellar function that decrease locomotion and coordination in TH compared to B6 mice.

Tumor-Suppressor Role of the Caveolar α1-Na/K-ATPase Signalosome in NASH related Hepatocellular Carcinoma

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Background: Hepatocellular Carcinoma (HCC) is the second cause of cancer- related mortality worldwide. In the Western countries, due to the epidemic of obesity, non-alcoholic steatohepatitis (NASH) has become the major cause of HCC

Study Hypothesis: We hypothesized that, the growing uncoupled metabolism during NASH progression to ESLD and HCC manifested by lower cell oxi-redox status and an apoptotic 'switch' activity, follows a disarrangement in the wild type NKA α1/Src signalosome with dysregulation of the caveolar-α1 subunit promoting an amplification of the pSrc →PI3K →Akt pathway as signaling for survivin overexpression.

Methods: Expression of Cav-1/Smac-Diablo/Survivin proteins was performed by confocal-microscopy on immunostained HCC cell lines (Hep3B and SNU475), and livers in both NASH and NASH-HCC rodent models, and in humans. Signaling pathway studies were explored in-vitro. Selective blockage of Src-p at its kinase domain was performed by administration of a synthetic peptide (pNaKtide). Significant differences among groups were established at $p<0.05$ using ANOVA/Turkey's post hoc test.

Results: Blockage of Src-p at the α1-NKA promoted apoptosis of Human HCC cell lines. pNaKtide at IC50 drove the downregulation and upregulation of Survivin and SMAC expressions ($p<0.05$), respectively. In-vivo, liver tumor burden was significantly lower in animals treated with pNaKtide vs non-treated animals ($p<0.01$). Furthermore, Cav-1 and Survivin expressions were significantly higher, while SMAC protein expression was significantly lower in livers from rodents with NASH/HCC vs animals treated with pNaKtide ($p<0.01$). Similar pattern of proteins expressions was noted in tumors from patients with NASH±HCC vs liver tissue from healthy subjects ($p<0.05$). In-vitro, Src-p at the α1-NKA activates PI3K/Akt pathway.

Conclusion: Src-phosphorylation by the α1-NKA at Caveola regulated Survivin/SMAC expressions, leading to cellular "switch" from apoptosis to cell division via the PI3K/Akt pathway. Therefore, point target blockage may be explored as a treatment for tumor regression.

Nephrotoxic Potential of Three Dichlorobenzene Isomers in Isolated Kidney Cells from Female Fischer 344 Rats

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Background: Dichlorobenzenes (DCBs) are used in the manufacture of many products. DCBs induce nephrotoxicity in male rats via the α2u-globulin nephropathy mechanism. The blood borne DCB complex with this protein accumulates in the kidney, promoting protein droplet formation, leading to cell death and tumor formation. A recent study from our laboratory showed that the decreasing order of direct DCB nephrotoxic potential was 1,4 DCB > 1,3 DCB > 1,2 DCB in isolated kidney cells (IKCs) from male Fischer 344 rats. However, whether DCBs are direct nephrotoxicants to IKC from female rats is unknown.

Study Hypothesis: Given that female rats don't make α2u-globulin, a similar result to our previous study would show that the mechanism of in vitro nephrotoxicity for DCBs may not involve just the α2u-globulin pathway seen in the male rats, but another pathway shared by both genders or by different pathways.

Methods: IKCs (4 million cells/ml, 3 ml) were incubated with a DCB (0.25-1.0 mM) or dimethyl sulfoxide (DMSO) for 30 or 60 minutes with shaking at 37 C° under a 95% oxygen/5% carbon dioxide atmosphere. Cellular death was determined by measuring lactate dehydrogenase

(LDH) release.

Results: DCB isomers were found to be cytotoxic to female IKC, with the order of decreasing nephrotoxic potential found to be 1,3 DCB > 1,4 DCB > 1,2 DCB in the IKCs from female Fischer 344 rats.

Conclusion: A different nephrotoxic potential order is seen with female IKC than with male IKC. This difference may signify that the nephrotoxicity induced by DCB isomers is governed by different pathways in females than in males. The direct cytotoxicity of DCB observed in female IKC suggests that the α2u-globulin pathway is not required for direct effects of these compounds on the kidney. This work was supported in part by NIH grant P20GM103434.

Novel Protocol for Dual Immunofluorescence of Growth Plate Cartilage

Authors: Brett Johnson, L.J.E. Grace Kesler, Cassaundra A. White, and Maria A. Serrat

Department & Institution: Biomedical Sciences, Marshall University Joan C. Edwards School of Medicine

Background: Immunostaining is commonly used to identify biomarkers in tissue sections. This technique is especially useful for localizing proteins in heterogeneous tissues such as cartilage plates of elongating bones. Standard single-antibody staining is time- and labor-intensive due to secondary antibody and chromagen steps that must be separately applied for each protein. Simultaneous detection of multiple biomarkers using direct fluorescently-labeled primary antibodies would improve efficiency by reducing time and materials. IGF-I is the major stimulator of chondrocyte proliferation and bone elongation in growth plates. Its phosphorylated receptor, pIGF-IR, is a biomarker of IGF-I signal activation. PCNA is a biomarker of cell proliferation. We developed a dual immunofluorescence protocol for pIGF-IR and PCNA in growth plate cartilage using commercial antibodies not yet validated for paraffin immunostaining.

Study Hypothesis: We hypothesized that double-label immunostaining would be viable for simultaneous detection of pIGF-IR and PCNA in growth plates.

Methods: Paraffin-embedded growth plates of juvenile mice (N = 112) were subject to standard antigen-unmasking and blocking. Commercially available fluorescently-conjugated primary antibodies against pIGF-IR (Alexa 647) and PCNA (Alexa 488) were applied (1:200) and incubated overnight. Fluorescence images (20X) were captured.

Results: PCNA and pIGF-IR fluorophores were both visible in the growth plate. Overlapping images confirmed colocalization in chondrocytes. Single-antibody stained images showed no spectral overlap between fluorophores, and the negative control lacked background staining.

Conclusion: Results support our hypothesis that double-label immunostaining with pIGF-IR and PCNA is a feasible approach for examining co-localization in growth plates. Our novel protocol saves time and reagents compared to traditional methods and will be useful in future bone research applications.

Sex, Age, and Exercise Effects on Bone Density and Muscle Mass in Rats

Authors: Autumn Pennington, Habiba Chirchir, Kumika Toma

Department & Institution: Exercise Science, Marshall University

Background: The musculoskeletal system responds to various internal and external changes to maintain its ability to support the body weight and provide motion. Bone mineral density (BMD) tends to decline due to aging and decreased physical activities. During inactivity, such as space travel, skeletal muscle contraction decreases causing decrease in BMD. Considering the length of time in space and the age of astronauts, significant BMD loss and adverse health effects are major concerns. One of countermeasures to microgravity-induced muscle atrophy and BMD loss is keeping muscle contraction during fight. However, it is not well-known the sex difference on BMD.

Study Hypothesis: The stretched leg (exercised) of male rats will have the highest BMD, and the control (HLS) leg female rats will have the lowest BMD. The exercised legs will have a higher BMD compared to the controlled legs among all group.

Methods: Old (average 30-month-old) male and female F344/BN rats were divided into 2 groups (control, 2-week HLS). During HLS, one leg was stretched while other leg was free-from resistance. Body weight, wet muscle mass (soleus and gastrocnemius), BMD at tibia shaft and femur condyle were measured using pQCT. Data was compared by one-way ANOVA and 2-way repeated measure of ANOVA.

Results: Two-week of HLS resulted in significant body weight loss for both sexes ($p > 0.001$). Weights of soleus and gastrocnemius were significantly reduced by HLS but not by stretching ($p>0.001$). BMD of tibia shaft was not significantly affected by HLS or stretch among both sexes, while BMD of femur condyle was significantly lowered by HLS among male rats ($p=0.034$).

Conclusion: Exercise attenuate muscle mass loss in both sexes but BMD may be affected by sex differences. These could be due to a short experimental duration, smaller sample size or unknown reasons and further investigation is necessary (partially supported by NNX15AI01H).

Western Diet Accelerates Initiation and Progression of Myelodysplastic Syndrome in Mice

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Department & Institution: Department of Pharmaceutical Science and Research, Marshall University School of Pharmacy

Background: While it has been well-established that obesity contributes to the pathogenesis of many solid tumors, recent studies suggest that obesity due to Western diet also contributes to hematologic malignancy.

Study Hypothesis: In this study, we tested the hypothesis that Western diet contributes to more rapid initiation and progression of cancer phenotypes in an established mouse model of del(5q) myelodysplastic syndromes.

Methods: Combined deletion of TIFAB and miR-146a, two genes associated with del(5q) MDS/AML, in mice recapitulates del5q MDS disease phenotypes. Tifab-/-;miR-146a-/- double knock out (DKO) mice are susceptible to MDS-like phenotypes.

Results: In this study, DKO placed on Western diet for 10 weeks display sex-specific weight gain and minor changes in phenotypes associated with MDS when compared to mice on a low-fat control diet. Mice placed on a Western diet for 15 weeks display weight gain, increased spleen weight, and alterations in blood and bone marrow immunophenotyping that suggest more rapid initiation and progression of disease.

Conclusion: These findings underscore the importance of diet in onset and progression of hematologic malignancies in individuals with increased susceptibility to disease.

Antimicrobial Activities of Secondary Metabolites from Model Bryophites

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Background: Many potent antibiotics have fallen victim to a rising bacterial resistance.

Study Hypothesis: The purpose of our research is to evaluate bioactive compounds produced by two model mosses and test their ability to inhibit growth of Gram-positive and negative bacteria. One model moss species we investigate is Ceratodon purpureus, which has a male (R40) and a female (GG1) strain. The second model moss is Physcomitrella patens, which has several different strains, including Gransden, Villersexel and Reute.

Methods: We have initiated analysis of moss methanol extracts and exudates (secondary metabolites secreted by the moss when grown in liquid culture). In order to test our samples, we perform two different tests. The first test is the disk diffusion method (DDM), which provides us with qualitative and semi-quantitative results on the inhibitory zones produced by our samples on bacterial lawns in Petri dishes. The second test, broth dilution method for minimal inhibitory concentration (MIC)

determination, is fully quantitative and allows us to determine the precise metabolite concentration needed to inhibit the growth of bacteria in liquid cultures.

Results: Thus far, our analyses indicate that moss methanol extracts do not harbor any substantial anti-bacterial activities. However, exudates from both moss species contain potent antimicrobial compounds and can inhibit the growth of several different gram-positive bacteria, such as Staphylococcus aureus, Streptococcus pyogenes and Enterococcus faecium.

Conclusion: Overall, our results suggest that bioactive compounds present in model moss exudates can potentially be used for treating infections caused by antibiotic resistant bacteria, such as methicillin-resistant Staphylococcus aureus (MRSA) or vancomycin-resistant enterococci.

microRNAs as Potential Biomarkers for Drug-induced Liver Damage

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Department & Institution: School of Pharmacy, Marshall University; Joan C Edwards School of Medicine, Marshall University

Background: Methamphetamine is a commonly abused psychostimulant. In the United States 1.6 million people were yearly users of Methamphetamine from 2015-2018. Aside from neuronal damage methamphetamine use increases the risk of HIV infection, Hepatitis B, and Hepatitis C. and is also known to result in liver dysfunction. The common diagnostic test for detecting liver damage involves looking at the activity of liver enzymes, but these values may be elevated by other non-liver related causes.

Study Hypothesis: The aim of this study is to assess microRNAs as potential liver-specific biomarkers of drug-induced liver damage. One common model for drug-induced liver damage is the overdose of acetaminophen. In this preliminary study, we used acetaminophen combined with methamphetamine to assess the feasibility of microRNAs as biomarkers for liver damage providing a non-invasive detection of liver damage.

Methods: Ten liver tissue samples from male balb-c mice, five from individuals administered intraperitoneal acetaminophen (300 mg/kg) and subcutaneous methamphetamine (3 mg/kg) and five were treated with saline and methamphetamine (3 mg/kg) as the control. RNA isolation was performed on the tissue samples using the mirVana® miRNA isolation kit (Applied Biosystems) as per the manufactures protocol. QPCR was performed using microRNA specific primers for mir-122, mir-29c, mir223, mir-155 and mir-192 using the TAQMAN® QPCR kit as per manufacturer protocol. Differences in QPCR Ct values were tested using one-way ANOVAs.

Results: RNA yields from 0.1 to 0.2 g of tissue yielded concentrations ranging from 71.6 to 1,015 ng/ul, sufficient for running multiple QPCR assays with replication for the 5 microRNAs. All five microRNAs were detectable in all samples. Two of the microRNAs, mir155 and mir223, showed significant increased expression in the treatment samples compared to the control treatment (mir155, $P>0.003$; mir233 $P<0.00007$).

Conclusion: These preliminary studies have identified two microRNA candidates for biomarkers for liver damage.

Impact of HSP90 inhibition on epigenetic drift in hematopoietic stem cells

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Background: "What are the molecular defects that promote age-related dysfunction?" The emerging importance of epigenetic gene regulation in the aging process necessitates not only our understanding of which genes are potential targets, but how the process of epigenetic drift results in dysfunctional stem cells. Understanding the mechanisms that promote evolution of the epigenome and its increasing dysfunction with age in stem cells are critical to therapeutic strategies targeting aging. Thus, we propose to investigate canalization as a mechanism of molecular evolution of the epigenome in a mammalian system of epigenetic drift and clonal hematopoiesis. The results of our investigations in the Drosophila and murine model systems indicate a very important gene in canalization, heat shock protein 90 (HSP90), is well connected to epigenetic gene regulation

and phenotypic plasticity.

Study Hypothesis: This proposal is designed to test the hypothesis that inhibition of HSP90 results in changes in histone acetylation that in turn prevent epigenetic drift and loss of phenotypic plasticity in hematopoietic stem cells.

Methods: The approach is to test the hypothesis in vitro using our epigenetic drift model. We will exploit the connection between epigenetics and cellular differentiation in studies of differentiation using our EML hematopoietic stem cell model. The major tool of investigation will be flow cytometry analysis of differentiation and epigenetic drift of cells from a stem cell state into a progenitor state. Modulation of HSP90 levels will be performed in this system followed by analyses of the ability of hematopoietic cells to functionally produce mature cells of the immune system.

Results: Prior treatment of EML cells with EC50 levels of the HSP90 inhibitor, AUY-922 prevented epigenetic drift as measured by the loss of the stem cell marker Sca-1.

Conclusion: The results of this study will provide a basis for an understanding of the contribution of evolutionary theory to age-related dysfunction.

Methamphetamines and the Effects on the Heart

Authors: Lauren Morilla

Department & Institution: Department of Kinesiology, College of Health Professions

Background: The chordae tendinea are an essential part of the heart and papillary muscles due to how it prevents mitral valve prolapse and regurgitation. The chordae tendinea originate from the fibrous heads of the papillary muscle and insert to the leaflet. The chordae anchor the atrioventricular valve to the wall of the ventricle which prevent the backflow of blood by stopping valve leaflets from inverting. Ruptured chordae affect the atrioventricular heart valves and is one of the primary causes of valve regurgitation. The use of methamphetamines causes an increase in catecholamine toxicity, which, causes alteration to the myocardium and remodeling/structural damage through electrical remodeling. The effects of methamphetamines on chordae tendinea and the heart is not yet fully understood.

Study Hypothesis: It was hypothesized that damage would occur on the chordae tendinea.

Methods: Rat hearts were obtained from a previous study of Dr. Hambuchen (School of Pharmacy). Rats were administrated 1mg/bw kg of methamphetamine, 3 times a day for 4 days. Then, rats were euthanized by opening the chest cavity to induce pneumothorax under isoflurane-induced anesthesia and stored in a -80o C freezer for further analyses. In this study the hearts were examined one at a time. The frozen hearts were defrosted in a cold phosphate buffer solution (in the refrigerator) overnight, then, carefully opened to expose the chordae tendinea. The opened heart was then placed in a buffer solution in a petri dish and observed under a stereomicroscope. The number and degree of chordae tendinea rupture and/or damage were recorded, and photographs were taken for each heart.

Results: A significant lack of chordae tendinea, along with significant thinning and ruptured chordae, was found.

Conclusion: The data collected supports the clinical observation that methamphetamine use is dangerous to the central nervous system but also to the cardiac function by changing the heart's structure.

The association of the α1-Na/K-ATPase signalosome with Chaperone Mediated Autophagy in NASH related HCC.

Authors: Levi Nolan, Joshua Keefer, Utibe Udoh, Pradeep Rajan, Yuto Nakafuku, Sodhi K, Pierre S, Xie Z, Shapiro J, Juan Sanabria

Department & Institution: Department of Surgery, Marshall Institute for Interdisciplinary Research, Marshall University Joan C Edwards School of Medicine

Background: Chaperone mediated autophagy (CMA) is a degradation process that helps regulate many cellular processes, such as DNA repair and apoptosis. CMA is drastically downregulated in hepatocellular carcinoma (HCC), the second most common cause of cancer death in the world. We aim to determine the association of the α1-Na/K-ATPase signalosome with CMA in NASH related

HCC. We are also observing the normalization of α1 by pNaktide (a synthetic 33 peptide synthetized for the NH2 side of the α1-subunit preventing its binding to Src) in healthy human hepatocytes and two human HCC cancer cell lines (Hep3B&SNU475).

Study Hypothesis: We aim to determine the association of the α1-Na/K-ATPase signalosome with CMA in NASH related HCC.

Methods: The methods of this experiment followed the procedures of the Autophagy Detection Kit by Abcam (ab139484). CMA activity of all cell lines was assessed by confocal and ELISA techniques under untreated and treated conditions. α1 subunit signaling was normalized by cell exposure to pNaKtide. Statistical significance was accepted at the 0.05 level.

Results: There was not an increase in CMA activity in the human normal hepatocytes after treatment compared to the untreated group ($p>0.05$). In contrast, both HEP3B & SNU475 showed significantly upregulated CMA activity in the treated group when compared to the untreated group (p0.05).

Conclusion: Normalization of the α1-Na/K-ATPase signalosome upregulated CMA activity in human HCC cell lines with no effect on normal human hepatocytes. This finding, if corroborated in vivo may see translation into target therapy for HCC.

The Effect of Continuous Sugar-Sweetened Beverage Consumption on Weight in Mice and Adipokine Expression in Preadipocyte Cells

Authors: Samuel Tetteh-Quarshie, Cynthia B. Jones

Department & Institution: Pharmaceutical Science and Research, Marshall University School of Pharmacy

Background: As obesity has escalated to epidemic proportions around the world, many causes, including high sugar consumption, has been suggested. Excessive caloric intake, especially for diets high in simple sugars such as sucrose and high-fructose corn syrup (HFCS), cannot be ruled out as contributors to this epidemic. Sugar-sweetened beverages (SSBs) are the highest single contributor to dietary fructose intake in the US diet. Consequently, the impact of excessive sugar consumption on health has become a debated topic among food experts. Hence, the objective of this study was to determine the effects of continuous SSB consumption on weight and adipokine expression in preadipocyte cells.

Study Hypothesis: We hypothesize that continuous consumption of SSB will induce weight gain in mice and alter adipokine expression in preadipocyte cells.

Methods: 3T3-L1 preadipocyte cells were cultured and differentiated in recommended differentiation medium supplemented with stock sugar solutions. Lipid accumulation was quantified with Oil Red O assay. C57BL/6 male mice were fed normal rodent chow and water with the following sugar sweetened beverages (SSB): 0% control, fructose (10% solution), glucose (10% solution) and a combined fructose/glucose (55% fructose and 45% glucose for a 10% solution) for 16 weeks. Animal weights were recorded weekly and sugar sweetened beverages were measured and replaced every two days.

Results: SSB exposed cells has significant intracellular lipid accumulation compared to the control. SSB-fed mice exhibited an addictive behavior with significant increase in SSB consumption, and weight gain. Mice exposed to glucose and fructose had similar weight gain. mRNA adipokine gene expression was greatly downregulated when preadipocyte cells were exposed to each SSB model.

Conclusion: Mice exposed to each SSB model consumed a significant volume of sugar beverage and gained significant amount of weight. Understanding the deleterious effects of over consumption of high sugar diet is paramount to avoiding overweight and obesity-related diseases.

Proteomic analysis provides evidence for metabolic reprogramming in clear cell renal cell carcinoma (ccRCC)

Authors: Anisha Valluri, Chelsea Thompson, Logan M Lawrence, Rebecca Russell, James Denvir, James C Jensen, Krista L Denning and Travis B. Salisbury

Department & Institution: Department of Biomedical Sciences, Department of Pathology, Department

of Oncology, Joan C Edwards School of Medicine, Huntington, WV

Background: Prior reports show ccRCC shows increased utilization of aerobic glycolysis and lactic acid accumulation to provide rapid energy for metabolism, however, the protein expression changes that mediate this shift in metabolic reprogramming in renal tumors is still an important knowledge gap.

Study Hypothesis: We hypothesized that specific proteins/enzymes that promote aerobic glycolysis and lactic acid production are upregulated in renal tumors compared with normal renal tissue.

Methods: The objectives of this study were to conduct reverse phase protein arrays (RPPA) to simultaneously detect the protein expression levels of 450 proteins and phospho-proteins that regulate cancer progression including changes in cancer cell metabolism. Using RPPA, we analyzed proteomic expression patterns of 44 clinical ccRCC samples (22 renal tumors and 22 pair-matched normal renal tissue controls). Samples were obtained from patients of the Edwards Comprehensive Cancer Center, Huntington WV.

Results: The RPPA results showed that of 450 proteins, 274 were statistically significantly differently expressed in renal tumors compared with normal renal tissue. Of these 274, 47 showed a fold change that was greater than 50%. Importantly, the expression of six enzymes in the oxidative glycolysis pathway were significantly increased in tumors compared with normal tissue, including a 4.8-fold increase in Hexokinase 2.

Conversely, the levels of four proteins in oxidative metabolism were significantly reduced in renal tumors, including a 5.86 fold reduction in the expression of Mitochondrially Encoded Cytochrome C Oxidase I (MTCO1). The results also showed tumors expressed higher levels of Lactate Dehydrogenase A (LDHA), which would promote lactic acid accumulation.

Conclusion: Collectively, the findings of this study support prior metabolic studies showing that tumors undergo a change in metabolism that drives oxidative glycolysis at the expense of reduced oxidative metabolism. We have identified the proteins in renal tumors that might mediate this metabolic reprogramming.

Salmonella Infection in Diabetic Mice

Authors: Cecilia G. Sierra-Bakhshi, Michael E. Smith, Lydia M. Bogomolnaya

Department & Institution: Biomedical Science, Joan C. Edwards School of Medicine, Marshall University

Background: In West Virginia, approximately 16.2% of the population is diagnosed with diabetes mellitus type 2 (T2D) compared to an average of 10.5% for the rest of the population. In addition to having a chronic condition, diabetic individuals are also at higher risk for developing severe, often life-threatening extraintestinal bacterial infections with non-typhoidal Salmonella. The underlying mechanism of this predisposition is not clearly understood, in this study, we utilized 8-week TALLYHO (TH) mice to establish a model of salmonellosis in a diabetic host.

Study Hypothesis: To establish a model that will form a foundation for the study of Salmonella pathogenesis in the diabetic host and to define options for preventing the extraintestinal spread of non-typhoidal Salmonella.

Methods: Eight weeks old TALLYHO mice were maintained on standard chow, or on a high-fat diet (45% fat) for 8 weeks to promote diabetes development. Mice were separated into groups based on their blood glucose level and infected with 10^6 colony forming units (CFU) of a fully virulent bioluminescent Salmonella Typhymurium strain to follow the pathogen spread in individual animals using the IVIS Lumina XRMS invivo imaging system.

Results: As expected, mice on the high-fat diet gained more weight compared to the animals on the standard chow. In addition to weight gain, by 16 weeks of age mice in the high-fat diet had developed diabetes. Mice from both groups developed clinical signs of salmonellosis. However, Salmonella spread in diabetic mice had an unusual pattern compared to healthy mice.

Conclusion: We collected feces from TALLYHO mice over the course of 8 weeks to understand the changes in microbiota composition during diabetes development, established the infection dose to study S. Typhimurium pathogenesis in TALLYHO mice, and utilized the power of in vivo imaging to study

Salmonella spread in TALLYHO mice, and found the altered pattern for the pathogen spread in a diabetic host.

Tissue-specific reduction of Na/K-ATPase in mice uncovers a new mechanism of regulation of sodium balance and systemic blood pressure by the renal proximal tubule

Authors: Kailey Stuart, Shreya T. Mukherji, Muhammad A. Chaudhry, Jiang Liu, Joseph I. Shapiro, Gustavo V. Blanco, Zijian Xie, Sandrine V. Pierre

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Background: Adequate reabsorption of salt and water in the renal proximal tubule (RPT) is an independent determinant of blood pressure (BP). Rather than the classic cardiotonic steroid (CTS)-mediated inhibition of Na+/K+ATPase (NKA) ion-transport in the RPT, low (physiological) concentrations of CTS initiate NKAα1/Srcmediated signaling to reduce apical Na+/H+-Exchanger-3 (NHE3) and transepithelial sodium flux in the RPT. Consistently, mice with genetic suppression of 70% of RPT NKAα1 (RPTα1-/-) lose signalingmediated inhibition of NHE3, which increases RPT sodium reabsorption by 65%. Remarkably, this sodium hyper-absorptive phenotype was not accompanied by hypernatremia at 4 months. However, sodium overload and elevated BP have not been excluded.

Study Hypothesis: We hypothesized that, although not hypernatremic at 4 months, the RPTα1-/- mice exhibit early manifestations of sodium overload and an elevated BP.

Methods: Four month old male RPTα1-/- and RPTα1+/+ (control) mice were examined using the CODA tail-cuff BP system (n=11). BP was collected from conscious mice following 3 days of training and 5 acclimation cycles were included each day. Serum collected from the mice were analyzed for aldosterone and atrial natriuretic peptide (ANP) levels by commercial ELISA.

Results: RPTα1-/- mice exhibited a decrease in serum aldosterone (172±22 vs 310±35 pg/mL in RPTα1+/+, $p<0.01$) and an increase in ANP (660±68 vs 386±27 pg/mL in RPTα1+/+, $p<0.01$, n=7), indicative of increased sodium retention. Consistently, RPTα1-/- mice had elevated systolic BP (122±4 vs 105±2 mmHg in RPTα1+/+, $p<0.01$), and mean arterial pressure (103±4 vs 86±2 mmHg in RPTα1+/+, $p<0.01$), with no difference in the heart rate (488±14 vs 497±14 beats/min in RPTα1+/+, $p=0.7$).

Conclusion: NKA receptor-mediated regulation of NHE3 and Na+ transport in the RPT is critical to systemic blood volume and pressure homeostasis. NKA signaling therefore provides a long sought-after mechanism for the natriuretic action of endogenous NKA ligands such as cardiotonic steroids.

The Promise of Exosome-Based Therapy for Central Nervous System Diseases

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Background: Exosomes are a major type of extracellular vesicles. In the central nervous system, they can be released from virtually all brain cells, including neurons, astrocytes, microglia, and endothelial cells. Over the past decades, increasing evidence shows that exosomes could serve as a novel type of cell-to-cell communicators via conveying of their carried biological cargoes, such as nuclear acids, proteins and lipids, thereby altering the biological functions of recipient cells in nearby and distal tissues or organs.

Study Hypothesis: Given the capability of exosomes to cross the blood-brain barrier and connect origin cells with target cells, exosomes hold great clinical application potentials for the central nervous system diseases by serving as biomarkers and therapeutic approaches.

Methods: Here, we reviewed the current state of the knowledge of exosomes, the roles and applications of exosomes as a viable pathological biomarker and exosome-based therapy for central nervous system diseases.

Results: Exosomes play a physiological role and are implicated in the pathogenesis of central nervous system diseases including stroke, vascular dementia, Parkinson's disease, Alzheimer's disease, and traumatic brain injury. Preclinical studies have suggested that administration of exosomes elicits neuroprotective and neurorestorative effects in rodent central nervous system models.

Compared to cell-based therapy, exosome-based therapy has the advantage of simple preservation and transfer, modification potential, blood-brain barrier permeability, inability to proliferate, and lack of risk of cellular injection-induced vascular occlusion.

Conclusion: Exosomes have a great promising of being used as biomarkers for diagnosis and prediction of central nervous system diseases, as well as a cell-free therapy for treating central nervous system diseases.

Sensing force dynamics is common to control of walking in humans, insects and robots

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Background: Force feedback could be valuable in adapting walking to diverse terrains but the effects of changes in substrate inclination on sensory discharges have not been examined. In insects, force feedback is provided by campaniform sensilla, mechanoreceptors that monitor forces as cuticular strains. In previous studies, we have examined how campaniform sensilla on the stick insect tibia encode 'naturalistic' force stimuli derived from joint torques measured in animals walking freely on level ground.

Study Hypothesis: Our major hypothesis is that insects, like humans, use sensory signals for force dynamics in motor control.

Methods: In the present study, we analyze the FT torques in more detail and record the responses of campaniform sensilla to the mean torques and the torques of individual steps that showed large variations (as naturally occurring 'perturbations').

Results: We find that in level walking, FT torques in different directions tend to occur in different ranges of joint angles (flexion torques in ranges of joint extension, extension torques in ranges of joint flexion). Extracellular recordings of campaniform sensilla in response to these torques indicate that sensory signals encode the torque direction and strongly reflect the rate of change of force (dF/dt) in walking on all substrates. Discharges also reflect the force magnitude on slopes but are modulated and follow dF/dt.

Conclusion: These findings are now being studied in a model that accurately simulates the properties of leg campaniform sensilla (Szczecinski et al., Living Machines, 2020). Future experiments are planned to examine the effects of sensilla on motor activities. Our working hypothesis is that sensory feedback indicating force dynamics (dF/dt) at the FT joint in the hindleg can be used to stabilize and adapt walking to diverse terrains.

The effect of umbilical cord essential and toxic elements, thyroid levels, and Vitamin D on childhood development.

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Background: The in-utero environment has been shown to have dramatic effects on childhood development. Little is known regarding the potential for adverse cognitive function and metabolic dysregulation for infants and children exposed to essential and toxic elements, thyroid levels, and Vitamin D during the prenatal period.

Study Hypothesis: We hypothesized that umbilical cord essential and toxic elements, thyroid levels, and Vitamin D effect childhood development.

Methods: From April 3, 2013 to January 30, 2014 umbilical cord was collected and analyzed at the time of delivery for 20 different essential and toxic elements, thyroid levels, and Vitamin D. A retrospective review was performed of well-child examinations from birth to 7 years old. Blood pressure, weight, and developmental milestones were extracted from the medical record and compared in infants and children whose cord blood was analyzed at the time of delivery.

Results: Data was available for 60 patients. There were associations with calcium and 9 month BMI (p=<.01), barium and 6 year old height (p=<.01), magnesium and 15 month gross motor skills (p=.01), mercury and 9 month weight (p=<.01), platinum with 12 month language development (p=<.01), and zinc with 4 year old fine motor skills (p=<.01) and 2 year old fine movement milestones (p=<.01). Elevated Vitamin D was associated with increased delay of 2 year old fine motor development (p=<.01). Thyroid function tests for free T3 were associated with multiple cognitive and physical milestones at less than 1 year old. T3 Uptake was associated with 7 year old weight (p=<.01) and systolic blood pressure (p=<.01). Total T4 was associated with 4 month old cognitive development (p=<.01) as well as 2 and 3 year old height (p=<.01).

Conclusion: There are multiple associations between umbilical cord essential and toxic elements, thyroid levels, and Vitamin D on childhood development.

Low dose Actinomycin D Predominantly Activates p53-p21 Pathway in Aerodigestive Tract Cancers: Implications for Cyclotherapy

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Background: Cyclotherapy is a recent concept to protect normal cells from chemotherapy-induced toxicities. p53-p21dependent cell cycle arrest of normal cells before adding chemotherapy drug is the backbone of cyclotherapy. Drugs capable of preferentially activating p53-p21 signaling are ideal candidates for cyclotherapy. Actinomycin D (ActD) is one of the earliest antitumor antibiotics discovered but its clinical application is severely compromised by toxicity at the recommended dose and its application in aerodigestive tract tumors is rare.

Study Hypothesis: Low-dose ActD predominantly activates the p53-p21 pathway in aerodigestive tract cancers.

Methods: SRB assay and annexin V-PE staining were used to measure cell growth and apoptosis, respectively. CalcuSyn and FlowJo software were used to calculate IC50 and analyze flow data, respectively. Protein expressions were measured by western blotting and mRNA by qPCR.

Results: The IC50 values of ActD spanned between 0.021-2.96nM and induced efficient apoptosis. Mechanistic studies revealed that ActD time- and dose-dependently increased the expression of total and phosphorylated p53 (ser15), and downstream targets p21 and Puma without significant effects on p27. Ablation of p53 expression

using shRNA decreased the expression of p21 and Puma in A549 cell lines but only p21 in H460 cell lines. Interestingly, ActD did not affect Puma mRNA expression in H460 cells and the expression of ActD-induced Puma protein expression increased after ablation of p53 in these cells suggesting posttranscriptional regulation of Puma by ActD in H460 cells. In cells with mutant p53 (PC-9), the effect of ActD on p21, p27, and Puma expression is very low as compared to cells with wild-type p53.

Conclusion: ActD is effective against aerodigestive tract cancers and context-dependently activates p53 target genes. p53-p21 is the predominant pathway activated by low-dose ActD and has implications for cyclotherapy.

Synovial fluid absolute neutrophil count a promising marker for diagnosing periprosthetic joint infection

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Department & Institution: Marshall Orthopaedics

Background: With no gold standard for diagnosing periprosthetic joint infection (PJI) clinicians who encounter a suspected PJI case have to use a combination of tests. Several studies have indicated the importance of absolute neutrophil count (ANC) in systemic infections. However, this test has not yet been investigated in synovial fluid (SF).

Study Hypothesis: In this study the performance of SFANC was assessed and compared to SFPMN% and SFWBC.

Methods: We conducted a retrospective multicenter study reviewing the clinical records of patients undergoing revision surgery from 2017 to 2020. Patients who had full set of SFWBC, SFPMN%, and SFANC were included in the study. Our cohort consists of 231 patients that were divided into two groups: aseptic revisions (N=136) and septic revisions (N=95). Sensitivity, specificity, positive and negative likelihood ratio (LR), and diagnostic odds ratio (DOR) were calculated for each test. The cutoff for SF absolute PMN was calculated using the Youden's Index (>1950 cells/μL).

Results: SFANC had a sensitivity of 88.4%, specificity of 85.2%, positive and negative likelihood ratio of 6.0 and 0.1, and a DOR of 44.2 (95% confidence interval [CI]: 20.1-97.3). SF WBC showed 84.2% sensitivity, 83.8 specificity, 5.2 +LR, 0.1 –LR, and 27.6 (95%CI: 13.5-56.5) DOR. Synovial PMN% had a sensitivity of 80.0%, a specificity of 80.8%, + and – LR of 4.1 and 0.2 respectively, and a DOR of 16.9 (95%CI: 8.732.7). SFANC with an area under the curve (AUC) of 0.93 was a significantly better predictor of PJI than both SF WBC (AUC=0.91, p=0.007) and SF PMN% (AUC=0.88, p=0.016). The AUC was comparable for SF WBC and SF PMN, p=0.16 .

Conclusion: Based on the findings of the current study, it appears that SFANC has a better performance for diagnosing PJI than SFWBC and SFPMN%. We recommend that the orthopaedic community to consider this test in diagnostic work up for PJI.

Effects of Physician Beliefs in Benevolent Sexism on Recommendations for Patients with Knee Arthritis

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Background: Women hold a greater prevalence of knee arthritis, experience greater arthritic disability, and report worse pre-operative pain and lower physical functioning than men (AHRQ, 2015). However, men are 22 times more likely to receive recommendations for total knee arthroplasty (TKA; Borkhoff et al., 2008). These results are concerning, given that women and men who undergo TKA gain equivalent postoperative functioning (Fitzgerald et al., 2004).

Study Hypothesis: The present research experimentally investigated the potential impact of physicians' beliefs in benevolent sexism on recommendations for patients with osteoarthritis. It was predicted that physicians who held beliefs consistent with benevolent sexism would be: 1) more likely to recommend invasive procedures to men patients, and 2) more likely to recommend conservative treatment options to women patients.

Methods: Participants included 130 orthopedic surgery residents, fellows, and attending physicians. A recruitment email directed participants to an online survey where they completed the Ambivalent Sexism

Inventory (Glick & Fiske, 1996), read a clinical case vignette developed and pretested by orthopedic surgeons to manipulate patient gender and elicit 50/50 recommendations for conservative treatment options vs. TKA, and made recommendations for care.

Results: The first hypothesis was not supported. The second hypothesis was supported. Specifically, physicians were more likely to recommend non-narcotic analgesics to the woman patient when they held beliefs consistent with benevolent sexism; no differences were found for the man patient.

Conclusion: Differences in TKA recommendations were not found. However, when examining the data of physicians who held more than five years of experience, we noticed a trend toward TKA recommendations being impacted by patient gender and physician beliefs in benevolent sexism. This trend may indicate that bias is acquired during physicians' years in practice; we intend to explore this further. The present research represents an important step toward understanding how physician bias might impact the allocation of healthcare for women.

Assessment of Elements Affecting Nutritional Status in Elderly West Virginians

Authors: Andrew S. Ferguson, Robert Walker, Courtney Wellman, Paris Johnson, Adam M., Franks

Department & Institution: Family Medicine, Marshall University Joan C. Edwards School of Medicine

Background: As advancements in medicine progress, the population aged 85 and older increases. Of this population not much is known about the relationship between nutritional status while living alone versus living with a partner or family, especially for those dwelling in the Appalachian area, as many of them live in rural areas. Studying the effects of isolation and living arrangements on nutritional status would enable healthcare providers to potentially better the health of this aging population.

Study Hypothesis: It is hypothesized that the nutritional status of the independently living population 85 years and older is correlated to: gender, BMI, living arrangement, and degree of isolation from society (suburb versus rural).

Methods: A random population of Oldest Olds living in Cabell, Wayne, and Lincoln counties in West Virginia were asked to answer a survey about the number of occupants in their home, their home setting (suburb, rural, isolated), and a Mini Nutritional Assessment.

Results: Of 170 surveyed, 8 were considered malnourished and 32 at risk of being malnourished. Of the malnourished, 7 are living alone, 6 are living in either a rural or isolated setting, and 6 are underweight. On average, those who are malnourished are older (87.88 years) than those at risk (85.66 years), or normal (84.65 years). Of the 130 with normal nutrition, 38 were obese, 51 overweight and 0 underweight.

Conclusion: The data accepts the hypothesis that nutritional status is correlated to living arrangement and BMI. The number of occupants living at home had a larger impact on BMI than home location did. The obesity rate was significantly higher in those with a normal nutritional status. Malnourishment carries a higher risk of organ failure, increased infection susceptibility, and perioperative complications. As expected, there is an increased risk of malnutrition as patients age.

Parental Attitudes on Epinephrine Use during COVID-19 Pandemic

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Department & Institution: Pediatrics, Marshall Health

Background: Currently if parents use an epinephrine autoinjector in response to their child's food anaphylaxis they are to seek medical care for evaluation. However, the recent COVID-19 pandemic resulted in major shifts in avoiding healthcare utilization. We sought to determine if parental attitude regarding epinephrine use and post-administration treatment was altered by the COVID-19 pandemic.

Study Hypothesis: Parents will show increase in health care avoidance during COVID after administering epinephrine.

Methods: Electronic medical record search of Allergy/

Immunology patients with a food allergy diagnosis was performed. Exclusion criteria were no epinephrine prescription, or not a current patient. Parents were interviewed regarding previous use of epinephrine and follow up care to that event. They were asked if and why their attitude during the COVID pandemic changed regarding post epinephrine medical care utilization. IRB approval was obtained.

Results: Of 392 patients, 84 were not reached, 129 declined, and 25 met exclusion criteria. Twenty-three of the 154 eligible patient parents had used an epinephrine autoinjector prior to the pandemic. Post pandemic, 96.1% indicated they would seek medical evaluation post-epinephrine use. Only six patients (3.9%) would avoid going to the hospital due to COVID exposure. Nine (5.8%) parents indicated that the COVID-19 pandemic had changed their thoughts on treating allergic reactions with epinephrine.

Conclusion: Parents of children with food allergy resulting in anaphylaxis would still overwhelmingly use their epinephrine and seek medical care after use during the COVID-19 pandemic. A minority would choose to stay home or an alternative route of care, with only two patients citing fear of exposure to COVID-19 and perceived increased treatment time

Identifying Factors that Predict Positive Testing during a Pandemic in a Division I Sports Medicine Program

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Background: In December 2019, the outbreak of SARS-CoV-2, a coronavirus causing a severe acute respiratory syndrome was discovered in Wuhan, China. Within months, a worldwide public health emergency was declared. The high infectivity of COVID shut down nearly all public gatherings and the medical system was pressed to provide personal protection equipment, hospital beds, and ventilators. The 'stay at home' orders specifically impacted athletes with practice restrictions, game cancellations, and season delays. This threatened the athletes conditioning, as well as scholarships and future livelihoods. Sports teams fall into high-risk populations with close contact during practices, living situations, and traveling to games. No established best practice protocol exists to monitor this high-risk population. To develop a protocol, understanding the factors that drive testing and positive results is the first step.

Study Hypothesis: A partially self-contained population, such as a collegiate athletic program, has identifiable factors that can predict COVID risk and testing volume that is independent from the community population.

Methods: The Marshall University Athletics sports medicine division created an evolving protocol revolving around prevention, testing, and follow-up to protect the staff and student athletes (May 2020). Testing numbers and positive test results within the athletic department were compared with local state numbers of positive cases.

Results: Superimposed linear trends of state positive cases did not correlate with linear trends of testing or positive cases within the athletic department. Similar analysis against local counties data will be conducted next.

Conclusion: A sports program was its own confined system, independent of state numbers. Peaks in positive tests were influenced by specific sport seasons and campus activities, rather than the positive cases in the community.

Antifungal Activity of Antabuse and its Primary Metabolite with Copper, against Fluconazole Resistant Candida strains

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Background: Candida is a normal yeast flora, but can cause severe infection as an opportunistic pathogen in immunocompromised patients. Hospital-borne infections involving fluconazole-resistant Candida species are often encountered with isolates of C. glabrata and C. auris. As a rapidly emerging pathogen, the incidence of multidrug-resistance is highest for C. auris and isolates have been found to be resistant to all three available antifungal classes. The focus of this research was to establish if disulfiram (Antabuse) exhibits antifungal activity with and without copper against C. glabrata and C. auris.

Study Hypothesis: We hypothesized that disulfiram (DSF) exhibits synergy with copper sulfate (CuSO4) against C. glabrata and C. auris resulting in decreased the minimum inhibitory concentrations (MICs).

Methods: The MICs were determined by the microdilution assay method in RPMI-1640 media. Overnight cultures of Candida sp. adjusted to an initial inoculum of 10^3 cells/mL were treated with two-fold dilutions of DSF or its primary metabolite DDTC ± 10 µM CuSO4. Following incubation at 35 °C, the MICs were recorded as the lowest drug concentration that conferred complete inhibition of visual growth after 24 hours.

Results: Copper supplementation to DSF yielded lower MIC values against C. glabrata and C. auris (MIC90 1 µg/mL). Similar results were obtained for DDTC, while CuSO4 addition to the fluconazole had no effect on the MICs.

Conclusion: The addition of CuSO4 to DSF and DDTC decreased the MICs against fluconazole-resistant strains of C. glabrata and C. auris. These results provide a basis for future research into evaluating alternative pharmacologic therapies indicated for resistant Candida infections.

Use of a web-based sex education tool to improve reproductive health knowledge

Authors: Danielle Roth, Jennie Yoost, Morgan Ruley, Kristin Sinning, Jodi Plumley, Maya Menking-Colby, Emma Nellhaus

Department & Institution: Marshall University Joan C. Edwards School of Medicine

Background: At 27.1 births per 1000 females aged 15 to 19 years, West Virginia's teen birth rate is considerably higher than the national average (18.8 births per 1000 females aged 15 to 19 years). This underscores the need for comprehensive sex education that is easily accessible to and intended for West Virginian teens. The purpose of this study is to assess the effect of an online sex education curriculum (marshallteentalk.org) on high school adolescents' reproductive health knowledge and self-efficacy. The sex education curriculum consists of short videos, which can be viewed in approximately 45 minutes, that cover anatomy, puberty, sexuality, healthy relationships, consent, birth control, and sexually transmitted disease prevention. English-speaking females aged 14 to 18 years who attend or recently graduated from a high school in West Virginia, Kentucky or Ohio were eligible to participate. Internet access and an email address were also required for inclusion in the study. Eligible participants were recruited from social media platforms and Marshall Health OB/GYN and Pediatric Clinics and offered a $25 Amazon gift card as an incentive for participation. Participants completed a pre-intervention survey, the sex education curriculum, then a postintervention survey within two weeks of finishing the curriculum. The pre-intervention survey included questions that collected demographic information and assessed reproductive health knowledge and self-efficacy. Reproductive health knowledge was assessed with the Reproductive Health Knowledge Index (RHKI), a previously validated tool. The post-intervention survey included questions that assessed the acceptability of Marshall Teen Talk website and curriculum in addition to the reproductive health knowledge and self-efficacy questions from the pre-survey intervention. Participant pre- and postintervention responses will be compared to quantify the impact of the sex education curriculum on reproductive health knowledge and self-efficacy.

Operative Rib Fixation at a Rural Trauma Center: A Single Institution Retrospective Review

Authors: Dylan Smith, Andrew Feyh, Timothy Kocher, Farzad Amiri, David Denning

Department & Institution: Surgery, Marshall

Background: Operative rib fixation is becoming more common in the management of rib fractures, with studies showing benefits of surgery in decreasing time on mechanical ventilation, ICU and total hospital length of stay, and mortality. This paper will review the data from our institution to determine if these benefits were seen in our trauma population.

Study Hypothesis: Rib fixation patients will be associated with decreased time on the ventilator, decreased ICU length of stay, decreased overall hospital length of stay, and decreased analgesia dependence during the hospital course and on discharge.

Methods: A retrospective review of the institution's trauma database was conducted, including operative patients (n=36), control patients (nonoperative patients from October 2018 to October 2019, n=207), and selected control patients based on similar injury severity score range as operative patients (n=181). Data reviewed included time on mechanical ventilation, ICU and total hospital length of stay, and disposition at discharge, including mortalities. Operative complications were also reviewed.

Results: The operative group had a higher average injury severity score (ISS) compared to both controls, longer average time on mechanical ventilation, longer average ICU and total hospital length of stay, and a higher percentage of patients discharged to inpatient facilities. The operative group had lower mortality compared to the control groups. The operative group had 3 surgical site infections requiring readmission and hardware removal.

Conclusion: Unlike other studies, our operative patients did not see improvements in time on mechanical ventilation, ICU and total hospital length of stay, or disposition at discharge, but did see a mortality benefit. Confounding factors include higher average ISS in the operative group, and oversedation in the ICU. Data collection is ongoing, and refinements are being made to perioperative and ICU management to minimize these confounding variables.

Effects of Increased Opioid Dosage for Geriatric Hip Fracture Patients in an Inpatient Rehabilitation Setting

Authors: Adam Franks, Chris Fitzpatrick, Rabah Boukhemis, Courtney Wellman, Erin Shaver, Lee Chafin, Paris Johnson

Department & Institution: Family and Community Health, Marshall University Joan C Edwards School of Medicine

Background: Hip fractures are a common concern for geriatrics patients. How well one recovers from a hip fracture can determine their future quality of life and longevity. Pain during the recovery and rehabilitation process must be managed and sometimes may be managed using opioids. The amount prescribed may affect the patient's ability to complete the program and, ultimately, recover.

Study Hypothesis: High MME (morphine milligram equivalents) among geriatrics patients in an acute inpatient rehabilitation hospital reduces ability to perform measured self-care and mobility activities of daily living.

Methods: A single-center retrospective chart review was conducted among patients who had a hip fracture and then were transferred to an acute inpatient rehabilitation center in Cabell County, West Virginia from 2020-2021 as part of a quality improvement project. MME was calculated by adding the daily amount of each opioid the patient is prescribed, converting it using a preexisting morphine conversion factor, then calculating the average daily rate. MME of each patient was then compared to their ability to perform self-care, cognition, and mobility activities, as measured by the Section GG standardized patient assessment on admission, throughout their stay, and on discharge. Patients were excluded based on baseline demographics and certain comorbidities.

Results: Data are being evaluated for a correlation analysis. Preliminary results appear to indicate that a higher MME led to better mobility results in this patient group.

Conclusion: It is reasonable to assume that the increased opioid utilization would provide adequate pain control to allow for effective rehabilitation. The implication of a substantial benefit from increased opiates is a disconcerting one. Further research is required to determine the implications of the project beyond the acute rehabilitation setting.

Effect of exogenous testosterone use on factors contributing to increased intraocular pressure and open angle glaucoma

Authors: Gavin Hayes, James Gigantelli MD, Manik Goel MD

Department & Institution: Marshall Eye Surgeons

Background: Open angle glaucoma is among the leading causes of blindness in the United states, with over 2.7 million cases documented in the year 2010. The primary risk factor indicated in this disease is elevated intraocular pressure, which contributes to glaucomatous damages such as optic neuropathy and retinal ischemia. Several case reports and clinical studies mention a link between serum testosterone levels and intraocular pressure, yet there is no quantifiable relationship nor clear mechanism for how this may occur. This study seeks to establish a more robust correlation between supraphysiologic exogenous testosterone use (an increasingly popular therapy), and changes associated with glaucoma or risk of glaucoma.

Study Hypothesis: In a cross sectional medical database search, there will be a significant number of patients who match keywords associated with both exogenous use of testosterone and increases in intraocular pressure or other risk/signs of glaucoma.

Methods: This study uses a deidentified medical database search to cross reference keywords related to testosterone-use to those associated with glaucomatous damage.

Results: The database search yielded 48 patients who met the criteria for both variables established in the hypothesis.

Conclusion: The number of patients who meet the criteria for both variables suggests a relationship between testosterone-use and elevated intraocular pressure/glaucoma, a relationship which was previously disregarded. The current data establishes a precedent for more research, in order to strengthen the validity of this correlation and establish a mechanism for which it might occur.

Retrospective Report of Antibiotic Resistance in Laboratory-Confirmed Bloodstream Infections in a Peruvian Children's Hospital from 2015-2017

Authors: Graham Sutherland, Matthew Murphy, Taryn Clark, Sassan Noazin, Jose Lopez, Carlos Santillan, Robert Gilman

Department & Institution: Department of International Health, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD.

Background: Many bloodstream infections (BSIs) demonstrate antimicrobial resistance (AMR), which is one of the greatest threats to human health. We performed a retrospective analysis of data from the Institución Nacional de Salud del Niño, San Borja (INSN-SB) in Lima, Peru, and report the resistance profiles of various organisms causing BSIs in INSN-SB from 2015 to 2017.

Study Hypothesis: Distinct antibiotic resistance profiles will be observed for various organisms causing LC-BSIs.

Methods: We identified 15,078 blood culture records from between January 1, 2015 and December 31. 2017 from INSN-SB's central database. 2,795 duplicate records, 10,024 negative (i.e., no BSI) records, and 1,688 records that did not meet Centers for Disease Control and Prevention criteria for laboratory-confirmed BSI (LC-BSI) guidelines were excluded. 571 LC-BSIs entered into the final analysis. Corresponding antibiogram data was assessed to determine AMR profiles for the following organisms: Acinetobacter spp., Enterococcus spp., Escherichia coli, Klebsiella spp., Pseudomonas aeruginosa, Salmonella spp., and Staphylococcus aureus.

Results: Below are the antibiotics for which resistance of ≥50% was observed, by organism:

Acinetobacter – Cefazoline, 100% (20/20); Amoxicillin-Clavulanate, 63.60% (14/22)

Enterococcus – Clindamycin, 100% (27/27); Cefazoline, 97.9% (47/48); Trimethoprim-sulfamethoxazole, 55.4% (36/65)

Escherichia coli – Amoxicillin-Clavulanate, 77% (47/61); Trimethoprim-sulfamethoxazole, 73.1% (49/67); Cefazoline 62.7% (37/59); Ampicillin-Sublactam, 61.1% (11/18); Ceftazidime, 57.8% (37/64); Cefepime,

56.7% (38/67)

Klebsiella – Ampicillin Sublactam, 77.8% (56/72);

Amoxicillin-Clavulanate, 60.2% (56/93); Cefepime,

59.3% (67/113); Trimethoprim-sulfamethoxazole, 57.8% (67/116); Ceftazidime, 54.4% (56/103); Cefazoline, 50.6% (40/79)

Pseudomonas aeruginosa – Amoxicillin Clavulanate, 100% (49/49); Cefazoline, 100% (42/42); Trimethoprim-sulfamethoxazole, 100% (49/49)

Salmonella – Gentamycin, 90.9% (10/11)

Staphylococcus aureus – None

Conclusion: Distinct antibiotic resistance patterns were observed amongst causative organisms. Properly managing antibiotic usage is crucial to curtailing resistance, and knowledge of the susceptibility profiles of various organisms could be clinically valuable and improve outcomes when initiating empirical treatment regimens.

Perception of Virtual Interviews in Vascular Surgery Fellowship Application

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Background: In 2020, the national resident matching program recommended virtual interviews for initial applicant screening, deviating from the more traditional in-person format. During that time, our fellowship program completed both in-person and virtual interviews to compare the two experiences and better understand perceptions of using the virtual modality for ranking.

Methods: Our survey involved vascular surgery fellowship applicants interviewed either in-person or virtually during the 2020 application cycle. Voluntary surveys were administered after applicants and programs completed their rank-order list but before match day. Data was collected via an anonymous online survey tool. Gender, age range, race, completed interview number, in-person and virtual interview percentage, overall modality satisfaction, areas of improvement, and attitudes towards virtual fellowship interviews were documented.

Results: 13/25 applicants completed the online survey (52%). 7/16 (43.8%) completed the in-person survey and 6/9 (66.6%) completed the virtual. Respondents were mostly male (9/13, 69.2%) and white (7/13, 53.8%). Most (11/13, 84.6%) felt virtual interviews can differentially influence candidate ranking. 8/13 (62%) participated in 11-15 interviews. Six reported that virtual interviews comprised <10% of their total while the remainder reported virtual interviews accounted for 11-40%. No virtual applicants reported that in-person follow-up visits would influence their ranking. Respondents from both groups cited virtual platform benefits including reduced cost and travel, but almost half (6/13) identified no advantage to virtual interviews. Inperson interview preferences included exploring surrounding towns, in-person tour, and technical issue avoidance. Given the option between in-person interviews and virtual, no respondent would choose virtual over in-person (0/13).

Conclusion: Given an in-person or virtual interview option, no applicant preferred virtual, and none planned an inperson follow up. Responders were mostly white males, limiting generalizability. This survey highlights the importance of making virtual interviews more appealing to fellowship applicants should they remain a future option. However, additional studies are required to corroborate this.

Early Detection for Women's Cancer: Disparities and Prevention in Vulnerable Women

Authors: Jessica Tall, Daniela M. Dinulescu, Ph.D.

Department & Institution: Brigham's Women and Children's Hospital STARS Program & Dana-Farber/ Harvard Cancer Center, Gynecologic Cancer

Background: Ovarian cancer is the most lethal gynecological cancer with current screening methods having little impact in reducing mortality. The only current viable option for high-risk BRCA mutation carriers with a family history of breast and ovarian cancer is to undergo genetic screening and risk reduction surgery by

age 3540 years or when childbearing is complete.

Study Hypothesis: Minority, immigrant, lower socioeconomic/educated women have lower rates of BRCA screening and early detection tests.

Methods: Review of medical literature databases (PubMed, Science Direct, and Google Scholar) using relevant keywords yielded 42 articles with 12 used for this review.

Results: While the mortality ratios for ovarian and breast cancer are higher in Black women, there are no significant differences in the percent of BRCA mutations found in various racial/ethnic groups. Psychological distress is a major barrier for minority women undergoing early screening due to fear and embarrassment associated with the procedure, severe cancer specific distress, and racial discrimination. Language barriers may have lower mammography screening rates and are less likely to receive genetic counseling and referrals for immigrant women. Having trained interpreters in the examination room may increase patient-provider communication, enable patients to trust their providers, and aid patients in making fully informed decisions, which can improve patient outcome. Women who have lower education levels and socioeconomic status may have barriers to early screening such as a lack of education concerning modifiable cancer risk behaviors which can affect their ability to take full advantage of available resources and services for preventive health care.

Conclusion: Vulnerable women who carry BRCA mutations are at a higher risk of diagnosis with ovarian cancer at a later stage, resulting in higher mortality rates. We need to increase diversity in medicine and to facilitate access to professional-trained translators and implement education programs to address disparities and promote advocacy.

Improving LARC Use in Women with Substance Use Disorder

Authors: Karagan Mulhall, Shelby Wellman, Hannah Rowe, Michelle Worthy, Joe Evans

Department & Institution: Pediatrics, Marshall University

Background: The purpose of this quality improvement study is to increase long acting reversible contraceptives (LARCs) awareness and placement in opioid addicted mothers through prenatal and postpartum education. Ultimately, the aim is to decrease the rate of second pregnancy neonatal abstinence syndrome (NAS) in this subpopulation.

Study Hypothesis: Investigators hypothesized that by using a multidisciplinary approach, the use of long acting reversible contraceptives can be increased and the number of babies born at Cabell Huntington Hospital with Neonatal Abstinence Syndrome can be decreased.

Methods: The goal of this quality improvement study is to increase the percentage of inpatient LARCs placed in opioid addicted mothers through physician, nursing, hospital staff and patient education. This is a combined project involving an initial retrospective chart review followed by a prospective quality improvement (QI) study. This will be a multidisciplinary approach involving Plan Do Study Act (PDSA) cycles.

Results: The initial data, collected through a retrospective review, determined that 10.99% of mothers who had a neonate admitted to the Neonatal Therapeutic Unit (NTU) at Cabell Huntington Hospital, went on to have subsequent neonates born with NAS admitted to the NTU. Overall 10.37% of these admissions (158 total) could potentially have been prevented if the opioid addicted mothers would have chosen to leave the hospital with some form of Long-Acting Reversible Contraceptive (LARC), after the first delivery.

Conclusion: Charts will be reviewed to determine the number and type of LARC placed, and if the subject had a diagnosis of substance abuse. The QI team will meet monthly or bi-monthly as data is reviewed to decide on further educational actions through PDSA cycles with the goal of increasing the number of LARCs placed in opioid addicted mothers, thereby reducing the subsequent numbers of babies born with NAS.

Adverse Outcomes of Intra-Articular and Peri-Tendon Corticosteroid Injections.

Authors: Kristin Forkapa, Brian Krusemark and Aaron Lear, Kassandra Flores

Department & Institution: Department of Family Medicine, Cleveland Clinic Akron General, Akron, Ohio

Background: Osteoarthritis (OA) of large joints such as the hips and knees is an extremely common problem in the United States. Significant morbidity can be associated with the incidence and complications arising from OA, particularly of the knee and hip joints. There is no consensus among various medical specialties as to when intra-articular corticosteroids (IACS) are indicated. Recently, several studies have shown an increase in the progression of OA, and adverse effects such as tendon tears, worsening pain, avascular necrosis, subchondral insufficiency fractures and more using IACS injections.

Study Hypothesis: The use of IACS can cause rapid progression of OA, failure of improvement, worsening of pain, soft tissue and bone pathology. Adverse outcomes may be linked to risk factors such as increased age, sex, high BMI, diabetic status, repeated injection into joint, co-morbid conditions and concurrent medication use.

Methods: This is a retrospective cohort study. Patients given an injection between 2016-2017, comprised the cohort, with chart follow up through 2018-2019. Data was collected from EPIC health record system and a baseline was established for each patient at the time of injection. Type/dosage and location of injection, baseline pain level, baseline OA scale (scored with Kelgren Lawrence), co-morbid conditions, medications and demographics were recorded. Outcomes of joint pathology and clinical care were recorded for following 2 years.

Results: Preliminary data analysis was calculated early in the study. Complete and final data analysis is set to be completed October 10, 2021. Preliminary results indicate 30% patients experienced worsening pain within 6 months of injection and 16% showed x-ray evidence of progressing OA within 1 year of injection. Final evaluation is in progress.

Conclusion: Preliminary data shows a positive correlation of worsening joint pain and progression of OA in patient who received intra-articular corticosteroid injections. However, no final conclusions have been drawn at this time.

A descriptive study assessing temporal changes in infective endocarditis in Huntington, West Virginia.

Authors: Kennedy Snavely, Brianna Canales, Kendell McNeely, Daniel Snavely, Todd Gress, and Mehiar ElHamdani

Department & Institution: Internal Medicine, Joan C. Edwards School of Medicine

Background: The rate of infective endocarditis (IE) in persons who inject drugs (PWID) has increased concomitantly with the use of heroin in the United States. Moreover, West Virginia has the highest age-adjusted rate of drug overdose deaths involving opioids. The purpose of this study is to analyze changes in the presentation of IE over a 10-year period in Huntington, West Virginia.

Study Hypothesis: We suspect that there will be a significant increase in the incidence of IE over time, particularly in those with opioid use.

Methods: Utilizing de-identified data from the Marshall Health Data Warehouse, we identified 444 hospitalized patients at a single hospital with an ICD-9/ICD-10 diagnosis of IE from years 2010 to 2019. We defined a variable for illicit opioid drug use and collected information on patient demographics, hospital charges, and echocardiography results. We obtained study approval from our local Internal Review Board.

Results: Among the 444 patients with IE, 272 (61.3%) met the criteria for illicit opioid drug use. There was a significant increase (P for trend < 0.01) in the number of annual admissions for IE among no opioid use cases and opioid use cases between 2010 and 2019. The proportion of opioid use cases comprising the total cases of IE steadily increased throughout the study period. Echocardiographic information was available on 339 patients (76.1%). Transthoracic echocardiograms were performed as the initial echocardiographic test in 93.8% of patients (N=318), and this did not differ by drug use.

Conclusion: Our findings reveal that annual admissions for IE at CHH have markedly increased throughout the past decade, and cases of IE associated with opioid use comprised a majority (61.3%) of total IE cases during this study period. Echocardiographic data suggest that standard IE diagnostic protocol was followed in a majority of cases (93.8%) with no statistically significant variations in protocol with regards to opioid use.

Flu Vaccine Administration before and after COVID-19

Authors: Jodi Plumley, Isabel Pino, Allyson Bias, Eric Mendenhall, Paige Lester, Deborah Preston

Department & Institution: Department of Pediatrics, Joan C. Edwards School of Medicine at Marshall University

Background: According to the CDC in 2018-2019, the influenza vaccine was given to an estimated 45.3% of adults in the US and prevented approximately 4.4 million influenza cases, 58,000 hospitalizations, and 3,500 deaths. The influenza vaccine reduces the risk of influenza illness by an estimated 40-60% when the variants circulating in the population are closely matched to the vaccine. During the COVID-19 pandemic, it is imperative to reduce strain on the health care system by encouraging patients to get their influenza vaccine.

Study Hypothesis: We suspected more influenza vaccines were administered during the 2020-2021 flu season compared to the 2019-2020 flu season.

Methods: A survey was distributed to parents of pediatric patients >6 months of age regarding past influenza vaccinations for the child, reasons for not having their child take the vaccine, and if they would take the COVID vaccine. It was distributed at the resident clinic from 10/01/2020 until 03/31/2021. Query obtained regarding number of influenza vaccines during the 2019-2020 and 2020-2021 seasons.

Results: season compared to the 19/20 season. According to the data query, less influenza vaccines were administered in the 20/21 season compared to the 19/20 season, 7715 and 9173, respectively. However, it is suspected that less patients were seen overall during the latest season due to the pandemic (pending query for unique patient visits during 10/2019 to 03/2020 and 10/2020 to 03/2021). Of parents surveyed, 49% said they would give their child the COVID vaccine if available to them.

Conclusion: There were less influenza vaccines administered during the latest flu season compared to the prior year. We will determine how many vaccines were administered relative to unique patient encounters pending query.

COVID-19 Impact on Hospital Presentation of Adolescent Suicide Ideation and Attempts

Authors: Kelly Melvin, Hillary Porter, Allyson Bias, Joshua Hall, Catherine Cavender, Paige Lester, Deborah Preston

Department & Institution: Departments of Psychiatry and Pediatrics, Joan C. Edwards School of Medicine at Marshall University

Background: The COVID-19 pandemic has had wide ranging negative indirect effects on the mental health of populations worldwide. Studies have documented increases in anxiety, depression, and suicide attempts in various groups. Pediatric populations are a potentially high-risk population as, pre-COVID, suicide is the second leading cause of death in the United States for individuals 15 to 19-years-old. Potential contributory factors include increased isolation due to quarantine, reduced contact with preferred peer groups, increased reliance on electronic media for communication, transition to virtual learning, fear of infection for self or loved ones, and many others. Studies examining the mental health consequences of the COVID-19 pandemic on youth are sparse, especially Appalachian youth. This study will contribute to a notable and important gap in the existing literature.

Study Hypothesis: The objective is to determine whether there has been an increase in pediatric hospital admissions for suicidal youth following the onset of the COVID-19 pandemic compared to the prior year.

Methods: A data query was submitted for hospital admissions and ER visits between 03/2019 to 03/2021 for patients 8 to 21 years old at the time of admission and diagnosed with suicidal ideation, intentional ingestion/ overdose, or suicide attempt. They were identified by MRN and DOB. Charts reviewed for age, sex, residential location, past psychiatric history, comorbid conditions, current and past medications, family history of psychiatric illness, CPS involvement, and situation leading to presentation to the hospital. Chart data collected and entered into REDCap then de-identified.

Results: Preliminary data not yet available due to continued data collection.

Conclusion: We anticipate there will be an increase in patient admissions for suicidal ideation and attempts during the COVID-19 pandemic compared to before it occurred.

Improving Pneumococcal Vaccination Rates Among Rheumatoid Arthritis Patients of the Rural Appalachian Population in an Academic Rheumatology Clinic

Authors: Lauren Clower, Austin Nichols, Tori Leader, Andenet Mengistu, Rajesh Gopalarathinam, Ralph Webb, Adenrele Olajide.

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Background: Invasive pneumococcal disease is associated with high mortality rates among rheumatoid arthritis (RA) patients. The Centers for Disease Control (CDC) and American College of Rheumatology (ACR) has current guidelines recommending pneumococcal vaccination for this patient population. In 2019, we conducted a retrospective chart analysis and found suboptimal vaccination rates of only 10% among the 107 RA patients in our study. We conducted a quality improvement project at the Marshall Health rheumatology clinic to improve vaccination rates among RA patients.

Study Hypothesis: We hypothesized that patient compliance for receiving pneumococcal vaccinations would improve if RA patients were educated about the importance of receiving these vaccinations during their clinic visit.

Methods: Adult RA patients were interviewed and completed a brief questionnaire after patient consent was obtained. During interviews, patients were educated about the importance of receiving pneumococcal vaccinations and were administered age-appropriate vaccinations. Vaccinations outside of Marshall Health were obtained through the West Virginia Statewide Immunization Information System (WVSIIS). Out of state vaccination records were not easily accessible, so our co-investigators conducted follow-up calls to these providers.

Results: Among the 100 patients enrolled in our study, 24 were previously up to date with the recommended vaccination series. Of the remaining 76 patients, 47 of these participants agreed to vaccination during their clinic visit. Findings indicated that 71% of subjects were compliant whereas 29% of subjects were not compliant. Common reasons cited by subjects for avoiding pneumonia vaccination included concern about side effects, lack of awareness, or a prior adverse reaction to another vaccine.

Conclusion: We concluded that higher rates of pneumococcal vaccination can be achieved among RA patients by education during clinic visits and recording vaccination history in their electronic health records. The role of both primary care physicians and specialists are crucial in discussing the importance of vaccination and resolving vaccine related misconceptions among patients.

Duration of Mother's Own Milk Feeding in Infants Post NICU Discharge and Maternal Perception of Reasons for Discontinuation

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Background: Mother's own milk (MOM) provides numerous short and long-term benefits to both infant and mothers, during and after NICU stay.

Study Hypothesis: Describe duration of mother's own milk feeding in infants post NICU discharge and maternal perception of reasons for discontinuation

Methods: This study included infants discharged from NICU on MOM diet and received at least one home follow-up visit. Group 1 (n=53) included infants born <33 weeks gestational age (GA) or birth weight <=1500g, Group 2 (n=163) included infants born >=33 weeks GA and birth weight >1500g. Kaplan Meier curves describe duration of MOM feeding. Reasons for discontinuing MOM feeding were summarized.

Results: This study included 216 infants born to 201 mothers. By 60 days post NICU discharge, 60% of Group 1 infants and 71% of Group 2 infants were still feeding MOM. Of those infants who stopped receiving MOM, discontinued at a median (IQR) of 27 (20, 54) and 36 (15, 62) days post discharge in Groups 1 and 2, respectively.

The most common reason for discontinuing MOM feeding was perception of low milk supply, followed by caregiver frustration.

Conclusion: Implications for practice: The first month after NICU discharge is the critical time for interventions to sustain MOM feeding. Early and frequent follow-up visits with intensive hands-on lactation to maintain milk supply and transition from bottle feeding to feeding at breast post NICU discharge is imperative in sustaining MOM feeding. Identifying the timing and reasons for stopping MOM feeding post NICU discharge is critical to develop targeted interventions to increase the duration of MOM feeding.

Implications for Research: Further research is needed to develop preventative strategies and early interventions to address the causes of early MOM feeding discontinuation in at risk populations.

Complications and Hospital Admissions Among Pregnant Women with Substance Abuse

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Department & Institution: Department of Obstetrics and Gynecology, Marshall University Joan C. Edwards School of Medicine, Huntington, WV

Background: In recent times, there has been an increase in drug abuse in not only the general population, but in women of reproductive age.

Study Hypothesis: Our objectives were to identify, classify, and describe the spectrum of complications, the average number of admissions, and length of hospital stay that occur among pregnant women with substance abuse. The aim was to obtain better understanding of complication prevalence in order to improve management in this ever-growing population.

Methods: A retrospective chart review was conducted of pregnant women ages 18-45 with a history of substance abuse treated at Marshall University's OB/GYN from 2013-2018. We collected the following data: demographics, medical history, specific substances abused, inpatient admission dates and diagnoses, and delivery information.

Results: A total of 411 patients met the inclusion criteria, with a total of 525 pregnancies. Out of 525 pregnancies, most patients used buprenorphine (i.e., Subutex) (71.6%) followed by the use of opiates, excluding heroin (43.4%); 35% of patients used heroin. Out of the 525 pregnancies, there were 714 inpatient antepartum admissions. Of these, 376 were admissions due to withdrawal symptoms (52.7%). A total of 263 pregnancies had at least one admission for withdrawal, drug abuse, overdose, or buprenorphine/methadone conversion (50%). The average length of hospital stay for withdrawal admissions was about 3.4 days. There were 62 admissions for infectious causes, 24 of these being due to pyelonephritis (38.7%).

Conclusion: This study has the potential to lead to many quality improvement measures in the future as it has given background information on a population that has much data lacking. For example, there is potential for improvements in the management of patients admitted for withdrawal. Another area for consideration includes a case series on infectious causes for admission in this population. The results highlight multiple areas for future studies and assessment.

Transitioning To Unfractionated Heparin In Treatment of NSTEMI patients on Direct Oral Anti-Xa Inhibitors

Authors: Mohamed Tashani, Raice Stevens, Hari Vishal Lakhani, Brittany Riley, Sharon Jones, Jason Mader

Department & Institution: Marshall Cardiology, Marshall University

Background: Direct oral anticoagulants (DOACs), such as rivaroxaban, apixaban, and edoxaban, use is becoming more prevalent throughout the United States. The use of such medications is related to coagulation lab value abnormalities, namely elevation in the heparin anti-Xa assay. This assay is commonly used in dosing heparin. Patients with non-ST-segment elevation myocardial infarction (NSTEMI) are recommended to receive heparin for ischemia-guided management. Problems arise when patients prescribed DOACs present to the hospital with a NSTEMI. Elevations in the anti-Xa assay may lead to delays in administration of heparin.

Study Hypothesis: The goal of this research is to

determine if heparin use is delayed in the setting of elevated anti-Xa levels in patients with a diagnosis of NSTEMI.

Methods: This is a single-center, retrospective, chart review study. Patients included were 18 years of age or older with a documented DOAC home medication and a diagnosis of NSTEMI. The study population consisted of adult patients admitted between during 2019. Data collection included patient demographics, activated partial thromboplastin time, prothrombin time, DOAC dose, and left heart catheterization. Data was also collected for Xa levels, in addition to reason of delay, if any, in the administration of heparin. Statistical analysis included determination of r-squared correlation and one-way ANOVA using Graphpad Prism 8.0.

Results: Patient population was divided in three groups based on baseline Xa levels of patients. Elevated Xa level was noted more in patients who were taking apixaban. Heparin infusion was delayed among this subgroup of patients. Elevated baseline Xa levels were significantly improved after 12 hours. Primary outcome of in-hospital mortality was not observed.

Conclusion: High sensitivity of heparin anti-Xa assay to DOACs affect assay accuracy and result in elevated heparin anti-Xa level with use of DOACs, hence, resulting in delayed start of heparin therapy in treating NSTEMI patients.

Increased risk of type 2 diabetes in Appalachian patients with bipolar disorder after exposure to antipsychotic medications: a Cox proportional hazard model

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Department & Institution: Psychiatry, MUSOM

Background: Patients with bipolar disorder (BD) have a high rate of co-morbid type 2 diabetes (T2D), and strong evidence indicates that initiation of antipsychotics precedes the development of type 2 diabetes. However, several studies suggest that type 2 diabetes and bipolar disorder may share a common pathophysiological mechanism (Charles et al. 2016). However, a degree of uncertainty regarding etiology remains due to confounding variables not eliminated in these studies.

In West Virginia, there is a high incidence of diabetes, and many studies have shown that type 2 diabetes and mood disorders may share a genetic link further influenced by environmental factors. High rates of obesity, lack of appropriate resources in the community, poor healthcare literacy, and geographically isolated populations may all play a role.

Study Hypothesis: Even when controlling for other factors, we propose that there will be a higher risk of developing type 2 diabetes with exposure to an antipsychotic medication.

Methods: Using data from 2,860 patients with bipolar disorder over a ten-year period, we developed a Cox proportional hazard model to determine the increased risk in development of type 2 diabetes after exposure to antipsychotic medications. The study design takes into account other risk factors for diabetes, including family history, obesity, older age, gestational diabetes, and gender to isolate the risk attributable to the medication. In addition, because the data is from multiple clinical encounters over time, we will also be able to draw conclusions about the timeline to onset of diabetes after exposure to the medication.

Results: With regard to the risk of developing diabetes while taking an antipsychotic, there was a Cox proportional hazard ratio of 1.384 with a 95% confidence interval of 1.125-1.702. The value for p was 0.002.

Conclusion: After analyzing the data, we did find an increased risk of developing type 2 diabetes with an exposure to antipsychotic medication.

Neonatal Abstinence Syndrome (NAS) treated inborns at Cabell Huntington Hospital (CHH) in the SARS- CoV2 pandemic era

Authors: Nana Bosomtowe, Jessica Haas, Jodi Plumley, Luke Damron, Rebecca Barnett, Lori Blackburn, Talisha Franklin, Emily Stacy, Cindy Massey, Balaji Govindaswami

Department & Institution: Pediatrics, Clinical and Translational Sciences, Marshall University School of Medicine

Background: Fetal exposure to narcotics and related substances is very dynamic. Changes in data acquisition, laboratory testing for urine and umbilical cord, and EMR have occurred concurrent to the pandemic.

Study Hypothesis: We studied infants treated for NAS, their prenatal exposure to narcotics and related substances in both the pre SARS-CoV2 era and compared it to post pandemic frequency to examine relevant differences, if any.

Methods: Data were collected prospectively, and examined retrospectively. Only inborn infants are included in this study. Maternal and infant demographic variables, maternal history of substance exposure, results of maternal urine toxicology during labor, infant cord toxicology reports were all examined for NAS treated infants. Data were entered electronically, after manual correction of errors. Pre and post pandemic fetal exposure to narcotics and related substances, neonatal abstinence syndrome (NAS) frequency will be analyzed using Chi-square for differences between groups, and run charts will be presented to examine quarterly frequency and trends using statistical process control.

Results: During the study period,(January 2018-September 2021) ~ 9500 consecutive newborn at CHH and data relevant to NAS treated infants were examined. The rate of NAS was > 8 % during the study period. A summary of substances and methods of detection for infants managed with NAS will be presented in Table 1. It was not possible to ascertain fetal exposure for non NAS treated inborn infants. This presents an opportunity for improvement. Prenatal Tetrahydrocannabinol exposure in NAS treated infants in 2020 was 48%.

Conclusion: Fetal exposure in narcotic and related substances in all inborns at CHH present challenges in ascertainment. NAS frequency continues to increase at CHH and is presently > 8% for inborns in four consecutive years. Changes in manual acquisition and electronic transformation may enable us to have improved clarity in data collected in 2020-21.THC frequency increased during the study period but its relation to the pandemic is unclear.

Sudden Cardiac Death in Heart Failure with preserved Ejection as compared to Heart Failure with reduced Ejection Fraction: A Nationwide Analysis

Authors: Rodrigo Aguilar MD, Daniel Vilchez MD, Kanaan Mansoor MD, Carlos Rueda MD

Department & Institution: Internal Medicine, Cardiology

Background: Prevalence and admissions to acute heart failure are rising nationwide and globally. Heart failure with reduced ejection fraction (HFrEF) has been linked to causing sudden cardiac death (SCD), however, recent studies are showing heart failure with preserved ejection fraction (HFpEF) may be associated with the development of SCD. Data and predictors of such risk factors are scarce.

Study Hypothesis: To compare mortality outcomes between HFrEF and HFpEF

Methods: Nationwide Inpatient Sample 2009 to 2012 database, using (ICD-9), was queried for the study. Patients with an age less than 21 were excluded, as well as patients with a diagnosis of hyperkalemia, restrictive cardiomyopathy, and hypertrophic cardiomyopathy. A primary cohort of patients having SCD was selected, these included ventricular tachycardia, ventricular fibrillation/flutter, cardiac arrest, and sudden death. Later subgroups were divided into HFpEF and HFrEF based on ICD9 coding. These 2 arms were compared.

Results: A total of 94,174 patients were included in the study, 25,894 accounting for HFpEF vs 68,278 with HFrEF. (27.50% vs 72.50%). Mean age was 74.5 (±12.5) years old in HFpEF whereas HFrEF mean age was 69.2 (±13.5) years old. Interestingly, 71.42% of the sample was white patients as compared to other races, including Latino, African American, Asian, and native. DM2 was more frequent in HFpEF (38.3% vs 35.2% p=0.001). HTN was similar (34.4% vs 33.6% p=0.02). Hyperlipidemia was frequent in HFrEF (41.9% vs 38.6% p=0.001). Cardiac

arrest was more frequent in patients with HFpEF (22.8 vs 14.1% p= 0.001). Ventricular tachycardia was frequent in HFrEF (83.2 vs 71.5% p= 0.001). Ventricular fibrillation was higher in HFrEF (10.8 vs 5.8% p= 0.001) Overall inpatient mortality was higher in HFpEF (17.83 vs 11.65% p= 0.001).

Conclusion: Inpatient mortality risk is higher in patients with HFpEF as compared to HFrEF. However, HFrEF is frequently more associated with SCD. Further studies are warranted.

In-hospital mortality of transcatheter versus surgical mitral valve replacement: A Nationwide Analysis

Authors: Rodrigo Aguilar, Kanaan Mansoor, Daniel Vilchez, Jason Mader

Department & Institution: Internal Medicine, Cardiology

Background: Transcatheter mitral valve replacement (TMVR) is an alternative to surgical aortic valve replacement (SMVR) for patients with moderate to severe mitral regurgitation

Study Hypothesis: To evaluate mortality difference in percutaneous vs traditional mitral valve repair

Methods: Nationwide inpatient sample (NIS) data from 2010 to 2014 was queried. Patients who had mitral regurgitation and underwent consequent either TMVR or SMVR were selected. Cases that received both techniques were excluded, patients were categorized by BMI, those with normal BMI were also excluded. We stratified the groups according to the propensity match score and used TMVR as a reference. Charlson comorbidity index (CCI) was used to assess the severity of comorbidities in both arms.

Results: Among 1,831 patients admitted for mitral valve replacement between 2010 and 2014, 68.32% underwent TMVR and 31.68% underwent SMVR. Mortality prior to propensity match was higher in SMVR (2.86% vs 2.08%, p= 0.008). The all-cause mortality was equal in both arms after adjusting commorbidities. Length of stay was shorter in TMVR (6.9 days vs 11.2 days). Charlson comorbidity index was higher in patients with TMVR. Both SMVR and TMVR was more commonly performed on obese patients (68.32%) before match and after match (66.6%), however mortality was higher among morbidly obese patients pre match (3.93% vs 2.40% vs 2.82%; p=0.21) as well as post match (3.85% vs 1.86% vs 0; p= 0.73). Overall morbidly obese undergoing SMVR had higher mortality rates (OR 0.26 95% CI 0.08-1.02)

Conclusion: Data analysis results demonstrated that the patients with higher mortality have occurred when performing SMVR as compared as TMVR despite those patients selected for TMVR have a higher CCI, however recent reports support improving techniques, faster recovery, shorter length of stay. Further studies are suggested to further stratify mortality across the years.

Evaluation of a pharmacy technician-based medication prior authorization program

Authors: Ron Carico Jr, Ryan West, Tiffany Miller, Jessica Brown, Dustin Baum, Sarah Dunaway, Alexander Hill, Will Finley, Jesse Bates, Jeff Fenerty

Department & Institution: Marshall Pharmacy

Background: The roles of pharmacy technicians in clinical practice are being explored. Medication prior authorizations (PAs) from insurers can lead to delays in pharmacotherapy.

Study Hypothesis: The objective of this study was to assess the efficiency of our clinical pharmacy technicians in processing PAs for medications.

Methods: Clinical pharmacy staff in 4 clinics recorded information about PA requests from January 21, 2020, to April 21, 2020. In 3 of the clinics, PA requests were primarily processed by clinical pharmacy technicians. In another clinic, requests were processed by a clinical pharmacist. Information collected included the date the request was received, outcomes (e.g., approval, therapy change, or nonapproval), and the date of final outcome. Descriptive statistics were prepared, including number of requests that were approved, number of business days between request and decision, and final outcome.

Results: Overall, 720 PA requests were received. Of these, 88.6% were approved with first response, and 673 (93.5%) were eventually approved. Median time to first response was 0 business days, regardless of clinic. In 75%

of cases, first response was within 1 business day. PA characteristics varied across clinics; however, PA approval percentages were comparable (91.2%–94.3%).

Conclusion: In an assessment of clinical pharmacy technicians' efficiency in responding to pharmacy plan PA requests, more than 90% were approved, often within one business day. Our results must be interpreted in light of local factors and a virus pandemic during the study. However, results of requests handled by technicians were similar to results when the requests were handled by a clinical pharmacist. Clinical pharmacy technicians can be efficient and cost-effective in this role.

Understanding biomarker expression in association with graded recoil analysis following shooting

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Background: The impact of traumatic brain injury (TBI) along proper diagnosis and treatment have been heavily highlighted recently in contact sports like football and rugby. However, other sports may also experience the same recoil force that propagates alterations in the brain and its cellular physiology. Competitive shooters experience regular contact with repetitive gun recoil, ultimately transferring multiple acceleration to the body and brain. Current research in the military suggests that chronic exposure to firing weaponry contributes to the development of TBI. In this study, we sought to understand if newly identified and approved neurobiomarkers of TBI are changed following graded shooting events.

Study Hypothesis: We hypothesize that increased recoil transferred to shooters will alter neurobiomarker expression in the plasma. Neurobiomarkers of interest include glial fibrillary acidic protein (GFAP), ubiquitin C-terminal hydrolase L1 (UCH-L1), and S100B. We postulate a graded response in more repeats of shots fired.

Methods: Subjects were recruited to shoot 25 or 100 rounds from a shotgun at a gun range. During the event, sensors were placed on the gun and subject to calculate recoil (G force). Prior to shooting, subjects underwent a blood draw (time 0). Blood was also drawn 1-hour and 24-hours post shooting to evaluate biomarker change. GFAP, UCH-L1 and S100B were analyzed via ELISA.

Results: Significant neurobiomarker changes were observed in patients. However, there was no statistically significant graded response or gender differences. G-force calculations averaged around 2 G's for the ear sensor per shot.

Conclusion: Alterations in neurobiomarker expression suggest some degree of brain injury, although it is unclear the depth of this injury. As competitive gun shooting is a fast-growing sport and hobby, it is important to note any health issues that can be combated to make shooting safer.

Assessing Factors that Impact COVID Testing Rates in a Regional Testing Center

Authors: Seth Bergeron, Justin Spradling, Tammy Bannister, Paris Johnson, Adam M. Franks

Department & Institution: Department of Family and Community Health, Joan C. Edwards School of Medicine, Huntington, WV.

Background: COVID-19 is a deadly disease caused by the SARs-CoV-2 virus, and is responsible for over 4.5 million deaths worldwide. Symptoms of COVID-19 are highly variable from each individual and range from asymptomatic to severe respiratory failure. The transmissibility for this virus is extremely high.

Transmission of SARs-CoV-2 can even occur from asymptomatic carriers. An important measure in trying to manage and reduce the spread of COVID-19 is the use of testing to identify and isolate individuals who have been infected before transmission occurs, or to limit transmission, however, this has its challenges. Mass testing is not feasible due to lack of time and leads to a potential waste of resources. In order to efficiently test populations, there needs to be an appropriate allocation of resources. This means that it is fundamental to identify where testing is needed the most. Current resource allocation for testing is designated by the state or local rates of COVID-19 cases.

Study Hypothesis: We believe the testing demands on a regional COVID-19 testing center are not completely explained by state or local rates of SARs-CoV-2 infection.

Methods: Longitudinal state and county study data was compared to the COVID-19 testing rates within the regional COVID-19 testing center operated by the department of Family and Community Health within the Marshall University School of Medicine.

Results: Analysis of state and county COVID-19 positive case data does not fully correlate with the rates of SARsCoV-2 infection. (Full analysis pending)

Conclusion: These results indicate that there are other factors responsible for demands on testing volume. These factors are essential to identify for better resource allocation in the current pandemic and for future outbreaks as well.

Impact of Setting Chapter Quizzes Benchmarks in a Capstone course on Exam Performance

Authors: Shadi Bashai, Craig Kimble, Angel Kimble, Omar Attarabeen

Department & Institution: Pharmacy Practice, Administration, and Research

Background: The final year in the PharmD program is key to graduates' competence and practice-readiness. This study aimed to assess the impact of setting minimum scores for students to meet throughout their rotation year.

Methods: Students complete multiple home practice exams as well as chapter quizzes during their rotation year. Only the 2020 class was required to achieve a minimum of 70% in chapter quizzes. For two consecutive classes (2019 and 2020), we calculated the improvement in student scores in exams by comparing the final exam scores to the first exam scores. Score improvement was compared between the two classes using independent sample t-test in order to check for statistical difference.

Results: Data from 79 students from the 2019 class and 65 students from the 2020 class were included in the analyses. On average, scores improved from the first home practice exam to the last home practice exam by 27 points for 2019 students and 15 points for 2020 class students. The t-test analysis demonstrated that the difference in score improvement between the two classes was statistically significant [$t(137) = 5.381$, $p < 0.001$].

Conclusion: Compared to 2019 class students, 2020 class students' knowledge improvement from exam 1 to exam 3 was smaller. Holding students responsible for meeting a 70% minimum does not seem to improve their knowledge and preparedness for the final home examination. Perhaps having met this minimum gave students a false sense of mastery of the material. Future research will explore the reason setting a 70% minimum for chapter tests is associated with this decline in exam scores.

Risk Factors for Arthrofibrosis and Primary and Revision Total Knee Arthroplasty: A Multicenter Study

Authors: Shane Taylor, Alisina Shahi, Vishavpreet Singh, Galen Berdis, Matthew Bullock, Ali Oliashirazi

Department & Institution: Risk Factors for Arthrofibrosis and Primary and Revision Total Knee Arthroplasty: A Multicenter Study

Background: The purpose of this study was to determine the incidence, timing, and risk factors associated with arthrofibrosis after primary TKA (pTKA) and revision TKA (rTKA) and provide guidance for perioperative risk assessment.

Methods: This is a multicenter retrospective study which reviewed patients who underwent pTKA and rTKA between 2008-2017. Patients who underwent ipsilateral manipulation under anesthesia (MUA) for arthrofibrosis were identified and time to MUA was recorded. Multivariate logistic regression was used to determine odds ratios for risk factors for MUA including RA, age, obesity, smoking diabetes, and revision TKA.

Results: 10,842 TKAs were included in the study of which 7595 were pTKA and 3,247 were rTKA. 1.9% of patients (n=206) underwent MUA after TKA (42 patients) and rTKA (164 patients). MUA was significantly greater in rTKA 5.1% vs 0.55% in pTKA ($p<.000001$). 72% of MUAs occurred

within the first 3 months postoperatively. Young patients (<50 years) had significantly higher odds of MUA after rTKA (6.5, P< .0001). The remaining risk factors were significant for patients who had undergone TKA or rTKA in descending order were: obesity (odds ratio [OR]: 5.1, 95% Confidence Interval [CI]: 3.8-6.9), diabetes (OR: 4.7, 95%CI: 3.5-5.8), smoking (OR: 3.9, 95%CI: 2.1-4.6), rTKA (OR: 3.6, 95%CI: 2.8-4.8), and rheumatoid arthritis (OR: 2.4, 95%CI: 1.5-3.3).

Conclusion: This study found age younger than 50 was a risk factor for arthrofibrosis in rTKA. Obesity, diabetes, smoking and rheumatoid arthritis are significant risk factors for MUA in both pTKA and rTKA. Patients undergoing rTKA are at higher risk for MUA compared to pTKA. This study provides surgeons with patient related risk factors for arthrofibrosis and may help to guide preoperative risk factor modification or to identify patients at risk for arthrofibrosis in the postoperative period.

Improving LARC Use in Women with Substance Use Disorder

Authors: Char-Leigh Arnold, Courtney Crain, Anne DeFruscio, Joseph Evans, Brandon Fazalare, Jennifer Gerlach, Ken Kurek, Sean Loudin, Karagan Mulhall, Hannah Ray, Kayla Rodriguez, Benjamin Russell, Shelby Wellman, Anna Wood, Michelle Worthy, Jennie Yoost

Department & Institution: Marshall Pediatrics, Cabell Huntington Hospital

Background: The purpose of this quality improvement study is to increase long acting reversible contraceptives (LARCs) awareness and placement in opioid addicted mothers through prenatal and postpartum education. Ultimately, the aim is to decrease the rate of second pregnancy neonatal abstinence syndrome (NAS) in this subpopulation.

Study Hypothesis: Investigators hypothesized that by using a multidisciplinary approach, the use of long acting reversible contraceptives can be increased and the number of babies born at Cabell Huntington Hospital with Neonatal Abstinence Syndrome can be decreased.

Methods: The goal of this quality improvement study is to increase the percentage of inpatient LARCs placed in opioid addicted mothers through physician, nursing, hospital staff and patient education. This is a combined project involving an initial retrospective chart review followed by a prospective quality improvement (QI) study. This will be a multidisciplinary approach involving Plan Do Study Act (PDSA) cycles.

Results: The initial data, collected through a retrospective review, determined that 10.99% of mothers who had a neonate admitted to the Neonatal Therapeutic Unit (NTU) at Cabell Huntington Hospital, went on to have subsequent neonates born with NAS admitted to the NTU. Overall 10.37% of these admissions (158 total) could potentially have been prevented if the opioid addicted mothers would have chosen to leave the hospital with some form of Long-Acting Reversible Contraceptive (LARC), after the first delivery. Mother's demographics are analyzed to study relationships between insurance type and LARC placement.

Conclusion: Charts will be reviewed to determine the number and type of LARC placed, and if the subject had a diagnosis of substance abuse. The QI team will meet monthly or bi-monthly as data is reviewed to decide on further educational actions through PDSA cycles with the goal of increasing the number of LARCs placed in opioid addicted mothers, thereby reducing the subsequent numbers of babies born with NAS.

Impacting Patient Perception of Medical Care and Communication in a Patient Centered Medical Home through an Informative Brochure

Authors: Tanner Gilbert, Willie Kimler, Tammy Wilson, Hyla Harvey, Robert Hall, Paris Johnson, Adam M. Franks

Department & Institution: Family Medicine, Joan C. Edwards School of Medicine

Background: A patient centered medical home (PCMH) model allows patients to have customized care tailored to their health by a team of physicians and clinical staff, who coordinate care across disciplines. The areas of concern determined for this project at first were the physicians' unawareness in their lack of communication, unawareness of patient's perception of care, lack of time to communicate with patients, and a lack of knowledge by patients of the services available by the PCMH. In order to address these concerns, interventions were implemented.

Study Hypothesis: We believe that improvement in patient satisfaction scores can be achieved to >/= 95% in four vital areas of quality of care (Access, Communication, Patient Centeredness and Continuity) by improving physician-patient communication in a patient centered medical home (PCMH).

Methods: A quality improvement project was conducted identifying key drivers that inhibit quality communication: physician awareness of the impact of their communication on patients' perception, lack of time for quality communication, and poor patient understanding of the services offered by the PCMH. Interventions included physician education of their patient perception scores (August 2019), an educational video of PCMH services was created for patients in the waiting room, and an educational PCMH brochure (Summer 2021).

Results: A minimal impact was seen by the educational video across the four measures (+1.23%). The awareness provided to physicians about their own performances provided a slight increase in satisfaction scores from June to December of 2020 (+0.97%). The educational brochure data is not yet available.

Conclusion: Effective communication is a complex process that has not been significantly improved with serial interventions at this time. The improvements demonstrated give hope that the quality improvement process will deliver a broad enough impact on communication barriers to affect a positive change.

Weight Loss in Community-living older adults during the Covid-19 pandemic

Authors: Thomas McIntosh, Adam Franks

Department & Institution: Family Medicine, Marshall University

Background: The population of Oldest-Old, or seniors 85 years or older, is projected to drastically increase over the next few decades. As it is the most rapidly increasing population in West Virginia, it is important to understand this group and how to improve their quality of life. The development of the pandemic caused by COVID-19 has forced strict precautionary measures on the population, causing an adjustment to individuals' daily lives. Maintaining optimal nutritional status is imperative for healthy aging, but the pandemic could have a deleterious impact on this aspect of this population's life. The Mini-Nutritional Analysis (MNA) is a standardized tool that assesses nutritional wellbeing.

Study Hypothesis: Poor pre-pandemic nutritional status predicts a poorer change in weight for Appalachian elders 80 years old and older during the first year of COVID-19.

Methods: The MNA was administered to 80 years and older Appalachian residents of Cabell, Wayne and Lincoln counties. Both pre-COVID and 6 months into the pandemic. Weights were obtained and BMIs were measured before COVID and both 3 months and 6 months into the pandemic.

Results: Of the 170 enrolled seniors, 130 (76.47%) were scored as well-nourished (scores 12-14), while 32 (18.82%) were at risk for malnourishment (scored 8-11) and 8 (4.71%) were malnourished (scored <8). Weight change at 3 months were -2.05 lbs for the malnourished group, -1.13 lbs for the at-risk group, and 0.01 lbs for the well-nourished groups. Changes at 6 months were -7.00 lbs, -5.22 lbs and -1.88 lbs.

Conclusion: Individuals with lower MNA scores lost greater amounts of weight. This study shows that the lockdown caused by COVID-19 may affect eating habits and overall nutritional status of the older population in rural West Virginia disproportionally. Future efforts should be to improve targeted nutritional support for the community-dwelling older population during a recurrent quarantine.

Coronary Artery Disease Status Modulates Non-Coding RNA Expression in Epicardial Fat

Authors: Tristan Burgess, Brendin Flinn, Nepal Chowdhury, Todd Gress, Nalini Santanam

Department & Institution: Research Service, Hershel "Woody" Williams VA Medical Center; Department of Cardiovascular and Thoracic Surgery, St. Mary's Heart Center, Huntington, WV; Department of Biomedical Sciences, Marshall University Joan C. Edwards School of Medicine, Huntington, WV

Background: Coronary artery disease (CAD) is

characterized by the buildup of atherosclerotic plaques within the coronary arteries, causing obstructions of blood flow among other serious complications. CAD affects a large portion of the US population, having age-adjusted prevalence of 7.2% and 4.2% for males and females, respectively. Epicardial adipose tissue (EAT) is known to both interact with the heart and vasculature and be altered under pathological conditions. Understanding alterations in the EAT of CAD patients is useful for both understanding mechanisms of EAT's interactions with the heart and vasculature and discovering novel biomarkers for CAD.

Study Hypothesis: Despite there being evidence for roles of noncoding RNAs (ncRNAs) in EAT, only small fractions of alterations within specific classes of ncRNAs have been characterized. Thus, a more comprehensive view of ncRNA expression in the EAT of CAD patients is needed to better characterize the transcriptomic alterations occurring.

Methods: For this aim, we isolated the microRNA, long non-coding RNA, and messenger RNA (mRNA) from the EAT and subcutaneous adipose tissue (SAT) of male and female CAD patients undergoing coronary artery bypass graft as well as from nonCAD patients undergoing aortic valve replacement/repair and analyzed differential expression of these RNAs between tissues and between disease status.

Results: We demonstrate that ncRNA and mRNA expression within the EAT of CAD patients is distinct from nonCAD patients and that the differentially expressed ncRNAs correlate with targeting the following functions/pathologies: inflammation, angiogenesis, apoptosis, diabetes, heart failure, etc.

Conclusion: These findings reveal potential biomarkers for CAD and elucidate possible pathological roles of EAT in CAD.

The Case for Using Composition Tomography to evaluate Perirectal Necrotizing Fasciitis: Is It Really Necessary?

Authors: Ty Bayliss, David Denning

Department & Institution: Department of Surgery, Cabell Huntington Hospital, Marshall University Joan C Edwards School of Medicine

Background: Fournier's Gangrene; a form of necrotizing soft tissue infection affecting the penis, scrotum, or perineum is a very serious and life-threatening disease where rapid surgical intervention is important to the survival of the patient.

Clinical reviews of Fournier's Gangrene state that diagnostic protocols include interpreting CT, MRI, or Ultrasound imaging along with the patient's presenting clinical symptoms, physical exam and results of lab values.

Study Hypothesis: We think that the use of imaging techniques as a diagnostic tool is not needed because Fournier's Gangrene is a specific disease that clinicians can diagnose using physical examination and laboratory values alone.

Methods: Cases of perirectal necrotizing soft tissue infection from November 23, 2016 to April 9, 2021 admitted to St. Mary's Medical Center were reviewed. Results of physical exams and imaging were compared using parameters of severity upon admittance and length of stay. Regarding the ongoing Sars-CoV-2 pandemic, we investigated if patients were delaying seeking treatment.

Results: It was found that no additional clinically significant data was gathered using imaging techniques, particularly CT scans, that wasn't discovered from a physical examination. The only instances where imaging provided useful were when the patient presented to the clinical setting very early in the disease process. It was also found that a higher number of patients delayed seeking treatment due to the SarsCoV-2 pandemic. Our sample size was too small to determine whether the number of delayed patients was statistically significant.

Conclusion: We conclude that CT scans are not needed in diagnosing Fournier's Gangrene unless the disease is in its early stages. This should result in faster diagnosis and treatment in the operating room. This is especially important when hospital resources are compromised and the patient presents in an advanced disease state; relevant to the ongoing Sars-CoV-2 pandemic.

The Utility Of Leukocyte Esterase Test In Diagnosing Culture Negative Periprosthetic Joint Infections

Authors: Vishavpreet Singh, Alisina Shahi, Matthew Bullock, Ali Oliashirazi

Department & Institution: Marshall Orthopaedics

Background: Diagnosis of periprosthetic joint infection (PJI) is very challenging especially when the cultures are negative. The Leukocyte Esterase (LE) strip test is an inexpensive marker that is now part of the diagnostic criteria for PJI.

Study Hypothesis: The purpose of this study was to assess the performance of the LE strip test in identifying culture negative PJIs.

Methods: We conducted a retrospective study and identified 394 revision arthroplasties for PJI that had negative cultures and available LE strip test results. Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), positive likelihood ratio (+LR), and negative likelihood ratio (-LR) were calculated using both the ++ and ++/+ cutoff for the LE strip test.

Results: Using the ++ threshold, LE test had a sensitivity of 47.8%, specificity of 97.1%, PPV of 41.1%, NPV of 95.0%, +LR of 10.3, and –LR of 0.7. When the ++/+ threshold was used the LE test had 95.0% sensitivity,

85.8% specificity, 32.8% PPV, and 99.58% NPV. The +LR and –LR were 6.67 and 0.05 respectively.

Conclusion: Based on the findings of this study, the LE strip test could effectively rule out PJI in culture negative patients given its high NPV and sensitivity. Our results demonstrated that the LE test with ++ threshold has a great specificity with low sensitivity whereas the ++/+ cutoff delivers both high sensitivity and specificity. In case of a high clinical suspicion with negative cultures LE strip test is a reliable marker to rule out PJI.

Serum ESR and CRP are not reliable markers for screening/diagnosing PJI

Authors: Wade Smith, Alisina Shahi, Matthew Bullock, Ali Oliashirazi

Department & Institution: Marshall Orthopaedics

Background: Based on the recommendations of the American Academy of Orthopaedic Surgeons serum ESR and CRP are the first lines for periprosthetic joint infection (PJI) work up. The studies that shaped these guidelines frequently contained small sample sizes and rarely used a standardized definition of PJI.

Study Hypothesis: The purpose of the paper is to reexamine the sensitivity of serological tests utilizing a contemporary definition of PJI.

Methods: A retrospective review of an institutional database of 689 total joint arthroplasties (368 knees, 321 hips) that underwent surgery for PJI. The 2018 ICM definition of PJI, and the defined thresholds for various parameters, were used to categorize patients into infected and non-infected (only the major criteria). Sensitivities were calculated for serum CRP among all PJIs, ESR for chronic infections, and for both tests together.

Results: The sensitivity of these markers for diagnosing chronic PJI (defined as infection occurring greater than 6 weeks from index arthroplasty), was 74.3% (95% CI: 67.7-80.9%) for CRP, and 80.0% (95% CI: 75.484.6%) for ESR. The sensitivity of these tests combined was 82.5% (95% CI: 73.3-85.7%) for ESR or CRP to be abnormal and 78.4% (72.7-90.6%) for both markers to be elevated. The sensitivity of CRP (threshold of >100mg/L) was 64.2% (95% CI: 61.3-67.1%) for acute PJIs.

Conclusion: It appears that serum ESR and CRP have a high false negative rate than previously reported, especially in patients with acute PJIs. Antibiotic administration, PJI with low virulent organisms, and the high thresholds for these tests than previously described may be some of the reasons for the high false negative rate. Current thresholds recommended by the ICM may need to be examined and possibly lowered to improve the sensitivity of these screening tests. Surgeons should be aware that PJI may still occur despite normal serological tests and should maintain a high clinical suspicion.

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