20th Annual
USF Health Research Day
Friday, February 19, 2010
20th Annual
RESEARCH DAY
FRIDAY, FEBRUARY 19, 2010

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20th Annual RESEARCH DAY
FRIDAY, FEBRUARY 19, 2010

AGENDA

**USF Health Rotunda**

07:30am - 08:00am  Registration and Poster Set Up

08:00am - 10:30am  Session “A” - Faculty and Staff Poster Presentations

**USF Health Auditorium**

8:30am – 10:30am  1st Annual Joseph Krzanowski, PhD USF Health Invited Oral Presentations Session

**USF Health Rotunda, CON Foyer, Atrium**

11:00am -12:30pm  Session “B” - Student Poster Competition Session

**USF Health Rotunda**

12:30pm - 01:00pm  Lunch for Presenters and Judges

**USF Health Auditorium**

01:00pm - 02:00pm  14th Annual Roy H. Behnke Distinguished Lectureship

02:00pm - 02:45pm  Awards Ceremony

**USF Health Rotunda**

3:00pm – 4:00pm  Reception in USF Health Rotunda
Dear Colleagues,

This year, we have another great Research Day planned for USF Health. This year’s event is especially important because it represents the “20th Anniversary” in which Research Day has been formally organized for all of our students, trainees, staff and faculty across USF Health. A total of 194 presentations will be delivered during the day. From this total, eight student proposals have been selected for oral presentation to be held during the morning of Research Day. This oral session is especially notable for it is to recognize Dr. Joseph Krzanowski, emeritus professor and former associate dean of the College of Medicine, who was instrumental in expanding Research Day into its current form. This part of the Research Day program is to be called the “The 1st Annual Joseph Krzanowski, PhD USF Health Invited Oral Presentation Session” and is a tribute to the dedication “Joe K” had for students across USF Health and for the importance of research in the successful development of their careers.

Another important aspect of our Research Day Program is the Annual Roy H. Behnke Distinguished Lectureship. Dr. Behnke, an early faculty member of the College of Medicine and founding chair of the Department of Internal Medicine, was a nationally recognized medical educator and consummate physician. His family established this lectureship to further research at USF Health and to invite an internationally recognized Health expert to provide the Research Day keynote address on a current topic of great relevance to USF Health’s students, trainees, staff and faculty. This year, Jean-François Rossignol, MD, PhD, FRSC is providing the Annual Roy H. Behnke Distinguished Lecture. Dr. Rossignol, a Sorbonne graduate, was trained as a synthetic medicinal chemist and physician leading to an accomplished career as a research scientist. During his academic career, Dr. Rossignol developed albendazole, the first broad spectrum, single dose anthelminthic, and the antimalarial drug, halofantrine. Dr. Rossignol has published more than 100 articles in scientific or medical journals and been awarded 25 US patents. For 11 years, he served first as a consultant and later as an expert in parasitic diseases for the World Health Organization in Geneva, Switzerland. He is currently affiliated with Stanford University School of Medicine and serves as Chairman & Chief Science Officer, Romark Laboratories, L.C. His featured presentation is “Translational Research: Myth or Reality or the Discovery of the Thiazolides.”

With this 20th Anniversary of Research Day, we continue to acknowledge the importance and value of the research enterprise to USF Health and the University. Our goal is to continue to recognize, grow and enhance the research efforts of our students, trainees, staff and faculty. Our USF Health community has been instrumental in the continuing growth of research at the University of South Florida. This last year, USF Health was awarded $234 million of extramural funding of the total USF funding of $380 million. USF Health certainly contributed extensively to the University recently being recognized as the most rapidly growing institution in regards to federal expenditures from 2000-2007 by the Chronicle of Higher Education. We are committed to the goal of expanding growth and quality of research at USF Health. Research Day is but one of those special events during the year that recognizes the hard work of our researchers and the great contribution they are making to the scientific advancement of their disciplines. It is an important day for this purpose and one we hope all will enjoy.

Sincerely,

Phillip J. Marty, PhD
Associate Vice President, Office of Research
USF Health
20th Annual
RESEARCH DAY

14th Annual
Roy H. Behnke Distinguished Lectureship

PLACE:
USF Health Auditorium

TIME:
1:00pm

SPEAKER:
Dr. Jean-François Rossignol
Stanford University Consultant Professor of Medicine
Research Fellow, Exeter College, University of Oxford, U.K.
Chairman & Chief Science Officer, Romark Laboratories, L.C.

TITLE:
Translational Research:
Myth or Reality or the Discovery of the Thiazolides

Sponsored by:
Department of Internal Medicine, College of Medicine, USF
Jean-François Rossignol, M.D., Ph.D., F.R.S.C.
Chairman of the Board of Managers
and Chief Science Officer

Dr. Rossignol, a Sorbonne graduate, was trained as a synthetic medicinal chemist and physician, leading to an accomplished career as a research scientist. He spent 20 years in academia in France and in the United States before joining the pharmaceutical industry with Squibb Corporation in Princeton, New Jersey, in 1987.

In 1993, Dr. Rossignol co-founded Romark Laboratories to develop one of his early inventions, nitazoxanide, the first of a new class of drugs called the thiazolides.

During his academic career, Dr. Rossignol developed albendazole, the first broad spectrum, single dose anthelminthic, and the antimalarial, halofantrine, both for SmithKline Beckman Corporation, today GlaxoSmithKline PLC. Both drugs received regulatory approvals in many countries, including the United States, and albendazole made the World Health Organization's Essential Drug List.

Dr. Rossignol has held numerous full-time and adjunct faculty positions, published more than 100 articles in scientific or medical journals and been awarded 25 US patents. For 11 years, he served first as a consultant and later as an expert in parasitic diseases for the World Health Organization in Geneva, Switzerland. He is currently affiliated with Stanford University School of Medicine.
20th Annual
RESEARCH DAY

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- College of Medicine
- College of Nursing
- College of Public Health
- School of Biomedical Sciences
- School of Physical Therapy and Rehabilitation Sciences
- Department of Internal Medicine, College of Medicine

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<td>Associate Vice President - USF Health Office of Research, Professor, Public Health, Internal Medicine, and Psychiatry and Behavioral Medicine, Interim Director, Lawton and Rhea Chiles Center for Healthy Mothers and Babies</td>
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<tr>
<td>Patricia A. Burns, PhD, RN, FAAN</td>
<td>Senior Associate Vice President – USF Health Dean, College of Nursing</td>
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<td>William S. Quillen, PT, PhD, SCS, FACSM</td>
<td>Associate Dean and Director, School of Physical Therapy and Rehabilitative Sciences</td>
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<td>Wilbur K. Milhous, PhD</td>
<td>Associate Dean for Research, College of Public Health</td>
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COLLEGE OF MEDICINE

Policy Committee
Committee on Research
2009-2010

COLLEGE OF MEDICINE FACULTY

Cesario Borlongan, PhD
Chairman Committee on Research

BASIC SCIENTISTS

Eric Bennett, PhD
Molecular Pharmacology and Physiology
Cesario Borlongan, PhD
Neurosurgery
Gianluca Del Rossi, PhD
Orthopedics
Kersti Linask, PhD
Pediatrics
Min You, PhD
Molecular Pharmacology and Physiology

DEAN’S APPOINTMENTS (AT LARGE)

David Birk, PhD
Pathology and Cell Biology
John Carter, MD
Internal Medicine
Esteban Celis, MD, PhD
Oncologic Sciences
Javier Cuevas, PhD
Molecular Pharmacology and Physiology
Chad Dickey, PhD
Molecular Medicine/Byrd Institute
Svitlana Garbuzova-Davis, PhD
Neurosurgery
John Mayer, PhD
Physical Therapy & Rehabilitation Sciences
Michael White, PhD
Molecular Medicine
Theresa Zesiewicz, MD
Neurology

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Mary Lien, MD
Dermatology
Melissa Loscalzo, MD
Pediatrics
Jorge Lujan-Zilbermann, MD
Pediatrics
Diane Straub, MD, MPH
Pediatrics
Kathleen Rockefeller, PT, MPH
Physical Therapy & Rehabilitation Sciences

OTHER COMMITTEE MEMBERS

Phillip J. Marty, PhD
Associate VP for Research, Ex Officio
COLLEGE OF NURSING

Research Committee

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Trudy Wittenberg, BS
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Shirley Fitzgerald, PhD
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Yiliang Zhu, Ph.D.
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20th Annual RESEARCH DAY

STAFF ACKNOWLEDGEMENTS

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Ashley Hudak

USF Undergraduate Research Office
Penny Carlton
Lauren Schumacher
Naomi Yavneh, PhD
20th Annual RESEARCH DAY

POSTERBOARDS

Location: USF Health Rotunda and College of Nursing Foyer and Atrium

Set up:
Session “A” 07:30am – 08:00am
Session “B” 10:45am – 11:00am

Tear down:
Session “A” 10:30am – 10:45am
Session “B” by 1:00pm

Poster Size: 4 ft. by 6 ft.

JUDGING OF POSTERS

Student posters will be judged by faculty members.
Students must be present at their poster to be eligible for judging.

Posters will be judged on:

1. Presentation of poster
   • Organization
   • Readability
   • Appearance
   • Sense Appeal

2. Presentation of data
   • Oral Communication

3. Knowledge of subject
   • Question and answer presentation

AWARD CEREMONY

Student Winners will be announced at the Awards Ceremony following the USF Health Keynote Lecture
2010 Research Day
Abstracts

Disclaimer: Abstracts printed within are for USF Health internal use only.
### 1st ANNUAL JOSEPH KRZANOWSKI, PhD, USF HEALTH INVITED ORAL PRESENTATIONS SESSION

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<th>The Latent Human Herpesvirus-6A Genome Specifically Integrates in Telomeres of Human Chromosomes In Vivo and In Vitro</th>
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#### Lehigh Valley Health Network Research

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#### Allergy, Immunology and Infectious Diseases Research

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<td>Thomas, John C.</td>
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1st ANNUAL

JOSEPH KRZANOWSKI, PhD

USF HEALTH INVITED

ORAL PRESENTATIONS SESSION
The Latent Human Herpesvirus-6A Genome Specifically Integrates in Telomeres of Human Chromosomes In Vivo and In Vitro

Jesse H. Arbuckle1, Maria M. Medveczky1, Janos Luka2, Stephen H. Hadley1, Dharam Ablashi3, Troy Lund4, Kenny De Meirleir5, Jose G. Montoya6, and Peter G. Medveczky1
1Department of Molecular Medicine, University of South Florida, Tampa FL; 2Bioworld Consulting Laboratories, Mt. Airy, MD; 3The HHV-6 Foundation, Santa Barbara, CA; 4Department of Pediatrics, University of Minnesota, Minneapolis, MN; 5Department of Physiology, Vrije, University of Brussels, Brussels Belgium; 6Stanford University School of Medicine, Stanford, CA.

Keywords: Latency, HHV-6, Telomere, Viral integration

Objective: Human herpesvirus-6 variants A and B (HHV-6A and HHV-6B) have been implicated in bone marrow graft rejection, co-factors in AIDS progression, and various neurological diseases. The objectives of this study was to 1) identify the integration site of HHV-6A in three families, 2) to determine if HHV-6A can reproducibly integrate in vitro, and 3) determine if HHV-6A can be reactivated from its integrated state.

Methods: Peripheral blood mononuclear cells (PBMCs) isolated from individuals of three independent families previously identified as having at least one million copies of HHV-6 per ml of peripheral blood.

Results: HHV-6A DNA was co-localized with telomeric regions of one allele of chromosomes of parents and siblings 17p13.3, 18q23, and 22q, suggesting vertical transmission of the viral genome. Integration of the HHV-6A genome into TTAGGG telomere repeats was confirmed by the methods of Gardella gel and direct sequencing of the viral-chromosomal subtelomere junction. Integration of the viral genome into telomeres was also demonstrated in latently infected HEK-293 cell clones and Jihan T-cells. No circular episomal forms were detected even by PCR in DNA isolated from patient's PBMCs or HEK-293 cells. Furthermore, reactivation of integrated HHV-6A virus from individuals' PBMCs was successfully accomplished by TPA plus hydrocortisone, while TSA induced viral DNA synthesis in latently infected T-cells and HEK-293 cells.

Conclusion: Taken together, the data suggest HHV-6 is unique among human herpesviruses as this virus specifically and efficiently integrates into telomeres of chromosomes during latency. However, further research is needed to understand the implication of telomere integration and its relation to disease.

Research supported by: HHV-6 Foundation and NIH
Enhancing the Degradation of Aβ Amyloid In APP+PS1 Mice Using Raav Vector Expression of Insulysin. Dana Cruite*, Nikisha Carty, Don Williams, Justin Rizer, Daniel Lee, Kevin Nash, Marcia N. Gordon, Dave Morgan Byrd Alzheimer's Institute, Department of Molecular Pharmacology and Physiology, University of South Florida College of Medicine, Tampa, FL 33612. *Member of the Scholarly Concentration in Research for the MD program at the USF College of Medicine

Keywords: insulin-degrading enzyme, Alzheimer’s disease, Aβ clearance, viral vector, Congo Red

Objective: The pathogenesis of Alzheimer’s disease involves the abnormal accumulation of Aβ amyloid within the brain due to an imbalance between its production and removal. Therefore, one way of reducing the amount of Aβ amyloid is to enhance its degradation. In this study, we evaluate the ability of recombinant adeno-associated virus (rAAV) vectors encoding insulysin (IDE) – an enzyme normally involved in the degradation of Aβ – to decrease the amount of Aβ deposits in the brains of APP+PS1 transgenic mice.

Methods: 32 transgenic mice were divided into 2 treatment groups, which were given unilateral, intracranial injections of rAAV vectors containing 2 different forms of the IDE gene, and 2 control groups, including 8 untreated mice, and 8 mice given injections of rAAV vectors expressing green fluorescent protein. After sacrifice, the brain tissue was analyzed using immunohistochemical staining of the Aβ amyloid and of the Hemagglutinin (HA) tag present on the C-terminus of the IDE viral vector, and with a Congo Red stain of the Aβ amyloid. Slides of the stained tissue were made and are being imaged and analyzed, focusing on the frontal cortex and hippocampus injection regions.

Results: We expect to find that within the 2 control groups, there are no appreciable differences in the degree of Aβ and Congo Red staining between the injected and non-injected hemisphere of each mouse. Also, within the 2 treatment groups, we expect to see less Aβ and Congo Red staining in the injected hemisphere as compared to the non-injected hemisphere.

Conclusion: If our results confirm our expectations, we will conclude that rAAV vectors encoding the insulysin enzyme are an effective means of reducing the Aβ amyloid load in the brains of APP+PS1 transgenic mice.

Research supported by: NIH AG25509
Urinary Angiostatin is Elevated in Patients with Epithelial Ovarian Cancer

Christina Drenberg, Beatriz Saunders, George Wilbanks, Ren Chen, Patricia Kruk, Santo Nicosia – Department of Pathology and Cell Biology, College of Medicine, University of South Florida, Roberto Nicosia - Veterans Administration Puget Sound Health Care System

Keywords: angiostatin, epithelial ovarian cancer biomarker, urine, VEGF

Objective: The poor prognosis associated with epithelial ovarian cancer (EOC) is due to the lack of overt early symptoms and the absence of reliable diagnostic screening methods. Since many tumors over-express angiogenic regulators, the purpose of this study was to determine whether levels of the angiogenic molecules vascular endothelial growth factor (VEGF), hepatocyte growth factor (HGF), endostatin (ES), and angiostatin (AS) were elevated in plasma and urine from patients with EOC.

Methods: VEGF, HGF, ES and AS were assayed by ELISA in samples from pilot cohort consisting of healthy women (N=48; pre-menopausal N=23, post-menopausal N=25), women with benign gynecological disease (N=54), patients with primary peritoneal cancer (PP) (N=2) and EOC (N=35). Wherever possible, parallel serum samples were measured for CA125 levels by ELISA.

Results: AS was the angiokine that independently discriminated EOC patients from healthy individuals. Levels of urinary AS (uAS) from healthy individuals or women with benign gynecological disease averaged 21.4 ng/mL±3.7 and 41.5 ng/mL±8.8, respectively. In contrast, uAS averaged 115 ng/mL±39.2 and 276 ng/mL±45.8 from women with Stage I (N=6) and late stage (N=31) EOC, respectively. Further, uAS was elevated in EOC patients regardless of tumor grade, stage, size, histological subtype, creatinine levels, menopausal status, or patient age, but appeared to complement CA125 measurements.

Conclusion: Levels of AS are elevated in the urine of patients with EOC and may be of diagnostic and/or prognostic clinical importance. Further studies of uAS as a biomarker for EOC alone or in combination with other markers is warranted.

Research supported by: Moffitt Cancer Center Focused Interest Group (#7P30CA76292-08) US DOD New Idea Award (#W81XWH-07-01-0276)
**Ethnic Differences in HPV knowledge and Intentions among Hispanic and Non-Hispanic Males**


Department of Community and Family Health, College of Public Health, University of South Florida

**Keywords:** HPV, Males, Ethnic Differences, Psychosocial

**Objective:** Human Papillomavirus (HPV) is a well-established causal factor in cervical cancers as well as other cancers and genital warts in both females and males. A natural history study of HPV in men is being conducted at Moffitt Cancer Center, where men are tested every 6 months for genital, skin and oral HPV. This study has provided a USF COPH team the opportunity to assess cognitive and emotional effects of receiving an HPV test result among men.

**Methods:** Hispanic and non-Hispanic men were compared on items assessing HPV knowledge and vaccine intentions. A knowledge index was created by summing the number of correct answers to 20 items on a validated knowledge scale. Vaccine intention was assessed on a four-point scale with higher scores reflecting greater intentions to receive the vaccine if it became available for males. Logistic regression was used to assess differences between Hispanic and non-Hispanic men.

**Results:** Of 505 participants, 86 (17.0%) identified as Hispanic. Generally, the men were knowledgeable, answering 15 of the 20 questions correctly, with no differences in knowledge scores between Hispanic and non-Hispanic men. There were significant differences in vaccine intentions: Hispanic men reported stronger intentions to receive the HPV vaccine (OR=1.88; 95% CI=1.09-3.27); however, they were less likely to have a place to get the vaccine (OR=0.34; 95% CI=0.12-0.92) and more likely to perceive getting time off work or school as a potential barrier (OR=2.03; 95% CI=1.00-4.13).

**Conclusion:** The HPV vaccine has been approved for males for the prevention of genital warts, but not for cancer prevention. Identifying positive motives for, and barriers against, vaccine uptake among men is an important public health activity.
CARDIA: Prevalence and Socio-demographic Predictors of Pulmonary Hypertension in Young Adults
Swathy Kolli (USF), Ren Chen (USF), Maya Guglin (USF), College of Medicine, Department of Cardiology, University of South Florida

Keywords: Pulmonary Hypertension

Objective: Risk factors for pulmonary hypertension (PH) are poorly understood. The aim of current study is to describe the prevalence, socio-demographic and clinical predictors of pulmonary hypertension in young adults.

Methods: Coronary Artery Risk Development in Young Adults (CARDIA) study is a longitudinal study of young adults to assess the risk factors for cardiovascular disease. Limited access dataset provided by NHLBI was analyzed. Doppler pulmonary artery acceleration time (DPAAT) was used to assess PH. Socio-demographic characteristics including age, race, gender, body mass index (BMI), smoking status, alcohol intake, physical activity and self reported clinical variables (hypertension, diabetes mellitus, cholesterol problem, heart problems and kidney problems) were assessed.

Results: Study subjects retained in year five were included in current analysis, 3986 males and females aged 23-35 years. The prevalence of severe PH (DPAAT ≤70 msec) and mild to moderate PH (DPAAT = 109.9-70.01 msec) was 1.1% and 14%, respectively. Multivariate analysis using linear regression revealed that age (p=0.011), Hispanic race (p=0.013), male gender (p<0.0001), overweight status defined as BMI 25-29.9 kg/m2 (P<0.0001), obesity defined as BMI ≥30 kg/m2 (P<0.0001), physical activity (P=0.04), and current smoking (P=0.0001) were statistically significant variables predicting PH. Multivariate analysis using ordinal logistic regression showed that gender, physical activity, BMI, current smoking status were statistically significant predictors of PH.

Conclusion: In young adults, the prevalence of pulmonary hypertension estimated by DPAAT is high (15%). Factors predicting pulmonary hypertension include age, obesity, male gender, smoking, and low physical activity.
Cord Blood Administration Induces Oligodendrocyte Survival by Altered Protein Expression

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Keywords: Stroke, White matter, Ischemia, Microarray, Anti-oxidant

Objective: Oligodendrocytes (OLs) are the predominant cell type found in white matter and are essential to structural integrity for proper saltatory conduction. Studies in vitro have demonstrated that human umbilical cord blood (HUCB) cells directly protect primary OL cultures from oxygen glucose deprivation (OGD). This study examines changes in OL gene and protein expression induced by HUCB cells to enhance survival after an ischemic insult.

Methods: Microarray was performed on RNA prepared from primary OL cultures subjected to OGD and treated with HUCB cells. Quantitative RT-PCR followed by immunohistochemistry was utilized to verify these results. Promoter region analyses of selected genes were analyzed utilizing Genomatix software.

Results: Microarray studies revealed increases in the following genes attributed to HUCB cell treatment: U2AF homology motif kinase 1 (Uhmk1), Insulin induced gene 1 (Insig1), Metallothionein 3 (Mt3), Tetraspanin 2, Peroxiredoxin 4 (Prdx4), Stathmin-like 2, Myelin Oligodendrocyte Glycoprotein (MOG), and Versican. qRT-PCR verified microarray results. In vivo, MOG, Prdx4, Uhmk1, Insig1 and MT3 protein expression was up-regulated in the white matter of rats infused with HUCB cells 48hrs following MCAO. Promoter region analysis of selected genes revealed common transcription factor binding sites: EVI1, MZF1, NKX6.1, Pax6, GATA1, GSH-2, SOX 5 and SRF.

Conclusion: The elucidation of genes and proteins altered by HUCB cell treatment following an ischemic insult will provide insight into the signaling mechanisms of cellular survival, consequently providing new targets for an effective stroke therapy.

Research supported by: NIH (R01 NS052839) and AHA (0715096B to A.A.H.)
MRSA Prevention in the Correctional Healthcare Setting Natalia Saunders, College of Nursing, University of South Florida

Keywords: MRSA prevention, correctional healthcare, infection control

Objective: Review current literature and evidence-based practices related to methicillin-resistant staphylococcus aureus in correctional healthcare.

Methods: literature review and current practices in correctional healthcare

Results: The potential for substantial cost to employers is evident in all areas of correctional health. MRSA infections have a inherent effect on turnover rate among corrections staff.

Conclusion: Education is the key to MRSA prevention in the correctional healthcare setting. Management involvement is also critical in the success of education programs and disease prevention. Legislative involvement regarding policy and funding play a vital role in prevention the spread of MRSA from the community to the corrections setting.

Research supported by: evidence-based practices, department of corrections policy and procedure manuals, local health department statistical reports
Molecular Characterization of Resistance to Artemisinin Drugs in Plasmodium falciparum
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Keywords: Plasmodium malaria artemisinin resistance drugs

Objective: Artemisinin (QHS) and its derivatives are effective against all stages of Plasmodium spp. and they provide faster clearance of parasitemia than any other drugs. Recent studies suggest resistance to these drugs may be emerging in the world. We have developed resistant parasites and are using these lines to determine the molecular basis of QHS resistance.

Methods: Discontinuous exposure to artelinic acid (AL) or QHS in vitro produced AL and QHS resistant progeny of P. falciparum lines W2, D6, and TM91c235. Using this method, we produced parasites that could tolerate 2400ng/ml of QHS (D6), 200ng/ml QHS (W2), and 280ng/ml of AL (TM91c235). We conducted molecular analyses and in vitro susceptibility testing to dissect resistance using the most drug selected lines.

Results: Resistant D6 and W2 lines were less susceptible to some standard antimalarial drugs, but not all. Resistant parasites exhibited similar susceptibility to artemisinin drugs as parental strains, with few exceptions. Copy number of pfmdr1 was approximately 1 in all D6 lines, but was increased in resistant progeny of W2 and TM91c235 (comp to par strains). Whole genome sequencing of D6 and D6.QHS2400 identified SNPs that may be involved in resistance as well as a 76 kb amplification event. Proteomic analyses found proteins that may be differentially expressed in D6 vs. D6.QHS2400 and W2 vs. W2.QHS200.

Conclusion: Subjecting P. falciparum to increasing amounts of artemisinin drugs produced resistant parasites that will serve as valuable reagents for molecular analyses and other studies. Future research will focus on dissecting whole genome sequence and proteomic data of parental and resistant parasites to further elucidate mechanisms of resistance to artemisinin drugs.

Research supported by: NIAID, R01 AI058973
SESSION A
FACULTY AND STAFF
Abstract #: LVH-1

Presented by: James Orlando, Faculty

Teaching Leader Series: An Interprofessional Approach to Sustained Faculty Development

Keywords: Faculty Development Interprofessionalism

Objective: The purpose of the Teaching Leader Series is to assess and build workforce capabilities for teaching and competency-based education. This interprofessional workshop series is intended for all new and experienced network members who have teaching responsibilities.

Methods: This CME/CNE accredited Series is evaluated using a mixed methods approach that includes post-course evaluations, mid-year surveys and focus groups.

Results: In AY08, 14 workshops were conducted. Over 300 clinical educators attended with a composition of 50% nurses, 25% physicians and 25% other educators. Based on post-course evaluations, participants significantly increased their teaching knowledge and skills. Six-month follow-up survey results and focus group data suggested that participants most frequently incorporate small group teaching guidelines and feedback skills as a result of attending the workshops. In addition, respondents acknowledged that the interprofessional nature of the workshops fostered greater collaboration during interdisciplinary teaching rounds. In AY10, the Series was approved for additional funding and doubled its number of workshops.

Conclusion: The Teacher Leader Series increases faculty's knowledge of teaching concepts and provides tools to help assess their trainees' learning needs and enhance their knowledge and application of diverse teaching methods. At the same time, our findings suggest that an interprofessional approach may not only help sustain faculty training efforts but may also facilitate greater collaboration of clinical educators across traditional boundaries.

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Abstract #: LVH-2

Presented by: James Orlando, Faculty

Impact Study of a Central Lines Simulation Program Using Kirkpatrick’s Four-Level Evaluation Model

Keywords: Simulation Assessment and Evaluation Measurement

Objective: The study evaluates the impact of central lines simulation training program from 2006-2009 on learner and patient outcomes within the framework of Kirkpatrick’s four-level evaluation model.

Methods: The study utilizes a mixed methods, quantitative/qualitative approach to: (a) determine the relationships between course outcomes, operator practices, and patient outcomes, (b) validate the accuracy of data collected on the procedural checklist, and (c) determine how changes in training, policies and protocols impact a hospital’s central line-associated infections trend rate.

Results: Chi-square and linear regression analyses suggested that operators' Institute for Healthcare Improvement (IHI) central lines bundle compliance rate predicted their complications rate. Focus group data suggested that having a nurse in the room had an unanticipated effect of reducing the number of attempts by course-taking residents and therefore, lowering complications rate. Time-series analysis suggested that central lines training and changes in policies and practices significantly reduced the hospital's central line-associated infection trend rate since April 2005.

Conclusion: This study produced evidence suggesting that interprofessional simulation training contributes to better resident adherence with IHI Central Lines Bundle and lower complication/infection rates. Performance support mechanisms introduced in the course and presented in the clinical setting, such as a central lines checklist and peer support, reinforce course learnings and enable skill transfer. Kirkpatrick’s Four-Level Evaluation Model is a promising framework for evaluating the impact of clinical training programs on resident performance.
Abstract #: A-1

**Fetal/placental Immune Regulation: Analysis of FOXP3+ Regulatory T cells in Human Gestation**

Ileana Arbona-Ramirez, M.D., Rene Ruiz, M.D., Morna J. Dorsey, M.D., M.M.Sc.; Department of Pediatrics, Division of Neonatology, College of Medicine, University of South Florida

**Keywords:** Tregs, cord blood infants

**Objective:** In this study the infant's cord blood Treg level will be quantified using Treg extracellular markers (CD4+,CD25+) and intracellular transcription factor fork head box protein 3 (FOXP3+). Also, the Tregs can be further characterized into naturally occurring (naïve cells) characterized by CD45RA marker, memory cells that presents after antigenic exposure and are characterized by CD45RO marker and Tregs of thymic origin characterized by the CD31 marker. It has been noted that the phenotypic characteristics in Tregs may differ between adults and infants, so we will characterize the phenotypic markers seen in the infant's cord blood Tregs. After quantification of the Tregs in infant's cord blood we will observe the relationship between the gestational age and the level of the Tregs and correlate with demographic and clinical data.

**Methods:** Umbilical cord blood is obtained from placentas from 24 to 42 weeks gestational age at the time of delivery. Infant’s with hx of maternal chronic steroid use or maternal HIV were excluded. Then, mononuclear cells are isolated by PBMC-Ficoll separation and Treg markers are identified by fluorescent staining and quantified by flow cytometry. Maternal medical and obstetric history was obtained to correlate with results.

**Results:** Preliminary data from 17 infants between 35 and 41 weeks gestational age show that Treg cells comprise 5.8% ± 0.2 (m ± SD) of the umbilical cord blood T cell populations. A significant (p=0.013) negative correlation between Treg % and GA.There is a significant decline in Treg / Total T cells with advancing gestation.

**Conclusion:** Expected outcome: We expect to find a Treg is a discrete population in cord blood that will decrease with advance in gestational age.

Abstract #: A-2

**Elevated IL-8 Levels in Allergic Airways Inflammation is Downregulated Following Aerosolized Brevetoxin Exposure**

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**Keywords:** Asthma, allergy, inflammation, cytokines, red tide

**Objective:** We examined cytokine production in patients with pre existing allergic airways inflammation following exposure to aerosolized brevetoxin.

**Methods:** Using an established method of collection with filter paper cytokines in nasal secretion were examined using cytokine bead array analysis on samples from allergic asthmatic subjects (n = 57) and healthy controls (n = 31) to establish baseline differences. Samples were collected from allergic asthmatic subjects (n = 26) and healthy controls (n = 13) before and after one hour of exposure to brevetoxin during a Florida red tide bloom. Ex vivo studies were performed on PBMCs cultured in purified brevetoxin.

**Results:** Baseline increased IL-8 levels were observed in allergic asthmatic subjects compared to healthy controls (p = 0.004). Brevetoxin exposure in allergic asthmatic subjects resulted in decrease in IL-8 (p = 0.018), while a trend toward significant decrease was seen in healthy controls. PBMCs in combined LPS and Pbtx culture showed significant decreases in IL-8, IL-6, and IL-10 production when compared to LPS culture alone.

**Conclusion:** Cytokine production within the airway epithelium following brevetoxin exposure does not appear to involve early inflammatory cell recruitment pathways. Pre existing Th2 immune deviation results in more pronounced absence of typical Th1 pro inflammatory response during acute phase of inflammation. Brevetoxin demonstrates anti-inflammatory effects on immune cells. Respiratory symptoms during brevetoxin exposure are not due to increased inflammation.

**Research supported by:** NOAA OHHI NA05NOS4781248
**Parainfluenza Virus Type 3 N-terminally truncated C protein, CNΔ25, is a potent Inhibitor of Viral Replication in Mice**

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¹Division of Allergy and Immunology, The Joy McCann Culverhouse Airway Disease center, Department of Internal Medicine, College of Medicine, USF,² Cleveland Clinic Foundation. ³Department of Molecular Medicine, University of South Florida, College of Medicine and VA Hospital, Tampa, Florida

**Keywords:** Parainfluenza virus, Mouse lung inflammation.

**Objective:** Human parainfluenza viruses (HPIVs) cause serious lower respiratory tract disease with repeat infection especially among the elderly and immunocompromised patients. Currently there is no vaccine. The C protein of HPIV-3 is a multifunctional accessory protein that inhibits viral transcription and interferon (IFN) signaling. The main goal of this study was to determine the effect of a truncated C protein, CNΔ25, on HPIV-3 infection in a mouse model.

**Methods:** We treated mice with a CNΔ25 expression plasmid and examined the effect on HPIV-3 titers, lung inflammation and cytokine profile. BALB/c mice were treated intranasally (i.n.) with chitosan nanoparticles alone or containing pCNΔ25 or vector pcDNA3 on days 1 and 2. On day 3, the three groups of mice were given PBS (vehicle, negative control) or 7x10⁶ MOI HPIV3 i.n. Mice were euthanatized on day 8 and bronchoalveolar lavage (BAL) fluid and lungs were taken. Differential cell counts in BAL fluid, histopathology in lung sections, cytokine levels and virus titers were determined.

**Results:** pCNΔ25-treated mice showed fewer eosinophils in BALF, less damage to lung mucosal epithelium and fewer infiltrating inflammatory cells. The HPIV-3 titer was significantly lower in pCNΔ25- treated mice compared to the vector control group. Interleukin-4, -5, and -13, IFN-g, and TNF-a were significantly decreased in pCNΔ25-treated mice.

**Conclusion:** Truncated C protein of HPIV-3, CNΔ25, significantly decreased virus titer and lung inflammation in a mouse model of HPIV infection. These findings suggest that CNΔ25 could be used as an antiviral agent to prevent HPIV3 infection in humans.

**Research supported by:** Division of Allergy and Immunology, The Joy McCann Culverhouse Airway Disease center.

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**Glutathione Metabolism in Young and Aged Ischemic Wounds**

Andrea N. Moor, PhD, University of South Florida, Dept. of Surgery, Evan Tummel, MD, James A. Haley Veteran's Hospital, Lisa J. Gould, MD, PhD, University of South Florida and James A. Haley Veteran's Hospital

**Keywords:** glutathione, ischemia, hyperbaric oxygen, wounds

**Objective:** Hyperbaric oxygen (HBO) is often used to treat chronic wounds, however the mechanism of action remains unclear. We examined the impact of HBO on the glutathione antioxidant defense system in ischemic wounds of young and aged Fisher rats.

**Methods:** Rats underwent surgery to create an ischemic flap with 6mm circular wounds made within (ischemic) and adjacent to the flap (non-ischemic). Daily HBO (90 minutes, 2.4 atm) was compared to normoxia. Wounds were harvested at 7, 14 and 21 days. Wound lysates were analyzed for the regulatory subunit of γ-glutamylcysteine synthetase (GCLM), glutathione reductase, glutathione peroxidase and total glutathione.

**Results:** Non-ischemic wounds were healed by day 14, while ischemic wounds remained open through day 21 despite HBO treatment. In young rats, non-ischemic wound glutathione decreased over time and GCLM did not change significantly. In aged rats, non-ischemic wound glutathione and GCLM peaked at day 14 and declined as wounds healed. Conversely, in ischemic wounds, glutathione was nearly undetectable at day 7, rose rapidly by day 14 and remained elevated. Aged ischemic wounds treated with HBO showed a marked but transient induction of GCLM at day 14 with a 10-fold increase in glutathione by day 21. Glutathione peroxidase and reductase levels were not different between groups.

**Conclusion:** These results suggest that wound healing requires induction of GCLM in aged rats. In contrast, there is minimal de-novo synthesis of glutathione in wounds of young rats, indicating sufficient pre-existing antioxidant defense. HBO treatment of ischemic wounds in aged rats results in a late but substantial increase of de-novo glutathione synthesis, suggesting a marked, albeit delayed adaptive response to oxidative stress.
A University-Corporate Partnership to Enhance Vaccination Rates among the Elderly: An Example of Corporate Public Health Delivery

John T. Sinnott (1), Helen Georgiev (1), Karina D’Souza (1), Leela Mundra (1), Robin Sistruk (2), Jennifer Pytlarz (2), Jacqueline Wooley (3)(1) Office of International Affairs, College of Medicine, University of South Florida (2) Publix Supermarkets, Inc. (3) Brown University

Keywords: Mass vaccination, university-corporate partnership

Objective: Public health campaigns often rely on government programs and government-sponsored clinics for vaccine initiation. However, travel is often required for these programs and the elderly may find accessing services difficult.

Methods: We employed the unique strategy of using a pharmaceutical chain in collaboration with a university to address the public health issue of mass vaccination and to see if the elderly were more easily accessed. Florida statutes were changed in 2005 to allow vaccinations by trained pharmacists. Publix Pharmacy and the University of South Florida (USF) jointly developed a handbook and training program to facilitate these vaccinations. The injections were provided “at cost.”

Results: Between 2008 and 2009, the Publix-USF partnership vaccinated 30,063 people against influenza A (H3N2), 29,404 of which resided in Florida. The age range was 1 to 105 years old and the median age was 65 years old. Seventy-six percent of the participants were over 50 years old. The reported side effects of the vaccine were not serious, but included vertigo, cold sweats, chills, vomiting, syncope, rash, nausea, stomach pain, elevated BP, injection site reaction, inflamed bursa, and bilateral thigh discomfort. No patients were hospitalized. An income by zip code analysis revealed 44% of those vaccinated resided in zip codes where the average household income was less than $50,000 while 56% had an average income of greater than $50,000 per year.

Conclusion: This unique university-corporate partnership successfully delivered H3N2 vaccine to a vulnerable cross-section of society at low cost with minimum side effects. University-corporate partnerships are effective at reaching the aging population and delivering vaccines with few side effects.

Activity of Ciprofloxacin Derivatives Against Facultative Intracellular Bacteria

John C. Thomas1*, Rebecca J. Kapolka1, Ryan Cormier2, Glenn Roma1, Edward Turos2 and Burt E. Anderson1,1 Department of Molecular Medicine, College of Medicine and 2Department of Chemistry, College of Arts and Science, University of South Florida, Tampa, FL

Keywords: Bartonella, Francisella, Ciprofloxacin, fluoroquinolone, drug-design

Objective: To determine the anti-bacterial activity of N-acyl ciprofloxacin analogues against intracellular bacteria.

Methods: In vitro testing of Bartonella was performed by Kirby-Bauer disk diffusion assays, agar dilution testing and in cell culture with HMEC-1 human endothelial cells to assess intracellular activity of each novel fluoroquinolone compound. Additionally, the activity of these ciprofloxacin compounds against F. tularensis subspecies holartica was determined in vitro by broth dilution testing.

Results: Antibacterial activity varied widely, but 7 compounds: RC4-17, RC4-28, RC4-29, RC4-32, RC4-48, RC5-58, and RC5-69, showed significant anti-Bartonella capabilities. Multiple ciprofloxacin derivatives were shown to exhibit activity against Francisella tularensis subspecies holartica. Activity of these novel compounds was shown to correlate with inhibition of DNA gyrase.

Conclusion: Results suggest that synthetically derived N-acyl ciprofloxacin derivatives may be effective therapeutic alternatives for the treatment of infections caused by facultative intracellular bacteria.

Research supported by: This work was supported by DOD/DARPA grant HR0011-08-1-0087; Entitled “Drug Discovery, Design, and Delivery”. Approved for Public Release, Distribution Unlimited.
Abstract #: A-7
Presented by: Sarah Thomas, MS, Staff

Episomally Encoded Antisense RNA as a Screening Tool for Potential Virulence Regulator Genes in Bartonella henselae
Sarah Thomas, Izabella Perkins, Colton Faza, Lisa Smith and Burt E. Anderson:
Department of Molecular Medicine, College of Medicine, University of South Florida, Tampa, FL

Keywords: Bartonella, henselae, antisense, virulence, gene

Objective: Knowledge of the mechanisms by which Bartonella henselae regulates genes required for association with eukaryotic cells is largely unknown. In an effort to identify regulatory circuits required for virulence by this bacterium, the entire genome of B. henselae was screened for potential gene regulators and several candidate genes were chosen. The construction of gene knockouts to assign gene function has proven very difficult for B. henselae. Therefore, we have developed an antisense RNA knockdown method for use in B. henselae as an alternative to chromosomal knockouts for more rapid screening of candidate genes.

Methods: Constructs generating antisense RNA corresponding to full length genes and the upstream ribosome binding site were employed. Antisense plasmids were created for five genes of interest in B. henselae: BH620, BH11650, BH14160, BH14230, and BH16140. Each plasmid was electroporated into B. henselae Houston-1, and phenotypes were assessed by infecting human microvascular endothelial cells (HMEC -1) with these strains. Each gene was evaluated by counting the number of viable intracellular bacteria.

Results: All antisense strains had a reduced ability to invade and/ or survive within the endothelial cells as shown by lower colony forming units (CFUs) compared to the empty vector control, pNS2Trc.

Conclusion: Methods used in these experiments provide a novel and rapid way of screening for genes that play a role in cell invasion.

Research supported by: This work was supported by NIH/NIAID grant AI038178.

Abstract #: A-8
Presented by: Doris Wiener, MS, Faculty

Effects of Carotenoids on Differential Expression of Scavenger Receptors and Cytokines in Human Monocyte-Derived Macrophage Subpopulations
Doris Wiener, Xiaoming Gong, Raju Marisiddaiah, Lewis Rubin, Dept. Pediatrics, College of Medicine, University of South Florida

Keywords: Carotenoid, macrophage, cytokine, scavenger receptor

Objective: Circulating monocytes differentiate into macrophage (MΦ) subpopulations in response to environmental signals. Certain carotenoids, biologically active dietary components, may reduce MΦ oxidant stress, alter the inflammatory response, and lower risk of atherosclerosis in vivo. These observations prompted us to determine the effects of carotene (β-carotene, lycopene) and xanthophyll (astaxanthin, lutein) carotenoid classes on differentiation of cultured normal human monocytes into MΦs (M1 and M2) phenotypes.

Methods: Monocyte-to-MΦ differentiation was driven by treatment for 6 days with GM-CSF or M-CSF ± individual carotenoids. M1 MΦs (+GM-CSF) were activated with LPS + IFNγ; M2 MΦs (+M-CSF) were activated with IL-4.

Results: We validated that M1 and M2 cells showed distinct morphologies and expression patterns (qRT-PCR) of scavenger receptors (SR-A, LDLR, CD36, SR-B1) and cytokines (IL-10, IL-12). M1 and M2 markers (CD80, CD163, CD36, CD14 and CD16) and cytokines (PGE2, IL-8, IL-6, IL-10, IL-12, TNFα, IFNγ) were confirmed by flow cytometry. Expression of CD14, CD16, CD163, and IL-10 was higher in M2 than M1 MΦs. Lycopene and astaxanthin decreased SRA, CD36 and IL-10 expression in M1 cells but had no significant effect on M2 cells. Lutein also reduced SRA, CD36 and IL-10 expression in M2. Conversely, β-carotene increased M1 and decreased M2 expression of SRA, CD36, IL-10 and IL-12.

Conclusion: In sum, these results show individual carotenoids can regulate MΦ scavenger receptor and cytokine expression. We speculate that dietary components (carotenoids) influence MΦ polarization, thereby altering tissue inflammation and, via scavenger receptors, inhibiting cholesterol accumulation in MΦ-derived atherogenic foam cells.

Research supported by: Pamela and Leslie Muma Endowment
Atrial Natriuretic Peptide Receptor Signaling Plays a Critical Role in Induction of Tolerogenic Dendritic Cells

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Keywords: Dendritic Cell, Atrial Natriuretic Peptide Receptor, TLR2, SOCS3

Objective: We sought to determine whether ANP receptor, NPRA, signaling modulates DC function and analyze the downstream signaling events.

Methods: The role of NPRA signaling in modulating DC function was examined using ANP and NPRA-signaling inhibiting peptide NP73-102. NPRA signaling modulating immune outcome, the potential downstream molecules and the protein interactions among these molecules involved in this signaling pathway were investigated using immunoblotting and immunoprecipitate assays based on human monocyte-derived DC (hmDC) model. Induced regulatory T cell phenotype and function were analyzed using flow cytometry and BrdU proliferation ELISA assay. To further determine whether NPRA signaling in DCs influences lung inflammation induction, bone marrow-derived DCs generated from wild-type C57BL/6 mice were incubated with ovalbumin (OVA) and injected i.v. into NPRA-/- C57BL/6 mice, which were sensitized and challenged with OVA. Lung sections were analyzed by H.E. staining and cytokines were measured in bronchoalveolar lavage (BAL) fluid collected from parallel groups of mice.

Results: The results demonstrate that down-regulation of NPRA signaling in DCs primes DCs to induce regulatory T cells and attenuation of lung inflammation in DC adoptive transfer mouse model. NPRA is associated with TLR-2, SOCS3 and STAT3 and affects induction of IL-6, IL-10 and TGF-β but not IL-12.

Conclusion: TLR2 and SOCS3 are integral to NPRA signaling in DCs leading to immunity or tolerance induction.

Research supported by: Veterans' Affairs Merit Review Award and US National Institutes of Health #5RO1HL71101-01A2.

Differential Expression of Carotenoid Metabolism Related Genes in Human Lung Epithelial Cells

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Keywords: Carotenoid, metabolism, gene expression, lung epithelial cell

Objective: Epidemiologic studies have shown that dietary β-carotene (BC) and total carotenoids are inversely associated with non-small cell lung cancer (NSCLC) risk. However, in clinical trials, high-dose BC supplementation for chronic smokers has been linked to increased lung cancer risk. Little is known about the carotenoid metabolomic alterations that characterize neoplastic progression.

Methods: Using two human pulmonary cell lines, we determined the expression of genes regulating carotenoid metabolism in normal bronchial epithelial BEAS-2B and lung adenocarcinoma A549 cells. We assayed gene pathways including ALDH1A2, ALDH8A1, CMO1, CMO2, Cyp26A1, Cyp26B1, Cyp26C1, LRAT, RHD5, RHD8, RDH11 ~ 14, RHD16 and SDR16C5 by qRT-PCR.

Results: Carotenoid metabolism-related genes are differentially expressed in BEAS-2B and A549 cells. BEAS-2B cells express high levels of CMO2 and LRAT but not CMO1, Cyp26A1 or Cyp26B1, indicating an active lycopene metabolic apparatus. In contrast, A549 cells highly express CMO1, ALDH1A2, Cyp26A1 and Cyp26B1. In A549 cells, we could detect only low levels of CMO2 and no LRAT expression, implying presence of active BC metabolism. We tested this hypothesis by treating BEAS-2B and A549 cells with lycopene. CMO2 and LRAT expression were increased but there were no effects on CMO1 expression. Finally, immunostaining of NSCLC tissues shows profuse CMO1 staining in the tumors but little in adjacent normal lung.

Conclusion: These findings suggest that specific beneficial effects of carotenoids on lung health may be linked to CMO2-dependence in tissue. A shift to decreased expression of CMO2 and LRAT and increased CMO1 and Cyp26 family genes can alter carotene metabolism and may contribute to lung carcinogenesis.

Research supported by: NIH HD42174, Muma Family Endowment
Information Seeking Behavior of Oncologists on Benefits and Harms of Cancer Treatments in the Context of Evidence-Based-Medicine - A Qualitative Study

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Keywords: oncologists, information-seeking, benefits, harms, cancer treatments

Objective: To assess the information seeking behavior of oncologists regarding questions on benefits and harms of cancer treatments, and to discern how medical oncologists judge scientific reliability of the accessed information.

Methods: Qualitative, semi-structured, in-depth interviews with 31 oncologists at a cancer center in the United States. Stratified random purposive sampling was used.

Results: Majority of oncologists used the Internet as a primary source to find information on benefits and harms of cancer treatments. In particular, PubMed and Up-to-date were accessed most often, followed by oncology association websites (ASCO, NCCN etc.) and Google. A small number of oncologists relied on peers and experts in the field. Majority of oncologists judged scientific reliability based on the reputation of authors, journal or website. Few judged reliability based on sample size, or data analysis methods, or by publication date. A small minority determined reliability by study design or methodological quality. Very few participants indicated considering hierarchy of evidence in determining scientific reliability.

Conclusion: To our knowledge, this is the first qualitative study on information seeking behavior of oncologists specifically in regards to benefits and harms of cancer treatments. Results show urgency for creating awareness among oncologists about Evidence Based Medicine issues and imparting skills for the same.

Research supported by: American Cancer Society's Institutional Review Grant # 60-14599-01-07

Tumor Location Does Not Impact Survival After Resection for Pancreatic Adenocarcinoma Despite Larger Tumors in the Tail

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Keywords: Pancreatic Adenocarcinoma Pancreatic Tail

Objective: Convention dictates that patients with adenocarcinoma arising in the tail of the pancreas have shorter survival than patients with adenocarcinoma in the head of the pancreas. We aim to determine the differences in tumor characteristics and survival between resectable adenocarcinomas of the pancreatic tail and head.

Methods: Since 1992, data has been collected on all patients undergoing pancreaticoduodenectomy (PD) or distal pancreatectomy with splenectomy (DPS) for adenocarcinoma. Survival comparisons were undertaken using Mantel-Cox survival curve analysis. Median data are presented.

Results: 220 patients underwent PD and 33 patients underwent DPS for adenocarcinoma. Tumors in the head or tail were similar by T stage, N stage, AJCC stage, and margin status though tumors leading to DPS were larger (4 cm) than tumors leading to PD (3 cm, p=0.005). Survival at each location was impacted by T stage, N stage, and AJCC stage (p<0.05). Margin status impacted survival in patients undergoing PD (p=0.004), but did not in patients undergoing DPS (p=0.73). Overall survival was similar after PD (17 months) vs. DPS (15 months; p=0.74). However, when comparing tumors in the head vs. tail, there were no differences in survival by T stage, N stage, AJCC stage or margin status.

Conclusion: Survival is unsatisfactory though similar after resection of cancers in the head or tail of the pancreas, despite generally larger tumors in the tail. Survival is improved, irrespective of location, by complete tumor extirpation (R0 resection). By most descriptors, adenocarcinomas in the head or tail of the pancreas are similar; margin status is the only operative factor over which surgeons have control and complete tumor extirpation should be aggressively sought.
Abstract #: A-13
Presented by: Asmita Mhaskar, Staff

Timing of First-line Cancer Treatments: Early versus Late – A Systematic Review of Phase III Randomized trials
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Keywords: Systematic review, Meta-analysis, cancer treatment, timing

Objective: If delaying cancer treatment results in outcomes comparable to early treatment, the window of opportunity created can be utilized by cancer patients to address time-sensitive issues. The objective of this research was therefore to conduct a systematic review and meta-analysis of phase III randomized controlled trials comparing efficacy of early versus late first-line treatments for cancer.

Methods: A comprehensive literature search of MEDLINE and Cochrane library databases was performed (1966-2008). Data was extracted on overall survival, progression-free survival, and response rate, and pooled as per the methods recommended by the Cochrane Collaboration.

Results: Of the 570 identified studies, 10 (3811 patients) met inclusion criteria: 3 each in prostate cancer and multiple myeloma (MM), 2 in chronic lymphocytic leukemia (CLL), and 1 each in lung cancer, and follicular lymphoma. The analyses showed no survival benefit with early treatment except in prostate cancer (hazard ratio [HR] =1.21, 95% CI 1.09 to 1.35 p<0.001). There was no survival difference in MM (HR =1.10, 95% CI 0.54 to 2.24 p=0.78), CLL (HR =0.76, 95% CI 0.56 to 1.04 p=0.09), lung cancer (HR =0.95, 95% CI 0.72 to 1.24 p=0.71), or follicular lymphoma (HR=1, 95% CI 0.55 to 1.83 p=0.99). No statistically significant difference in response rate between early and late treatment was detected in any cancer type.

Conclusion: Data shows that delaying cancer treatments does not necessarily compromise therapeutic outcomes except possibly in locally advanced prostate cancer. These findings provide a unique window to oncologists and patients to address time sensitive issues if desired by patients.

Abstract #: A-14
Presented by: Rahul Mhaskar, MPH, Staff

Bisphosphonates in Multiple Myeloma. Systematic Review and Meta Analysis.
Rahul Mhaskar 1, Jasmina Redzepovic 2, Keith Wheatley 3, Otavio Augusto Camara Clark 4, Branko Miladinovic 1, Axel Glasmacher 5, Ambuj Kumar 6, Benjamin Djulbegovic 7.  1 Center for Evidence-based medicine and Health Outcomes Research, University of South Florida, Tampa, Florida, USA  2 Bayer Schering Pharma AG, Berlin, Germany  3 Cancer Research UK Clinical Trials Unit, School of Cancer Sciences, University of Birmingham, Birmingham, UK  4 EVIDENCIAS, Scientific Solutions in Healthcare, San Paolo, Brazil  5 Haematolgy and Oncology, University of Bonn, Bonn, Germany  6 Moffitt Cancer Center, Tampa, Florida

Keywords: Bisphosphonates, Myeloma, Systematic review, Meta-analyses, Osteonecrosis

Objective: The exact clinical role of bisphosphonates which are specific inhibitors of osteoclastic activity in multiple myeloma (MM) remains unclear. Hence we conducted the systematic review and meta analysis to address the role of bisphosphonates in MM.

Methods: Randomized controlled trials with a parallel design addressing bisphosphonate in MM and observational studies examining bisphosphonates related osteonecrosis of jaw (ONJ) were selected. All pooled data are reported using either hazard ratio or risk ratio.

Results: Seventeen trials were included with 1520 patients in bisphosphonates groups, and 1490 in control groups. In comparison with placebo / no treatment, the pooled analysis demonstrated the beneficial effect of bisphosphonates on prevention of pathological vertebral fractures [RR= 0.74 (95% CI: 0.62 to 0.89), P = 0.001], total skeletal related events (SREs) [RR= 0.80 (95% CI: 0.72 to 0.89), P < 0.0001] and on amelioration of pain [RR = 0.75 (95% CI: 0.60 to 0.95), P = 0.01]. The indirect meta-analyses did not find the superiority of any particular type of bisphosphonate over others. The identified observational studies suggested that ONJ may be a common event (range: 0% to 51%).

Conclusion: Adding bisphosphonates to the treatment of MM reduces pathological vertebral fractures, SREs and pain but not mortality. Assuming the baseline risk of 20% to 50% for vertebral fracture without treatment, we estimate that between 8 to 20 MM patients should be treated to prevent vertebral fracture(s) in one patient. Also, with the baseline risk of 35% to 86% for SREs without treatment, we estimate that between 6 to15 MM patients should be treated to prevent SRE(s) in one patient. No bisphosphonate appears to be superior to others.

Research supported by: University of South Florida
**Abstract #:** A-15

**Presented by:** Connor Morton, BS, Staff

**Deregulation of the Rb/E2F Pathway via Loss of p16 Expression in Pancreatic Adenocarcinoma**


Dept. of Surgery, College of Medicine, University of South Florida

**Keywords:** Deregulation Rb/E2F P16 Adenocarcinoma

**Objective:** Deregulation of pathways governing cell cycling via chromosomal deletion or mutational inactivation may predict clinical outcomes for patients with pancreatic adenocarcinoma. We aim to interrogate relationships between tumor genetics and clinical outcomes.

**Methods:** Cytogenetic analysis was undertaken on ten cell lines. PCR analysis, SSCP analysis, and northern blotting were used to determine the status of regulators of the Rb/E2F pathway. Immunohistochemical staining for p16 was performed on FFPE tissues, and correlated with clinical outcomes.

**Results:** Pancreatic adenocarcinoma cell lines frequently possessed chromosomal abnormalities in the following locations; 3p, 6p, 8p, 9p, 17p, and 18q. Given the high frequency of deletions involving chromosome 9p, we sequenced the 3 exons of CDKN2A and found homozygous deletions for 7 of 10 cell lines. DNA sequence mutations were detected in the other 3 cell lines expressing CDKN2A mRNA. In order to determine the clinical significance of p16 protein expression, immunohistochemical staining was undertaken on 26 patients who underwent resections for pancreatic adenocarcinoma [AJCC Stage IB (3), IIA (8), and IIB (15)] and completed adjuvant gemcitabine-based chemotherapy. Median overall survival was 20 months. p16 protein expression was absent in 96% of patients. Pan-INSs were identified with adenocarcinoma in two patients; both cancers were p16 negative while the associated Pan-IN was p16 positive.

**Conclusion:** Deregulation of the Rb/E2f pathway, specifically via deletions or mutations of CDKN2A (p16), appears to be a near ubiquitous finding among pancreatic adenocarcinomas making it a possible target for immuno-diagnosis / therapy. Furthermore, p16 may play an integral role in tumorigenesis.

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**Abstract #:** A-16

**Presented by:** Kejal Patel, MS, Staff

**Curcumin-Genistein Nanocomplex: a Potent Drug for Prostate Cancer**

Kejal Patel1, Xiaoqin Wang1, Ronil Patel1, Vikas Sharma1, Sandhya Boyapalle2,3, Julio Garay2,3, Shyam Mohapatra2,4 and Subhra Mohapatra1,4

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**Background & Objectives:** Curcumin, an ingredient of Indian curry spice, has anti-inflammatory, antioxidant and anticarcinogenic activities. It inhibits proliferation of tumor cells in culture, prevents carcinogen-induced cancers in rodents and inhibits the growth of tumors in animal models. However, curcumin is only slightly water soluble and its bioavailability is poor. Genistein is a soybean isoflavone antioxidant that inhibits tyrosine kinase activity. Prostate cancer (PCa), is the second leading cause of cancer-related death among U.S. men. Identification of nontoxic agents that delay the onset and/or progression of PCa is the goal of this project. Here we test nanocomplex formulations containing curcumin and genistein for increasing water solubility and anticancer potential to treat PCa.

**Methods:** We prepared curcumin-genistein nanocomplexes and tested their cytotoxic effects on different PCa cell lines and normal cells by WST proliferation assay. Apoptosis induced by curcumin-genistein nanocomplexes was determined by terminal transferase dUTP nick end labeling (TUNEL) assay and poly-ADP polymerase (PARP) cleavage.

**Results:** Formulation of curcumin-genistein nanocomplexes increased curcumin’s water solubility and resulted in significantly greater cytotoxicity on PCa cells than curcumin alone. Treatment of PCa cells with nanocomplex caused more apoptosis than free curcumin and genistein as measured by TUNEL assay and PARP cleavage.

**Conclusion:** Nanocomplex formulations of a curcumin-genistein mixture show enhanced water solubility of curcumin and anticancer activity against prostate cancer cells. Nanocomplex therapy may prove to be very effective in preventing or treating PCa.

**Research supported by:** NIH
**Abstract #**: A-17

**Presented by**: Ronil Patel, MS staff

**Association of Natriuretic Peptide Receptor A Expression with Prostate Cancer Progression.** Ronil Patel¹, Xiaoqin Wang¹, Kejal Patel¹, Gary Hellermann², Sonya Kong³, Alex Lopez³, Domenico Coppola³, Shyam Mohapatra²,³ and Subhra Mohapatra¹,³,⁴ Departments of ¹Molecular Medicine and ²Internal Medicine, University of South Florida College of Medicine, ³H. Lee Moffitt Cancer Center, ⁴JAH VA Hospital, Tampa, FL

**Objective**: Natriuretic peptide receptor A (NPRA) is expressed on human prostate cancer (CaP) cell lines; however its expression in human CaPs is unknown. To evaluate the role of NPRA in CaP progression, its expression was examined in benign prostatic hyperplasia (BPH), high grade PIN (prostatic intraepithelial neoplasm) and prostatic adenocarcinoma.

**Methods**: NPRA expression was examined at the Moffitt Cancer Center Tissue Core using a human CaP tissue microarray (TMA) containing 240 samples. The TMA samples included BPH (n = 24), regular prostatic intraepithelial neoplasm (PIN-R) (n = 21), high PIN (PIN-H) (n = 14), prostate carcinoma (PC) with a Gleason score of 6 (n = 33), PC with a Gleason score of 7 (n = 82), PC with a Gleason score of 8 and up (n = 51) and androgen-independent (AI) PC (n = 15). The TMA slide was stained using an in-house human NPRA antibody in a Ventana Discovery XT automated system and the data statistically analyzed.

**Results**: Epithelial cell NPRA staining was weak for the majority of BPH and PIN-R samples, weak to moderate in PIN-H, moderate to strong in Gleason-6 and uniformly strong in epithelial tumors of Gleason-7 and -8 and in AI samples. Moderate staining was seen in stromal and inflammatory cells. Analyses of medians by chi-square and P-K test showed a strong association between intensity of NPRA staining and CaP stage.

**Conclusion**: There is a significant difference in staining intensity between BPH, PIN and prostatic adenocarcinoma, suggesting that NPRA expression is strongly associated with CaP progression.

**Research supported by**: Bankhead Coley Cancer Research Program

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**Abstract #**: A-18

**Presented by**: Athanasios Tsalatsanis, PhD, Staff

**Small World Networks and Treatment Discovery Process in Cancer** Athanasios Tsalatsanis¹, Izotk Hozo², Benjamin Djulbegovic¹, ¹USF Health Center for Evidence-Based Based Medicine and Health Outcomes Research, University of South Florida, ²University of Indiana, Department of Mathematics

**Keywords**: Social networks, clinical trials, small worlds, oncology

**Objective**: In this work we study the impact of social interactions between randomized controlled trials (RCTs) on treatment discovery in cancer. We hypothesize that treatment discovery is a function of the extent of interactions between RCTs.

**Methods**: Social network analysis is utilized to model the role of RCT interactions to treatment discovery. We study 280 RCTs enrolling 91,847 patients conducted by NCI co-operative groups (COGs) from 1955 to 2006. Each trial is described by three components: the COG that proposed the trial, the type of disease and the type of treatment studied. Interactions between trials occur for trials that share common components.

**Results**: The pattern of interactions between trials follows the small-world network model: trials are connected through a small number of ties. In comparison to a random model, the information exchanged within the COG improved by 45%. Similarly, the COG network exhibited greater than 10 times tendency to cluster together. We further found that trials with the highest centrality measures were the ones that tested curative/definitive treatments in solid tumors. The most influential group within the NCI network was the ECOG COG. However, the observed higher level of connectivity in the network did not translate into predictably higher treatment success as measured by improvement in survival.

**Conclusion**: The pattern of therapeutic discovery adheres to the small world network model. On surface it appears counterintuitive that there is no direct relationship between treatment success and RCT interactions. We explain our findings by the role of social network to maintain equipoise in RCTs, which in turn preserves unpredictability of the results at individual RCT level.

**Research supported by**: NIH Grant: 1R01CA133594-01
**Natriuretic Peptide Receptor A regulates Macrophage Inhibitory Factor expression in Prostate Cancer**  
Xiaoqin Wang, Hongyu Zheng, Murali Kanakenahalli, Ronil Patel, Kajel Patel, Vikas Sharma and Subhra Mohapatra  
Department of Molecular Medicine, College of Medicine, University of South Florida

**Objective:** We have recently demonstrated that mice deficient in atrial natriuretic peptide receptor A (NPRA-KO) cannot support the growth of implanted prostate tumor cells and downregulation of NPRA expression by siNPRA or NPRA inhibitor induced apoptosis in PCa cells and reduced tumor burden in mice. However, the precise mechanism of NPRA action in PCa remains unclear. Macrophage migration inhibitory factor (MIF), a proinflammatory cytokine, is overexpressed in prostate cancer and unique for its functions in many processes associated with tumor survival. In this study, we investigated the effect and associated mechanisms of NPRA and MIF pathways in a transgenic adenocarcinoma of mouse prostate (TRAMP) model.

**Methods:** In vivo expression of NPRA and MIF was checked by RT-PCR, Western blot and ELISA assay in prostate tissues of TRAMP mice as well as TR-C1 cell transplanted C57/BL6 mice. The role of NPRA deficiency in modulating MIF signaling was examined in PCa cell lines derived from TRAMP mouse prostate (TRAMP-C1) and in TRAMP-C1 xenograft treated with NPRA inhibitor.

**Results:** NPRA expression co-related with MIF expression in TRAMP mice during PCa progression. Downregulation of NPRA expression by siNPRA significantly reduced MIF expression. Moreover, treatment of TRAMP-C1 xenografts with NPRA inhibitor also reduced MIF expression.

**Conclusion:** Our results suggest that NPRA is an upstream regulator of MIF signaling during PCa progression, and NPRA promotes PCa development by modulating MIF pathway.

**Research supported by:** NIH and Bankhead Coley Cancer Research Program

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**Folate Protection of Fetal Alcohol Syndrome Related Cardiac Defects by Modulation of Wnt/β-Catenin Signaling**  
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**Keywords:** Folate, Wnt-β-catenin signaling, Fetal Alcohol Syndrome, gene expression, myo-inositol.

**Objective:** Alcohol (ethanol) consumption during pregnancy is linked to congenital heart defects associated with Fetal Alcohol Syndrome (FAS). Because reports have linked ethanol (EtOH) exposure with Wnt/β-catenin signaling, we defined whether a similar relationship exists between EtOH and Wnt-β-catenin signaling.

**Methods:** Exposure of stage 4 chick and quail embryos to 30% EtOH suppressed Wnt/β-catenin modulated gene expression of Hex (a marker of the primary heart field) and of Islet-1 (a marker for the second heart field) within the cardiogenic crescent. Exposure of pregnant mice similarly during gastrulation to an accepted binge-drinking dose of EtOH on ED 6.75 induced atrioventricular and semilunar valve defects, as determined noninvasively by echocardiography on ED 15.5.

**Results:** We had shown previously that folic acid (FA) supplementation acts by overriding Wnt/β-catenin inhibition of the induction of cardiac gene expression in the heart fields. Thus, FA, known to protect against neural tube defects, was tested for protective effects against ethanol potentiation of Wnt/β-catenin signaling during cardiac specification. Culture medium supplementation with FA, with and without myo-inositol, resulted in normal expression of the cardiac markers upon 30% EtOH exposure in the avian model. In the mouse, FA supplementation (10.5 mg/kg) of diet on morning of vaginal plug date or FA in combination with myo-inositol, resulted in normal valve development (100%) after EtOH exposure on ED 6.75, as assessed by echocardiography on ED 15.5.

**Conclusion:** In conclusion, FA supplementation at a high dose, or in combination with myo-inositol, prevents alcohol potentiation of Wnt/β-catenin signaling allowing normal gene activation and cardiogenesis.
Melphalan Induced Atrial Arrhythmias: Incidence and Risk Factors

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Keywords: Atrial arrhythmia; Atrial fibrillation; Supraventricular tachycardia; Chemotherapy; HSCT

Objective: Cardiology consults in cancer centers are often generated by atrial arrhythmias (AA). It is not well established which chemotherapy is associated with the highest AA rate.

Methods: We retrospectively reviewed the data on patients who received hematopoietic stem cell transplant (HSCT) in 1998-2005 and compared the proportion of patients who developed AA based on the drug regimen. Fisher’s chi square test and Student’s t-test were used for comparison of categorical and continuous variables, respectively.

Results: In 1998-2005 there were 1221 HSCTs, 62 (5.1%) of which were complicated by AA. Only melphalan demonstrated significantly higher rate of AA than any other chemotherapy. Out of 438 patients who received melphalan, 48 (11%) developed atrial fibrillation (AF) (35) or supraventricular tachycardia (13) during same hospital admission, and 390 did not. Patients with AA were older (62.8 ±7.9 versus 55.5 ±10.2, p<0.001). Concomitant hypertension was associated with higher rate of AA. Length of stay was greater (24.9 ±8.9 days versus 19.6 ±5.8 days, p<0.0001) in those who developed AA, even after adjustment for co-morbidities.

Conclusion: Atrial arrhythmias, mostly AF, complicate about 5% of chemotherapeutic treatments used with HSCT. Melphalan is most arrhythmogenic and is associated with atrial arrhythmias in 11% of patients. Atrial arrhythmias are more common in patients with increased age and concomitant hypertension and result in about a four day increase in length of hospital stay.

Insulin Coordinately Regulates Nitric Oxide Synthase and Argininosuccinate Synthase to Maintain Vascular Endothelial Function

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Keywords: argininosuccinate synthase, nitric oxide synthase, citrulline nitric oxide cycle, diabetes, insulin

Objective: The pathogenesis of diabetes includes impairment of vascular endothelial cell nitric oxide (NO) production. In this report, we demonstrate that insulin not only maintains endothelial NO production through regulation of endothelial nitric oxide synthase (eNOS), but also via the regulation of argininosuccinate synthase (AS), which is the rate-limiting step of the citrulline nitric oxide cycle.

Methods: The methods used in this research include Western blotting for protein determinations, QPCR for mRNA quantitation, and the DAN assay for measuring nitric oxide levels. Aortic vessel reactivity was measured in the presence of increasing concentrations of acetylcholine.

Results: First, using serum starved, cultured vascular endothelial cells, we show that insulin coordinately up-regulates both eNOS and AS expression, supporting NO production. Second, using a rat model, we show that diminished eNOS and AS expression in rats with streptozotocin-induced diabetes is reversed by insulin treatment, delivered via slow release pellets. Impaired endothelium-dependent vasorelaxation is also reversed with insulin treatment.

Conclusion: Thus, insulin supports endothelial function not only through regulation of eNOS, but also through the coordinate regulation of AS.

Research supported by: Supported by the James and Esther King Biomedical Research Program, DOH Florida; NIH RO1 HL083153-01A2
Endogenous and Diet-induced Hypercholesterolemia in Nonhuman Primates

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Keywords: Rhesus, cholesterol, diabetes

Objective: Nonhuman primates (NHPs) share with humans many features of lipid metabolism. We have previously reported that rhesus monkeys develop features of diabetic dyslipidemia, including hypertriglyceridemia and low HDL cholesterol. a) To define and characterize spontaneous hypercholesterolemia in rhesus monkeys, and b) To examine the detailed lipoprotein profiles in middle-aged and aged rhesus monkeys fed a lifelong low fat/low cholesterol (LFLC) diet and compare these profiles to those induced by a high fat/high cholesterol (HFHC) diet.

Methods: Adult rhesus monkeys (N=217) were studied, including 123 normal healthy animals used to establish the average normal levels of total cholesterol (TC), the LDL and HDL fractions, and triglycerides (TG). In addition, 22 monkeys (out of 217) were divided into two matched groups, and were fed either a LFLC or a HFHC diet for 16 weeks.

Results: The mean TC in healthy non-diabetic monkeys was 146 ± 2.7 mg/dl. Hypercholesterolemia was identified as the 95th percentile of the normal cholesterol distribution: ≥200 mg/dl. The LDL levels were: normal 65 ± 1.9 mg/dl and the 95th percentile: ≥107 mg/dl. The HDL levels were: normal 67.6 ± 1.6 mg/dl with low HDL, 5th percentile: ≤38 mg/dl. Neither age nor adiposity contributed significantly to hypercholesterolemia. In addition, severe hypercholesterolemia developed in the HFHC fed group, however, no weight gain occurred with this high fat diet.

Conclusion: Despite ingesting LFLC diet, approximately 10 - 20% of middle-aged NHPs develop hypercholesterolemia, reflecting lipoprotein patterns very similar to humans, and this lipid profile of spontaneous hypercholesterolemia differs significantly from the hypercholesterolemia induced by a HFHC diet.

Research supported by: NIH NO1AG31012 to BCH

Molecular Basis of X-linked Dominant Protoporphyria Due to Mutations in 5-aminolevulinate Synthase

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Keywords: Aminolevulinate Synthase, X-linked Dominant Protoporphyria.

Objective: 5-Aminolevulinate synthase (ALAS) catalyzes the decarboxylative condensation of glycine and succinyl-coenzyme A (sCoA) yielding 5-aminolevulinic acid (ALA). Recently two defects in the C-terminal coding region were found to cause a previously unrecognized disease, X-linked dominant protoporphyria (XLDPP), which is characterized by porphyrin accumulation and photosensitivity. It is hypothesized that these deletions result in an intrinsic increase in activity or stabilize the enzyme against degradation. This investigation will use molecular biological and biochemical studies to investigate the role of these deletions in substrate binding and catalysis of ALAS.

Methods: The delAT and delAGTG mutations were introduced into human erythroid specific ALAS using site-directed mutagenesis.

Results: Steady-state kinetic parameters of wild-type human ALAS and the variant enzymes were obtained, in summary the catalytic activities of the mutant enzymes were more than three-fold greater in comparison to wild-type ALAS. In addition the mutant enzymes had a ten-fold greater catalytic efficiency for succinyl-CoA in comparison to wild-type ALAS. The absorption spectra of wild-type human ALAS and the variants had maxima at 420 and 330 nm, with a greater A420/A330 ratio for the mutant enzymes. Transient kinetic analysis of the formation and decay of the quinonoid intermediate from glycine and succinyl-CoA indicated that the rate of quinonoid intermediate decay of the delAGTG mutant-catalyzed reaction was ten-fold greater than that of the wild-type.

Conclusion: The enhanced kinetic properties of these mutant enzymes could be responsible for the XLDPP phenotype. Further studies will involve pre-steady state kinetics, circular dichroism and structural characterization.
**Abstract #**: A-25  
**Presented by**: Mallory Gillam, MS, Staff  

**Multiple Inhibitory Binding Sites in Ferrochelatase May Regulate Activity by Modulating Enzyme Dynamics**  
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**Keywords**: ferrochelatase erythropoietic protoporphyria heme enzyme dynamics pi-helix

**Objective**: Ferrochelatase unites the porphyrin and iron-transport pathways, and catalyzes insertion of ferrous iron into protoporphyrin IX to produce heme at the inner mitochondrial matrix membrane in eucaryotes.

**Methods**: A conserved helix extending away from the active site contains a short stretch of amino acids forming the unusual pi-helix structure, which is characterized by the presence of 4.5 residues per helical turn rather than the 3.7 observed in alpha-helices. Crystal structure studies indicate that the pi-helix is quasi-stable, and is unwound in the product bound structures.

**Results**: When either a conserved histidine or glutamate residue in the pi-helix is mutated the severe metal ion inhibition observed with the wild-type enzyme is eliminated, albeit with substantial loss of activity.

**Conclusion**: We propose that these residues may be crucial to the quasi-stable nature of the pi-helix, and comprise a inhibitory metal ion binding site that stabilizes the pi-helix and prevents the unwinding which is presumably required for product release.

**Research supported by**: National Institutes of Health

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**Abstract #**: A-26  
**Presented by**: David Mitchell, PhD, Faculty  

**A Family of Bioactive Lipids Inhibits the Saccharomyces cerevisiae Erf2/4 Palmitoyl Acyl Transferase (PAT)**  
Linda Chan2, Jaron Swift1, Yiping Ling1, Gayatri Mitchell1, Fuyu Tamanoi2, Robert J. Deschenes1, and David A. Mitchell1,3  
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**Keywords**: Palmitoylation, Fluorescence, High Throughput Screen, Enzyme Assay, NAD

**Objective**: Protein palmitoylation refers to the posttranslational addition of a 16-carbon fatty acid to the side chain of cysteine forming a thioester linkage. The central role of palmitoylation in the subcellular localization and function of numerous signaling proteins involved in cancer has made it necessary to devise assays to study palmitoylation. To date, no specific palmitoylation inhibitors exist nor have these enzymes been systematically studied as a drug targets. The goal of this research is to establish an in vitro fluorescence-based high throughput screening (HTS) protocol which can be used to identify inhibitors of protein palmitoylation.

**Methods**: Each 50 μl reaction contains dH2O, 12.5μl 4X PBS (containing 8 mM α-ketoglutarate, 1mM NAD, 0.80 mM Thiamine Pyrophosphate, and 1 mM DTT), 2.5μl KDH, 6.3μl 200 μM Palmitoyl-CoA in dH2O, and 2μg of the palmitoyl acyltransferase (PAT), Erf2/Erf4. Denatured Erf2/4 is used as a negative control (and all reactions contain 1% DMSO). The reaction was allowed to proceed for 30 minutes at 30°C. The reaction was excited at 340 nm and the fluorescence monitored at 464 nm (emission) on a Flex Station II (Molecular Devices).

**Results**: After screening 7224 compounds, a family of lipid compounds similar in structure to arachidonic acid showed inhibitory effects in the fluorescence-based assay and an independent radioactivity-based assay. The effect of these inhibitors could be “washed-out”, demonstrating that the inhibitors were not disrupted the enzyme structure.

**Conclusion**: Unsaturated lipids appear to affect the activity of Erf2/Erf4. These results suggest that there may be a regulatory role for these lipids in palmitoyl acyl transferase reactions in vivo.

**Research supported by**: NIH
Large-Scale Purification and Enzymatic Characterization of a Yeast Membrane Protein Acyl Transferase Complex, Erf2/Erf4
Gayatri Mitchell, Yiping Ling, Laura Motran, David Mitchell and Robert J. Deschenes
Department of Molecular Medicine, College of Medicine, University of South Tampa, FL

Keywords: membrane protein, protein acyl transferase, DHHC domain

Objective: DHHC protein acyl transferases are a new class of membrane proteins that transfer palmitate (C16:0 lipids) to proteins including Ras, G-proteins, ion channels and other signaling molecules. The significance of palmitoylation is multifold from controlling trafficking of molecules to their stability and ability to transduce signals in the cell. Here we have developed a large scale protein purification system and novel assays to characterize the enzyme activity.

Methods: Using galactose-inducible vectors, we subcloned 6xHIS:Erf2/Erf4 (yeast PAT for Ras2) into the multiple cloning site. Cells were induced for protein expression and disrupted to isolate the protein extract. With 1% dodecylmaltoside the membrane protein was solubilized and using metal chelation affinity chromatography, the protein was purified. Purified protein was used in a coupled assay to measure the release of reduced CoA from Palmitoyl CoA.

Results: 6xHIS:Erf2/4 was purified to 80% purity using one step affinity column. The turnover of the enzyme was measured via the reduction of NAD+ to NADH through the release of reduced CoASH. The kinetic parameters were obtained (Vmax of 120 pmol/min/ug of protein and Km of 54+/- uM). This method allowed us to measure the activity of various catalytic mutants and give a deeper insight into the mechanism of palmitoylation.

Conclusion: DHHC protein acyl transferases are a novel class of enzymes and to our knowledge this is the first report of isolating pure protein. Based on our enzyme assays and comparison between the different mutants in the conserved regions of the protein, we conclude that the DHHC residues are necessary for in vivo and in vitro activity. In the light of the current results a model has been discussed.

Research supported by: NIH

Two Thyroid Response Elements are Required for the Rapid Increase in Transcription of Rat Hepatic LDL Receptor Mediated by Thyroid Hormone
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Department of Molecular Medicine, College of Medicine, University of South Florida

Keywords: LDL-R, Thyroid Hormone, Transcriptional Regulation

Objective: Thyroid hormone acts rapidly, within 60 min, to increase transcription of the LDL receptor. Using H4IIE rat hepatoma cells, we have previously identified elements at -156 and -612 that bind TRβ1 and RXRα. When these hepatoma cells are transfected with a LDL receptor construct bearing mutations in both of these elements, the stimulation by thyroid is negated. We wished to determine whether these same elements also mediate the thyroid hormone induction of the LDL receptor in rats.

Methods: To investigate this premise, we utilized the technique of in vivo electroporation to directly introduce LDL receptor constructs into livers of hypophysectomized rats. LDL receptor-luciferase constructs were introduced in duplicate into each of three liver lobes so that the wild type and two different mutant constructs could be directly compared in the same animal.

Results: Serum T3 levels were increased from less than 1 pg/ml in the Hx rats to 49.9 +/- 4.1, 60 min after T3 injection. Luciferase activity of the wild type construct was increased from 0.49 +/- 0.13 to 7.54 +/- 3.06 by the T3 treatment. Mutation of the -156 element had no effect while mutating the -612 element decreased luciferase activity to 3.06 +/- 1.51. Mutating both elements further decreased luciferase activity to 1.6 +/- 0.9.

Conclusion: The data suggest that both of these elements are necessary for the T3 promoted increase in hepatic LDL receptor transcription.

Research supported by: This work was supported by a grant from the National Institutes of Diabetes and Digestive and Kidney Diseases (R01DK075414) and does not necessarily represent the official views of the NIDDK or the National Institutes of Health.
Abstract #: A-29
Presented by: Cesar Borlongan, PhD, Faculty

National Incidence and Prevalence of Adult-Onset Brain Disorders  Cesar V. Borlongan, Seong Jin Yu, Jesus Recio, Jack Burns, Steven Amatangelo, Eunkyung Cate Bae, Hideki Shojo, Paul R. Sanberg, Yuji Kaneko, Dwaine F. Emerich Department of Neurosurgery and Brain Repair, College of Medicine, University of South Florida, Tampa, FL; InCytu Inc, Lincoln, RI

Keywords: Aging, public health, novel therapeutic interventions

Objective: Aging, especially within the brain, has been a major importance to the field of public health. Aging-related disorders will increase dramatically in the US over the next decades because of aging baby boomers. A review of the literature over the last 10 years reveals that the incidence and prevalence of adult-onset brain disorders have been not reported as a group, but rather conveniently mentioned in the introduction section of many research articles dedicated for each specific disorder, thereby limiting vis-a-vis comparisons between diseases. A handy reference guide summarizing current incidence and prevalence of aging-related disorders is lacking.

Methods: The most recent available data on the incidence and prevalence of adult-onset brain disorders in the US were obtained via PUBMED and NIH websites and compiled into tabular forms to generate US nationwide figures. A Google search was also conducted to retrieve similar data from 10 years ago.

Results: Summary tables were created detailing the results of the most current incidence and prevalence of adult-onset brain disorders in the US. These commonly diagnosed brain disorders reveal obvious increments in incidence and prevalence over the last 2 years compared to 10 years ago.

Conclusion: The updated tabular representations of adult-onset brain disorders will be a helpful, quick reference for physicians and scientists engaged in this field. That the incidence and prevalence have demonstrated continued increase over the last decade put into perspective the current state of these debilitating diseases which remain as significantly unmet clinical need despite our efforts for advancing novel therapeutic interventions from the laboratory to the clinic.

Research supported by: Department of Neurosurgery and Brain Repair Funds

Abstract #: A-30
Presented by: Vedad Delic, MS, Staff

Treatment of In Vitro Traumatic Brain Injury by Transcription Factor Modulation  Vedad Delic, Haris Hatic, Carrie L. Butler, John S. Dennis, Jessica N. Chang, Bruce A. Citron Bay Pines VA Healthcare System and Department of Molecular Medicine, College of Medicine, University of South Florida

Keywords: Traumatic Brain Injury, Transcription Factor, Nrf2, neurite, neurons

Objective: Currently, there are no effective treatments for traumatic brain injuries (TBI) and they can be devastating to the individual and their families. Worldwide incidence of TBI is approximately 0.5% per year, and even mild injuries frequently result in long lasting effects. Furthermore, lifelong care required by TBI sufferers is placing a great deal of burden on our healthcare system. The clinical consequences of TBI are due to a combination of diffuse axonal injury and neuronal loss. We are seeking a better understanding of the molecular mechanisms underlying the effects of traumatic brain injury. Transcription factors appear to play a coordinating role in the balance between cell death and survival. For example, Nrf2 acts on the antioxidant response element (ARE) sequence, shared by several protective genes whose products confer antioxidant properties.

Methods: We have developed an in vitro biaxial stretch injury, mimicking the torsional stress of a TBI. With this system we are able to injure a hippocampal neuronal cell line and subsequently measure neurite numbers and length. This system also allows us to rapidly test compounds that could protect neurons.

Results: Pretreatment of a hippocampal cell line with tert-butylhydroquinone (tBHQ) was demonstrated to be neuroprotective against severe traumatic brain injury. tBHQ is an activator of transcription factor Nrf2 which promotes strong antioxidant activity. Neurite retraction occurred to a significantly lower degree, following injury, in the treated cells compared to those receiving only vehicle.

Conclusion: We are able to model injury induced neurite loss in vitro and we have demonstrated neuroprotection with an Nrf2 transcriptional activator.

Research supported by: Department of Veterans Affairs and Bay Pines Foundation.
**Cotinine: a Dual Action Drug with Multiple Benefits against Alzheimer’s Disease**  
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1 Bay Pines VA Healthcare System, Bay Pines, FL, 2 Department of Molecular Medicine, University of South Florida, 3 Department of Chemistry, University of Miami, FL, 4 Department of Biology, Boston College, Chestnut Hill, MA, 5 MRC Functional Genomics Unit, Department of Physiology Anatomy and Genetics, University of Oxford, Oxford, OX13QX, UK, 6 Florida Alzheimer’s Disease Research Center, University of South Florida, Dept. of Cell Biology, Microbiology, and Molecular Biology, University of South Florida, Tampa, FL

**Keywords**: Tobacco, neurodegeneration, beta amyloid

**Objective**: Epidemiological studies show that smoking negatively correlates with the incidence of Alzheimer’s disease (AD). We investigated the effects of cotinine, the main metabolite of nicotine, against AD and its effect on plaque formation, memory and amyloid beta (Ab) aggregation as well as on the activity of the nicotinic acetylcholine receptors (nAChRs).

**Methods**: The effect of cotinine on Ab aggregation in vitro was studied by using Western blot, dot-blot immunoassays and atomic force microscopy. To investigate the effect of cotinine in vivo, we treated transgenic Tg6799 and control mice with cotinine (2.5mg/kg) and tested them using radial arm water maze, circular platform, and interference tests. The analysis of Ab levels and deposition was performed using immunohistochemical and ELISA techniques. The effect of cotinine over nAChR activity was evaluated using voltage clamp and heterologously expressed nAChRs. Molecular dynamics (MD) simulation of the cotinine-Ab interaction was performed.

**Results**: Cotinine stabilizes the monomeric form of the peptide and decreases Ab aggregation. MD simulation of the cotinine/Ab interaction showed that cotinine interacts with key residues that affect Ab aggregation. Behavioral analyses showed that cotinine improves memory and decreases plaque formation. Voltage clamp analysis showed that cotinine is a modulator of the alpha7nAChR.

**Conclusion**: Cotinine reduces Ab aggregation, improves memory and reduces amyloid burden in the Tg6799 mice. Cotinine is a positive modulator of the alpha7nAChR.

**Research supported by**: The James and Esther King New Investigator grant (to VE), the Johnnie B. Byrd, Sr. Alzheimer’s Center and Research Institute pilot grant (VE and GA), and Alzheimer’s Association, (VE). Bay Pines VA Healthcare System, Bay Pines Foundation.

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**The Duality of Heat Shock Proteins in Tau Pathogenesis**  
Jeffrey Jones, Umesh Jinwal, John Koren III, Qingyou Li, Laura Blair, Ying Jin, Jose Abisambra, Yoshinari Miyata, Lyra Chang, Labeena Wajahat, Grant Mauk, Jason E. Gestwicki, Chad Dickey  
Dept. of Molecular Medicine, College of Medicine, University of South Florida

**Keywords**: Alzheimer's Disease, Tau, Heat Shock Proteins.

**Objective**: Tau is a microtubule-associate protein implicated in Alzheimer’s disease and many other neurodegenerative diseases. Heat shock proteins are chaperones that help to either refold or target certain aberrant proteins for degradation. The relationship of two different heat shock proteins to tau was investigated for this project.

**Methods**: The primary tool utilized for this project was recombinant proteins. Western blotting, dot blotting, ELISA, and sucrose cushions were utilized for the in vitro data. Cell culture and western blotting were utilized for in vivo data.

**Results**: In vitro, Hsp27 was able to maintain the solubility of the tau protein and prevent aggregation. Conversely, Hsp70 provided a conducive environment for the formation of a stable tau conformation known as MC1.

**Conclusion**: The research presented for this poster, demonstrates the ability of heat shock proteins to play a key role in the prevention or development of various tau pathologies; thus presenting a duality in the role of heat shock proteins relative to tau.

**Research supported by**: NIH/NIA, AFAR, Alzheimer’s Association, CurePSP, Dept. of Molecular Medicine/USF
**Abstract #: A-33**

**Presented by:** Yuji Kaneko, PhD, Faculty

**DJ-1, A Key Neuroprotective Protein Against Parkinson’s Disease, Ameliorates Neuronal Cell Death in Ischemic Stroke Possibly Via Mitochondrial Pathway**

Yuji Kaneko, Hideki Shojo, SeongJin Yu, Eunkyung Cate Bae, Cesar V. Borlongan. Department of Neurosurgery and Brain Repair, College of Medicine, University of South Florida, Tampa, FL

**Keywords:** Stroke, Neurotrophic Factors, Brain Repair

**Objective:** DJ-1 is an important redox-reactive neuroprotective protein implicated in regulation of oxidative stress after ischemia. However, the molecular mechanism, especially the mitochondrial function, by which DJ-1 protects neuronal cells in stroke remains to be elucidated. The aim of this study was to reveal whether DJ-1 translocates into the mitochondria in exerting neuroprotection against oxidative stress. In particular, we examined DJ-1 secretion from primary rat neurons exposed to experimental stroke.

**Methods:** Primary rat neuron/astrocyte cells were exposed to the oxygen glucose deprivation (OGD), an established ischemic stroke model. After this period, DJ-1 translocation was measured by immunocytochemistry and its secretion from the primary neuronal cells detected by ELISA.

**Results:** Under hypoxic-ischemic condition, DJ-1 translocated into the mitochondria. Moreover, significant levels of DJ-1 protein were secreted by hypoxia-ischemic primary rat neurons. Altogether, these results revealed that DJ-1 principally participates in the early phase of ischemic stroke involving the mitochondrial pathway.

**Conclusion:** These results revealed that DJ-1 is intimately associated with early phases of disease progression in stroke. That DJ-1 is detected immediately after stroke and efficiently translocated into the mitochondria offer a new venue for developing neuroprotective and/or neurorestorative strategies against ischemic stroke.

**Research supported by:** Department of Neurosurgery and Brain Repair Funds

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**Abstract #: A-34**

**Presented by:** Luis Munoz, MD, Staff

**Intravenous Ibuprofen Treatment for Patent Ductus Arteriosus in Preterm Infants Does Not Affect Cerebral Blood Flow Velocity**

Luis Munoz, MD Jane Carver, PhD, Dawn Bruton, RN Stacey Stone, MD, Roberto Sosa, MD and Rajan Wadhawan, MD All Children’s Hospital & University of South Florida

**Keywords:** Ibuprofen and CBF

**Objective:** Patent ductus arteriosus (PDA) is a common clinical problem in very low birth weight (VLBW) infants. PDA is often treated with medications such as indomethacin or ibuprofen. The effects of indomethacin on systemic circulatory beds have been widely studied, while effects of ibuprofen are largely unknown in this regard. Clinical trials have shown that indomethacin administered within 24 hours of birth is effective in intra-ventricular hemorrhage (IVH) prophylaxis in preterm infants. Ibuprofen, however, does not have this beneficial effect. We propose that the reason ibuprofen may not provide IVH prophylaxis is lack of an effect of the drug on the cerebral vasculature in preterm infants.

**Methods:** Ongoing prospective study of VLBW infants (<1500g bw) at a level III NICU at All Children’s Hospital. Infants are enrolled when they are diagnosed by an echocardiogram as having a PDA needing medical therapy. Doppler ultrasound is used to measure middle cerebral artery blood flow velocity (MCABFV) and ductal size. The measurements are made before and 30 minutes after the administration of the first dose of ibuprofen, and are repeated at the same time intervals with the third dose of ibuprofen.

**Results:** Preliminary data are available for 15 patients. Mean gestation and birth weight are 25.5 ± 1.5 wks and 852 ± 258 g, respectively. Interim data analyses indicate that ibuprofen has no effect on MCABFV. For dose 1, the mean pre and post-dose MCA peak systolic velocity was 38.7 ±11 and 40.6 ±10 cm/sec, respectively (p=0.40). For dose 3, the mean pre and post-dose MCA peak systolic velocity was 39.5 ±14 and 40.1 ±13 cm/s, respectively (p=0.81).

**Conclusion:** Preliminary results support the hypothesis that there is no reduction of MCABFV after ibuprofen administration.
**Abstract #**: A-35

**Presented by**: Kevin Nash, PhD, Faculty

**Title**: rAAV transduction of Murine Brain Neuronal Precursor Cells

**Authors**: K. Nash¹, M. Berg², P. Reid¹, M. C. Cardenas-Aguayo², N. Marks², M. Gordon¹, D. Morgan¹

**Institution**: Byrd Alzheimer Institute, Mol. Phar. And Phys., University of South Florida, Tampa FL; Centers for Dementia and Neurochemistry Nathan Kline Institute, New York University, Orangeburg, NY

**Keywords**: adeno-associate virus neuronal precursor

**Objective**: Objectives: Recombinant Adeno-associated viruses (rAAV) provide tools to manipulate gene expression of proteins relevant to neurodegeneration. Stem cells provide a potential method for cell replacement therapy. Here we examine rAAV’s ability to transduce neural precursor cells (NPC) from murine E 13 embryonic brain areas destined to differentiate into areas vulnerable to neurodegeneration. Transduction with rAAV enables modification of gene expression in NPC cells for potential therapeutic applications.

**Methods**: Methods: We evaluated transduction efficiency for rAAV serotypes 1, 2, 5, 8, 9 encoding green fluorescent protein (GFP) with a CMV/chicken beta-actin promoter, in cultured NPCs derived from anterior brain destined to become primary neurons or glia. Transduction efficiency was assessed by fluorescence/phase-contrast, cell differentiation by immunocytochemistry, and toxicity by colony counts on long-term culture. NPCs were cultured 0-30 days in serum free media plus EGF to limit terminal differentiation in presence of 103-105 viral genomes (VG) per cell.

**Results**: Results: Transgene expression was observed 72 h post infection with rank order of efficiency of 1>2>9>8>5. This compares with minimal detection after 10 d with intracerebral injections. Stable expression of the transgene was detected for up to 30 d and significant toxicity was only seen at higher viral titers.

**Conclusion**: Conclusions: Data provides an in vitro model to rapidly determine infectivity of viral preparations for which none currently exist for most serotypes. We demonstrate that rAAV are excellent vehicles for modification of gene expression in NPC for potential therapeutic applications.

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**Abstract #**: A-36

**Presented by**: Yasser Saloum, MD, Faculty

**Title**: Catheter-free Esophageal pH Monitoring: Are We Getting Useful Clinical Information?

**Authors**: Jeffrey Gill, MD, Yasser Soloum, MD

**Institution**: Digestive Diseases and Nutrition, University of South Florida, Tampa, FL.

**Keywords**: Catheter-free esophageal pH monitoring

**Objective**: The aim of our study is to compare the upright versus supine pH values using catheter-free esophageal pH monitoring. Secondary objectives are to compare DeMeester scores on days 1 and 2, patient compliance with reporting symptoms, and correlation of acid reflux, defined as pH<4, during symptoms.

**Methods**: Chart review of 222 catheter-free esophageal pH monitoring studies at Tampa General Hospital between Jan. 2007 and Sept. 2008

**Results**: The prevalence of any acid reflux was greater in the upright (93.1%) vs. supine (62.8%) position (p<.001). The mean % of time pH<4 was greater in the upright (8.23%) vs. supine (6.79%) position (p=.002). The number of longest episodes were also higher in the upright (5.73) vs. supine (2.42) position (p<.001). There was a tendency for day 1 DeMeester scores to be higher than day 2 DeMeester scores (p<.023). Only 36/192 (18.7%) of patients reporting symptoms were experiencing acid reflux (pH<4) greater that 20% of that time, and 8/192 (4.2%) of patients reporting symptoms were experiencing acid reflux (pH<4) greater than 50% of that time. For all patients during symptoms, acid reflux occurred only a mean of 10.6% of the time. The symptoms with the highest correlation of acid reflux was regurgitation and heartburn and the lowest was cough. Regarding compliance, despite all tests being done for symptoms, 30/222 (13.5%) did not indicate any symptoms during testing, 76/222 (34.2%) did not record when they were supine or upright, 52/222 (23.4%) did not record meal times.

**Conclusion**: Our study recognizes many flaws in the use of catheter-free esophageal pH monitoring, some of which can be improved with better patient education. Others may require changes in the analysis of the test, or the test itself.
**Promotion of Brain Self-Repair Mechanisms by Stereotaxic Micro-lesions**

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Department of Neurology, College of Medicine, University of South Florida

**Keywords:** Micro-injury; Neurogenesis; CNS Regeneration; Bone marrow stem cell.

**Objective:** Greater understanding of mechanisms that regulate neurogenesis may ultimately lead to enhanced brain self-repair following trauma, stroke and neurodegenerative diseases. In this report, we studied the effects of stereotaxic micro-lesions on neurogenesis in multiple regions of adult mice brain.

**Methods:** The acupuncture needle was inserted briefly with stereotaxic guidance into specific brain regions of normal mice and in brains of chimeric mice with green fluorescent protein-expressing bone marrow. The primary objective was to determine the extent to which insertion of a micro-needle would increase generation of new neurons and glia in non-neurogenic zones. BrdU was used simultaneously to birth-date newly born stem cells. To determine the extent to which newly born neural cells in various brain regions were derived from bone marrow, we used GFP+ chimeric mice.

**Results:** After transient stereotaxic insertion of the sterile micro-needle, mice were euthanized at 1, 2, and 3 wks and processed for immunohistochemistry. Antibodies directed against neuronal (TuJ1, NeuN), glia marker (GFAP) and BrdU or GFP revealed many double-labeled neural cells in brain, suggesting that neural stem cells or bone marrow stem cells could be induced to differentiate into neurons or neuron-like cells.

**Conclusion:** This method generates a micro-lesion that results in increased migration of bone-marrow derived cells to the site; many of these cells appear to differentiate into neurons in regions where new neurons generally do not form. Understanding the signaling induced by transient micro-injury may shed light on the regulation of neurogenesis but may also elucidate possible neuroprotective mechanisms attributed by some researchers to deep brain stimulation.

**Research supported by:** VA Merit Review grant.

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**Dog Placenta Cells Express Embryonic Stem Cell Markers and Exert Neuroprotection Via Hsp27 in Experimental Stroke Model**

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Dept. of Neurology, College of Medicine, University of South Florida

**Keywords:** Dog, placenta, apoptosis, stem cell, stroke, Hsp27

**Objective:** Stroke is a major cause of death and disability in adults, which primarily cause brain cell death. Stem cell transplantation has been proposed for treating brain diseases, including stroke. The placenta has been recognized as a source of non-embryonic stem cells. Here, we examined dog placenta cells (DPCs) as an equally good source of stem cells, and also explored their potential benefits in an in vitro model of stroke.

**Methods:** Dog placenta cells were grown in well-defined cell culture media. Established phenotype markers of embryonic stem cells (e.g., Oct4, Nanog, SSEA4, CXCR4) and neural lineage (e.g., nestin, MAP2, NeuN, GFAP and O4) were employed to reveal stemness and cell fate over time. In parallel studies, primary rat neurons/astrocytes were exposed to oxygen-glucose deprivation (OGD), an established in vitro stroke model, and subsequently co-cultured with DPCs. Cell viability was measured by immunocytochemistry and trypan blue to determine neuroprotective effects of DPCs. To reveal the mechanism of action underlying therapeutic benefits of DPCs, we examined the expression of heat shock protein (Hsp) 27, a recently discovered member of the heat shock protein family with neuroprotective effects against stroke.

**Results:** At early and prolonged (>8) passages, DPCs expressed stable embryonic stem cell markers, and when grown in defined media, started to exhibit neural phenotypes. Under the OGD condition, DPCs dose dependently reduced cell death in primary rat cells. Moreover, Hsp27 was strongly expressed under the OGD condition.

**Conclusion:** These results indicate that DPCs possess stem cell properties, differentiate towards the neural lineage, and promote neuroprotection against stroke which is likely mitigated by Hsp27 induction.
Racial/Ethnic Disparities in the Identification of Developmental Disabilities and Insurance Status
Heather Agazzi, PhD. Pediatrics, COM, USF, Mulubrhan Mogos, MSC, Nurse Research Resident, CON, USF; Jillian Williams, PhD, Clinical Instructor, Child Development, Pediatrics, COM, USF

Keywords: health disparities, developmental disabilities, Autism, Latinos/Hispanics (L/H)

Objective: We sought to examine racial and ethnic disparities in the recognition of developmental disabilities or delays, including Autism Spectrum Disorders (ASD) and health insurance status.

Methods: Data were obtained from a population (N = 668) of clinic-referred caregivers of children (0-6 years) with challenging behavior who participated in a group delivered parent training program. By utilizing crude and adjusted odds ratios, we estimated the association between race/ethnicity and caregiver-reported diagnosis status and insurance status of the child, adjusting for caregiver age, educational level, and insurance status (only for the diagnosis analyses). It was hypothesized that the exposure L/H would be a risk factor for not having a diagnosis and for not having health insurance.

Results: Sixty percent of children had a diagnosis, 14.5% had an ASD; and 93% had health insurance (private or Medicaid). In crude analyses, children who were L/H (OR=1.10; 95% confidence interval [CI] = 0.80, 1.52), were more likely than non-L/H children to have a diagnosis. For insurance status, L/H children (OR=0.80; CI = 0.55, 1.14) were less likely than non-L/H children to have health insurance. Similar non-significant results were obtained for adjusted analyses.

Conclusion: Early intervention is often contingent upon a documented delay or diagnosis and is critical to improved long-term outcomes for children with developmental disabilities and ASD and the literature is mixed as to whether there are disparities in diagnosis by race/ethnicity. This information is useful for continuing education for clinicians and preservice training of medical residents, and psychology and nursing students.

Research supported by: The Children’s Board of Hillsborough County

Charles SC, Lakis MG, Schocken DM, Monroe ADH. Office of Educational Affairs, College of Medicine, University of South Florida

Keywords: Standardized Patients, medical education, simulation

Objective: Standardized patients (SPs) are used throughout the healthcare curriculum to provide students an opportunity to practice clinical skills and be assessed in a clinical environment. A trained SP provides feedback to the student to enhance their understanding of patient management. The inter-observer reliability and validity of SP responses is key to the validation of an assessment activity. This research was designed to measure the inter-observer reliability and validity of the SP responses in order to develop a certificate program for SP training.

Methods: Faculty developed core competencies to measure students' skills during a clinical assessment to include physical exam skills, history taking and communication skills. Each SP completed an evaluation checklist to measure these observed skills. All 120 MS IIIIs were evaluated following each clinical clerkship. Observation and data measurements were collected following these events to measure the inter-observer reliability and validity of each encounter.

Results: The inter-observer reliability was not statistically significant either between cases or among SPs. The validity scores for these encounters demonstrated significant validity in measurements of physical exam and history taking skills, but not for communication skills.

Conclusion: Based upon these results, several recommendations were made. The student encounter needed to be longer to significantly improve the validity of the evaluation of communication skills. The SPs needed additional training to address inter-observer reliability among SPs and between cases. The Certificate Program for Standardized Patients was developed for the specific purpose of improving the clinical assessments of all our students.

Research supported by: Office of Educational Affairs
Abstract #: A-41

Presented by: Micki Cuppett, PhD, Faculty

Linebacker Stan: Use of High Fidelity Simulator to Teach Emergency Care of Spine Injured Athlete

Micki Cuppett EdD ATC Orthopaedics & Sports Medicine, Fred Slone MD Center for Advanced Clinical Learning, Dawn Schocken MPH Center for Advanced Clinical Learning Barbara Morris DHSc ATC Ortho & Sports Med.,College of Medicine, University of South Florida

Keywords: simulation, simulators, spine injury,

Objective: To provide a realistic and safe environment for the teaching and evaluation of on-field care of a spine injured football player in respiratory or cardiac arrest. To measure students confidence levels in caring for a spine injured athlete.

Methods: 5 groups of 2nd year Athletic Training students (n= 28) participated in the simulation utilizing a tetherless high-fidelity simulator (iStan, METI Corp) dressed in football equipment lying prone on the “field” in respiratory or cardiac arrest. The team was required to assess the downed “athlete” and provide life-saving measures while maintaining spinal stabilization. The groups were debriefed while watching a video recording of their performance. A self-confidence survey was completed by each student at three different points in the simulation: pre, post, and post-debrief.

Results: There was no significant difference in overall pre-sim confidence scores between teams (Mean 57.75 out of 80 possible points) There was a significant difference between teams in the post-sim confidence levels (mean 61.92 sd=7.789). Successful teams had higher post-sim confidence levels less successful teams. Post-debrief confidence levels between teams was also significantly different (mean 57.39, sd= 9.578) and decreased across all groups from the post-simulation confidence, presumably because students saw mistakes on the video that were previously not realized. Teams that performed inadequately (according to instructor observation) had lower confidence scores post-debrief than teams who performed well.

Conclusion: This simulation provided a realistic safe environment to practice team skills in the care of the spine injured athlete that could not be simulated with low fidelity models or standardized patients.

Research supported by: CACL

Abstract #: A-42

Presented by: Daryl DeNittis, Staff

Investigator-Initiated Studies in the Tampa General Hospital Emergency Department

Daryl DeNittis, Div.of Emergency Medicine, Internal Medicine, COM, USF, Charlotte Derr USF/TGH, Richard Paula USF/TGH, Larry Land USF EM Residency, Megan Lasseter USF EM Residency, Tamas Gaspar USF EM Residency, Aaron Osborne USF EM Residency, Matthew Fucarino USF EM Residency, Lawrence Neuman USF MSIII, Gillian Bayley University of Ottawa MSIV

Keywords: investigator-initiated; ultrasound; wound irrigation; cardiac imaging

Objective: This posterboard aims to showcase the efforts of TGH Emergency Medicine physicians/residents and USF Medical Students in designing and carrying out clinical studies. Our students, residents and attendings are at the front line of emergency care and are constantly looking for ways to improve patient care in the ER.

Methods: Dr. Charlotte Derr is the Ultrasound Fellowship Director. Working with residents Larry Land, Megan Lasseter and Matthew Fucarino, she developed a protocol to demonstrate the utility of ultrasound in placing central venous catheters. Dr. Richard Paula, Research Director, worked with resident Tamas Gaspar to produce a pilot protocol looking at the efficacy of a new device for wound irrigation. Dr. Richard Paula helped third-year medical student Lawrence Neuman to create a chart review to study the prescribing patterns of ER physicians. A study currently being developed involves new cardiac imaging technology for stratifying risk in cardiac patients.

Results: These studies are currently active or pending activation in the TGH ER. We anticipate finalising data collection throughout 2010.

Conclusion: Viewers of this posterboard will gain an in-depth understanding of the opportunities to initiate clinical studies while collaborating with USF/TGH Emergency Medicine Research.

Research supported by: USF/TGH
Benefits and Harms of Phase I Trials in Oncology

Sanja Galeb, Moffitt Phase I Trials Collaborative Group1, 1H. Lee Moffitt Cancer Center and Research Institute and Center for Evidence-Based Medicine and Health Outcomes Research, University of South Florida

Keywords: Phase I Trials, Survival, Toxicity

Objective: The ethical issues of conducting phase I trials revolve around the purported aim of phase I trials to assess safety/toxicity (i.e. harms) of new cancer therapies without including clinical benefit to patients as one of the objectives. The existing, albeit limited, empirical data do suggest that patients enrolled in phase I trials can potentially benefit from novel therapies. The extent to which important outcomes, such as survival, can be affected in phase I trials has not been extensively evaluated.

Methods: We reviewed all consecutive, single-center, nonpediatric, phase I oncology trials conducted at the H. Lee Moffitt Cancer Center and Research Institute between 1997 and 2007. Our primary objective was to assess overall survival in patients participating in phase I trials, while our secondary objective was to assess response rates, grade III and IV toxicities, treatment-related mortality, elderly participation, and publication rates.

Results: We extracted data for 39 trials enrolling 1295 patients. Median overall survival (OS) was 1.169 years (range: 0.01-11.82 years). The overall response rate was 24%. The rate of any grade III or IV toxicity was 64%, while the treatment-related mortality rate was 1%. Overall elderly participation rate was 34%; however, only one trial exclusively enrolled elderly patients. Of the completed trials, publication rate was 85%.

Conclusion: Contrary to previous belief, phase I trials do not enroll dying patients with no prospect of benefit. In fact, our results show that a substantial number of patients have a meaningful survival.

Research supported by: H. Lee Moffitt Cancer Center and Research Institute

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Title: Parents' Healthy Weight Perceptions and Preferences For Obesity Counseling in Preschoolers: Pediatricians Matter

Raquel G Hernandez, MD, MPH1,2, Tina L Cheng, MD, MPH1,3 and Janet R Serwint, MD1,3. 1Pediatrics, Johns Hopkins University School of Medicine, Baltimore, MD, United States; 2Pediatrics, University of South Florida College of Medicine, Tampa, FL, United States and 3Johns Hopkins University Bloomberg School of Public Health, Baltimore, MD.

Keywords: Obesity, Preschoolers, Perceptions, Visual Sketches

Objective: To compare parental report of child body image to perceived healthy weight body image in preschoolers and describe weight-counseling preferences.

Methods: Parents of preschoolers receiving well-child care in an urban pediatric clinic were interviewed and asked to select body images that best resembled: 1) their own child’s current weight, 2) a healthy weight preschooler and 3) friend and family report of a healthy weight preschooler. Those indicating that their overweight (age-gender specific BMI ≥ 85th ≤ 94th percentile) or obese (BMI ≥ 95th percentile) child resembled a healthy weight image were considered to misclassify their child’s weight. Logistic regression was used to identify predictors of misclassification. Card-sorting exercises explored preferences for weight counseling.

Results: Of the 150 children in our sample, 32.7% (n=49) were overweight or obese. Misclassification occurred in 71.4% (n=35) of parents in this subgroup with some indicating a desire for a heavier child by sketch report. Absence of pediatrician comment on child weight strongly predicted misclassification (OR: 12.3, 95% CI 1.74-87.2). Pediatricians ranked as the most valued weight advisor.

Conclusion: Pediatricians’ guidance is highly valued and strongly associated with parental accuracy in classifying child weight. Informing providers that their advice matters may promote more effective clinical discussions surrounding early childhood obesity.

Research supported by: The Thomas Wilson Sanitarium for Children of Baltimore City
**Abstract #: A-45**  
**Presented by:** Branko Miladinovic, PhD, Staff  

**Correcting For Bias in Per Protocol Data Using Instrumental Variable Analysis**  
Branko Miladinovic, Benjamin Djulbegovic, Office of Clinical Research, Center for Evidence Based Medicine and Health Outcomes Research, College of Medicine, University of South Florida  

**Keywords:** Instrumental variable analysis, per protocol data, intention to treat, physician preference  

**Objective:** Intention to treat (ITT) is the standard method of data analysis in randomized controlled trials in which departures from randomization exist. It compares outcomes between the groups as randomized, ignoring the actual treatments received, and so averages treatment effects across all randomized patients regardless of whether they received the treatment or not. Per protocol (PP) analysis excludes any data collected from a participant after they have departed from randomized treatment, but suffers from selection bias in that the groups being compared have different characteristics. Methotrexate (MTX) is used as a preventative treatment for graft-versus-host-disease (CGVHD) in transplant patients. In nine separate trials, 573 patients were prescribed four doses of MTX and 534 patients were prescribed three doses of MTX. Using individual patient data, a method based on instrumental variable (IV) analysis will be used to correct for selection bias.  

**Methods:** IV analysis exploits a variable (instrument) that is related to the treatment but not the outcome, except through the treatment itself. Physician prescribing preference (PPP) has been identified as a good IV because if patients choose their doctors without a sense of prescribing preference, then PP introduces a “quasi-randomization” that takes care of the selection bias in PP data. The Generalized Method of Moments, two-stage logistic and probit models are used.  

**Results:**  
Probit: OR = 1.10 (0.95, 1.21)  
Logit-Logit Model OR = 1.68 (1.41, 2.02)  
Linear-Logit Model: OR = 1.22 (0.89, 1.67)  
GMM: 1.20 (0.62, 2.34)  

**Conclusion:** Using per protocol data and PPP as the instrument, the odds in favor of four doses of MTX used as prophylaxis for CGVHD are significantly increased.

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**Abstract #: A-46**  
**Presented by:** Jessica Morgan, BA, Staff  

**A Comprehensive Analysis of Skin-picking in Prader-Willi Syndrome**  

**Keywords:** skin-picking, Prader-Willi Syndrome, children  

**Objective:** The purpose of this study was to gain a comprehensive understanding of skin picking in youth with PWS. Prevalence, frequency, and the nature of skin picking in youth with PWS are addressed (e.g., areas from which children are picking, hours spent picking, antecedents, etc), as well as behavioral correlates of skin-picking.  

**Methods:** Parents of 67 youth (aged 5-19 years) with PWS were recruited to complete an internet-based survey that included measures of: skin-picking behaviors; the automatic and/or focused nature of skin-picking; severity of skin-picking symptoms; anxiety symptomology; developmental functioning; symptoms of inattention, impulsivity, and oppositionality; and quality of life.  

**Results:** Results indicated that skin-picking was endorsed in 95.5% of youth with PWS. Direct associations of moderate strength were found between skin-picking severity and symptoms of anxiety; impulsivity, inattention, and oppositionality; developmental functioning; and quality of life. Other descriptive data, such as areas picked, cutaneous factors, antecedents, and consequences related to skin-picking are reported.  

**Conclusion:** The prevalence and consequences associated with skin-picking in PWS indicate a greater need for clinician awareness of the behavior and interventions tailored to meet the needs of this population.
Citizen Perceptions of Less Surgery and Notes: The Impact of Age, Gender, and BMI

Keywords: LESS NOTES Age BMI Perception

Objective: Laparo-Endoscopic Single Site (LESS) surgery and NOTES have received significant recognition in the lay press. This study was undertaken to determine public attitudes toward LESS surgery and NOTES and to determine how these attitudes are impacted by age, gender, and obesity.

Methods: College educated Americans completed a validated questionnaire with unbiased guidance utilizing a Likert scale (1=disagree to 5=agree). Statistical analyses utilized Spearman regression. Data are presented as median (mean ± SD).

Results: 152 people, 56% female, of median age 29 years and BMI 24 kg/m2 completed the survey. They liked their physique (4, 4 ± 1.0), felt they were attractive (4, 4 ± 1.0), and felt that others found them attractive (4, 4 ± 0.8). LESS surgery was appealing only if it involved no more risks, minimally more pain, minimally longer operative time, no prolonged recovery, and minimally more cost. Older persons were more interested in reduced risk, pain, and recovery time and less interested in scarring/appearance. Heavier persons were more interested in reduced pain, operative time, and pain medication use and were less interested in scarring/appearance. With regard to NOTES, only 32% were willing to consider it and only with no more risks, minimally more pain, minimally longer operative time, and minimally more cost. Lack of visible scarring with NOTES was considered most important by only 30%.

Conclusion: With increased public awareness, acceptance of LESS surgery and NOTES depends upon no additional risk, pain, recovery time, and cost. Improved cosmesis is not generally a priority, particularly in older or heavier people. Safety, pain, and recovery time remain major issues in deciding operative choices by most Americans.

Identifying Local Faculty Development Needs at a Regional Medical School Campus

Keywords: community faculty development needs assessment

Objective: As medical schools move into the community-based practice setting for preparing students in clinical rotations, meeting the training needs of community-based teaching faculty presents curricular challenges. This study identified faculty development needs at a community-based, regional medical school.

Methods: In 2006, formative assessment methods, including stakeholder meetings and focus groups guided development of a main survey. Primary care and specialty physicians, along with third- and fourth-year medical students (N=302) responded to this anonymous survey in which they rated teaching characteristics, teaching tools, and faculty development services. Data analysis included descriptive statistics and independent means t-tests.

Results: Feedback from focus group interviews identified themes including the importance of enthusiasm and feedback. The main survey questionnaire supported focus group findings identifying enthusiasm, humanism, professionalism, and clinical competence as important teaching characteristics, and evaluation, clear expectations, and feedback as key teaching tools. Highly rated faculty development services included medical library resources. The process provided opportunity for ownership of the study purpose thereby building support for decisions impacting the health care of the community.

Conclusion: A systematic identification of local faculty development needs of community teaching physicians at regional medical school campuses is important for medical education curricular design. The study’s methods and results strengthened collaborations and fostered community-building strategies, contributing an important educational framework for future community-based teaching models.

Research supported by: Sarasota Memorial Healthcare Foundation Sarasota Florida
Abstract #: A-49
Presented by: Bradley Peckler, MD, Faculty

**Social-Behavioural Studies with Students and Residents in Emergency Medicine**


**Keywords:** Emergency Medicine resident/s; medical student/s; hi-fidelity simulation; triage communication

**Objective:** This posterboard aims to showcase the past and ongoing social-behavioural studies initiated by USF/TGH Emergency Medicine physicians.

**Methods:** Dr. Bradley Peckler has produced several papers on various aspects of resident and medical student education, including the use of hi-fidelity simulation in clinical exams and skills retention in Advanced Cardiac Life Support training. Jason Wilson, PGYII Emergency Medicine resident, is currently collaborating with Dr. Eric Eisenberg of the College of Arts and Sciences in a project looking at power roles, sensing-making and storytelling during the triage process in the Emergency Room at Tampa General Hospital.

**Results:** We will provide summaries of the results of completed studies.

**Conclusion:** Viewers will gain an in-depth understanding of the social-behavioural studies at USF/TGH Emergency Medicine Research.

**Research supported by:** University of South Florida College of Medicine & Tampa General Hospital

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Abstract #: A-50
Presented by: Jeannette Reid, MS, Staff

**Clinical Correlates and Treatment Response of the Yale-Brown Obsessive Compulsive Scale Auxiliary Items**

Jeannette M. Reid (Department of Pediatrics, USF), Eric A. Storch (Departments of Pediatrics and Psychiatry, USF), & Tanya K. Murphy (Departments of Pediatrics and Psychiatry, USF)

**Keywords:** obsessive compulsive disorder; Yale-Brown Obsessive Compulsive Scale; clinical correlates; treatment response; auxiliary features

**Objective:** The current study examined clinical correlates and treatment response as they relate to auxiliary clinical characteristics (i.e., insight; avoidance; indecisiveness; sense of responsibility; pervasive slowness; pathological doubt; duration of obsession-free and compulsion-free intervals) in 172 adults with OCD.

**Methods:** A trained evaluator administered the Yale-Brown Obsessive Compulsive Scale and the Anxiety Disorders Interview Schedule for DSM-IV. As well, patients completed the Obsessive Compulsive Inventory-Revised; Beck Depression Inventory-Second Edition; State-Trait Anxiety Inventory; RAND 36-Item Health Survey; and Sheehan Disability Scale.

**Results:** Results indicated numerous associations between auxiliary OCD features and both depressive and anxious symptoms as well as impaired health and functioning. All auxiliary features, excluding insight, were reduced following cognitive-behavioral therapy. In particular, changes in symptom-free intervals; avoidance; and indecisiveness were most reliably associated with reductions in core OCD features. The lack of a relationship with insight did not come as a surprise, as insight is considered a relatively stable construct in those with OCD.

**Conclusion:** The present study marks the first known investigation into the clinical import of Y-BOCS auxiliary items in the presentation and treatment of OCD. The above findings offer practical implications regarding the assessment and treatment of patients with OCD.
Abstract #: A-51

Presented by: Dawn Schocken, MPH, Faculty

The Hybrid Use of High-Fidelity Simulation in Advanced Cardiac Life Support (ACLS) Enhances Retention
Schocken DM, Slone FA, Monroe ADH. Department of Internal Medicine, College of Medicine, USF, Office of Educational Affairs, College of Medicine, USF

Keywords: Medical Education, Simulation, ACLS

Objective: The teaching of Advanced Cardiac Life Support (ACLS) is a core competency for most health care providers. This research was designed to determine if using a high-fidelity simulator with confederates would enhance a medical student's retention of the complex algorithms embedded in ACLS compared to traditional teaching methods.

Methods: Core competencies were developed by the faculty to include CPR, teamwork, and leadership skills. The AHA ACLS course was taught to 120 MS IIIs over a two week time period. Practice simulation sessions and final assessments were done with confederates on high-fidelity simulators. A follow-up survey was conducted at two months to measure ACLS retention. A post-test was sent out at month six. During month ten, all MS IIIs completed a year end clinical exam with a station designed to retest the students on ACLS with simulation and confederates.

Results: Of the initial 120 MS IIIs, 115 passed the criteria to be certified. At month 2, a survey evaluation found 87% of the students were able to recall the ACLS guidelines. 70% of these students were able to recall ACLS at month six with a passable score. During the year end exam, 50% of the students successfully recalled the ACLS guidelines. When compared to the previous year's students, overall retention of ACLS by the medical students taught with the hybrid methodology was 30% higher.

Conclusion: Opportunities to participate in learning ACLS guidelines abound. Students who learn via the hybrid approach demonstrated greater retention than students who learned ACLS in the traditional method. Further studies will include a review of the student's retention during actual clinical practice in the hospital setting.

Research supported by: Office of Educational Affairs.

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Abstract #: A-52

Presented by: Erin Stirling, Staff


Keywords: ProCESS; Beriplex; oseltamivir; BEACON registry; critical care; VIDAS

Objective: This posterboard aims to highlight the large-scale and multi-discipline clinical trials, currently in various stages, being conducted at Tampa General Hospital by USF staff and residents. These are national emergency and acute care studies in areas that will contribute greatly to the body of knowledge for emergency and acute care medicine.

Methods: USF/TGH Emergency Medicine Research is a site for the following national studies: - ProCESS: Protocolized Care for Early Septic Shock is the world's largest study of Early Goal-Directed Therapy. - Beriplex: a Phase IIIb clinical trial to assess the efficacy, safety and tolerance of Beriplex P/N (concentrated prothrombin complex) for rapid reversal of coumarin-induced coagulopathy. - IV Oseltamivir: a Phase II study to evaluate the effectiveness and safety in the treatment of H1N1 and seasonal influenza. -VIDAS: is a study aimed at using Vidas Brahms PCT test to guide the initiation of antibiotic therapy in patients with lower respiratory tract infections. -BEACON: Best Expert Agreement for Care of Occult MI Nationally is a registry with the goal of over 70,000 patients.

Results: As large national studies, the final results and publications will be in the future. However, in all that are actively recruiting patients, TGH-USF Emergency Medicine Research is a top enroller.

Conclusion: Viewer will gain an understanding of the various large-scale and national studies which are being conducted by the Emergency and Acute Care Medicine attendings and residents.

Research supported by: University of South Florida and Tampa General Hospital

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**Abstract #**: A-53

**Presented by**: Maureen Groer, PhD, Faculty

**Suppression of Natural Killer Cell Cytotoxicity in Postpartum Women**  
Groer, M., Van Eeopel, J., Hasty, B., Harrington, M., (College of Nursing), El-Badri, N. (College of Medicine)  
University of South Florida, College of Nursing

**Keywords**: Natural Killer Cells, Cytotoxicity, Postpartum

**Objective**: 1. To determine the trajectory over postpartum months 1-12 of Natural Killer (NK) cell numbers and cytotoxicity in postpartum women

**Methods**: Postpartum women (N=35) were measured 9 times over 12 months of the postpartum. A standard Cr51 release assay from K562 cells cultured with peripheral blood mononuclear cells (PBMC) was performed and lytic units calculated. Initially these assays were done with fresh cells, but extremely low cytotoxicity was observed, so recombinant Interleukin-2 preincubation for 18 hours was done before culturing the PBMCs with labeled K562 cells. Cytotoxicity was compared to a control group of aged matched women. Peripheral blood mononuclear cells were isolated by Ficoll, and stored in RPMI with 10% DMSO ad 10% fetal calf serum. Cells were stained for flow cytometry and NK cells were measured through the use of CD56 and CD16.

**Results**: Data indicate suppression of NK cytotoxicity. Lytic units from fresh and pre-incubated cells were lower than those from reported norms, and from controls. NK cell percentages were lower than controls in postpartum women.

**Conclusion**: These data suggest that the postpartum, like pregnancy, is characterized by decreased NK cytotoxicity activity. This suppressed NK cytotoxic effect may result as a response to interaction with fetal tolerized microchimeric cells accumulated during pregnancy in maternal blood and tissues.

**Research supported by**: NIH (RO1-NR05000)

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**Abstract #**: A-54

**Presented by**: Carla Nye, Faculty

**What do Students Really Want From Their Faculty?**  
Carla Nye, DNP, RN, CNE VANA College of Nursing, University of South Florida,  
Kae Livsey, PhD, MPH, RN, School of Nursing, University of North Carolina-Wilmington

**Keywords**: nursing student perceptions faculty interactions

**Objective**: 1. What faculty teaching strategies contribute to student satisfaction with their nursing educational experience? 2. What faculty behaviors do students perceive contribute to professional role development? 3. How do faculty-student interactions impact student satisfaction with their nursing education program? 4. What teaching strategies do students perceive as contributing to learning in the classroom?

**Methods**: After institutional IRB approval was received, two qualitative focus group sessions were held to gather data from a convenience sample of six junior and six senior nursing students.

**Results**: Students reported that key faculty interaction strategies included exhibiting passion and enthusiasm in the classroom, setting high standards for student performance, encouraging, supporting, and empathizing with students lived experiences; providing timely, respectful and constructive feedback; and role modeling quality nursing care and professional behavior. Effective faculty teaching strategies included integrating classroom and clinical learning, weaving case studies and NCLEX style questions throughout lecture content, and using the Socratic Method to stimulate discussion in the classroom.

**Conclusion**: Students described clear desires for both interactions with and teaching methods from faculty. Faculty-student interaction and teaching strategies that capture and support the needs of the students are more likely to lead to program satisfaction.

**Research supported by**: Richard J. Corbett Charitable Trust
Abstract #: A-55


Global Health Infectious Disease Program, Department of Global Health, College of Public Health, University of South Florida, Tampa, Fl

Keywords: Brugia malayi, phage display, RNA recognition motif

Objective: There is little information available about gene regulation in the human filarial parasite Brugia malayi. Only two Brugia malayi promoters have been mapped in detail so far, BmHSP70 and BmRPS12. The BmRPS12 promoter contains a 44- nucleotide tandem repeat sequence, the deletion of which results in 80% loss in promoter activity in the transient tranfection assays. This essential promoter domain lacks the binding sites for most general transcription factors present in other eukaryotic promoters but contains several GATA transcription factor binding sites encoded within this repeat. In the present study, we employed the T7 phage display technique to identify putative transcription factors that interact with this repeat domain.

Methods: Brugia malayi adult female T7 phage display library was made and screened using RPS12 repeat probe.

Results: Using a B. malayi adult female T7 phage cDNA library, we have identified 5 different candidate proteins that were represented ≥5 times out of total 100 clones sequenced after final round of biopanning. Two of these proteins contain the RNA recognition motif (RRM) and constituted the most abundant group when equal number of phages displaying all five proteins was subjected to selection using stringent conditions.

Conclusion: RRM-domain containing proteins have been shown previously to bind to single stranded as well as double stranded DNA, and have been found in several transcription factors. The RRM containing proteins that we have identified could thus constitute a new class of transcription factors in B. malayi.

Research supported by: NIAID grant # R01AI048562

Abstract #: A-56

Mom's Mouth Matters: Implications of Oral Health Misconceptions and Care Barriers for Women’s Health

Linda A. Detman, Ph.D., Lawton & Rhea Chiles Center for Healthy Mothers and Babies, University of South Florida; Barbara H. Cottrell, ARNP, MSN, CNE, Florida State University; Marie Denis-Luque, MSPH, MPH, Lawton & Rhea Chiles Center for Healthy Mothers and Babies, University of South Florida

Keywords: oral health, prenatal care, anticipatory guidance

Objective: Exploring Florida women’s experiences in obtaining dental care prior to and during their pregnancies.

Methods: Data for this study were obtained from 272 recently pregnant African American women, 18-35 years of age who were residents of one of three Florida counties were approached in the hospital after giving birth and invited to take part in an in-depth, face-to-face interviews one month after the birth of the baby. Interview questions on obtaining oral health care before and during pregnancy and recall of guidance on oral health care during prenatal care were transcribed and analyzed using MAXqda2007, a qualitative data management program. Through subject level content analysis, key themes regarding the interviewees’ perspectives on obtaining oral health care before and during pregnancy were assessed. Participants self-selected to take part in the interviews and their views may not be representative.

Results: About half of the participants did not seek oral health care and over half did not recall receiving information during prenatal care about getting dental care. Barriers to obtaining dental care during pregnancy included lack of insurance, difficulty finding a provider, other life priorities, general misconceptions about the safety and appropriateness of dental care during pregnancy, and sporadic anticipatory guidance during prenatal care.

Conclusion: Patients and practitioners need guidance on appropriate oral health care in the perinatal period. Improved insurance coverage and funding for oral health care can enhance access and use. Further work is needed to understand and address misconceptions about dental care during pregnancy.

Research supported by: Florida's Agency for Health Care Administration
Abstract #: A-57

**Contribution of a Winged Phlebotomy Device Design to Blood Splatter**

**Shawn Applegarth, MSME, PhD**

Student; **Donna Haiduven, PhD, RN, CIC**; **Christine McGuire-Wolfe, MPH**; **Meredith Tenouri, MPH, MLIS**, Dept. of Global Health, College of Public Health, University of South Florida

**Keywords:** occupational health evaluation of sharps devices blood exposures

**Objective:** The primary objective of this study was to evaluate the blood splatter potential of six different designs of winged phlebotomy devices.

**Methods:** A laboratory-based experiment assessing blood splatter was conducted using a vascular system with simulated veins containing mock venous blood. A total of 18 devices, representing six device designs (A-F), were tested. Scientific filters were positioned around the devices and weighed with an analytical scale, both before and after the venipuncture. Data were analyzed using descriptive statistics and complementary log-log transformation models.

**Results:** The percentages of devices and gloves with visible blood in addition to filters with measurable blood splatter among the six device designs ranged from 0-20%. Overall, there was a statistically significant association between device design and visible blood on the device (p< 0.0001), and between device design and filters with measurable blood splatter (p<0.0001), but not between device design and visible blood on gloves. A wide range of associations between device design and visible blood on gloves/device, as well as blood splatter incidence, were demonstrated when comparing designs.

**Conclusion:** Data from this experiment illustrate that the design of winged phlebotomy devices can contribute to the likelihood of blood splatter during the venipuncture procedure. Blood splatter has important implications for the occupational safety of healthcare workers. It is recommended that future studies focus on evaluating additional designs of intravascular devices (intravenous catheters, other phlebotomy devices) for blood splatter.

**Research supported by:** Veterans Administration Division of Occupational Safety & Health

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Abstract #: A-58

**Targeted Method for the Detection of Eastern Equine Encephalitis Virus from Mosquitoes in Florida**

**Christy L. Ottendorfer**, **Greg S. White**, **Hassan Hassan**, **Kevin Hill**, **Lillian M. Stark**, **Thomas R. Unnasch**

GHIDR Program, Dept. of Community & Family Health, College of Public Health, University of South Florida

**Keywords:** arbovirus, surveillance, EEE, mosquitoes

**Objective:** Eastern equine encephalitis virus (EEEV) is a serious arboviral infection of humans and horses, with case mortality rates approaching 50%. In Florida, EEEV maintains year-round enzootic transmission foci monitored by a statewide sentinel chicken surveillance program.

**Methods:** During 2008 - 2009, historical sentinel seroconversion rates and mosquito collection data were used to place mosquito traps for year round surveillance. Furthermore, EEEV seroconversions were used to target mosquito collection and cloacal sampling of chickens for up to 2 weeks at sites with recent transmission activity for virus isolation. County agencies trapped mosquitoes using either CDC light traps or pickle jar traps, baited with CO2, 1 – 2 nights per week.

**Results:** Culex nigripalpus was the most abundant species trapped. Mosquitoes belonging to several genera (Aedes, Anopheles, Coquillettidia, Culiseta, Culex, Ochlerotatus, Uranotaenia) were speciated, pooled in groups of 50, and processed for molecular RT-PCR and Vero cell culture assays. EEE viral RNA was detected in pools of Cx. erraticus and Cx. quinquefasciatus. Other virus(es) were cultured from both mosquito pools and cloacal swabs of sentinel chickens at targeted EEEV sites.

**Conclusion:** Further characterization of these viral isolates is needed as they were negative by a standard RT-PCR panel used for identification of suspected arboviral isolates in Florida. The targeted method successfully detected EEEV and resulted in the isolation of additional virus(es) from mosquitoes collected at sites with recent sentinel seroconversions. This technique enhanced surveillance and characterization of arboviral pathogens in Florida.
In Vitro Pharmacodynamic and Cytocidal Activity of 1,2,3,4-tetrahydroacridone and 4(1H)-quinolone Antimalarials

Anupam Pradhan, Matthew Cross, Tina Mutka, Roman Manetsch, and Dennis E. Kyle

Abstract #: A-59
Presented by: Anupam Pradhan, PhD, Faculty

In Vitro Pharmacodynamic and Cytocidal Activity of 1,2,3,4-tetrahydroacridone and 4(1H)-quinolone Antimalarials

Anupam Pradhan, Matthew Cross, Tina Mutka, Roman Manetsch, and Dennis E. Kyle

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Keywords: Malaria, antimalarial screening, tetrahydroacridone, quinolone

Objective: To determine in vitro pharmacodynamic properties of selected derivatives from the THA (RMMC_93) and 4Q series (RMMC_95 and RMMC_105)

Methods: Active compounds determined by in vitro antimalarial assay were further analysed for rate of action and in vitro accelerated resistance to multiple drug (ARMD) phenotype assay.

Results: These chemotypes manifested excellent activity versus either atovaquone susceptible or resistant parasites and even showed more than 10,000-fold specificity towards parasites than mammalian cell lines. The three tested compounds manifested a similar trend in the rapidity of antimalarial action in W2 as well as 3D7 P. falciparum. The inhibition of nucleic acid synthesis starts from 24 to 36 hours depending on the concentration exposed, with the most significant inhibition observed with highest concentration tested (120 ng/ml). The 50% growth inhibition time (TI50) was similar irrespective of concentration to which the parasite was exposed. In comparison, TI90 was similar with that of atovaquone (10 ng/ml) for concentrations 60 ng/ml and above. The rate of antimalarial action of these RMMC compounds was similar to the rapidity of action of atovaquone. Importantly the THA and two 4Qs displayed not only remarkable erythrocytic stage activity, but resistance could not be selected for any of these drugs in an accelerated resistance to multiple drugs (ARMD) phenotype assay.

Conclusion: These studies suggest the THA and 4Q derivative have outstanding potential as new antimalarial drugs with nM potency for blood and liver stages of malaria.

Research supported by: MMV

Evaluation of Novel Arylimidamides in Hamster Model of Visceral Leishmaniasis

Anuradha Srivastava, Brian Vesely, Dennis E. Kyle, Department of Global Health, College of Public Health, University of South Florida, 3720 Spectrum Blvd. Tampa, Florida

Abstract #: A-60
Presented by: Anuradha Srivastava, PhD, Faculty

Evaluation of Novel Arylimidamides in Hamster Model of Visceral Leishmaniasis

Anuradha Srivastava, Brian Vesely, Dennis E. Kyle, Department of Global Health, College of Public Health, University of South Florida, 3720 Spectrum Blvd. Tampa, Florida

Keywords: Visceral Leishmaniasis, Kala Azar, Screening assays, Arylimidamides, Hamster model

Objective: Objective 1. Evaluation of In vitro and In vivo antileishmanial activity of arylimidamides (AIAs),

Methods: After primary screening against L. mexicana axenic amastigotes and Secondary screening in infected macrophage with beta lactamase transfected L. donovani or L. amazonensis, compounds were tested in in-vivo 11 day and 39 day hamster model. Drug efficacy was detected by LDUs and Real Time PCR

Results: Submicromolar IC50 values were obtained in in-vitro assays. Promising in-vivo activity was obtained for all AIAs.

Conclusion: Both day 11 and day 39 Hamster model has been validated and established successfully. Excellent activity of AIAs have opened a new avenue in anti leishmanial the drug discovery efforts.

Research supported by: Bill and Melinda Gates Foundation to Consortium for Parasitic Drug Discovery (CPDD)
**Phylogeny of Eastern Equine Encephalitis Virus in Florida**

**Gregory S. White** GHIDR Program, College of Public Health, USF; **Lillian M. Stark** Florida Department of Health, Bureau of Laboratories-Tampa; **Christy L. Ottendorfer** GHIDR Program, College of Public Health, USF; **Thomas R. Unnasch** GHIDR Program, College of Public Health, University of South Florida

**Keywords:** Eastern Equine Encephalitis Virus, phylogeny

**Objective:** The state of Florida has more documented Eastern Equine Encephalitis virus (EEEV) activity than any other state. Furthermore, it has been recently suggested that Florida may serve as a reservoir from which EEEV might be periodically introduced into the Northeastern USA. To study the evolutionary patterns of the EEEV in Florida, the majority of the genome (~11.6 kb) was sequenced from seven isolates from north Florida (Volusia County).

**Methods:** These data were used to identify 4 regions in the genome exhibiting high divergence among the Florida isolates. Over 20 additional Florida EEEV isolates, chosen from distinct years and geographical areas, were then sequenced in these 4 regions, together encompassing roughly 3kb.

**Results:** A phylogenetic analysis of these data revealed the existence of three distinct clades in Florida. Two clades showed temporal and spatial clustering, while the third contained strains from different locations and years. When published partial genomic sequences (roughly 1kb) derived from isolates Northeastern USA were analyzed with the corresponding regions from the Florida isolates, some, but not all of the NE isolates grouped with particular isolates from Florida.

**Conclusion:** In these cases, the isolation of the Florida isolate predated the NE isolate, supporting the hypothesis that these particular NE isolates may have been derived from Florida, implicating Florida as a potential source for the introduction of EEEV into the Northeastern USA.
Development of Therapeutic antitumor HPV E7 vaccine
Kelly Barrios, Department of Molecular Medicine, University of South Florida and Immunology Program, Moffitt Cancer Center, Tampa, Florida, Esteban Celis. Immunology Program, Moffitt Cancer Center, and Department of Oncologic Sciences, University of South Florida, Tampa, Florida

Keywords: HPV, Tumor, CTL, Vaccine, TLR

Objective: Our aim is to generate a therapeutic peptide-based vaccine against viral-induced cancers. We focus on human papillomavirus (HPV) because of its etiology in cervical carcinoma and in head & neck cancers.

Methods: We are using a mouse model to test a vaccine (TriVax) containing a CD8 T cell epitope, administered in combination with a Toll-like receptor (TLR) agonist and an antibody against the CD40 molecule as adjuvants to generate specific high affinity cytotoxic T lymphocytes (CTLs) against the HPV-E7 antigen expressed by tumor. We are assessing the immunogenicity of HPV-E7 TriVax by cytokine production using ELISPOT, ELISA and flow cytometry assays.

Results: we have tested the ability of several peptide compositions and formulations containing the HPV-E7 epitope within the TriVax vaccine to generate CTLs specific responses against an HPV-E7-expressing tumor cell. From these studies we have selected the most potent vaccine formulation to assess its therapeutic effect against established tumors.

Conclusion: The results from these experiments will allow us to compare conventional vaccines with TriVax for the induction of effective anti-tumor responses against established viral-induced tumors.

Research supported by: The Moffitt Cancer Center.

Traumatic Brain Injury Induces Inflammatory Gene Expression in a Rat Model. Mahasweta Das, PhD, Postdoc, Xiaoyuan Kong, James Musso III, Subhra Mohapatra, Keith R. Pennypacker, Lisa A. Collier, and Shyam Mohapatra. Departments of Internal Medicine (Division of Allergy and Immunology), Molecular Physiology and Pharmacology, and Molecular Medicine, Nanomedicine Research Center, College of Medicine, University of South Florida, Tampa, Fl

Keywords: Traumatic brain injury, inflammatory markers, cytokines, chemokines, qPCR

Objective: Traumatic brain injury (TBI) causes a complex spectrum of symptoms in which leukocyte migration into the brain and release of inflammatory cytokines play a crucial role in its pathophysiology. However, the nature of these inflammatory molecules and the mechanism of their action is unclear. The goal of this study was to determine the expression profile of inflammatory cytokines, chemokines and receptors (inflammatory markers) in the spleen after TBI.

Methods: A percussive trauma device was used to produce moderate brain injury in adult male SD rats. 24 h after the injury, surviving animals were deeply anesthetized and the spleens were removed and frozen at -80°C. The animals were then transcardially perfused, brains were removed and sections analyzed by Fluoro-Jade staining. Total RNA was isolated from spleens and converted to cDNA. Quantitative real-time PCR was performed on the cDNA to measure the expression of an array of inflammatory markers.

Results: Histopathology after Fluoro-Jade staining showed moderate injury to the rat cerebral cortex. qPCR arrays revealed expression of 84 inflammatory genes in the spleen at 24 hr post-injury. 20 genes were up-regulated with 6 of these being >2-fold up-regulated. 64 genes were down-regulated with 10 of those >2 fold down-regulated.

Conclusion: Our results indicate that the spleen is involved in mounting an immunologic response to TBI as early as 24 hrs post-trauma. Further studies are required to elucidate the exact immunologic pathway evoked by the spleen in the course of TBI.
**Biochemical and Pharmacological Characterization of Cytochrome b5 Reductase in Candida albicans**

Mary Jolene P. Holloway, Chris Laird, Kamisha Woolery, Michael J. Barber, Andreas Seyfang, Department of Molecular Medicine, College of Medicine, University of South Florida

**Keywords:** Candida albicans, cytochrome b5 reductase, antifungal drug targets

**Objective:** *Candida albicans* is a commensal member of the human microflora, the most common causative agent of fungal-related disease. Emerging drug resistance is a major problem in *Candida*, contributed by enzymes involved in the detoxification of xenobiotics and pharmacological agents. One such enzyme, cytochrome b5 reductase (cb5r), has high pharmacological significance owing to its role in fatty acid elongation, ergosterol biosynthesis, and cytochrome P450-mediated detoxification of xenobiotics.

**Methods:** Phylogenetic analysis revealed *Candida cb5r* is closest to plant homologues, and a recent patent listing plant cb5r as a novel herbicidal target renders these plant inhibitors promising candidates as a new class of antifungals against *Candida cb5r*.

**Results:** Kinetic analysis of the *Candida cb5r* isoform CBR1 revealed similar substrate affinity but lower catalytic rate ($K_m=6.6\mu M; k_{cat}=473s^{-1}$) when compared to the rat enzyme ($K_m=6.0\mu M; k_{cat}=800s^{-1}$). Thermodynamic analysis showed a decreased $T_{50}$ value and activation energy (44°C; $E_a=27.5 kJ/mol$) for *Candida CBR1* in comparison to rat cb5r (55°C; $E_a=35.9 kJ/mol$). Furthermore, we tested 17 substrate analogues to determine effects on enzymatic activity and found that ADP and ADP-ribose have the highest inhibitory effects on both rat and fungal cb5r, while GDP showed a higher inhibitory effect on *Candida CBR1* than its mammalian counterpart (48% versus 19% competitive inhibition).

**Conclusion:** We further developed *in silico* ribbon models of both *Candida* enzyme isoforms CBR1 and MCR1 based on the crystal structure of rat cb5r. These data now serve as the basis for *in silico* docking models and fragment-based drug design as subsequent steps in our antifungal drug discovery of *Candida cb5r* as a novel therapeutic target.

**Research supported by:** FCoE-BITT seed grant and NIH F31 Fellowship

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**Monosodium-Luminol Attenuates Ethanol- Or Lps- Induced Ros and Tnfa in Cultured Kupffer Cells**

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**Keywords:** Inflammation, Alcohol, Liver, ROS, SIRT1, NRF2

**Objective:** Monosodium-luminol (MSL) is a phthalazinedione compound. Studies have demonstrated that MSL has the ability to restore redox balance, regulate proteostasis, and up-regulate cell survival factors. Alcoholic liver disease is clinically associated with the development of steatosis, inflammation, fibrosis and cirrhosis in humans. In the present study, the effects of MSL on ethanol-induced oxidative stress and free radical generation were observed in cultured Rat Kupffer cells (RKC).

**Methods:** RKC cells were cultured for 4 hrs in media alone, or media containing various combinations of MSL, ethanol and LPS. After treatment, levels of reactive oxygen species (ROS) were determined using a DCFDA fluorescent assay, and the TNFα concentrations from supernatant were measured using a mouse TNFα ELISA kit.

**Results:** RKC cells exposed for 4 hours to either ethanol or LPS displayed marked increases in intracellular ROS levels and/or TNFα secretion. Co-incubation with MSL resulted in significant reductions of ROS and TNFα in a dose dependant manner to levels at, or below, controls.

**Conclusion:** MSL protects RKC cells against oxidative damage by reducing initial free radical production, and may reduce inflammatory pathways associated with LPS-stimulated TNFα secretion. Our findings suggest that MSL may have therapeutic potential in the treatment of human alcoholic liver disease.

**Research supported by:** National Institute on Alcoholism and Alcohol Abuse Grants AA-015951 and AA-013623
The Zebrafish Embryo as a Model for the Study of Bartonella henselae Infection and Host Response

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Keywords: Bartonella henselae, Angiomatosis, Zebrafish, Angiogenesis

Objective: Bartonella henselae (Bh) is a fastidious Gram-negative, facultative intracellular bacterium that is the causative agent of bacillary angiomatosis (BA) and cat scratch disease. BA is characterized by abnormal proliferation of the small blood vessels in some infected patients (angiogenesis). There is a need for a practical in vivo model in which the virulence factors of Bh and the response to its infection can be studied. Zebrafish (Danio rerio) embryos have previously been used as an animal model to study the pathogenesis and host response to bacterial infection. The zebrafish model has also been used to study angiogenesis. Therefore, we wanted to test if zebrafish can become infected with Bh.

Methods: Bh carrying the plasmid pNS2T5 expressing the red fluorescence protein gene (pNS2T5DsRed) were microinjected into 24-hour old zebrafish embryos. Infected embryos were examined by microscopy and homogenates of infected zebrafish were plated to determine the number of viable bacteria/zebrafish.

Results: Results show that zebrafish are infected with Bh as determined by; 1) the sustained presence of specific red fluorescence in the zebrafish for up to one week, 2) an increase in the number of viable bacteria/zebrafish over the first days of infection, 3) the detection of viable bacteria in zebrafish over the entire week of infection.

Conclusion: These results suggest that zebrafish model system is a suitable animal model to study Bh pathogenesis and host response.

Research supported by: This research is supported by the National Institute of Health grant AI038178 to BA.

Gene Regulatory Networks in the AIDS Pathogen, Toxoplasma gondii Have a Plant Origin

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Keywords: Toxoplasma gene regulation AIDS

Objective: The malaria-related protozoan, Toxoplasma gondii is an obligate intracellular parasite of mammals. Toxoplasmosis, the disease caused by this pathogen, exhibits a severe and sometimes fatal outcome for immunocompromised patients. It is estimated that 25% of the world population is chronically infected with T. gondii. T. gondii has a complex life cycle. The proliferative stage (tachyzoite) can be cleared by the immune system. Contrastingly, the parasite evades the immune system through its life-long chronic tissue cyst stage (bradyzoite). In infected immunocompromised patients, repeated rounds of recrudescence lead to pathogenesis. Unfortunately, current drugs are not well tolerated. Bradyzoite and tachyzoite stages can be differentiated by their gene expression profiles. What parasite factors control these stages and transitions have been a mystery until the recent discovery of a family of transcription factors related to the plant AP2 family. Plant-like AP2 genes are present in related parasites like Plasmodium and 68 AP2 genes were identified in T. gondii.

Methods: We hypothesize that AP2 transcription factors drive the tachyzoite/bradyzoite developmental transition, opening the door to targeting these AP2 proteins with small molecules.

Results: We have identified the bradyzoite-specific AP2 genes. Identification of their DNA binding motifs through protein binding array generated a strong candidate DNA sequence that was over-represented in the promoter regions of bradyzoite-specific genes.

Conclusion: We will discuss ongoing studies aimed at developing genetic models of induced expression of these factors as well as biochemical and structural studies to define the promoter mechanism regulated by these novel bradyzoite transcription factors.

Research supported by: NIH
Abstract #: B-7  
Presented by: Josh Radke, BS, Graduate Student

**Characterization of a Cell Cycle Transcription Factor in the AIDS Pathogen Toxoplasma gondii.** Josh Radke, Olivier Lucas and Michael White, Molecular Medicine Department, USF Health, College of Medicine, University of South Florida, Tampa, FL.

**Keywords:** Toxoplasma, cell cycle regulation, Apetela2

**Objective:** Toxoplasma gondii is an obligate intracellular parasite that causes severe disease in people with underdeveloped or compromised immune systems. Toxoplasmosis can be a lethal infection for people with AIDS, those undergoing chemotherapy and recent transplant recipients. Pathogenesis in this disease is the result of uncontrolled parasite growth in conjunction with significant tissue damage and inflammation.

**Methods:** Given that parasite growth and division is critical to disease, it is important to understand the mechanisms that regulate the progression through the parasite cell cycle. Approximately ~2500 mRNAs show cyclic patterns of expression during parasite division, however, the transcriptional mechanisms that regulate periodic gene expression are largely uncharacterized.

**Results:** The recent discovery of a class of plant-like transcription factors (Apetela2) in Apicomplexa has revealed an important set of proteins that play a critical role in parasite development and division. Over one-third of the AP2-like genes in Toxoplasma are cell cycle regulated during parasite replication with their timing of expression distributed across the cell cycle.

**Conclusion:** The AP2-factor, TgAP2VI-1, is a tightly regulated protein expressed at maximum levels during S phase of the cell cycle. We have verified cell cycle expression of this protein in live parasites, determined its DNA binding specificity, and developed a genetic model of conditional expression in order to pinpoint the group of genes regulated by this cell cycle transcription factor.

**Research supported by:** National Institute of Health

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Abstract #: B-8  
Presented by: Hilary Seifert, MS, Graduate Student

**Interferon Gamma in the Brain and the Spleen Following Ischemic Stroke, A Potential Therapeutic Target**

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**Keywords:** MCAO, HUCB cells, IFN-γ, pro-inflammatory cytokines

**Objective:** The immune system plays a critical role in delayed neural injury following ischemic stroke. Systemic treatment with human umbilical cord blood (HUCB) cells 48 hours (hrs) following middle cerebral artery occlusion (MCAO) decreases inflammation and neural injury. HUCB cells reduce inflammation by migrating to the injured brain and the spleen following MCAO; reducing the levels of the pro-inflammatory cytokine interferon gamma (IFN-γ) in both these organs.

**Methods:** Immunohistochemistry (IHC), for IFN-γ quantification, was performed on brains from rats which received sham, MCAO only, or MCAO + HUCB cell treatment after 48 hrs. All IHC was performed on brain sections from 3, 24, 48, 72, and 96 hrs post MCAO. Brain sections from 96 hr after MCAO were double stained for IFN-γ and immune cell surface markers. ELISA for IFN-γ was performed on spleens from rats which received sham, MCAO, or MCAO + HUCB cell treatment at 48 hrs. The ELISAs were run using eBioscience’s READY-SET-GO! kit.

**Results:** Cells expressing IFN-γ were present in the brain and the spleen following MCAO and levels peak 72 hrs post MCAO. Administering HUCB cells 48 hrs after MCAO reduced the amount of IFN-γ levels in both the brain and the spleen at 72 hrs following MCAO. T cells, B cells, and NK cells were identified as the cells producing IFN-γ in and around the infarcted area.

**Conclusion:** IFN-γ plays a significant role in the inflammatory signals leading to delayed neural injury following a stroke. First, it is present at the same physiological sites that HUCB cells migrate to when administered after experimental stroke. Secondly, HUCB cells decrease the amount of IFN-γ at these sites. Therefore effective therapies for stroke should target IFN-γ.

**Research supported by:** NIH RO1 NS052839
**Abstract #**: B-9  
**Presented by**: Kenrick Semple, MS, Graduate Student

**Efficient and Selective Prevention of Graft-versus-Host Disease (GVHD) by Antigen-Specific Induced T Regulatory Cells (Tregs)**  
Kenrick Semple, Department of Pathology and Cell Biology, University of South Florida, Claudio Anasetti, Blood and Marrow Transplantation & Immunology Programs, H. Lee Moffitt Cancer Center & Research Institute, and Department of Oncologic Sciences, University of South Florida, Xue-Zhong Yu, Blood and Marrow Transplantation & Immunology Programs, H. Lee Moffitt Cancer Center & Research Institute, and Departments of Pathology and Cell Biology, University of South Florida.

**Keywords**: nTregs, induced-Tregs, GVHD, Foxp3, TGFβ

**Objective**: To control GVHD by antigen-specific induced Tregs (iTregs)

**Methods**: Antigen-specific iTregs were generated by stimulating CD4+CD25- naïve cells from OVA-specific, OT-II TCR transgenic, foxp3/gfp knock-in mice with OVA peptide, IL-2 and TGFβ in the presence of syngeneic antigen-presenting cells (APCs). iTregs were isolated by FACS sorting CD4+CD25+GFP+ cells. GVHD was induced by transferring CD4+ effector T cells plus marrow cells from B6 mice into lethally irradiated OVA-expressing (B6 x bm12)F1 recipients. Additional OVA-specific iTregs were added into donor graft for evaluating their activity in the suppression of GVHD development using polyclonal iTregs as controls.

**Results**: Polyclonal naturally-derived Tregs (nTregs) have been shown to prevent GVHD with low efficacy and selectivity. In this study, we have tested the hypothesis that antigen-specific iTregs will prevent GVHD with high efficacy and selectivity. Using in vitro generated antigen-specific iTregs, we found that OVA-specific iTregs (CD4+GFP+) efficiently prevented GVHD lethality induced by T effector cells in OVA+ recipients, but not in OVA- recipients. As controls, CD4+GFP- cells had no effect on GVHD development in OVA- recipients, and even exacerbated GVHD in OVA+ recipients. Furthermore, the efficacy of these antigen-specific iTregs was significantly higher than polyclonal iTregs generated from C57B/6 donors and is more effective than polyclonal iTregs in preventing GVHD because they were suppressive at a Treg: T effector cell ratio up to 1:8 while polyclonal iTregs were suppressive at 1:2 ratio.

**Conclusion**: These results reveal the therapeutic potential of TGFβ induced antigen-specific iTregs to prevent GVHD efficiently and selectively.

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**Abstract #**: B-10  
**Presented by**: Lisa Smith, PhD, Postdoc

**Potential Regulators of the VirB Operon Encoding a Type IV Secretion System in Bartonella henselae**  
Lisa Smith, Kellie Larsen, Sarah Thomas, and Burt Anderson, Department of Molecular Medicine, College of Medicine, USF

**Keywords**: bartonella, virulence, antisense RNA, transcriptional regulation

**Objective**: Bartonella henselae is a facultative intracellular pathogen that colonizes human vascular endothelium causing inflammation and vasoproliferative lesions. Many of the virulent properties of B. henselae have been shown to be mediated by the VirB Type IV secretion system. However, little is known about the mechanisms that regulate VirB expression in Bartonella.

**Methods**: We constructed individual ihfA and spoT antisense RNA knockdown strains to determine whether either of these genes plays a role in B. henselae virulence. First, we assessed the relative ability of the knockdown strains to invade human endothelial cells compared to wild-type by gentamicin protection assay. We then measured the relative level of transcription of representative genes from the VirB operon in the knockdown strains compared to wild-type by real-time PCR.

**Results**: We identified two potential regulators based on homology to recently-discovered regulators of the VirB operon in the closely-related genus Brucella: integration host factor (IHF) and stringent response mediator SpoT. Knockdown of either of these two genes in B. henselae by antisense RNA causes decreased ability to invade human microvascular endothelial cells and reduced transcription of VirB2 and VirB5 compared to wild-type.

**Conclusion**: We have shown that knockdown by antisense RNA of either ihfA or spoT genes decreased the ability of B. henselae to invade and/or survive within human endothelial cells. Further, we showed that the relative levels of VirB mRNA in these knockdown strains were greatly reduced compared to wild type. These results suggest that each of these genes may individually play a role in regulating Bartonella virulence, namely through modulation of VirB operon expression.

**Research supported by**: NIH grant AI038178
Genomic Analysis of the Toxoplasma gondii Cell Cycle. Elena S. Suvorova, Department of Molecular Medicine, College of Medicine, University of South Florida, Tampa, FL. Margaret Lehmann, Department of Veterinary Molecular Biology, Montana State University, Bozeman, MT. Michael W. White, Department of Molecular Medicine, College of Medicine, University of South Florida, Tampa, FL.

Keywords: Toxoplasma gondii cell cycle genetic complementation

Objective: The AIDS pathogen, Toxoplasma gondii causes widespread infections in humans and animals. In the US the risk of permanent infection by this microorganism is 1:2 by age 50. Systemic growth of this pathogen leads to severe encephalitis in immunocompromised patients and current medical treatments for this disease are limited and not well tolerated by patients.

Methods: To provide new targets for drug discovery against this pathogen, we have developed a forward genetic approach to identify genes that are essential for parasite growth. More than 60,000 chemical mutants were generated and screened for conditional growth at permissive (34°C) and non-permissive (40°C) temperatures; 165 temperature-sensitive mutants emerged from this high-throughput screen.

Results: To date the primary phenotype has been determined for nearly half of this collection and mutants fall into six classes according to the cell cycle stage that was found defective: G1, S, mitotic/cytokinetic, early and late budding, and general growth mutants. To link mutant phenotypes with the responsible defective gene, we applied a robust genetic complementation approach that employed a genomic cosmid library.

Conclusion: The list of identified genes includes regulatory enzymes, structural components, and nucleic acid binding proteins. A summary of the mutant collection, details on the genetic technology and studies of selected mutants will be presented.

Characterization of a Putative Argininosuccinate Synthase Regulatory Phosphorylation Site Implicated in Vascular Endothelial Nitric Oxide Production. Ricci Thompson1, Karen D. Corbin2, Laura C. Pendleton1, and Duane C. Eichler1 1Molecular Medicine, University of South Florida, 12901 Bruce B. Downs Blvd. Tampa, FL 33612; 2Nutrition Research Institute, UNC Chapel Hill, 500 Laureate Way Suite 1302, Kannapolis, NC 28081

Keywords: argininosuccinate synthase, nitric oxide, phosphorylation, VEGF

Objective: Argininosuccinate synthase (AS) catalyzes the rate-limiting step of the nitric oxide (NO) cycle in bovine aortic endothelial cells (BAECs), converting citrulline to argininosuccinate. Much has been elucidated concerning the regulation and localization of endothelial nitric oxide synthase (eNOS); however, little is known about AS in terms of its regulation.

Methods: In our laboratory, we have developed a strong evidential case supporting the proposal that AS is regulated by phosphorylation. Using in silico, as well as proteomic analysis, we have identified putative phosphorylation sites in the AS amino acid sequence. Of particular interest was the phosphorylation site at serine 328 (S328) that juxtaposes a putative caveolin binding motif in a central region of AS tertiary structure. In this study, we examined the effects of mutating the S328 site to an alanine (S328A) or to the phospho-mimetic aspartate (S328D) in order to determine any differential effect on supporting NO production in BAECs. These mutant AS constructs were over-expressed, and NO production was measured after stimulation by either addition of calcium ionophore and ortho-vanadate, or by the addition of VEGF.

Results: With either condition, the S328D mutant produced significantly higher NO than over-expressed wild-type AS (n=3, p<0.05) while NO levels for the S328A mutant did not exceed background. Significantly, the S328A mutant also did not support viability when the BAECs were serum starved for 18 hours.

Conclusion: These data suggest that the S328D substitution supports stimulated NO production as well as cell viability during serum starvation. These results also support the proposal that S328 may be a biologically significant site for regulation of AS by phosphorylation.
Respiratory Syncytial Virus Non-structural Protein 1 (NS1): Nuclear Localization and Gene Regulation in Infected Epithelial Cells

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Keywords: respiratory syncytial virus (RSV), interferon response, transcriptional regulator

Objective: Nonstructural protein 1 (NS1) of respiratory syncytial virus (RSV) has been implicated in suppressing IFN activity and development of the antiviral immune response. Our lab investigated NS1’s role in RSV survival through the suppression of the antiviral response during early infection. The aim of this study was to determine the subcellular localization of NS1 and examine it’s ability to bind to genomic DNA as a transcriptional regulator.

Methods: A549 (human lung epithelial carcinoma) cells were transfected with pNS1-FLAG. A549 nuclear and cytoplasmic extracts were immunoprecipitated with anti-FLAG, separated by SDS-PAGE and immunoblotted with anti-FLAG. Transduction with adenoviral vector AdNS1-FLAG enabled indirect immunofluorescent visualization of FLAG. To assess the ability of NS1 to bind to genomic DNA and suppress IFN-inducible gene expression, genomic DNA from WT or NS1-deletion mutant RSV infected A549 was sheared and analyzed by chromatin immunoprecipitation (ChIP) assay. Precipitated DNA was used as template for qualitative and real-time PCR with primers specific for IFN-inducible genes.

Results: NS1 was found in the nucleus and cytoplasm using immunoblot and immunofluorescence. Immunoprecipitation of NS1 and ChIP analysis showed that NS1 binds to genomic DNA and may reduce transcription of IFN-inducible genes. Cells infected with NS1-deletion RSV yielded mRNA expression of IRF-7 comparable to mock-infected cells.

Conclusion: NS1 translocation to the nucleus may suppress the host immune response during early RSV infection by binding to, IFN-inducible genes and blocking transcription. To understand tight regulation of NS1-mediated RSV-dependent and independent gene regulation an inducible system is being established.

Research supported by: VA Merit Review Grant to SSM.

Beta-catenin Expression in Matched Pre-treatment and Post Neoadjuvant Chemotherapy Breast Cancers

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Keywords: B-catenin, Neoadjuvant Chemotherapy, Breast Cancers

Objective: Beta-catenin is a cell adhesion molecule that is associated with E-cadherin and is a pivotal part of the wnt signal pathway. The objective is to verify that beta-catenin is activated in residual breast carcinoma cells indicating resistance to neoadjuvant and have frequent activation of the wnt signaling pathway.

Methods: Twenty-nine matched pre-treatment and post neoadjuvant chemotherapy breast carcinomas were subjected to immunohistochemical stains with anti-beta-catenin antibody. Normal beta-catenin stain was defined as crisp membrane staining in >90% tumor cells; aberrant expression was nuclear staining in >5% tumor cells. Reduced membranous staining and cytoplasmic staining was also recorded. Clinicopathological data including tumor type, tumor size, tumor necrosis, lymph node status, predictive and prognostic marker studies (ER, PR and HER2) and clinical stage were available.

Results: Normal beta-catenin expression was observed in all pre-treatment and post neoadjuvant samples except 5 cases of invasive lobular carcinoma, which are negative for beta-catenin stain. Mild to moderate reduced membranous staining was seen in 2 post-treatment samples. Less than 5% cytoplasmic staining is seen in some cases but there is no difference in pre- and post-treatment specimens.

Conclusion: There is no difference in the expression pattern of beta-catenin in pre- and post-neoadjuvant chemotherapy specimens. Lobular carcinoma has complete absence of beta-catenin immunoreactivity. Except for the tumor type, there is no difference in the expression pattern of beta-catenin. Beta-catenin's role in activating the wnt signaling pathway and thereby possibly conferring neoadjuvant chemotherapy resistance, warrants further investigation.
Bcl-2 as a Potential Modulator of Angiogenesis in Ovarian Cancer

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**Abstract #**: B-15  
**Presented by**: Nicole Anderson, MS, Graduate Student

**Keywords**: Ovarian Cancer, Bcl-2, angiogenesis

**Objective**: Angiogenesis is the growth of new blood vessels from preexisting blood vessels. Ovarian cancer (OC) tumors undergo extensive vascularization via angiogenesis, requiring a balance between angiogenic stimuli (i.e. VEGF) and angiogenic inhibitors (i.e. angiostatin). B-cell lymphoma 2 (Bcl-2), an antiapoptotic protein, is elevated in many solid tumors, including OC tumors. Bcl-2 is typically localized to the membranes of the endoplasmic reticulum, nuclear envelope, and mitochondria. However, upon culturing ovarian cancer cell lines in vitro, we saw Bcl-2 present in their conditioned media. The purpose of this study was to determine if secreted Bcl-2 affects angiogenesis.

**Methods**: Conditioned media (CM) from OC cells was immunodepleted using Bcl-2 and/or VEGF antibodies. CM was used to culture HUVECs in an in vitro angiogenesis assay. Tube formation was observed using an inverted microscope and images were acquired and analyzed using the Image Pro Plus (IPP) digital imaging system. IPP was used to measure tube length and the number of polyhedral chambers.

**Results**: Results show that Bcl-2 significantly affects in vitro HUVEC tube formation as well as the complexity of tubal networks. Secreted Bcl-2 from OC cells appeared to have an inhibitory effect on HUVEC cell migration and their ability to produce long tubes that connect with one another.

**Conclusion**: The data suggest that the antiapoptotic Bcl-2 protein may play a role in regulation of OC tumor angiogenesis. Bcl-2 may assist in controlling the growth and development of new blood vessels as well as maintaining the integrity of established blood vessels. Therefore, these findings could be clinically useful in the development of therapeutic angiogenic inhibitors.

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Use of Anti-Phosphohistone H3 Immunohistochemistry to Determine Mitotic Rate in Thin Melanoma

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**Abstract #**: B-16  
**Presented by**: David Casper, MD, Resident

**Keywords**: melanoma, phosphohistone h3, mitotic rate

**Objective**: The 7th AJCC melanoma staging system will introduce mitotic rate (MR) as a primary criterion for staging thin melanomas. Accurate counts are essential because the finding of a single mitotic figure (MF) will alter the staging and management of these patients. The traditional method to determine MR is based on locating MFs in H&E stained sections, and counting them in five 40X high power fields (HPF) for a total area equal to 1 mm². Although reproducible, this technique can be time consuming and prone to inter-observer variability. We developed and tested a protocol for phosphohistone H3 (pHH3) immunohistochemistry (IHC) to more easily quantify MFs in these lesions.

**Methods**: MR was determined by anti-pHH3 IHC at 20X in 30 melanomas, 0.45 to 1.2 mm in depth, and the results were compared with traditional H&E methodology at 40X, in a double-blinded fashion. MFs could be easily visualized and counted on anti-pHH3 stained slides at 20X power by using the micrometer lines to form a cross and 1 mm² area. The MR on H&E was obtained in the traditional method by finding the “hot spot” on a 40X field and extending the count to an additional 4 HPF for a total of 1 mm².

**Results**: The mean MR was 1.63 by anti-pHH3 and 0.67 for H&E, an increase of 243%. The Spearman correlation coefficient was 0.88 (p<0.0001). When melanomas were designated as “mitotically active” if the MR by anti-pHH3 was > 2, and > 1 using H&E, the correlation coefficient was 1.0. No thin melanomas were mitotically “active” on H&E, but “inactive” on anti-pHH3 IHC.

**Conclusion**: Results indicate that anti-pHH3 can accurately label MFs in thin melanomas, which simplifies quantification of MR. We predict that this new staining technique will be a useful adjunct in melanoma staging.
A Retrospective Review of Extraintestinal Gastrointestinal Stromal Tumors: A Single Institution Experience

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Keywords: *Extraintestinal gastrointestinal stromal tumors *Sarcoma *c-kit

Objectives: Gastrointestinal stromal tumors (GIST) are the most common mesenchymal neoplasm of the gastrointestinal tract. Rarely, GISTs occur outside the GI tract and are known as extraintestinal GIST (EGIST). The clinicopathologic data and outcomes of EGIST and GIST patients were investigated.

Methods: We reviewed 205 consecutive patients with a histologic diagnosis of GIST from 1999 to 2009. Final pathology was confirmed at our institution. Age, primary tumor location, size, mitotic rate, imatinib therapy and overall survival of EGIST patients were compared with gastrointestinal primaries.

Results: Of 205 patients, there were 9 patients (4.4%) with EGIST: 5 retroperitoneum, 2 pelvis, 1 abdominal wall and 1 mesenteric/omentum. EGIST and GIST patients were similar age at diagnosis (median 72 vs. 63 yrs, p=0.09). The median tumor size of EGIST was larger than that of GIST (17.5cm vs. 8cm, p=0.002). A mitotic rate of >5/50 HPF was seen in 67% of EGIST vs. 48% of GIST (p=0.12, Χ²). The incidence of metastatic disease was similar in EGIST (44%) and GIST (48%) (p=0.83). Imatinib therapy was equivalent for GIST (61%) vs. EGIST (67%). After a median follow up of 33 months (1-163 months), the median overall survival was 18 months for EGIST vs. 49 months for GIST patients (p=0.003). Univariate analysis suggests EGIST (p=0.003), size>10cm (p=0.006), mitotic rate>5/50 HPF (p=0.05) and metastatic disease (p<0.001) at presentation were associated with worse overall survival.

Conclusion: EGIST patients demonstrate a rare subpopulation of all patients with GIST. In our experience these tumors tend to be more locally advanced on presentation relative to intestinal primaries. EGIST, large size and high mitotic index are associated with worse overall survival in GIST patients.

Alarming Trends Continue in Young Patients with Colorectal Cancer in the Early 21st Century: A Retrospective Review of One Institution’s Patients Less Than 50 Years of Age

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Keywords: Colorectal Cancer Screening; Colorectal Cancer in the Young; Colorectal Cancer Incidence

Objective: The objective of this study is to determine if trends observed in previous studies, including increasing incidence and aggressiveness, continue in young patients with colorectal cancer (CRC).

Methods: The electronic medical record system was queried for all patients under 50 with a new diagnosis of CRC from 2001-2008. This data was used to determine age, year of diagnosis, gender, tumor location, symptoms, family history, medical history, TNM staging, and histology.

Results: The query resulted in 100 patients with no clear trend in incidence per year. There did appear to be an exponential increase beginning between 36-40, however. The most common presenting symptom was nonspecific abdominal pain. Most patients did not have a significant medical or family history. Less than a third of the cancers were well-differentiated, and 72% were Stage III or worse. These tumors were well distributed.

Conclusion: CRC appears to substantially increase beginning at the age of 36-40. These tumors continue to be advanced at diagnosis, and occur throughout the colon. If these trends are confirmed through larger studies, colonoscopic screening should begin at an earlier age.
**Growth and Differentiation of Cancer Cells on Three-Dimensional (3D) Scaffolds as a Model to Study Tumor-Stroma Interactions**

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3. SIPAIID
4. Nanomedicine Research Center

**Keywords:** 3D, scaffold, tumor, stroma, PLGA

**Objective:** The goal of this study was to develop artificial 3D scaffolds with mechanical and topographical features of in vivo stromal matrix to study tumor-stroma interactions.

**Methods:** Scaffolds were made by electrospinning poly(lactic-co-glycolic) acid (PLGA) nanofibers which provide a large surface area and high porosity for cell growth and differentiation. An additional 3D matrix was made by self-assembly of the oppositely charged polyelectrolytes, chitosan and alginate to form a chitosan/alginate hydrogel. PC3 prostate cancer cells and WMPY stromal cells were cocultured on the scaffolds and proliferation was measured by WST (mitochondrial activity) and Ki67 (nuclear antigen) assays.

**Results:** On the PLGA scaffold, conditioned media from PC3 cultures stimulated WMPY proliferation while conditioned media from WMPY cultures stimulated PC3 proliferation. Cocultured PC3/WMPY cells showed greater proliferation than cells grown separately in conditioned media. Cells cocultured on chitosan/alginate hydrogels grew slower than on PLGA fibers indicating that differences in microenvironment are important in tumor-stroma interactions.

**Conclusion:** Tumor cells growing with stromal cells on tissue-engineered 3D scaffolds provide a pseudo-in vivo model for studying cancer progression and testing new anticancer agents.

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**A Retrospective Study of Patients with Melanoma Brain Metastases Receiving Concurrent Whole Brain Radiation and Temozolomide**

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**Keywords:** Temozolomide, Melanoma brain metastasis, whole brain radiation therapy, melanoma

**Objective:** Metastatic melanoma is the second most common cancer to metastasize to the brain and it is typically treated using stereotactic radiosurgery with or without whole brain radiation therapy. Recently, the alkylating agent temozolomide (which has demonstrated activity in patients with brain metastasis) has been suggested to be used with concurrent whole brain radiation (WBR) to delay metastasis recurrence, increase survival rate, and improve the quality of life of patients. Thus, the combination of WBR with temozolomide may provide additional benefit for patients with melanoma brain metastases compared to WBR alone. This retrospective study was intended to review the use of temozolomide in combination with WBR for patients with melanoma brain metastases.

**Methods:** This study used a retrospective chart review of all patients diagnosed with melanoma brain metastases treated at Moffitt Cancer Center with concurrent whole brain radiation therapy and temozolomide from January 2006 through June 2009. Data collected from PowerChart included the dates of WBR and temozolomide treatment, the date of diagnosis of metastatic melanoma, side effects and toxicity of the treatments. PFS (Progression Free Survival) and an estimation of KPS (Karnofsky Performance Scale) throughout treatment. Progression is defined as the appearance of new brain lesions or an increase in size of known lesions as measured by MRI. PFS was measured from the date of the first treatment of the concurrent WBR and temozolomide until the occurrence of progression.

**Results:** Kaplan-Meier method and Cox proportional model will be used to assess PFS and OS; data is still being collected.

**Conclusion:** The data is still in preliminary evaluation.

**Research supported by:** USF College of Medicine, H. Lee Moffitt Cancer Center
The Impact of Axillary Ultrasound and Needle Biopsy on Sentinel Lymph Node Biopsy Procedures in Breast Cancer Patients

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Dept. of Oncologic Sciences, College of Medicine, University of South Florida

Keywords: Axillary ultrasound, fine needle aspiration

Objective: To determine the impact of axillary ultrasound (AUS) and needle biopsy on performance of sentinel lymph node biopsy (SLNB) in patients with operable T2 or greater breast cancer tumors.

Methods: A retrospective review of Moffitt Cancer Center’s Breast Database from 2004 to 2008 was performed. Patients with breast cancer who underwent axillary ultrasounds with a T2 or greater tumor were identified. 212 patients received AUS. Patients with clinical tumor size <2cm, incomplete records, or with post-operative AUS were excluded. Patients with clinical T2 or greater tumors without AUS (2004-2008) were identified as a control group.

Results: 153 patients were analyzable for this study. All patients were female and the median age was 53.7 yrs (range 22.8-85.8) compared to 53.8 yrs for the controls. The median tumor size was 3.8cm (range 1.0-20.0) compared to 2.5cm. Of 153 patients with AUS, 120 were abnormal/positive (pos) and 33 were normal/negative (neg). Of the pos AUS results, 88 had pos FNA results (true pos) and 32 had a neg FNA (false pos). 8/32 (6.7%) neg FNA patients had pos SLNB. Of the 33 neg AUS results, 15 had pos SLNB (false neg). The sensitivity and specificity of AUS was 86.2% and 40.5%. AUS combined with FNA had a sensitivity of 89.3% and a specificity of 100%. 88/120 (73.3%) were spared the SLNB procedure (p<0.0001).

Conclusion: AUS and FNA significantly reduced the number of node pos breast cancer patients undergoing the SLNB procedure. This spares patients the time, expense, and discomfort of having the SNLB, and may avoid an additional operation.

Research supported by: Research Scholarly Concentration

Chemoradiotherapy without Elective Nodal Irradiation for Limited-Stage Small Cell Lung Cancer

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Keywords: small-cell lung cancer, chemoradiotherapy, elective nodal irradiation

Objective: Traditionally, treatment of limited-stage small cell lung cancer (L-SCLC) has involved large mediastinal radiation fields. More recently, involved-field radiation without elective nodal irradiation (ENI) in non-small cell lung cancer has been shown to have less toxicity and low risk of isolated nodal relapse. This has led to the question of treatment volume reduction for patients with L-SCLC. We set out to describe the treatment details and patterns of failure of the initial cohort of PET-staged L-SCLC pts treated without ENI at Moffitt.

Methods: We retrospectively identified 18 consecutive L-SCLC patients treated at Moffitt between 3/2006 and 4/2008. All patients received a platinum-based doublet and were treated with conformal technique to involved-fields (by PET and CT imaging), without ENI, to a dose of 4500 cGy in 30 BID fractions of 150cGy. Patient demographics, risk factors and treatment details were collected, and timing, type, and location of first failures were analyzed through review of the medical record and followup imaging studies.

Results: 16 patients(88.9%) completed ≥ 4 planned cycles of chemotherapy; all completed radiotherapy over a median of 21 days. With median follow-up of 15.5 months, 5 patients(27.8%) were alive with no evidence of disease(NED);3(16.7%) were alive with disease, and 10(55.6%) had died, including one who died NED. First failure was local in 2 patients(11.1%), distant in 6(33.3%), locoregional in 2(11.1%), and local, regional and distant in one(5.6%). Only one patient(5.6%) had an isolated regional failure.

Conclusion: Chemoradiotherapy without ENI for L-SCLC allows for dose reduction to uninvolved regional nodes with few isolated regional failures. Local failure rates suggest the need for dose escalation.
**Abstract #**: B-23  
**Presented by**: Steven Finkelstein, MD, Resident

**At the Confluence of Radiation Therapy and Immunotherapy: Combination of External Beam Radiation (EBRT) with Intratumoral Injection of Dendritic Cells (DC) as Treatment of Sarcoma Patients D.I. Gabrilovich, M.M. Bui, M.J. Cotter, D.Cheong, R.J. Gonzalez, R.V. Heysek, B.C. Lenox, V.K. Sondak, J.S. Zager, G.D. Letson, S.J. Antonia Department of Radiation Oncology, College of Medicine, University of South Florida**

**Keywords**: radiation immunotherapy sarcoma

**Objective**: 1) To determine if combined cell death inducing EBRT + intra-tumoral DC induces a T lymphocyte immune response specific for sarcoma antigens, in humans 2) To study functional activity of T cells, and presence/function of DCs 3) To assess toxicity and primary tumor responses

**Methods**: Patients with clinical stage T2N0M0 high grade soft tissue sarcoma of the extremity/trunk/chest wall were treated with standard neo-adjuvant EBRT 5040 cGy / 180 cGy coordinated with additional, experimental DC therapy consisting of DC progenitor apheresis, ex-vivo expansion and culture, and intratumoral injection of 10 million DC per injection. Preparation of autologous DC product was performed following standardized GMP laboratory procedures.

**Results**: Preclinical in-vivo data in a murine model demonstrated tumor directed immunity was curative. Clinically, targeted accrual was reached with 18 patients completing EBRT with experimental intratumoral DC therapy; all patients have completed full immunologic assessment. Ten of the patients (56%) were induced to produce significant anti-autologous tumor cell immune responses, as determined using ELISPOT assays on the patients’ PMBC before and after treatment. This response became stronger (in some cases a 4-fold increase over control) after the last DC injection. These responses persisted even 30 weeks starting treatment but can abate over time. No clinical toxicity has been observed. With clinical follow-up >1 year available on 14 patients, 1/7 patients developed progressive disease with robust response (detected at more than one time point), and 4/7 patients developed progressive disease with weak or no response.

**Conclusion**: Multifaceted immunological assessment suggests significant anti-autologous tumor cell immune responses.

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**Abstract #**: B-24  
**Presented by**: Thomas Hayman, BS, Med II Student

**Body Mass Index and Overall Survival in Esophageal Adenocarcinoma**

Thomas Hayman, Dept. of Oncologic Sciences, College of Medicine, USF, Ravi Shridhar, Sarah E. Hoffe, Jill Weber, Matthew Biagioli, Thomas J. Dilling, Dung-Tsa Chen, Richard C. Karl, James S. Barthel, Kenneth Meredith, Moffitt Cancer Center, Moffitt Esophageal Research Team

**Keywords**: esophageal cancer adenocarcinoma

**Objective**: The purpose of this study is to evaluate the association between pre-operative BMI and survival in esophageal adenocarcinoma.

**Methods**: An institutional database review was conducted of patients who underwent surgery for esophageal cancer from 1994 to 2008. Patients were grouped into two BMI categories: BMI > 25 or BMI ≤ 25. Patients were excluded if they had a pre-operative BMI < 20. The primary endpoints were overall survival (OS) and disease-free survival (DFS) which were analyzed using the Kaplan-Meier method and log-rank analysis. Univariate and multivariate analysis (MVA) were performed using Cox proportional hazard regression model. Variables included BMI, tumor grade, AJCC T and N-stage, number of nodes dissected, gender, type of surgery, and age.

**Results**: Four hundred thirty patients were included in the analysis. Univariate analysis showed a correlation between patients with a BMI > 25 and lower tumor grade (p < 0.0001). Individuals with BMI > 25 had increased OS (p = 0.0001) and DFS (p = 0.0003). The 3-year and 5-year OS for patients with BMI > 25 was 59.4% and 48.8% versus 39.7% and 30.6% for patients with BMI ≤ 25 respectively. Independent prognostic variables for increased mortality were male gender, increasing age, transthiatal surgery, and grade 2 or 3 tumors. BMI > 25 was an independent prognostic variable for increased survival on MVA (HR 0.618; 95% CI 0.411 - 0.930; p = 0.0209). T-stage, N-stage, surgery type, tumor length, number of nodes dissected, and use of neoadjuvant chemoradiotherapy were not prognostic for overall survival.

**Conclusion**: In our analysis, BMI > 25 correlated with lower grade tumors and increased overall survival and should be considered as a prognostic factor in patients with esophageal adenocarcinoma.
Abstract #: B-25

**Regulation of ERα activity by Fe65 in Breast Tumor Development**

Ming Hu, Junying Bao, Yintao Zhang, Jinfu Tang, Anfernee Kaiwing Tse, Xiaohong Zhang, Santo V. Nicosia, Wenlong Bai. Department of Pathology and Cell Biology, College of Medicine, University of South Florida

**Keywords:** ERα, Fe65, Tip60, E2

**Objective:** To investigate the pivotal function of Fe65 in ERα action and breast cancer development.

**Methods:** GST-pull down and co-immunoprecipitation assays were used to verify the interaction between Fe65 and ERα and to define the domains of Fe65 that mediates the interaction with ERα. Luciferase reporter assays were used to assess the effect of Fe65 on the transcriptional activity of the ERα. Knock down strategy is being pursued to determine the role of endogenous Fe65 in the growth response of breast cancer cells to estrogens and anti-estrogens. We are also in the process of establishing Fe65 transgenic mouse lines under the control of mammary specific promoter MMTV to test the oncogenic potential of Fe65 in mammary tumorigenesis.

**Results:** Fe65 and ERα forms a complex in non-neuronal cells and the complex formation is mediated through the PTB2 domain. Fe65 but not PTB2 mutant increases ERα Activity. Fe65 appears to be overexpressed in breast cancer cell lines and human breast tumor samples.

**Conclusion:** Fe65 may function as a positive regulator for estrogen receptor and potentiate the estrogen stimulation of breast cancer cell growth. Fe65, besides its defined role in neuronal cells, may stimulate mammary tumorigenesis and represent a novel target for breast cancer therapeutic treatment.

**Research supported by:** DOD funding

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Abstract #: B-26

**Automated Ki-67 Immunohistochemistry Quantification by Conversion of Tissue Staining into Two-dimensional VirtualFlow Cytometry Histogram**

F. Kalantarpour, G. Blanck, V. Adams, M.C. Lloyd and H.D. Cualing. Oncologic Sciences/ Hematopathology, H.L. Moffitt Cancer Center, USF Department of Pathology and Cell Biology, College of Medicine, USF

**Keywords:** flow cytometry, Ki-67, virtualflow, thymus

**Objective:** We present a novel method of converting tissue Ki-67 nuclear immunostain to flow cytometry-like result using VirtualFlow technic in thymic and lymph node tissue. There is a need to more accurately analyze Ki-67 paraffin tissue immunostaining beyond visual estimation for both diagnostic and prognostic value in lymphoid tissue.

**Methods:** Fourteen adult mantle cell lymphoma lymph nodes and a 3 randomly sampled pediatric thymic tissue (T1 to T3), were analyzed for Ki-67 AP-DAB immunostaining by automated Ventana (Tucson, AZ) method. Digital Virtual Flow analysis (beta testing by IHCFLOW Green-Great, Inc.) on 20x field, 10 frames in average, with 329 - 811 cells/ frame ) were done on 4 thymic regions: medullary, cortico-medullary, subcortical, and cortical-lobular. We examined the correlation coefficient to show applicability of the method using two different tissues, and in lymph nodes, using a small and large image frame, with different microscope systems, and varied rheostat settings with different light intensities (0.4 to 7.0 rheostat) and different operators.

**Results:** Mean Ki-67 expression in cortical regions were similar in two of the thymus glands, however in medullary regions all three cases, T1, T2, T3 showed a similarly lower Ki-67 expression. In lymph nodes the blastic variant of mantle cell lymphoma showed a uniformly higher Ki-67 expression in compare to the lymphocytic variant.

**Conclusion:** These results suggest Virtualflow cytometry microscopy could be used by pathologists and scientists when analyzing immunohistochmeical stain for Ki-67.

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Post ERCP Pancreatitis: A Single Center Experience

Deepika L. Koya, MD, MSCR Adel Daas, MD Prieto Ricardo, MD Jonathan Keshishian, MD Haim Pinkas, MD Jay Mamel, MD Yasser Saloum, MD Patrick Brady, MD
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Keywords: pancreatitis ERCP ampullectomy

Objective: The aim of our study is to determine the incidence of post ERCP pancreatitis and predictors associated with this complication at a single referral center.

Methods: Between May 2007 and February 2009, data were collected prospectively from 800 consecutive ERCPs performed at a single referral center and entered into a database. Univariate (Fisher’s exact test) and multivariate (multivariate logistic regression) analyses were used to identify patient and procedural risk factors associated with post-ERCP pancreatitis.

Results: Pancreatitis was the most common complication (18/800; 2.3%) and in 72% of cases it was mild pancreatitis. Risk factors significantly associated with post-ERCP pancreatitis identified by univariate analysis are female gender, sphincter of Oddi manometry, ampullectomy, pancreatic sphincterotomy and biliary sphincterotomy. After adjusting for relevant confounders ampullectomy and pancreatic sphincterotomy were identified as independent risk factors for post-ERCP pancreatitis. Of 15 ampullectomy cases, 6 had prophylactic pancreatic duct (PD) stents placed after the procedure. No pancreatitis occurred in the stented group whereas two patients with ampullectomy but no prophylactic PD stent placement developed pancreatitis (0% vs. 22%).

Conclusion: The most frequent ERCP-related complication was pancreatitis. The incidence of pancreatitis is similar to that reported from other major ERCP centers. We identified ampullectomy and pancreatic sphincterotomy as independent risk factors for post-ERCP pancreatitis. It appears that prophylactic PD stents confer a protective effect in minimizing post ERCP pancreatitis in patients with ampullectomy. Future large sample size studies are warranted to confirm these findings.

Anti-Cancer Activity of HIV Viral Protein R (Vpr)

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Keywords: Viral Protein R (Vpr) cancer cytotoxicity Protein Transduction Domain (PTD)

Objective: To determine the active region within the carboxy-terminus of the HIV-1 Vpr protein that is responsible for inducing anti-cancer activity in a number of cell lines including B16.F10 melanoma, ZR-75 breast duct, Panc-1 pancreatic, and LNCaP prostate carcinomas.

Methods: Overlapping 15 amino acid peptide sequences from the carboxy-terminus of Vpr were incubated with 20,000 cells of various tumor lines at concentrations ranging from 20µM to 100µM. In addition to the unmodified Vpr peptides, peptide transduction domains (PTDs) were conjugated to the peptides. One specific PTD consisted of an amino acid sequence from the HIV protein Tat. After incubation of cells at various time points, viability/cytotoxicity was determined using a WST-1 or MTT colorimetric assay. Percent viability/toxicity in the Vpr peptide treated wells were calculated using OD values at 450nm.

Results: Cells treated with several carboxy region Vpr peptides induced significant cytotoxic effects compared to control cells treated with a non-biologically active protein of similar molecular weight. Additionally, Tat-conjugated peptides were even more potent at similar concentrations and time-points than unconjugated peptides alone.

Conclusion: Vpr is able to induce cytotoxic effects in several tumor cell lines, with the activity likely mediated through the carboxy terminus of the protein.

Research supported by: FCoE-BITT, Inovio, Inc.
The 185delAG Mutant Protein, BRAT, Enhances MMP-1 Expression in Ovarian Surface Epithelial Cells
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Keywords: BRCA1, MMP-1, ovarian cancer

Objective: Studying risk-associated BRCA1 truncation mutations, such as the founder mutation 185delAG, may reveal signaling pathways important in hereditary OC etiology. Human ovarian surface epithelial (HOSE) cells expressing the 185delAG mutant, BRAT, exhibit enhanced chemosensitivity and up-regulation of the tumor suppressor maspin. In the current study, we wish to determine BRAT’s impact on expression and activity of matrix metalloprotease 1 (MMP1), another key player in invasion and metastasis, and the mechanism of this modulation.

Methods: HOSE, breast epithelial cells, and breast cancer cells expressing the 185delAG mutation were analyzed for differential target gene expression by semi-quantitative and real time PCR. Cells were transiently transfected with GFP, constitutively active Akt (CA-Akt), control siRNA, or si c-Jun. Western blotting was used to evaluate protein levels. FRET ELISA of conditioned media (CM) was used to evaluate levels and functionality of MMP1 in CM. Cells were treated with cisplatin and subjected to MTS assay to evaluate chemosensitivity.

Results: BRAT-expressing HOSE cells exhibited enhanced MMP1 mRNA expression and protein levels in CM. FRET ELISA of CM revealed total MMP1 (pro and active) was higher in BRAT cells, and that MMP1 was functional. CA-Akt and c-Jun knockdown diminished MMP-1 expression in HOSE, and c-Jun knockdown decreased MMP1 in CM. Expression of BRAT in normal and breast cancer cell lines did not alter chemosensitivity or MMP1 levels.

Conclusion: Normal HOSE cells expressing the BRAT mutation exhibit Akt and c-Jun-dependent changes in expression of MMP1. Further investigation is warranted since increased expression of this gene may represent an initial step forward on the continuum of malignant transformation.

Intracellular Accumulation and Metabolism of Lycopene in Human Prostate Cancer Cells Expressing Carotene 9', 10'-Monooxygenase (CMO2)
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Keywords: Carotene 9',10'-Monooxygenase 2, High Performance Liquid Chromatography, Lycopene, Metabolism, Prostate Cancer,

Objective: Lycopene has been reported to have protective and therapeutic effects in prostate cancer, but it is unclear if these effects are due to the parent carotenoid or to lycopene metabolites. Carotene 9',10'-monooxygenase (CMO2) is a putative lycopene cleavage enzyme, possibly producing cell type-specific lycopene metabolites. The range and specificity of CMO2-mediated physiological functions is under active investigation. CMO2 expression is higher in normal prostate tissues than in prostate cancers.

Methods: We (1) determined CMO2 protein and mRNA expression in androgen-sensitive (C4-2) and androgen-resistant (DU-145) human prostate cancer cells and (2) analyzed the accumulation of lycopene and its possible metabolites by HPLC.

Results: After 24h incubation with 1 and 3 µM lycopene, C4-2 and DU-145 cells accumulated 54 and 87 pmol and 45 and 132 pmol lycopene per million cells, respectively. By 48h, lycopene accumulation was reduced in C4-2 cells and increased in DU-145 cells. These findings are inversely related to CMO2 expression in these cell lines and, therefore, may indicate presence of CMO2-dependent lycopene metabolism. After 72h of lycopene incubation, several lycopene isomers and polar (lycopenoids?) compounds were identified in C4-2 cells.

Conclusion: Our data suggest that lycopene uptake, metabolism and accumulation by prostate cancer cells may depend on androgen status and CMO2 expression. We speculate that CMO2 may, in part, suppress prostate cancer growth by generating biologically active lycopene metabolites. Further studies are required to characterize unambiguous lycopenoids and their functions.

Research supported by: NIH HD42174, Muma Family Endowment
Abstract #: B-31  

Presented by: Abhishek Mathur, MD, Resident

**CT Measurement of Pancreatic Steatosis and Visceral Fat; Markers for Lethality of Pancreatic Adenocarcinoma**  
Abhishek Mathur MD, Jonathan Hernandez MD, Fawad Shaheen MD, Miloni Shroff MD, Sujat Dahal MD, Connor Morton BS, Thomas Farrior BS, Raj Kedar MD, Alexander Rosemurgy MD  
Department of Surgery, College of Medicine University of South Florida

**Keywords:** pancreatic steatosis, visceral fat, obesity, pancreatic adenocarcinoma

**Objective:** Pancreatic steatosis promotes lymphatic metastases and decreased survival for patients with pancreatic adenocarcinoma after pancreaticoduodenectomy (PD). However, a correlation between preoperative adipose measurements by CT scanning and patient outcomes remains ill defined. We aim to determine the utility of preoperative CT measurements of pancreatic steatosis and visceral fat as prognostic indicators for patients with pancreatic adenocarcinoma.

**Methods:** CT scans of 42 patients undergoing PD for pancreatic adenocarcinoma were reviewed. CT attenuation of the pancreas, liver, and spleen were measured in Hounsfield units. Perirenal adipose tissue was measured in mm. Pathology slides were reviewed for tumor differentiation and invasion. Data are presented as mean ± SD.

**Results:** Lymphatic metastases were absent (N0) in 43% and present (N1) in 57% of patients. Age, gender, tumor size, and margin status were similar between patients with and without nodal metastases. Outcomes for patients stratified by nodal status and perirenal adiposity are depicted below.

<table>
<thead>
<tr>
<th>Nodal Status &amp; Fat Pad Depth</th>
<th>Perirenal Adiposity (mm)</th>
<th>Pancreatic Body (HU)</th>
<th>Liver (HU)</th>
<th>Peripancreatic Fat Invasion (%)</th>
<th>Survival (months)</th>
</tr>
</thead>
<tbody>
<tr>
<td>N0</td>
<td>13±2</td>
<td>35±4</td>
<td>58±4</td>
<td>53</td>
<td>21±4</td>
</tr>
<tr>
<td>N1</td>
<td>18±1*</td>
<td>23±2*</td>
<td>50±2*</td>
<td>90*</td>
<td>11±2*</td>
</tr>
<tr>
<td>N1 &amp; &lt;10mm</td>
<td>9±1</td>
<td>27±2</td>
<td>53±4</td>
<td>80</td>
<td>16±2</td>
</tr>
<tr>
<td>N1 &amp; &gt;10mm</td>
<td>21±1†</td>
<td>25±2</td>
<td>48±2</td>
<td>91</td>
<td>7±1†</td>
</tr>
</tbody>
</table>

**Conclusion:** With resected pancreatic adenocarcinoma, increased pancreatic and liver steatosis, as well as increased visceral fat stores are associated with lymphatic metastases. Increased visceral fat is associated with an abbreviated survival for patients with lymphatic metastases. Therefore, we conclude that CT measurements of visceral fat predict the dissemination and lethality of pancreatic adenocarcinoma.

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Abstract #: B-32  

Presented by: Nishit Patel, MS, Med IV Student

**Folliculocentric Basaloid Proliferation: A Potential Pitfall for Mohs Surgery**  
Nishit Patel MS, Ronald Johnston MD, Brooke Baldwin MD, Jane Messina MD, Basil Cherpelis MD  
Department of Dermatology, University of South Florida

**Keywords:** Basal cell carcinoma, Folliculocentric basaloid proliferation, Mohs, BCC, FBP

**Objective:** The objective of this investigation is to aid in recognition of an unusual basaloid growth known as folliculocentric basaloid proliferation (FBP) that is commonly mistaken for a basal cell carcinoma (BCC) during Mohs surgical excisions. Awareness of this benign entity, and the ability to differentiate it from a BCC by the Mohs surgeon, is critical to avoid unnecessary tissue excision for margin control.

**Methods:** The case of a patient that underwent Mohs excision of a biopsy-proven BCC, which was terminated after several stages showed an unending basaloid growth, is reviewed. This unusual entity is most consistent with the diagnosis of FBP. An overview is given of the proposed histological criteria for FBP and how it differs from the criteria for BCC.

**Results:** FBP is characterized by a multifocal, multi-shape basaloid proliferation that involves the follicular epithelium. It frequently appears in aggregates with a smooth or uneven outline with fairly uniform individual cells and with possible peripheral palisading. FBP maintains a vertical orientation, is folliculocentric, and has normal surrounding stroma.

**Conclusion:** Both BCC and FBP are often difficult to differentiate, as both are multifocal-appearing basaloid growths with aggregates of varying shapes, sizes and borders with peripheral palisading. However, FBP’s vertical arrangement is in contrast to the horizontal layout of BCC. Secondly, the surrounding stroma is normal in FBP versus the myxoid stroma seen with BCC. Third, FBP has an axial distribution and is centered on the follicle with elongated epithelial cord attachments creating a “Head of Medusa” configuration. Finally, FBP has a prominent hyaline basement membrane versus the threadlike one seen in BCC.
Abstract #: B-33  Presented by: Chetna Purohit, MD, Postdoc

**FNA Biopsy of an Osteoclast-rich Undifferentiated Urothelial Carcinoma- A Cytology Case Report**

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3. Department of Pathology, University of South Florida, Tampa, FL  
4. Department of Cell Biology, University of South Florida, Tampa, FL

**Keywords:** 1) Osteoclast-rich, 2) giant cell tumor, 3) undifferentiated urothelial carcinoma, 4) urinary bladder and 5) fine needle aspiration biopsy

**Objective:** We report a first cytology case of metastatic osteoclast-rich undifferentiated carcinoma of urinary bladder (ORUCUB), diagnosed by fine needle aspiration biopsy (FNAB). ORUCUB is an extremely rare variant of high-grade urothelial carcinoma (UC) and has an aggressive behavior and poor outcome. Recognizing its cytomorphological features will be helpful in an accurate diagnosis. The differential diagnoses include giant cell carcinoma, foreign body/granulomatous reaction, trophoblastic carcinoma, sarcomatoid carcinoma and giant cell tumor of bone.

**Methods:** A 74 year-old male with a history of high-grade UC and prostatic cancer came to GU clinic with painful left groin lump for 2 weeks. FNA biopsy of the groin mass was performed by the cytopathologist.

**Results:** The specimen was hypercellular and consisted of two distinct cell populations: predominantly smaller, highly pleomorphic, dyscohesive, spindle-ovoid to polygonal mononuclear cells in the background of abundant large benign appearing multinucleated giant cells (MGC). The mononuclear cells were malignant and immunohistochemically positive for vimentin, cytokeratins, Ki-67, and p53. MGCs were scattered and were morphologically similar to osteoclasts and were positive for CD-68, vimentin. A final diagnosis of metastatic ORUCUB was rendered after comparing with previous surgical resected specimen.

**Conclusion:** ORUCUB is a rare but specific diagnosis that can be recognized by cytology. This study demonstrated the cytological, histological and immunohistochemical features of ORUCUB.

Abstract #: B-34  Presented by: Jacob Scott, MD, Resident

**Prognostic Factors for Glioblastoma (GBM) in Individuals 70 Years and Older: A Study of 446 Patients from Two Tertiary Centers.**

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**Keywords:** Glioblastoma, Elderly, RPA

**Objective:** To develop a prognostic algorithm for patients over the age of 70 years diagnosed with Glioblastoma.

**Methods:** The MSKCC and CCF institutional databases were used to identify patients 70 years or older at diagnosis with GBM. Demographic variables, performance status, clinical presentation, tumor characteristics and extent of therapy were evaluated as potential prognostic factors. Multivariate analyses were performed using Cox model to identify variables independently predictive of survival outcomes in a stepwise fashion. RPA sequentially divided patients into homogenous groups in terms of prognosis in a stepwise fashion; standard tree pruning methods were then applied.

**Results:** 446 GBM patients with a median age of 75 years (range, 70 to 99 years) were included. The median OS was 5.7 months (95% CI, 5.3 to 6.4). 36% underwent biopsy only while 43% had partial resection and 21% gross total resection; 72% received RT and 34% upfront adjutant chemotherapy. In multivariate analysis, older age (HR=1.06 per 1-year increase, 95% CI: 1.03-1.08, P<0.0001), KPS ≤ 60 (HR=1.71, 95% CI: 1.37-2.13, P<0.0001), mental status changes at presentation (HR=1.28, 95% CI:1.04-1.57, P=0.02), multifocal disease (HR=1.37, 95% CI:1.00-1.88, P=0.05) and corpus callosum location (HR=1.86, 95% CI:1.19-2.90, P=0.006) were associated with increased death risk while partial resection (HR=0.46, 95%CI:0.37-0.59, P<0.0001), GTR (HR=0.33, 95% CI:0.24-0.45, P<0.0001) and radiation to 50 Gy or higher (n=205, HR=0.68, 95%CI:0.51-0.92, P=0.01) were associated with improved survival. A RPA model will be presented.

**Conclusion:** Elderly patients are often left out of similar prognostic studies but advancing age, KPS and extent of therapy are important prognostic factors in this population.
Modulation of Nicotinamide Phosphoribosyltransferase Expression in Colorectal Carcinogenesis

RE Shackelford¹, A Hakam¹ and D Coppola¹. ¹Pathology, H. Lee Moffitt Cancer Center & Research Institute, Tampa, Florida, United States

Keywords: Nicotinamide phosphoribosyltransferase, Colon Cancer, NAD+

Objective: NAD+ is required for cell division, angiogenesis, survival, and DNA repair. Nicotinamide phosphoribosyltransferase (Nampt) catalyses NAD+ synthesis, regulating cellular NAD+ levels. Nampt expression also increases the expression of genes promoting cell survival. To date, there are no published studies on Nampt expression in colorectal cancer progression. We examined Nampt protein expression in benign colonic epithelium compared to colorectal adenomas and carcinomas, using tissue microarray.

Methods: We determined Nampt protein expression in 123 human colorectal adenocarcinomas (CRC), 23 adenomas (AD), and 27 normal colonic mucosa samples (NR) taken adjacent to the colorectal cancer. Formalin-fixed paraffin embedded core section in a tissue array were immunostained with Nampt murine anti-pan-visfatin monoclonal antibody (AdipoGen, Incheon, South Korea, at 1/1000 dilution), using the avidin-biotin-peroxidase method. A semiquantitative measure of Nampt protein expression was determined as the product of immunostain intensity and percent of cells stained, with both scored on a 0-3 scale, with 3 being maximal.

Results: Nampt protein expression was moderate (IHV scores 4 or less) in NR. Nampt expression increased significantly in AD (IHC between 6 and 9). No significant difference was detected between AD and CRC.

Conclusion: Nampt protein expression is increased in colon adenomas and carcinomas compared to benign epithelium, with the greatest difference seen between benign epithelium and adenomas. Thus increased Nampt expression likely plays a role in the early stages of colorectal carcinogenesis. The use of Nampt inhibitors, now in clinical trial, may prove of benefit for colorectal cancer patients.

Research supported by: The Moffit Cancer Center Department of Pathology

Bystander Vaccine Therapy in Mantle Cell Lymphoma (MCL): Phase II Clinical Results

Shah B.D., Tao J., Sokol L., Chervenick P., Tomblyn M., Pinilla-Ibarz J., Moscinski L., Antonia S., Sotomayor E.M. and Dessureault S. **Affiliation for all authors: H Lee Moffitt Cancer Center, Dept. of Oncologic Sciences, College of Medicine, University of South Florida

Keywords: Mantle Cell Lymphoma, Vaccination

Objective: A Phase I study showed that vaccination of cancer patients with irradiated autologous tumor cells + GM.CD40L bystander cells (engineered to secrete GM-CSF and express CD40L) is safe, recruits/activates dendritic cells, and elicits tumor-specific T cell responses. We tested this strategy in patients with MCL, an aggressive and incurable B-cell malignancy.

Methods: After lymph node resection (for tumor harvest), 4-6 cycles of chemotherapy, and restaging, patients with usable vaccine who obtained PR or CR lasting 1 month were vaccinated x4 at 28d intervals with IL-2 (0.5 x 106 U SC BID x 14d). Patients were monitored for toxicity, tumor response, tumor-specific immune responses, and PFS/OS.

Results: 43 were enrolled (21 relapsed MCL, 20 with int/high MIPI, and 6 with blastoid MCL). 20 were not vaccinated (2 withdrew, 7 failed tumor harvest, 1 progressed too fast to harvest, 10 progressed during chemotherapy). The unvaccinated were older (68.4y vs 62.8y; p=.026) but otherwise did not differ significantly by stage, LDH, MIPI, relapse status, or number of prior regimens. Among 23 treated, 10 had relapsed disease (median 3.6 prior regimens), 10 had an int/high MIPI, and 2 had blastoid MCL. Pre-vaccination response after chemotherapy included 7 CR and 16 PR. At 6 months after vaccination, 2 pts in PR converted to CR, 10 progressed (including 3 who progressed after only 1-2 vaccines), and 11 had no change. Patients receiving at least one vaccine were analyzed. This high risk cohort demonstrated median PFS of 9 mo, but an unexpectedly prolonged median OS (not reached, median follow-up 30 mo, range 8-68 mo).

Conclusion: Bystander vaccination may abrogate the aggressiveness of MCL leading to prolonged survival.

Research supported by: The Lymphoma Research Symposium, Novartis
**Rapid Frozen Section Immunostaining of Melanocytes by Microphthalmia-Associated Transcription Factor**

**Abstract #:** B-37  
**Presented by:** Donald Stranahan, MD, Resident

**Keywords:** Microphthalmia-associated transcription factor, immunostaining, melanocytes, Mohs

**Objective:** To present a novel 35 minute protocol using MITF immunohistochemical (IHC) staining for identifying melanocytes in frozen tissue for its potential use in Mohs Micrographic Surgery (MMS) for the removal of melanoma in situ (MIS).

**Methods:** Frozen sections of chronically sun damaged skin (CSDS) obtained from surplus tissue from 11 patients undergoing MMS for basal cell or squamous cell carcinomas of the head and neck were stained with MITF IHC on both frozen and FFPE tissue specimens. Melanocyte nuclear diameter and density were then compared between frozen and formalin fixed paraffin embedded (FFPE) sections stained with MITF. Comparison of nuclear diameter between MITF and melanoma antigen recognized by T-cells (MART-1) stained permanent and frozen sections was also performed using a previous dataset of MART-1 stained frozen and FFPE sections.

**Results:** We obtained equivalent measurements of melanocyte density and diameter in frozen sections compared to permanent sections using MITF, and similar nuclear diameter values to those with MART-1. Density values, however, were lower for MITF than with MART-1.

**Conclusion:** MITF, a nuclear stain, avoids the background staining of highly dendritic melanocytes by MART-1, and facilitates identification of melanocytes in frozen section. Our results suggest that MITF IHC staining may be useful in MMS for MIS on CSDS.

**Research supported by:** University of South Florida Department of Dermatology and Cutaneous Surgery

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**Role of Rb Family in Vitamin D Receptor-Mediated Anti-Tumor Effects in Ovarian Cancer Cells**

**Abstract #:** B-38  
**Presented by:** Jinfu Tang, PhD, Postdoc

**Keywords:** Vitamin D, Vitamin D receptor, Rb family, Ovarian cancer

**Objective:** The purpose of current study is to investigate the molecular mechanism underlying the resistance of human ovarian tumors to VD induced growth suppression and lay the groundwork for the application of VD-based hormonal therapy to the clinical management of OCa.

**Methods:** Multiple human ovarian cancer cell lines and mouse embryonic fibroblasts (MEF) in which one or all three of the Rb family was knocked out were chosen as experimental model systems to assess the role of Rb family in vitamin D action. Various techniques such as MTT assay, Western Blot, immuno-staining, co-immunoprecipitation and so on were used in this study.

**Results:** We found that about half of OCa cells are resistant to 1,25VD induced growth suppression. The transcriptional activity of the VDR is suppressed in VD-resistant OCa cells as comparison to sensitive ones. The loss of Rb function or expression in OCa cells contributes to VD resistance, which is associated with decreased VDR levels. Consistently, the level of VDR expression and vitamin D response was found to be decreased by shRNA of Rb family in OVCAR3 cells in Rb family triple knock-out MEFs. Further, co-immunoprecipitation analyses revealed that the VDR protein forms a complex with all three members of the Rb family and the complex formation was enhanced by VD treatment.

**Conclusion:** The function of Rb family is essential for VDR-mediated anti-tumor effects. Drugs that restores the functional status of Rb proteins can be used in combination with active vitamin D for OCa prevention and treatment.

**Research supported by:** R01 grant from NCI and a team science project (TSP) from the Florida Department of Health
**Abstract #**: B-39  
**Presented by**: Awet Tecleab, BS, Graduate Student

**Differential Requirement of RalA and RalB Proteins on Survivin Expression**

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Department of Molecular Medicine, College of Medicine, University of South Florida; Department of Drug Discovery, H. Lee Moffitt Cancer Center and Research Institute, Tampa, Florida.

**Keywords**: Survivin, Ral, Ras

**Objective**: Survivin is a member of the inhibitors of apoptosis proteins (IAP) that play an important role in cell cycle and apoptosis. It is highly expressed in cancer and associated with oncogenesis, chemoresistance and blocking of apoptosis. How survivin is re-expressed in cancer cells remains largely unknown, but loss of tumor suppressor genes and gain of oncogenes have been implicated. Constitutive active Ras (CA-H-Ras) has been shown to up-regulate survivin expression. However, effector molecules downstream of Ras that are involved in this regulation remain elusive. The RalGDS/Ral pathway is one of the canonical effector pathways that have not yet been studied for its possible role in the modulation of survivin.

**Methods**: We over expressed CA-Ras and CA-Ral in HEK293 and in T80 cells (a genetically defined cell line model) to determine the role of Ras and Ral on survivin expression and we used siRNA to RalA, RalB and K-Ras to determine if these genes are required for survivin expression in cancer cells.

**Results**: Transient over expression of Ral proteins in HEK293 and in T80 cells induces survivin expression, but only knockdown of RalA not RalB reduces survivin expression in cancer cells with K-Ras mutation. This suggests, Ras may induce survivin expression through RalA, which was previously shown to mediate Ras induced oncogenesis. Although knockdown of RalB induces apoptosis in panc-1 cells, it is not accompanied by a reduction in survivin expression.

**Conclusion**: Despite their sequence homology, RalA and RalB have been shown to have different function in cancer. Similarly our findings show that the two proteins also differ in regulating survivin expression.

**Research supported by**: NIH, grant number CA67771

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**Abstract #**: B-40  
**Presented by**: Anfernee kai-wing Tse, PhD, Postdoc

**Characterization of a New Natural Compound Evodiamine as a Selective Estrogen Receptor Modulator**

Anfernee kai-wing Tse, Jinfu Tang, Ming Hu, Xiaohong Zhang, Santo V. Nicosia, Wenlong Bai, Department of Pathology and Cell Biology, College of Medicine, USF, Tampa, FL

**Keywords**: Breast cancer, Evodiamine, natural compound, SERM, anti-estrogen

**Objective**: The purpose of current study is to investigate the molecular and pharmacological anti-estrogenic properties of evodiamine, a natural compound from Evodia fruits (Evodia rutaecarpa), including its ability to bind to estrogen receptors, its impact on breast and uterine endometrium cancer cell growth, and its inhibitory effects on the estrogen target genes, compared with other antiestrogenic agents such as tamoxifen and fulvestrant.

**Methods**: The antitumor activity of evodiamine was evaluated in estradiol-sensitive MCF-7 breast and Ishikawa endometrium cancer cells by MTT assay. Its ability to bind ERα was assessed by in vitro hormone binding assays. The antagonistic activity was compared in the ER-responsive reporter gene assays in MCF-7 cells or ER-negative Hela cells transfected with ERα or β. The effects of evodiamine on the ER level, stability, cytoplasmic-nuclear-nuclear matrix translocation and the expression of estrogen target genes were examined by Western blot analysis.

**Results**: Our results showed that evodiamine blocked the 17β-estradiol (E2)-induced human breast and endometrial cancer cell growth. Evodiamine also inhibited the E2-induced ERE-luciferase reporter gene activity and estrogen target genes such as c-myc and cyclin D1 in MCF-7 cells. We also found that evodiamine bound to and induced nuclear matrix immobilization and degradation of ERα and regulated ERα expression by decreasing its protein stability, which indicates that evodiamine can inhibit ERα signaling through competing ERα binding with E2 and causing the ERα degradation.

**Conclusion**: We conclude from this data that evodiamine is a selective ER modulator that inhibits estrogen signaling in human cancer cells through the reduction of E2-ER binding and the degradation of ERα.

**Research supported by**: RO1,TSP
Abstract #: B-41  Presented by: Xiaoqin Wang, MD, PhD, Postdoc

**Natriuretic Peptide Receptor A Regulates Macrophage Inhibitory Factor Expression in Prostate Cancer**
Xiaoqin Wang, Hongyu Zheng, Murali Kanakenahalli, Ronil Patel, Kajel Patel, Vikas Sharma and Subhra Mohapatra; Department of Molecular Medicine, College of Medicine, University of South Florida

**Objective:** We have recently demonstrated that mice deficient in atrial natriuretic peptide receptor A (NPRA-KO) cannot support the growth of implanted prostate tumor cells and downregulation of NPRA expression by siNPRA or NPRA inhibitor induced apoptosis in PCa cells and reduced tumor burden in mice. However, the precise mechanism of NPRA action in PCa remains unclear. Macrophage migration inhibitory factor (MIF), a proinflammatory cytokine, is overexpressed in prostate cancer and unique for its functions in many processes associated with tumor survival. In this study, we investigated the effect and associated mechanisms of NPRA and MIF pathways in a transgenic adenocarcinoma of mouse prostate (TRAMP) model.

**Methods:** In vivo expression of NPRA and MIF was checked by RT-PCR, Western blot and ELISA assay in prostate tissues of TRAMP mice as well as TR-C1 cell transplanted C57/BL6 mice. The role of NPRA deficiency in modulating MIF signaling was examined in PCa cell lines derived from TRAMP mouse prostate (TRAMP-C1) and in TRAMP-C1 xenograft treated with NPRA inhibitor.

**Results:** NPRA expression co-related with MIF expression in TRAMP mice during PCa progression. Downregulation of NPRA expression by siNPRA significantly reduced MIF expression. Moreover, treatment of TRAMP-C1 xenografts with NPRA inhibitor also reduced MIF expression.

**Conclusion:** Our results suggest that NPRA is an upstream regulator of MIF signaling during PCa progression, and NPRA promotes PCa development by modulating MIF pathway.

**Research supported by:** NIH and Bankhead Coley Cancer Research Program

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Abstract #: B-42  Presented by: Shengyan Xiang, PhD, Postdoc

**Histone Deacetylase 6 Promotes p53 Ubiquitination and Degradation**
Shengyan Xiang, Mu Zhang, Huiqin Dong, Santo V. Nicosia, Wenlong Bai and Xiaohong Zhang; Department of Pathology and Cell Biology, College of Medicine, University of South Florida

**Keywords:** p53, histone deacetylase 6, ubiquitination, protein degradation

**Objective:** Histone deacetylase 6 (HDAC6) is a unique HDAC which contains two functional HDAC domains and one ubiquitin binding domain. Our recent results show that knockdown of HDAC6 in a lung cancer cell line, A549, causes the accumulation of p53, a key tumor suppressor, compared to the control cells. Therefore, we propose that HDAC6 may promote p53 ubiquitination and degradation.

**Methods:** shRNA against HDAC6 was used to stably knockdown HDAC6 in lung cancer A549 cell line. Western blot was used to detect protein expression level. Immunoprecipitation was used to detect mono-ubiquitinated and polyubiquitinated p53. Cycloheximide blocking method was used to determine p53 half-life.

**Results:** HDAC6 was stably knockdown in lung cancer A549 cell line. Total p53 level is dramatically increased in knockdown stable pool than in control stable pool. Simultaneously, CDC25c, a downstream gene of p53, is upregulated in HDAC6 knockdown stable pool. The half-life of p53 is significantly increased when HDAC6 is knockdown. Immunoprecipitation data of A549 stable pools show that p53 is highly monoubiquitinated and polyubiquitinated in control stable clones. Furthermore, phosphor-Ser15 of p53 accumulates more in HDAC6 knockdown pool under the cisplatin, UVC and gamma-radiation treatments. So HDAC6 may promote ubiquitination of p53 and lead to p53 degradation.

**Conclusion:** Total p53 level is increased when HDAC6 is knockdown in lung cancer A549 cell line. Our results indicate that HDAC6 can promote p53 monoubiquitination and polyubiquitination and degradation.

**Research supported by:** Bankhead-Coley Florida Biomedical Research Program, James & Esther King Florida Biomedical Research Program and Moffitt Lung Cancer SPORE Career Development Grant.
Role of Histone Deacetylase 6 in Chemotherapeutic Response Through DNAMismatch Repair Proteins
Mu Zhang, Shengyan Xiang, Huiqin Dong, Santo V. Nicosia, Wenlong Bai and Xiaohong Zhang
Departments of Pathology and Cell Biology, College of Medicine, University of South Florida, Tampa, FL
Keywords: Histone deacetylase 6 (HDAC6), Mismatch protein 2/6 (MSH2/6), 6-TG, ubiquitination
Objective: Previous data from our laboratory have shown that HDAC6, a cytoplasmic deacetylase, contributes to cell survival under stress conditions such as drug treatment and low temperature. To gain further insight into this phenomenon, we co-purified HDAC6 and its partners MSH2/6 (MutSα), two DNA mismatch repair proteins. Here, we propose that HDAC6 promotes ubiquitination of MSH2/6 and weakens chemosensitivity in tumor cells.
Methods: Affinity purification was used to isolate the binding proteins of HDAC6. G418 selection was used to establish stable cell lines. Immunoprecipitation and Western were used to determine total and modified proteins. Ubiquitination assays were used to detect ubiquitinated MSH2 and MSH6. 6-TG and cisplatin were the drugs for treatment.
Results: HDAC6 co-purified with MSH2/6 as a complex in Hela cells’ nuclear extract, and DAC1 domain of HDAC6 bound to the N-terminals of both MSH2 and MSH6; Accumulation of MSH2/6 in knockout or knockdown HDAC6 cells induced apoptosis after treatment with 6-TG or cisplatin and such chemosensitivity was abrogated by restoring MSH2; HDAC6 enhanced ubiquitination and degradation of MSH2/6 both in vivo and in vitro through the proteasome pathway; Results from stable cell lines showed that both DAC and UBP domains of HDAC6 are necessary for ubiquitination of MSH2/6.
Conclusion: Inhibition of HDAC6 prolongs MSH2/6 stability through the ubiquitin-proteasome pathway and increase in mismatch proteins triggers apoptotic signaling. These data suggest that inhibitors of HDAC6 may provide new therapeutic tools in chemotherapy-refractory cancer patients.
Research supported by: Bankhead-Coley Florida Biomedical Research Program, James & Esther King Florida Biomedical Research Program and Moffitt Lung Cancer SPORE Career Development Grant.

Short Term Prognostic Value of B-Type Natriuretic Peptide in Comparison to Troponin Following Percutaneous Coronary Interventions
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Keywords: B-type natriuretic peptide; Percutaneous coronary intervention; Troponin
Objective: Elevated B-type natriuretic peptide (BNP) level has both diagnostic & prognostic values in heart failure and acute coronary syndrome. We aimed to evaluate the prognostic value of post-PCI BNP levels in predicting 1 month MACE (death, myocardial infarction, repeat revascularization) and its correlation with any concurrent troponin elevation.
Methods: We retrospectively analyzed patients who had BNP levels before and 24 hrs after PCI between July 2007 and June 2008. Information regarding pre and post procedural troponin (TnI) levels and 30 day follow up in terms of MACE were available.
Results: Among 31 patients with mean age of 62+/-8 yrs and mean LVEF of 53+/-12%, all were males with 45% diabetics, 81% hypertensives, and 61% with prior coronary artery disease. 10% underwent multivessel PCI; 19% received bare metal stents; 81% received drug eluting stents; 38% received LAD intervention; 22% had left circumflex intervention, 32% had RCA interventions, and 10% had vein graft interventions. Seventeen patients had troponin elevation while 6 patients had BNP elevation 24 hrs post-PCI. We saw a trend in the occurrence of BNP elevation among patients who also had troponin elevation (R=0.35; P=0.065). During 30 day follow up, 3 patients had NSTEMI. All the 3 patients had shown post-PCI TnI elevation while only 1 had shown BNP elevation. Post-PCI TnI or BNP elevation did not predict 30 day MACE in our patients (P=1.00).
Conclusion: We found no significant correlation between post-PCI BNP and troponin elevations. Besides, post-PCI TnI or BNP elevations did not predict 30 day MACE.
**AFFIRM Update**

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**Keywords:** AFFIRM Update

**Objective:** It is known that obesity is associated with new onset of atrial fibrillation. However, effect of obesity on recurrence of atrial fibrillation is not studied.

**Methods:** We used limited access dataset from the AFFIRM trial, provided by NHLBI. Using number of cardioversion, both electrical and pharmacological, and number of visits patient currently in atrial fibrillation or flutter as surrogate marker for recurrence of atrial fibrillation, and BMI as marker of obesity we studied relationship of obesity with recurrence of atrial fibrillation.

**Results:** From 4060 patients at the baseline, 1542 were excluded as they did not have baseline BMI information. From remaining 2518 patients, 1255 were in rate control arm and 1263 were in rhythm control arm. For BMI in relation to number of cardioversions these 2518 patients had 22753 visits. Using linear mixed model higher BMI had odds ratio (OR) of 1.017 [95% CI 1.005-1.029, p 0.006] requiring cardioversion. If BMI increase by 5 units OR is 1.088 [95% CI 1.024-1.155, p 0.006], and by 10 units OR is 1.183 [95% CI 1.049-1.334, p 0.006] of requiring cardioversion. For BMI in relation to patient being in persistent atrial fibrillation or flutter these 2518 patients had 22374 records (379 records missing by not having data). Using linear mixed model OR of having patient in persistent atrial fibrillation or flutter with higher BMI was 1.020 [95% CI 1.002-1.038, p 0.0283]. If BMI increase by 5 units OR is 1.104 [95% CI 1.011-1.205, p 0.0283] and by 10 units OR is 1.218 [95% CI 1.021-1.452, p 0.0283] of patient being in persistent atrial fibrillation or flutter.

**Conclusion:** Obesity is strongly associated with high incidence of recurrence of atrial fibrillation.

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**Intracardiac Tumors At Tampa General Hospital: A Five Year Retrospective Analysis**

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**Keywords:** incidence, etiology, intracardiac masses, tumors, myxomas,

**Objective:** 1) Determine the incidence and etiology of cardiac tumors at Tampa General Hospital 2) Determine the prevalence of various cardiac tumors among different age groups 3) Compare the incidence of various intracardiac tumors at TGH to well published data from the Mayo Clinic

**Methods:** Using a word search program we reviewed data from 2003-2008 on patients with cardiac tumors diagnosed with confirmation by pathologic specimens. Cardiac tumor diagnoses were reviewed and subdivided into the following categories: benign, malignant, primary, and metastatic. We then calculated the incidence of these cardiac tumors, median age and proportion of female versus male patients.

**Results:** Our results showed the most common intracardiac masses confirmed by histological specimen were lipomas (31.5%), followed by thymomas (25.9%), lymphomas (13.0%) and myxomas (7.4%). The mean ages for the four most common intracardiac masses were 53.2, 49.3, 34.1 and 58.7 years old respectively. There were significantly more female patients diagnosed with lipomas than males. There was no statistically demonstrable predilection of one sex over another for the other intracardiac tumors.

**Conclusion:** The data published from the Mayo Clinic reports myxomas as the most commonly excised benign mediastinal mass. In contrast, our data showed lipomas to be the most commonly excised mediastinal mass. There were no significant differences between the mean age of Tampa General Hospital patients when compared to those reported by the Mayo Clinic. The difference in incidences of the various cardiac and mediastinal masses may represent discrepancies in referral patterns as the Mayo Clinic is a tertiary referral center for intracardiac masses.
Can Heart Failure Medications Prevent Trastuzumab-Induced Cardiotoxicity? Christopher C. Reynolds, MD; Gregory Hartlage, MD; Ren Chen, MD, MPH; Vinod Patel, MD; and Maya Guglin, MD, PhD. Dept. of Cardiology, College of Medicine, University of South Florida, Tampa, FL; Emory University, Atlanta, GA, and H. Lee Moffitt Cancer Center, Tampa, FL.

Keywords: Heart Failure Chemotherapy Cardiotoxicity Breast Cancer

Objective: To determine if concomitant heart failure (HF) medications reduce the risk of trastuzumab-induced CTX.

Methods: We retrospectively studied the charts of 156 women with breast cancer who received trastuzumab for metastatic or adjuvant therapy. Ejection fraction (EF) was evaluated at baseline and then every 3 months. CTX was defined as a decrease of EF by 10%, or to <50%, or symptoms of HF. Relative risk was calculated for CTX in adjuvant patients with cardiovascular comorbidities and on concomitant HF medications.

Results: CTX occurred in 33.3% of patients. Presence of one or more cardiac comorbidities was related to an increase of CTX (RR=1.85, p=0.023). Symptomatic HF was associated with the presence of comorbidities (RR=5.00; p=0.007), in particular hypertension (RR=3.5, p=0.025). However, being on concomitant HF medications (ACE-Inhibitors Beta-Blockers, Statins) was associated with less decrease in EF (RR=0.69; p=0.092) and HF symptoms (RR=0.38; p=0.086). In the presence of hypertension, these medications reduced CTX (RR=0.35; p=0.063) and HF symptoms (RR=0.13; p=0.003).

Conclusion: Concomitant HF medications are associated with a decreased risk of trastuzumab-induced CTX in patients with breast cancer and cardiac comorbidities. Their potential to prevent or alleviate CTX needs further investigation.

Research supported by: USF Department of Cardiology

Treatment of Type II Endoleaks Through a Transarterial Technique with Onyx

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Keywords: Type II endoleaks, Onyx, ethylene-vinyl-alcohol copolymer, EVAR

Objective: Single center experience in treating 26 type II endoleaks (T2EL) post endovascular repair of abdominal aortic aneurysms via a transarterial approach using ethylene-vinyl-alcohol copolymer (Onyx).

Methods: Retrospective review revealed that we repaired 44 T2EL in 41 patients over 3 years with Onyx. 38 (93%) were male and 3 (7%) were female. Mean age was 79 years (R=65-93 years). Patients met one of the following criteria for treatment: sac growth > 5 mm in 6 months, aneurysm sac > 6 cm, change in aneurysm morphology or position of stent graft. 18 (41%) of the endoleaks were embolized through a translumbar (TL) approach, and 26 (59%) through a transarterial (TA) approach.

Results: Of the 26 T2EL embolized with Onyx via a TA approach, complete occlusion of the T2EL was achieved in 19 (73%) of the endoleaks. Of the 22 T2EL embolized via TL access, a left-sided approach was used for 20 (91%), a right-sided approach was used for 2 (9%). Technical success was achieved in all TL cases, defined as the complete embolization of the endoleak nidus with no persistent flow to the aneurysm sac.

Conclusion: The ideal agent for a T2EL repair would completely occlude the endoleak nidus and the origins of all inflow and outflow vessels, and could be delivered in a controlled fashion, limiting the risk of non-target embolization. Onyx is currently the best available agent for the treatment of T2EL. The TA route of embolization is technically feasible using Onyx. The negative features of Onyx are relative high cost, availability at most centers, and the streak artifact that degrades follow-up CT images. Long term follow-up is needed to determine the effectiveness of Onyx embolization of T2EL with regards to the rate of endoleak recurrence and aneurysm size stabilization.

Research supported by: AHA
A Novel Mutation CHS1 (LYST) Mutation: Osteomyelitis in a Child with Chediak-Higashi Syndrome

Presented by: Woei Yeang Eng, MD, Resident

Keywords: Novel Mutation CHS1 Chediak-Higashi Syndrome

Objective: Chediak-Higashi syndrome (CHS) is rare, autosomal recessive immunodeficiency disorder characterized by hypopigmentation, neutropenia and risk for hemophagocytic lymphohistiocytosis (HLH). We report a novel mutation in a child with CHS.

Methods: NK cell and cytotoxic lymphocyte functions, expressions of perforin and granzyme B on CD8, NK and NKT cells, and IL-2R expression as well as complete genetic sequencing of CHS1 were performed.

Results: Our patient with oculocutaneous albinism presented with osteomyelitis at the left lumbosacral junction, and MRSA was positive from blood culture. Peripheral blood smear showed absolute neutropenia with giant abnormal cytoplasmic granules in granulocytes, monocytes and lymphocytes. WBC was 6,780/uL with ANC of 678/uL. Absent NK cell and decreased cytotoxic lymphocyte function at 24% (Ref. >35%) were noted. IL-2R expression was elevated at 8288 units/mL (Ref. <2126). Genetic mutation analysis of all 55 coding exons of the CHS1 gene showed a homozygous mutation in exon 49. It is an insertion of an A which causes the introduction of an early stop codon: c.10883-10884insA, p. Tyr3628X. A novel heterozygous insertion in exon 49 of CHS: c.10883-10884insA, p. Tyr3628X was also found in patient’s mother. Patient later developed HLH from Ebstein-Barr virus infection successfully treated with rituximab and alemtuzumab followed by hematopoietic stem cell transplant (HSCT).

Conclusion: We report a novel homozygous mutation in CHS1 that results in absent NK cell and decreased cytotoxic lymphocyte functions leading to lethal infections and HLH. Early diagnosis and HSCT improved overall survival.

Research supported by: Self-funded.

Propranolol - A Novel Treatment for Orbital Infantile Capillary Hemangiomas

Presented by: Gretta Fridman, MD, Resident

Keywords: propranolol, hemangioma

Objective: To evaluate the treatment of orbital infantile capillary hemangiomas (ICH) with systemic (oral) propranolol.

Methods: This study was a retrospective observational case series of five infants who were diagnosed with orbital ICH between March and July of 2009.

Results: The average age at presentation was 3.1 months (3 weeks to 9 months). The patients received oral propranolol, dosage varying among study centers. The patients were treated for an average of 7.1 months. Treatment of our 5 ICH patients with oral propranolol produced a significant reduction in the size of the hemangioma in 4 (80%) of the patients and a minimal improvement in 1 patient. No patient had significant adverse events during the treatment period.

Conclusion: Oral propranolol may be an effective treatment for orbital ICH.

Research supported by: n/a
**Abstract #:** B-51  
**Presented by:** Jeffrey Gill, MD, Resident

**Safety of Conscious Sedation During Endoscopy for Patients with Obstructive Sleep Apnea**
Jeffrey Gill, MD, University of South Florida, Department of Internal Medicine, James A. Haley VA Hospital, Gitanjali Vidyarthi, MD, James A. Haley VA Hospital, Judy Parow, RN, James A. Haley VA Hospital, Prasad Kulkarni, MD, James A. Haley VA Hospital, William Anderson, MD, James A. Haley VA Hospital, William Boyd, MD, James A. Haley VA Hospital

**Keywords:** Obstructive sleep apnea, endoscopy, conscious sedation, anesthesia, safety

**Objective:** Obstructive sleep apnea (OSA) is a common medical disorder affecting up to 26 percent of adults. Due to the presumed higher risk of cardiopulmonary complications in these patients, many endoscopy centers consider OSA a relative contraindication to using conscious sedation. Our goal was to methodically evaluate the safety of conscious sedation during endoscopy for patients with moderate to severe OSA in a veteran population, and compare this to patients without OSA.

**Methods:** Polysomnography studies were reviewed from 2004 to 2009. Patients were considered if their study was positive for moderate or severe OSA. Endoscopy reports of these selected patients were then reviewed for cardiopulmonary complications. Data recorded included age, sex, BMI, level of OSA, indication and type of endoscopy, sedation used, and complications. A control group of our last 200 endoscopies, excluding those with a diagnosis of OSA, were then reviewed for the same complications.

**Results:** 200 and 200 endoscopy procedures were reviewed of patients with and without OSA, respectively. Of the 200 with OSA, 2 complications were identified. The first was a drug rash after meperidine prior to an upper endoscopy. The second was oxygen desaturation to the 80s during an upper endoscopy, requiring oxygen supplementation. Both procedures were completed and required no extended stay in the endoscopy suite.

**Conclusion:** Our study demonstrated that endoscopy can be done safely in OSA patients using conscious sedation, and that the complication rate is not significantly different than that of a population without OSA. Given these results, a significant amount of resources could be saved if conscious sedation was more broadly utilized during endoscopy in patients with OSA.

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**Abstract #:** B-52  
**Presented by:** Margarita Hernandez, MD, Resident

**Auditory Brainstem Responses in Preterm Infants**  
Margarita Hernandez, USF Health Neonatology Dept, Janet Sullivan Neonatology Department, College of Medicine, University of South Florida

**Keywords:** Auditory Brainstem Responses Neonate

**Objective:** The primary objective is to establish means and standard deviations for auditory brainstem conduction time from 34 to 35 weeks PCA and to determine the impact of gestational age (GA), PCA, gender and multiple gestation on this measure of myelination.

**Methods:** ABRs are performed at 34 and 35 weeks PCA. Subjects are screened for middle ear dysfunction, and wave I latency is measured as an additional index of peripheral hearing status. Informed consent is obtained from the subjects’ legal guardians. Measures: 1) Mean level of OAEs at 7 frequencies; 2) Wave I (8th nerve latency) and 3) I-V interpeak latency (ABCT). Subjects: Preterm infants (28 – 34 wks GA) in TGH NICU. Groups: Subjects are divided by GA and PCA, gender and type of gestation (singleton vs. twin). Inclusion criteria: Viable preterm infants (28 to 34 weeks gestation) from Tampa General Hospital’s NICU. Exclusion criteria: Major malformations, problems with skin integrity, inconsistency in gestational age estimates, significant brain hemorrhage, indirect hyperbilirubinemia requiring exchange transfusion, chronic lung disease, necrotizing enterocolitis and failed OAE screen in both ears.

**Results:** Data were analyzed for 16 infants. 1) Gestational age had a significant effect on ABCT at 34 wks (p<.003) but not at 35 wks PCA; 2) As expected PCA had a significant effect on ABCT (p<.001); 3) ABCTs were longer in males than females at 34 wks but the difference did not reach significance (p<0.07). There were too few males at 35 wks PCA to compare means; 4) Multiple gestation could not be analyzed due to small sample size

**Conclusion:** Gestational age and gender may temporarily affect ABCT in preterm infants. GA differences may disappear as term is approached

**Research supported by:** USF Health Neonatology Department
Abstract #: B-53

Presented by: Barrett McCormick, MS, Med II Student

**Title:** Graft Size May Not Influence Recurvature in SIS Patch Grafting for Peyronie's Disease (research scholarly concentration)

**Objective:** We wished to analyze the significance of graft size in penile recurvature incidence in patients treated surgically for Peyronie's Disease (PD) via plaque excision (PE) and small intestinal submucosa (SIS) grafting.

**Methods:** 120 patients with PD were treated surgically with PE and SIS between Jan 2005 to June 2009. Pre and Post operative functional outcomes were assessed using the SHIM and EPIC questionnaires for sexual function. Patients were all given detailed post operative rehabilitation protocols. Patients were also asked about result satisfaction, willingness to repeat the procedure, incidence of recurvature, and feelings about the surgery with respect to sexual capability, phallic length, and cosmetic effect (much better, better, the same, or worse).

**Results:** 93 patients with a median age of 60 years (33-72) and a followup of 19 months (6-53) completed the analysis. 27 were excluded due to incomplete records. Median SIS graft size was 4 cm (1.5-7) x 4 cm (1.25-6). A subset of 14 patients was found that self-identified as having any type of recurvature post-operatively. Of these patients, the mean age was 60 (55-69) and the mean SIS graft size was 4 cm (2.5-6) x 4.5 cm (2.5-5) with a mean graft size of 4.1 cm x 3.7 cm.

**Conclusion:** Preliminary data suggests that patients self-identifying as having any type of recurvature did not demonstrate SIS graft sizes of significantly differing sizes than patients with no recurvature. Our series may help to suggest other factors may be causative for recurvature incidence, perhaps including compliance with rehabilitation protocols as other preliminary data suggests. Further study is necessary to demonstrate other potential factors in the incidence of recurvature.

Abstract #: B-54

Presented by: Timothy Miller and Joshua Smith, Med II students

**Title:** Redefining the Natural History of Moderate Grade Asymptomatic Carotid Artery Stenosis

**Objective:** Moderate grade carotid artery stenosis (CAS) is defined as a 50-75% reduction in cross-sectional area of the internal carotid artery and its progression rate has not been completely defined. Current classification of carotid stenosis severity and associated stroke risk relies on measurement of diameter reduction caused by obstructing plaque using duplex ultrasonography to measure the flow rate in cm/sec through the internal carotid artery. Other techniques include MRA, CT angiography and/or constrast arteriography. Adverse event risk increases for greater degrees of stenosis severity. Surgical intervention is only recommended for higher degrees of narrowing (> than 60-80%). Current guidelines recommend annual observation of moderate grade stenosis by methods previously mentioned. A large, multivariate analysis may help in defining the natural history of moderate grade carotid artery stenosis.

**Methods:** A 511 patient cohort (aver. age 75.4, SD = 8.7, min=50, max=98, 501m, 10f) was observed for an average 46.1 months. Of these, 210 had bilateral moderate grade internal CAS and 301 had unilateral moderate CAS; producing a total of 721 carotid arteries followed. Adverse neovascular events were determined to be transient ischemic attack (TIA), stroke, progression of stenosis to high-grade (>75% reduction in cross-sectional area), and intervention by surgical carotid endarterectomy or percutaneous carotid stenting. All cause mortality rate was also assessed.

**Results:** 29 transient ischemic attacks occurred in the 511 patients during follow-up, giving an annual progression rate of 1.5%. 14 patients experienced a stroke without experiencing a TIA, giving an annual progression rate of 0.71%. 51 patients' carotid arteries progressed to either occlusion or to greater than 75% stenosis, giving an annual progression rate of 2.6%. 39 patients required intervention by either carotid endarterectomy (28) or percutaneous carotid stenting (11), for an annual progression rate of 2.0%. Total number of patients experiencing any of the adverse events during follow-up was 90, giving an overall annual progression rate to any event of 4.5%. The all-cause annual death rate was 4.9%. A multivariate analysis of various prognostic factors revealed that coinciding congestive heart failure in patients with moderate grade carotid artery stenosis was the only meaningful risk factor for progression to any of the adverse events or death (Relative Risk = 1.98, 95% CI = 1.30-3.00, p<0.0001).

**Conclusion:** The natural history of moderate grade CAS is fairly benign. Results show an overall annual progression rate of 4.5%, with a large number of these events accounted for solely by progression of stenosis to occlusion or greater than 75% stenosis. TIA or stroke incidence was very low (2.21%/year). All-cause mortality rate exceeded the adverse event rate, even when including benign events such as progression of stenosis to greater than 75%. We conclude annual follow-up of moderate grade CAS using duplex ultrasonography is adequate to monitor the progression of stenosis.

**Research supported by:** AHA Medical student summer research fellowship, and rSC
**Abstract #**: B-55  
**Presented by**: Mai Otsuka, MS, Med II Student

**Daptomycin Therapy for Vancomycin-Resistant Enterococcus Bacteremia in Neutropenic Patients** Mai Otsuka, MS (COM, USF), Mohamed A. Kharfan-Dabaja, MD Dept. of Blood and Marrow Transplantation; Bone Marrow Transplant Fellowship Program, Moffitt Cancer Center and Research Institute; Dept. of Oncological Sciences, COM, USF), Gene A. Wetzstein, PharmD, BCOP (Clinical Services; Moffitt Cancer Center & Research Institute), Rod E. Quilitz, PharmD, BCOP (Infectious Disease, H Lee Moffitt Cancer Center), Ramon L. Sandin, M.D., M.S., F.C.A.P. (Clinical Microbiology and Virology Laboratories, Moffitt Cancer Center and Research Institute; Dept. of Pathology and Oncological Sciences, COM, USF), John N. Greene, M.D. F.A.C.P. (H Lee Moffitt Cancer Center and Research Institute; Dept. of Internal Medicine and Oncological Sciences, COM, USF)

**Keywords**: Daptomycin, bacteremia  
**Objective**: Vancomycin-resistant Enterococcus (VRE) are rapidly emerging nosocomial pathogens in the US and worldwide. Vancomycin resistance is recognized to be an independent risk factor for increased morbidity and mortality among patients with enterococcal bloodstream infections (BSI), thus early appropriate therapy is vital in improving outcomes. We report our experience in treating VRE BSI in patients with a hematologic malignancies with daptomycin, a cyclic lipopeptide antibiotic with bactericidal activity against VRE.

**Methods**: We performed retrospective chart review for patients with a hematologic malignancies treated with daptomycin for a VRE BSI from January 2005 and May 2009. Patients were assessed for microbiological cure and survival. Patients were identified using a computerized clinical microbiology laboratory records.

**Results**: Out of 74 patients with a VRE BSI, 40 patients (31 male patients; median age, 61 years) met criteria and were included in the study. In this series, 35.6% of the patients had received a hematopoietic stem cell transplant (HSCT) and 57.8% of the patients were neutropenic. VRE BSI occurred at a median of 20 (4-109) days of neutropenia among these patients. Patients were treated with daptomycin monotherapy (62.5%) or daptomycin plus other antimicrobial (s) (37.5%). For those who were successfully treated with daptomycin alone, the median duration of therapy was 9 (1-36) days. Overall, 92.3% of patients achieved a microbiological cure which was evident at a median of 1 (1-16) days after the initiation of therapy with no VRE attributed mortality.

**Conclusion**: Daptomycin is an effective agent for the treatment of VRE BSI in patients with a hematologic malignancy, most of whom were neutropenic.

**Research supported by**: COM Research Scholarly Concentration

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**Abstract #**: B-56  
**Presented by**: Andrew Pepper, BS, Med II Student

**Non-invasive vs Invasive Pediatric Expandible Limb Salvage -- A Cost Analysis** Eric R. Henderson, MD*  
Andrew M. Pepper, BS* German A. Marulanda, MD* Odion T. Binitie, MD* Justin D. Millard, MD* David A. Johnson, PA-C** David Cheong, MD** G. Douglas Letson, MD ** *Department of Orthopaedics and Sports Medicine, USFCOM. Tampa, FL **Moffitt Cancer Center & Research Institute. Tampa, FL

**Keywords**: Cost Analysis Expandable Limb Salvage  
**Objective**: Non-invasive lengthening prostheses for pediatric limb salvage allow children to undergo limb lengthening at regular intervals in the clinic setting without the risk of repeated surgeries. Despite these advantages, the cost of these devices is cited as a deterrent. We sought to determine the financial costs associated with implantation and lengthening of these devices in the US and compare to invasive devices.

**Methods**: Current pricing information for the Stanmore non-invasive prosthesis and Stryker’s prosthesis were obtained from the manufacturers. A retrospective review of billing records was conducted for patients undergoing invasive expansion. Expenses associated with non-invasive lengthening were reviewed. Total costs for each device to achieve symmetric leg lengths were then calculated.

**Results**: Cost of the non-invasive prosthesis was $34,200 vs. $20,000 for invasive devices. Mean cost of a non-invasive lengthening was $269 vs. $9,821.82 invasively, including fixed-cost contracted expansions, otherwise the mean cost was $10,453.68. Mean age at implantation was 10.4 years, requiring a mean 4.1 cm of expansion before skeletal maturity. Total cost of 11.7 non-invasive expansions was found to be $3,147.30 vs $40,269.45 for invasive lengthening. Net estimated cost of reaching equal leg lengths with the non-invasive device is $37,347.30 vs. $60,269.45 invasively.

**Conclusion**: Initial savings with use of an invasive device were offset by the costs incurred with subsequent surgical expansions for patients requiring more than one expansion, assuming a 1 cm expansion. The implications of lost patient school time and parent work time deserve further attention. Cost saving could be realized by using the invasive device in children approaching skeletal maturity.
Abstract #: B-57  Presented by: Erika Reese, BS, Med II Student

**Comparing BMI, FAST Score and Energy Expenditure in Division I Intercollegiate Female Athletes**  
Erika Reese, MSII, University of South Florida College of Medicine, Frances Sahebzamani, P.h.D., A.R.N.P., University of South Florida, College of Medicine, Department of Family Medicine, Eric E. Coris, M.D., University of South Florida, College of Medicine, Department of Family Medicine Division of Sports Medicine

**Keywords:** Disordered eating, BMI, Exercise, Females

**Objective:** The purpose of this study was to examine relationships between Body Mass Index (BMI), disordered eating as defined by the Female Athlete Screening Tool (FAST) score, and calculated daily and weekly energy expenditure in elite female athletes calculated from responses to the Cooper Physical Activity Questionnaire.

**Methods:** One hundred female athletes consented to be screened with validated survey instruments including the FAST instrument and the Cooper Exercise Questionnaire. These surveys were used to examine 1) prevalence of clinical eating disorders as defined by the FAST score, and 2) energy expenditure calculated as metabolic equivalents (METs) of physical activity from the Cooper Exercise Questionnaire.

**Results:** There were no significant relationships between FAST score and daily exercise expenditure (r = .012, p = .919) or weekly exercise energy expenditure (r = .238, p = .01). There were no significant differences between BMI category on mean FAST score (F = 1.460, p = .221), mean daily calculated energy expenditure (F = .891, p = .474), or mean weekly calculated energy expenditure (F = .824, p = .515); or FAST score diagnostic category on mean daily (F = .315, p = .815) or mean weekly (F = .291, p = .832) calculated energy expenditures.

**Conclusion:** It was hypothesized that higher FAST scores would be positively associated with higher daily and weekly energy expenditures. The significant relationship between BMI and FAST score may be due to multiple psychological factors involved in disordered eating and warrants further study to examine this association.

**Research supported by:** Dr. Eric Coris and Dr. Frances Sahebzamani, USF COM Department of Family Medicine Division of Sports Medicine

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Abstract #: B-58  Presented by: Timothy Saunders, MD, Resident

**Traumatic Enucleation Series**  
Timothy Saunders MD, Dr. Charles Slonim MD, Department of Ophthalmology, College of Medicine, University of South Florida

**Keywords:** Traumatic enucleation

**Objective:** The purpose of this report is to review two new cases of unilateral traumatic enucleation and to compare these findings to previously published case reports.

**Methods:** The records of two traumatic enucleation cases were retrospectively reviewed. The results of our literature search (English language) of all cases of unilateral traumatic enucleation that provided both a description of the optic nerve that was transected and a contralateral visual field exam were compared in a meta analysis.

**Results:** A 47 year old male sustained enucleation injury to his left eye, the globe and the optic nerve were not found on exploration. They were presumed to have been transected at least 40-50mm from the globe. A follow-up Humphrey visual field demonstrated a complete contralateral temporal hemianopsia. A 38 year old male sustained enucleation injury to his left eye. The globe retained 50 millimeters in length of optic nerve still attached to it. A follow-up Humphrey visual field demonstrated a complete contralateral temporal hemianopsia. A thorough review of the literature produced 15 other cases of unilateral traumatic enucleation with a description of the injured optic nerve following the transection and an accompanying contralateral visual field exam. Ten cases reported contralateral temporal field loss to the injured eye and all 10 described an optic nerve transection 40 mm and longer. Two of the cases resulted in a superiortemporal field loss, eight of the cases however displayed a temporal hemianopsia.

**Conclusion:** Traumatic enucleation of the eye with an accompanied optic nerve transection greater than or equal to 40mm in length is associated with a contralateral temporal visual field anopsia. At least two patterns of visual field loss can be identified.
Abstract #: B-59

Evaluation of Prescribing Patterns of Psychotropic Agents in the Treatment of Eating Disorders
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Keywords: psychotropic drugs and eating disorder

Objective: To evaluate prescribing patterns of psychotropic agents in patients with a DSM-IV-TR Axis I eating disorder diagnosis. Determine if a relationship exists between the use of psychotropic agents and eating disorder diagnosis or outcome measures such as length of stay and BMI.

Methods: Single-site, prospective chart review

Inclusion Criteria
Adolescent and adult patients ≥ 12 years of age who were in residential treatment at Fairwinds Treatment Center
Axis I diagnosis of anorexia nervosa, bulimia nervosa or eating disorders not otherwise specified

Statistical Analysis
Descriptive statistics were utilized to describe psychotropic prescribing patterns. t tests were utilized to assess differences between groups.

Results:
Most common drugs prescribed: fluoxetine (28%), alprazolam (27%-given before meals), buspirone (25%), escitalopram (22%), olanzapine (15%) LOS was found to be longest for patients prescribed buspirone Patients who had lower BMI at admission were more likely to be prescribed olanzapine Patients with a higher BMI at admission (mean BMI 20.7) were more likely to be prescribed fluoxetine (p=.011).

Conclusion:
Patients with low BMI’s were more likely to be prescribed olanzapine. Fluoxetine was prescribed more often to patients with bulimia nervosa Buspirone was found to be correlated with longer length of stay. This may be because it was often added as an adjunctive medicine and many patients had drug abuse history thus leading to benzodiazepines not be chosen for anxiety. Many psychotropic medications are prescribed to treat co-morbid disorders associated with ED and more studies are needed.

Research supported by: Fairwinds Treatment Center, USF

Abstract #: B-60

Limb Preservation with Isolated Limb Infusion for Unresectable Non-melanoma Cutaneous and Soft Tissue Malignancies
Kiran K. Turaga1, Georgia M. Beasley2, Ricardo J. Gonzalez1, Keith A. Delman3, G. Douglas Letson1, David Cheong1, Douglas S. Tyler2, Jonathan S. Zager1
1: Department of Surgery, College of Medicine, University of South Florida, Moffitt Cancer Center, Tampa, Florida
2: Duke University, Durham, North Carolina
3: Emory University, Atlanta, Georgia

Keywords: ILI, sarcoma, limb preservation

Objective: Advanced, unresectable non-melanoma cutaneous and soft tissue malignancies (CSTM) including sarcomas (STS) of the extremities can pose significant treatment challenges for limb preservation. We report our experience including response and limb preservation using Isolated Limb Infusion (ILI) in CSTM.

Methods: We identified 16 pts with CSTM who underwent 17 ILI’s with melphalan and dactinomycin from 2004-2009 in 3 institutions. Outcome measures included limb preservation and in-field response rates. Regional toxicity was measured using the Wieberdink (WBD) scale and serologic toxicity via serum creatinine kinase (CK) levels.

Results:
Median age was 71 years (range, 19-92 years), and 50% were female. Five pts (31%) had pleomorphic undifferentiated sarcoma, 4 pts (25%) had Merkel cell (MCC), 3 (19%) had epitheliod sarcoma, and 4 patients (25%) had miscellaneous CSTM. The median length of stay was 7 days (range, 3-10). Twelve of 17 (71%) of the ILIs resulted in WBD grade I-II toxicity, 4 ILIs (24%) led to grade III toxicity; 1 (5%) patient developed grade IV toxicity. All but one pt had successful limb preservation resulting in 94% limb preservation. Of the 14 evaluable pts at least 3 month follow up, the in field overall response rate was 78% (36% CR and 42% PR). One pt had a repeat ILI after progressive disease after an initial partial response. One pt has stable disease and 2 have progressed in field. The overall 3 month progression free response rate is 86%; 100% in pts with MCC and SCC, while it is 78% for pts with STS.

Conclusion: ILI provides an attractive alternative therapy for regional disease control and limb preservation in pts with limb threatening CSTM. Short term response rates appear encouraging yet durability of response is unknown.
Abstract #: B-61
Presented by: Matthew Witmer, MD, Resident
Optic Neuropathy in Acute Retinal Necrosis Syndrome: A Case Series and Review
Matthew T. Witmer, MD, Resident
Department of Ophthalmology, COM, USF, Peter R. Pavan, Department of Ophthalmology, COM, USF

Keywords: Acute Retinal Necrosis, Optic Neuropathy, Papillitis, Herpes Virus

Objective: Acute retinal necrosis (ARN) syndrome is characterized by severe intraocular inflammation, occlusive vasculopathy, and peripheral retinal necrosis. Vision threatening complications of this syndrome include retinal detachment, macular edema and ischemia, and optic neuropathy. Traditionally, retinal detachment has been thought to be the principal cause of severe vision loss in ARN syndrome. With the advent of antiviral therapy, this complication has severely decreased. We investigate the significance of optic nerve disease in ARN syndrome and its effects on visual outcome.

Methods: We report four patients with ARN syndrome that presented with optic nerve inflammation at the time of diagnosis. We review optic nerve involvement in the ARN syndrome and present its incidence, pathogenesis, differential diagnosis, and treatment.

Results: Optic neuropathy in the course of ARN syndrome typically ranges from 47 to 57%. Several pathologic mechanisms including vasculitis, optic nerve ischemia, and direct invasion and destruction by a herpes virus may cause this complication. Treatment involves rapid diagnosis and treatment with antiviral therapy.

Conclusion: Optic nerve disease occurs frequently in ARN syndrome and causes severe vision loss. In the age of antiviral therapy, ARN associated optic neuropathy has likely become the most common cause of severe vision loss in this condition. ARN syndrome should be included in the differential diagnosis of acute optic nerve inflammation.

Abstract #: B-62
Presented by: Irfan Ahmed, MD, Resident
Novel Image Guided Visualization of Experimental Cellular-Based Vaccines, In-Vivo, During Combined Radiation/Immunotherapy Protocols
I. Ahmed, D. Gabrilovich, D. Cheong, R.J. Gonzalez, R. Heysek, D. Johnson, D. Letson, V. Sondak, S. J. Antonia, S. E. Finkelstein, Moffitt Cancer Center, Tampa, FL, Department of Oncologic Sciences, College of Medicine, University of South Florida

Keywords: image guided visualization Cellular based vaccine radiation

Objective: Novel technique for imaging dendritic cell (DC) vaccine migration, in-vivo.

Methods: DCs are labeled with 1mCi Indium Oxine. Cells are washed and the amount of radioactivity incorporated into the cells measured. A gamma probe which is pre-set to optimize detection of Indium energy peaks is used for localization purposes. Probe non-invasively obtains negative control baseline measurements of: the center of the tumor mass, tumor periphery, draining lymph nodes and background tissue to directly compare with the post injection, and intra-operative readings. Labeled DC's are administered via intra-tumoral injection under image guidance using sonography. Immediate post injection non-invasive measurements are obtained. Next, the patient is imaged using a standard gamma camera at 20 minutes post injection and immediately prior to surgery. The camera has a 20% window at the 171 kev Indium energy peak and a 20% window at the 245 kev Indium energy peak. Images of the injection site, regional lymph node stations, and the area between the injection site and regional basins are acquired in two projections. Any sites identified with the gamma camera are marked for localization at the time of surgery. Intra-operatively, the gamma probe is used to determine activity. The number of radioactive counts per 10 seconds within site are identified and recorded

Results: Ratios of various sites including tumor to background and lymph nodes to background can be compared. The ratio of counts of cases performed at different times can be compared following correction for decay and correlated with the level of specific immune response

Conclusion: This is a safe, minimally invasive technique and can further advance combined radiation/immunotherapy protocols
**Gene Expression Differences in Mouse Tendons**

Timothy Goede, College of Medicine, University of South Florida, Michael Mienaltowski, DVM, Ph.D., Department of Pathology & Cell Biology, College of Medicine, University of South Florida, David Birk, Ph.D., Department of Pathology & Cell Biology, College of Medicine, University of South Florida

**Keywords:** tendon, microarray, gene expression

**Objective:** The aim of this study was to characterize possible differences in gene expression between various mouse tendons to test the hypothesis that type-specific differences exist.

**Methods:** Achilles, Flexor Digitorum Longus (FDL), Extensor Digitorum Longus (EDL) tendons were harvested from four P30 C57Bl/6 wild-type male mice and then pooled based on type. Total RNA from pooled tendons was isolated using a QIAGEN RNeasy Lipid Mini Kit and then confirmed suitable for downstream array applications. Transcripts within RNA samples were hybridized to Affymetrix Mouse Gene Expression Arrays. Array data were normalized with Affymetrix software, and then analyzed using Expression Analysis Systematic Explorer (EASE).

**Results:** Transcripts for all of the genes that are typically considered biomarkers in tendon biology were detected on the array with greater signal intensities noted especially for procollagens, small leucine-rich proteoglycans, and housekeeping genes related to cytoskeletal structural components, transcription and translation machinery. Of the top 200 probe sets for each tendon type based on signal strength, a total of 269 distinct probe sets from the array were represented with 132 of those being shared across all three tendons.

**Conclusion:** Pilot gene expression data for Achilles, EDL, and FDL tendons of P30 C57Bl/6 wild-type male mice were quite similar. The hypothesis that tendon differences exist in P30 mice is not supported, and thus essentially a tendon is a tendon. Further analyses with more replicates are required to better detect differential expression and to validate the data. Future studies would also benefit from the inclusion of more age groups, such as mature and geriatric mice.

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**Cyanide Levels in Patients with Smoke Inhalation Injury**

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**Keywords:** Burn; Smoke inhalation injury; Cyanide

**Objective:** Smoke inhalation injury is a serious health threat resulting from aspiration of superheated gasses including steam and noxious products of combustion, involving the entire respiratory tract. Inhalation injury complicates the clinical course of burn injury patients by increasing risk for pneumonia and mortality. Hydrogen cyanide is a common component of smoke resulting from the combustion of common household items such as clothing, fabric, furniture, and plastics/polymers. Cyanide is extremely toxic to humans causing tissue hypoxia, organ failure, and if progressive; death.

**Methods:** We performed a pilot study to measure the levels of cyanide in smoke inhalation injury patients utilizing gas chromatography-mass spectroscopy analysis of blood samples taken from burn inhalation injury patients (n=15) within 24 hours of injury. Demographics and outcome information and data were collected.

**Results:** Cyanide levels were significantly increased in inhalation injury. Mortality was increased with significantly elevated cyanide levels, especially in conjunction with significant cutaneous burn injury.

**Conclusion:** Improved analysis and treatment of cyanide toxicity injury would be of benefit in patients with smoke inhalation injury.
Abstract #: B-65  
Presented by: Erinn Kellner, MS, Med I Student  

**The Interferon Signature is Not Associated with Depression or Fatigue in Systemic Lupus Erythematosus**  
Erinn Kellner, Pui Lee, Westley H. Reeves, M.D. University of Florida, Gainesville, Fl., Dept. of Internal Medicine, College of Medicine, University of South Florida  

**Keywords:** lupus, interferon, depression, fatigue  
**Objective:** Patients with SLE often suffer from depression and fatigue in addition to the physical manifestations of the disease. Elevated production of type-I interferons (IFN-I) has been found in lupus patients and recombinant IFN-I therapy is known to trigger a variety of neuropsychiatric side effects including depression and fatigue. This study was conducted to evaluate the relationship between dysregulated IFN-I production and the manifestations of depression or fatigue in lupus patients.  

**Methods:** Depression and fatigue in patients with SLE (n = 58) were assessed by Beck’s Depression Inventory and the Multidimensional Fatigue Index (MFI), respectively. Anxiety, pain and energy levels were evaluated by visual analog scales. Serum IFN-I levels were assessed using the expression of interferon-stimulated genes (ISGs) in peripheral blood mononuclear cells (quantitative PCR).  

**Results:** Depression and fatigue were reported by more than half of SLE patients in our study but these neuropsychiatric findings were largely unexplained by disease-related factors such as clinical manifestations, complement levels, medications, or autoantibody profile. In line with previous findings, elevated ISG expression was present in about two-thirds of SLE patients. However, through both cross-sectional and longitudinal studies analysis we found no significant correlation between ISG expression and the levels of depression or fatigue.  

**Conclusion:** Although recombinant IFN-I (IFN-alpha) therapy is associated with a wide-range of neuropsychiatric side effects including depressive illness, elevation of endogenous IFN-I levels is unlikely to be responsible for the depression and fatigue experienced by lupus patients.  

**Research supported by:** Research scholarly concentration and the Lupus Foundation of America.

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Abstract #: B-66  
Presented by: Charles Knapp, MD, Resident  

**Decreasing the Risk of Suture Breakage with Rotational Suturing,**  
Charles Knapp, MD; Basil S Cherpelis MD  
Department of Dermatology, College of Medicine, University of South Florida  

**Keywords:** suture breakage rotational suturing  

**Objective:** INTRODUCTION AND PURPOSE: Excision of cutaneous lesions on areas such as the scalp and back can often be difficult to close primarily, even with adequate undermining of the wound edges. Using traditional suturing techniques in such high-tension areas often leads to breaking of suture, thus resulting in a highly frustrating experience for both the dermatologist and the patient. “Rotational suturing” is a simple technique that enables the cutaneous surgeon to consistently secure knots without breaking the suture.  

**Methods:** METHODS: The technique involves beginning the closure with a simple “instrument tie”. With the needle driver clasped down on the suture, the needle driver is rotated two to four full turns toward the knot to wrap a few turns of suture around it. The opposite ends of the suture are then pulled to tighten the knot.  

**Results:** RESULTS/CONCLUSIONS: Traditional tightening of a knot to approximate wound edges involves simply clasping down on the blind end of suture with the needle driver and pulling. However, this allows a great deal of friction to occur between the suture and the serrations of the needle driver, often resulting in fraying or breaking of the suture. With the method of rotational suturing described above, the energy of this frictional force is transferred to a tensional force against the smooth surface of the needle driver’s outer walls, thus reducing the risk of suture breakage. Rotational suturing is a simple yet effective technique that dermasurgeons can employ in their practices to allay some of the frustration of large wound closures.  

**Conclusion:** See "Results" section
**Abstract # : B-67**

Presented by: Latha Kumaraswamy, MD, Resident

**Longitudinal Comparison of Infant Body Composition in Small- (SGA) and Appropriate-for-Gestational Age (AGA) Infants Using Air Displacement Plethysmography (ADP)**

Latha Kumaraswamy, M.D, Jane Carver, PhD,MS,MPH, 1Pediatrics, Nicole Nations, Judy Zaritt, Lewis P. Rubin, MD, Univ.of South Florida, Tampa, FL

**Keywords:** Infant body composition body fat Newborn Obesity

**Objective:** To use ADP instrumentation for longitudinal assessment of infant body composition and for assessment of the relationship between body composition and anthropometric measurements in infants between 1 and 8 kg.

**Methods:** SGA infants were matched to AGA infants of a similar gestational age. Infants with significant anomalies were excluded. Body composition and anthropometric variables were measured at hospital discharge and monthly thereafter until subjects reached a body weight of 8 kg.

**Results:** Demographic data were similar between groups. Preliminary findings show increasing % fat mass and decreasing % fat free mass over time. % fat mass is significantly lower and % fat free mass is significantly higher in SGA infants at the first measurement. Several anthropometric measurements remain lower in SGA infants at the second measurement. However, there were no significant differences found on subsequent measurements.

**Conclusion:** SGA infants of different gestational appear to catch up to their AGA counterparts in adiposity & anthropometric measurements within 8-10 weeks after birth. On-going studies assess the relationship between body composition and clinical/demographic variables.

**Research supported by:** Muma Family Endowment, Univ of South Florida

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**Abstract # : B-68**

Presented by: Qiyuan Liu, PhD, Med II Student

**Differentiation of Human Embryonic Stem Cells to Megakaryocytes in Feeder Cell-Free Cultures**

Qiyuan Liu, PhD, Department of Malignant Hematology, H. Lee Moffitt Cancer Center and Research Institute, Department of Internal Medicine, College of Medicine, University of South Florida

**Keywords:** human embryonic stem cells embryoid bodies megakaryocytes platelets

**Objective:** One important field in human embryonic stem cells (hESC) research is to produce blood cells in vitro in numbers that are clinically useful. We have developed a tissue culture system for hESC growth and expansion on feeder cell-free matrix. hESC were then induced to form embryoid bodies (EBs), which further proliferated and differentiated into megakaryocytes that produced platelet-like particles.

**Methods:** We coated the culture dishes with extracellular matrix by lysis of human foreskin fibroblasts growing for 5 days in the dishes. hESC cells were collected, and plated to the ultra-low attachment culture dishes to form EBs in medium with various growth factors. The EB morphology was observed microscopically, and differentiation of cells at each stage was tested by immunofluorescence.

**Results:** We found that expression of hESC markers Oct4 and Sox2 was decreased by EB day 7 and was undetectable by EB day 10. By EB day 14, a 10-fold expansion of cell numbers was counted compared to the beginning of the EB culture, and outside the EBs, there were exclusively a large numbers of mononuclear cells. Virtually 100% of these free floating cells were strongly CD34+/GATA1+. About 10% of them expressed very strongly megakaryocytic lineage markers, CD41, CD42a, and CD42b, with most of the remaining cells expressing low levels of these markers. Differentiation toward megakaryocytes was further confirmed by the observation that the cells started to produce apparent platelet-like particles by EB day 21.

**Conclusion:** We are able to induce the differentiation of hESC to mature megakaryocytes in the presence of appropriate growth factors in a feeder-free, serum-free culture setting.

**Research supported by:** American Society of Hematology Research Scholarly Concentration
Amnion-Derived Cellular Cytokine Solution (ACCS) Promotes Macrophage Phagocytosis and Bactericidal Activity

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Keywords: Amnion-derived multipotent progenitor (AMP) cells; Amnion-derived Cellular Cytokine Solution (ACCS); Macrophage; Infection; Wound Healing

Objective: Wound healing is a complex process requiring the collaborative efforts of different cell lineages and entails the coordinated interplay of several phases of proliferation, migration, matrix synthesis and contraction. Infection may disrupt this normal cascade of wound healing. Inflammatory cells play key roles in clearing the wound of invading microbes, thus defending against infection and allowing for the normal wound healing. Various studies have demonstrated the important role of cytokines for the accompanying inflammatory reaction as well as for repair processes during wound healing. Studies demonstrate that cytokines present during the healing of a wound are similar to those found in Amnion-derived Cellular Cytokine Solution (ACCS), a secretory product of Amnion-derived Multipotent Progenitor (AMP) cells. The goal of this study was to determine if the use of ACCS will stimulate the anti-microbial abilities of macrophages.

Methods: Three groups of macrophages were exposed to different concentrations of ACCS solution (10% / 20% / 40%). The killing and phagocytic activity of each group was compared to the control after one hour of exposure to Escherichia coli.

Results: Macrophage activity following activation by ACCS demonstrated increased phagocytosis and bactericidal activity proportional to concentration exposure.

Conclusion: Macrophage activity increased significantly when stimulated by ACCS in this in vitro model. These inflammatory cells, critical to normal wound healing, may be influenced to decrease bacterial load and allow for normal wound healing in the infected wound.

Research supported by: Stemnion, inc.

Amnion-Derived Cellular Cytokine Solution (ACCS) Accelerates Healing of Experimental Partial-Thickness Burns

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Keywords: Amnion-derived multipotent progenitor (AMP) cells; Amnion-derived Cellular Cytokine Solution (ACCS); Burn Wound; Wound Healing

Objective: Amnion-derived multipotent progenitor cells (AMP cells), unlike most stem cells, have been demonstrated to be non-tumorigenic and non-immunogenic. Amnion-derived cellular cytokine solution (ACCS), a secreted product of AMP cells, is a cocktail of cytokines existing at physiological levels. This study evaluates the influence of ACCS to accelerate epithelialization of experimental partial-thickness burns.

Methods: Using modifications of Zawacki’s guinea pig partial-thickness scald burn model, a total of 65 animals were treated with ACCS, ACCS + AMP cells, unconditioned medium (UCM) + AMP cells, or UCM alone or saline as controls. Dosage times ranged from every other day to once a week. Percent epithelialization was serially determined from acetate wound tracings. Histology was performed on wound biopsies.

Results: ACCS, UCM+AMP cells, and ACCS+AMP cells improved epithelialization compared with the two control groups (P<0.05). When ACCS was delivered more frequently, statistically significant more rapid epithelialization occurred (P<0.05). By day 7, all groups treated with ACCS had reached at least 90% epithelialization; whereas, control groups were between 20-40% epithelialized (P<0.05). Histology showed excellent regeneration of the epidermis with rete ridge formation. Hair growth occurred in ACCS-treated animals, but not in the control group.

Conclusion: Amnion-derived cellular cytokine solution accelerates the healing of experimental partial-thickness burns. Based on these findings a multicenter clinical trial is underway.

Research supported by: Stemnion, Inc.
**Workshop Using Simulators to Teach Medical Students Large Joint Injection**
Aasim Rehman, Ashley G. Sterrett, Vanessa Osting, John D. Carter, Louis Ricca, Joanne Valeriano-Marcet, Helen Bateman, Department of Internal Medicine, College of Medicine, University of South Florida

**Keywords:** Joint Injection Arthrocentesis

**Objective:** Teach medical students the proper technique of large joint injections and assess the effect of such a workshop on their comfort levels when performing these procedures.

**Methods:** All 4th year medical students attend the joint injection workshop at the beginning of their rheumatology rotation. The workshop consists of a teaching didactic session followed by the joint simulator injection session. The simulators are pre-wired anatomical models of the knee and shoulder. A pre and post workshop self assessment survey is given regarding student’s comfort level with examination and injection of knee and shoulder joints, on a scale of 1 to 5 (1= not comfortable to 5= fully comfortable).

**Results:** A total of 68 students completed the workshop. 23 students had performed knee injections, 6 had performed shoulder injection and 10 students had performed other musculoskeletal injection prior to attending the workshop. Table 1: Pre and post workshop comfort levels. (N=68) Pre-Workshop Mean Comfort Level (+/- SD) Post-Workshop Mean Comfort Level (+/- SD) p-value* Knee Palpation 2.81 (+/- 0.90) 3.93 (+/- 0.74) <0.0001 Detecting Knee Effusion 2.75 (+/- 0.90) 3.51 (+/- 0.91) <0.0001 Knee Injection 1.87 (+/- 1.02) 3.66 (+/- 0.84) <0.0001 Shoulder Palpation (sub-acromial) 2.59 (+/- 0.85) 3.88 (+/- 0.86) <0.0001 Shoulder Injection 1.44 (+/- 0.63) 3.59 (+/- 0.83) <0.0001 Selection of Corticosteroid Preparation 1.55 (+/- 0.68) 3.15 (+/- 1.10) <0.0001 * Paired t-test of post-workshop comfort level compared with pre-workshop comfort level.

**Conclusion:** Our joint injection workshop using simulators increases the comfort level of medical students when performing large joint injections.

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**Alzheimer Amyloid-Beta Inhibits Mitotic Kinesins: Implication for Aneuploidy and Neuronal Plasticity**
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**Keywords:** Alzheimer’s disease, cell cycle, microtubules, Eg5, long-term potentiation

**Objective:** Aneuploidy and impaired neuronal plasticity implicated in Alzheimer’s disease (AD) may result from Amyloid-beta (A-beta) induced microtubule (MT) dysfunctions. MT-based motors are critical for the proper organization and functions of MTs during a variety of cellular processes, including mitosis and the development of neuronal processes. We hypothesize that A-beta abrogates functions of specific MT motors, thereby leading to aneuploidy and impaired neuronal plasticity. Based on this, we aimed to study whether A-beta inhibits mitotic motor proteins and, if so, whether this inhibition affects the mitotic spindle and neuronal functions.

**Methods:** A-beta’s interaction with motor proteins was studied by co-immunoprecipitation and affinity purification. A-beta’s effects on motors’ activities were studied in an in vitro MT-dependent ATP-ase assay. A-beta’s effects on the mitotic spindle were studied by using Xenopus egg extracts. Effects of the Eg5 inhibitor, Monastrol, on neuronal processes were studied by using an LTP assay.

**Results:** A-beta induced defective spindle phenotypes, which are indicative of malfunctioning Eg5, KIF4A and MCAK motor proteins. Consistently, A-beta directly associated with these mitotic motors, inhibited their activities in vitro and abrogated their association with mitotic spindles. Interestingly, A-beta inhibited also neuronal Eg5. Importantly, treatment of hippocampus slices with the Eg5 inhibitor Monastrol significantly reduced LTP readings to levels comparable to the exposure with A-beta.

**Conclusion:** The data herein support a unified explanation for aneuploidy and defective neuronal plasticity associated with AD as resulting from A-beta direct inhibition of specific MT motors, including Eg5, KIF4A and MCAK.

**Research supported by:** USF start-up
Neuroprotection by Transcription Factor Modulation in Traumatic Brain Injury
Jessica N. Chang, Vardit Rubovitch, John S. Dennis, Vedad Delic, Harris Hatic, Chaim G. Pick, Bruce A. Citron
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Keywords: traumatic brain injury, transcription factors, neuroprotection, apoptosis, neurons

Objective: Traumatic Brain Injury (TBI) affects 1.4 million people annually in the United States, resulting in substantial neuronal loss for which there is currently no effective treatment. Even mild injuries have long-term consequences. Unfortunately, TBI is the signature affliction of recent combat operations with an incidence of approximately 15%. The molecular mechanisms that regulate neuronal death following TBI are incompletely understood. In an effort to elucidate these mechanisms and to identify novel therapeutic targets, we are investigating transcription factor signaling.

Methods: We, and others, have observed transcription factor dysregulation in several CNS disorders, including TBI, disrupting the balance of harmful and protective gene expression. These transcription factors provide therapeutic targets as slight adjustments can alter downstream genes and improve neuronal survival. By chemically modulating their expression increased neuroprotection was observed in vitro. We are testing these modulators with a mouse model of TBI monitoring behavioral and molecular improvements.

Results: We found that pharmacologic activation of a protective transcription factor reduced the level of activated caspase-3 in the injured region of the brain and we are quantifying the cognitive changes in the treated vs. untreated mice.

Conclusion: Transcription factor modulation seems a promising approach to protect the health of neurons following TBI.

Research supported by: This study was supported by the Department of Veterans Affairs (Veterans Health Administration, Office of Research and Development, Biomedical Laboratory Research and Development) and the Bay Pines Foundation.

Functional Analysis of Cochlear BK Channel-associated Proteins Using RNA Interference
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Keywords: proteomics, interactome, BK channel, protein-protein interactions, cochlea

Objective: Large conductance Ca2+-activated K+ (BK) channels regulate several important physiological processes, such as neuronal excitability, signal transduction, smooth muscle contraction, metabolism, and immunity. Thus, these channels will likely partner with a number of different types of proteins. We identified 174 BK-Associated Proteins (BKAPs) in mouse cochlea (Kathiresan et al., 2009) and more recently, 125 in chick, using BK coimmunoprecipitation (coIP), 2-D gel electrophoresis, and shotgun proteomics. Here, we selected 8 BKAPs involved in chaperoning, signal transduction, and cell growth to study their function.

Methods: These proteins were downregulated by using siRNA or upregulated by over-expressing BKAPs, to determine BK regulation in CHO cells.

Results: Knockdown of the chaperonins: calrecticulin, GRP78, and HSP60, by 15% – 50%, altered BK expression by 25% – 30% in CHO cells. Cell signaling BKAPs, such as 14-3-3 and annexin A5, increased BK expression by 25% – 30%, when downregulated by ~30%. BK decreased 25% – 60% when these same proteins were overexpressed by 15% – 30%. Silencing of development-regulated BKAPs, such as VCP and lamin A/C by 40% and 60%, decreased BK by 40% and 50%, respectively. A potential node in these pathways is Akt1, since a decrease of 20% caused a 15% increase in BK. Moreover, silencing 14-3-3 and annexin A5 decreased Akt expression by ~19% – 23%, whereas overexpression of both proteins increased Akt by 10% – 22%. Additional coIP experiments showed that Akt interacts with BK, which has a potential binding site at the C-terminus.

Conclusion: These experiments have begun to identify potential therapeutic targets in the regulation of BK.

Research supported by: Supported by NIDCD grant R01DC004295 To BHAS.
Outcomes of a Group Behavioral Parent Training Program for Families of Children with Autism Spectrum Disorders

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Keywords: Autism spectrum disorder (ASD), parent training, behavior, development

Objectives: To investigate the attendance rates, knowledge gains, and consumer satisfaction of caregivers of young children with a diagnosis of ASD who participated in a group-delivered, behaviorally-based parent training program

Methods: Descriptive and analytic methods were used with a population of caregivers of children with challenging behaviors who participated in the HOT DOCS program between 8/06-10/09 and provided information about their child’s diagnoses (n=723). The sample was restricted to those participants who indicated their child was diagnosed with ASD at the time of participation (n=124). It was hypothesized that caregivers of children with ASD would demonstrate significant increases in knowledge.

Results: A dependent means t-test showed significant increases in participant knowledge from pre- to posttest, t(1, 70)=.836, p<.001. The majority of participants (~99%) reported high levels of satisfaction with various aspects of HOT DOCS.

Conclusion: Access to early intervention has been shown to improve academic, behavioral, and social outcomes for children with ASD (Guralnick, 1997). Most research is on child-focused interventions, limited research is available on parent-focused programs (Boulware et al., 2006). Findings from this research are important in expanding the services available to families with young children with ASD diagnoses. HOT DOCS appears to be a promising early intervention program for families of children with ASD, while maximizing available resources (group format). Future directions include comparing participation and outcomes for families of children with ASD to families of children with no preexisting diagnoses and/or with various other diagnoses who participate in HOT DOCS.

Research Supported by: Children's Board of Hillsborough County

NT 020, a Natural Therapeutic Approach to Optimize Spatial Memory Performance and Increase Neural Progenitor Cell Proliferation and Decrease Inflammation in the Aged Rat.

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Keywords: Aging, natural compounds, NT 020, spatial memory, neurogenesis

Objective: The process of aging is linked to oxidative stress, microglial activation, and pro-inflammatory factors, which decrease cell proliferation and neuroplasticity. Natural compounds such as polyphenols from blueberry, and green tea, and amino acids like carnosine are high in antioxidant and anti-inflammatory activity that decreases the damaging effects of reactive oxygen species, in the blood, brain, and other tissues of the body. We have shown that the combination of these nutrients (NT-020) creates a synergistic effect that promotes the proliferation of hematopoietic cells in vitro. In this in vivo study, we investigated the natural therapeutic potential of NT-020 for improving learning and memory and neurogenesis in aged rats.

Methods: Three groups were treated with either NT-020 or water gavage. Aged (20 month old) male Fisher 344 rats were treated with 135.0 mg/kg per day (n = 13) of NT-020. Young (3 month old) (n = 10) and Aged control (20 month old) (n = 13), male Fisher 344 rats were treated with water gavage. All groups were treated for a period of 4 weeks. Rats were tested for learning on a Morris water maze. Brains were collected and immunostained.

Results: There were fewer aged impaired animals in the treatment group compared with controls. Decreased number of OX6 MHC II positive cells, increased neurogenesis, and increased number of proliferating cells were found in rats treated with NT-020 in comparison with aged control rats.

Conclusion: In sum, NT-020 may promote health, proliferation and maintenance of neurons in the age animals and exert anti-inflammatory actions which promote function in the aged stem cell niche.

Research supported by: USPHS grant AG04418 and the VAMRS. Conflict of Interest: PCB and PRS are founders of NaturaTherapeutics.
**Abstract #**: B-77  
**Presented by**: Antoinette Bailey, BS, Graduate Student

**sAPP-α and Neurodevelopment: A Potential Mouse Model of Autism**  
Affiliations: Dept. of Molecular Pharmacology and Physiology, Rashid Laboratory for Developmental Neuroscience, Silver Child Development Center, University of South Florida College of Medicine; Center for Aging & Brain Repair, University of South Florida College of Medicine; Department of Psychiatry & Behavioral Medicine, University of South Florida

**Keywords**: amyloid precursor protein alpha, neurodevelopment, autism,

**Objective**: Autism is a neurodevelopmental disorder characterized by impaired social interaction, deficient communication and stereotypic behavior. Recent studies report high levels of soluble amyloid precursor protein alpha (sAPP-α) in the plasma of children with severe autism. Several reports describe the neurotrophic functions of sAPP-α which promotes neurite outgrowth, differentiation and synaptic plasticity. These reports lead our group to believe that high levels of sAPP-α in the brain contributes to the development of autism and other such neurodevelopmental disorders. Our objective with this study is to explore the effects of elevated sAPP-α levels on neurodevelopment.

**Methods**: In order to investigate our hypothesis, our group has created a transgenic mouse model that over-expresses human sAPP-α. We are currently studying this model using molecular biological methods such as Western blotting and ELISA, immunohistochemical techniques and animal behavior tests.

**Results**: To date, we have discovered that the sAPP-α transgenic animals display increased tau phosphorylation, as well as increased expression of synaptophysin and impaired social behavior in males when compared to age-matched controls.

**Conclusion**: While further studies are required to identify the role of sAPP-α in neurodevelopment, our findings suggest that sAPP-α may enhance tau-associated neurite outgrowth and, additionally, cause an increase in synaptogenesis. Our results also imply that high levels of sAPP-α decreases social tendency in male mice. Further study of these and other roles of sAPP-α on the brain and behavior will elucidate a novel phenotype occurring in autism and similar neurodevelopmental disorders and may provide a new approach for diagnosis and treatment.

**Research supported by**: The Silver Foundation.

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**Abstract #**: B-78  
**Presented by**: Laura Blair, BA, Graduate Student

**In Vivo Administration of Heat Shock Protein 27 Improves Hippocampal Plasticity**  
Laura Blair1,4, Jose Abisambra1,4, Clara Kraft1,4, Jeffrey Jones1,4, Justin Rogers3,4, Jessica Banko4, John Koren III1,4, Umesh Jinwal1,4, Amelia Johnson1,4, Grant Mauk2,4, Edwin Weeber3,4, Chad Dickey1,4  

**Keywords**: Hsp27, Tau, Alzheimer’s disease

**Objective**: Tau, a factor in many neurodegenerative diseases, is a soluble protein involved with the assembly and stability of microtubules. Tau is phosphorylated by many kinases, which can cause it to be released from the microtubules. In an AD mouse model, rTg4510, tau becomes hyperphosphorylated and forms abnormal tangles in the brain causing cell death. During neurodegeneration, the small heat shock protein Hsp27 promotes neuronal survival. Hsp27 has been shown to be associated with tangles and reduce the amount of hyperphosphorylated tau.

**Methods**: To better understand the mechanism for this phenomenon, we investigated the impact of Hsp27 on tau levels in cells through over-expression and knock down. In order to further explore the relationship, adeno-associated viral particles expressing Hsp27 were directly injected into the hippocampus of rTg4510 mice. We performed electrophysiology on the hippocampus. We also stained like tissue using tau and Hsp27 antibodies.

**Results**: There is a correlation between tau reductions and Hsp27 over-expression both in cells and in mice. Furthermore, rTg4510 mice show an LTP (Long Term Potentiation) deficit, which is rescued by the injection of Hsp27 particles.

**Conclusion**: Molecular chaperones, like heat shock proteins, can be used to regulate tau expression. Hsp27 prevents tau aggregation and reduces its levels both in cells and in our mouse model. The rTg4510 mouse model has deficits in LTP, Hsp27 rescues this phenotype by improving hippocampal plasticity.

**Research supported by**: NIA grant R00AG031291
The Effect of an Alcohol Prime on ERPs to Alcohol Expectancy-Related Sentences
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Keywords: ERP Alcohol Expectancies

Objective: Expectations that individuals hold about the outcomes of consuming alcohol influence their information processing and drinking behavior. Priming these expectancies can influence subsequent behavior in expectancy-congruent ways. In an Event Related Brain Potential (ERP) study (Fishman et al. 2008), our lab reported that sentence terminations that violate a subject's alcohol expectancies elicit a P300, a component of the ERP that is sensitive to expectancy violations. We extended this paradigm to examine the extent to which priming alcohol expectancies would alter the ERP responses to alcohol-related sentence terminations.

Methods: Fifty undergraduates completed the study which included completing an alcohol expectancy questionnaire, viewing either an alcohol-related or neutral video, followed by an ERP task in which participants attended to two alcohol related sentence categories: Alcohol Positive & Alcohol Negative.

Results: Similar to our previous findings, positive expectancy scores were positively correlated with ERP scores, such that higher expectancy scale scores were associated with lower or less positive scores on the P300-like component of the ERP, which indicates less expectancy violation. Since this effect remained after controlling for priming condition, and the main effect of the alcohol prime was not significant, priming did not appear to influence ERPs to alcohol-related sentence endings.

Conclusion: ERPs appear to index violations of subjectively held expectancies concerning alcohol and serve as an upstream measure of a construct that has typically been measured via self-report questionnaires.

Research supported by: NIAAA Grants R01 AA008333 & R01 AA016091 to Mark Goldman

TRAF6 and Neuron Survival Carrie L. Butler, Jessica N. Chang, John S. Dennis, and Bruce A. Citron, Bay Pines VA Healthcare System and Department of Molecular Medicine, USF College of Medicine, University of South Florida

Keywords: TRAF6, ALS, neurodegeneration, wobbler mouse and neurons

Objective: TNF receptor-associated factor 6 (TRAF6) can activate c-Jun and NF-kB pathways, which are known to play a role in inflammatory responses and apoptosis. We sought to determine whether the expression of TRAF6 was altered in spinal cords undergoing motor neuron loss from human Amyotrophic Lateral Sclerosis (ALS) samples and a murine model of ALS.

Methods: We analyzed gene expression changes in neurons with Affymetrix gene arrays. Expression of a candidate gene was confirmed by qRT-PCR and Western immunoblotting. Localization of TRAF6 was examined in neurons. In addition, we tested cause and effect relationships in cultured cells by subcloning TRAF6 into an expression vector.

Results: We examined gene expression changes in neurons mechanically injured in culture and in vivo traumatic-impact-injured rat spinal cords and found TRAF6 mRNA to be upregulated. These results were extended with a two-fold increase in TRAF6 mRNA levels in human ALS and in the wobbler mouse model of ALS spinal cords. Protein levels in nuclear extracts of wobbler spinal cords were elevated. Over expression of TRAF6 through transient transfection was protective in HT22 cells in culture when injured with hydrogen peroxide as compared to controls.

Conclusion: Our data indicates that TRAF6, implicated in immune pathways, is also important in cell loss in the central nervous system (CNS). The identification of this and other factors that regulate neuronal survival may lead to attractive targets for pharmacological intervention.

Research supported by: The Office of Research and Development, Department of Veterans Affairs, and the Bay Pines Foundation.
**The Relationship Between Treatment Adherence and Child Behavioral Outcomes in a Parent Training Program**

**Presented by:** Jason Hangauer, Graduate Student

**Objective:** To determine the rate of adherence (i.e., attending & implementation of skills) to a parent training program and whether adherence is related to child behavioral outcomes.

**Methods:** Parents/caregivers (N=913) of children (M = 42 months) attended a group-administered parent training program. Data were analyzed to determine if participants who adhered to treatment reported greater reductions in challenging behavior. Adherence was operationalized as attendance & rate of weekly implementation of skills. Child outcomes were measured via parent report of behavioral symptoms (externalizing/internalizing). Descriptive analyses of adherence and correlational analyses examining the relationships between adherence and child outcomes were conducted.

**Results:** Mean attendance at parent-training sessions was 4.3 (SD = 1.7) out of 6. Mean number of days participants reported implementing skills was 16 (SD = 12) out of 35. When 80% is used as a standard of treatment adherence, only 26.4% of participants met criterion. Relationship between adherence variables (attendance & implementation of skills) and child change from pre to post in child outcome variables (internalizing & externalizing T-score change) was investigated. Correlations were small in size and insignificant at .01 alpha.

**Conclusion:** Treatment adherence is important to establish a functional relationship between a treatment and outcomes (Gresham, 2004). Treatment adherence for a majority of participants was below the 80% criterion used as a standard, but consistent with previous research (e.g., Pettinati et al., 2003). Plausible explanations (e.g., self-report of skill implementation) for the insignificant relationship between these variables will be discussed.

**Research supported by:** Children’s Board of Hillsborough County

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**Simultaneous Intracellular Measurement of Superoxide and Membrane Potential in Solitary Complex.**

**Presented by:** Michael Matott, MS, Graduate Student

**Objective:** The purpose of this study was to develop procedures for intracellular loading of dihydroethidium (DHE), a fluorescent probe for measuring the rate of superoxide production, while simultaneously measuring electrical activity via whole-cell recording at different O2 tensions.

**Methods:** Transverse rat brainstem slices (300-400µm) were prepared and incubated in either 95% O2 5% CO2 or 40% O2 5% CO2 (balance N2, 36°C) 24 cells were patched in the solitary complex using the whole-cell technique. Four concentrations of DHE (5,10, 15, 20 µM) were added to standard pipette solution. Pictures were taken every 3 minutes (2s exposure) for ≥ 1 hour.

**Results:** 10µM was the optimal concentration for single-cell labeling based on intensity of DHE fluorescence (FIDHE). DHE labeled neurons fired spontaneously with similar electrophysiology to unloaded neurons. The membrane potential of neurons patched with DHE was generally lower than non-DHE cells. FIDHE increased steadily over time but the slopes varied between cells and did not show a significant difference, on average, between cells in 40% or 95% O2.

**Conclusion:** This is the first study to combine fluorescent measurement of superoxide and electrical properties in a single neuron. Advantages of this technique are a reduction in background fluorescence and ability to correlate changes in superoxide production simultaneously with changes in electrical activity. FIDHE was not impacted by control oxygen concentrations. However, preliminary studies of acute O2 changes have shown a noticeable increase in FIDHE during anoxia.

**Research supported by:** ONR, NIH
**Abstract #**: B-83

Presented by: John O'Leary, BA, Graduate Student

**Methylene Blue Ameliorates the Cognitive Deficits in a Mouse Model of Tauopathy**

John C. O'Leary

III

Umesh Jinwal, John Koren III, Jose F. Abisambra, Yoshinari Miyata, Laura Blair, Clara Kraft, Amelia G. Johnson, Mindy Peters, Jason Gestwicki, Edwin J. Weeber and Chad A. Dickey

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**Keywords**: tau, alzheimer, drug discovery, hsp70, chaperone, methylene blue

**Objective**: Characterize the effect of chronic methylene blue treatment in a mouse model of tauopathy.

**Methods**: Standard histological, biochemical and behavioral methods.

**Results**: Here we show that peripheral administration of MB in sufficiently high doses can improve the cognitive abilities of transgenic mice that over-express P301L mutant human tau (rTg4510 mice). While the rTg4510 mice were eventually able to learn where the escape platform was located in the Morris water maze task, recall of this location is impaired during the probe trial. Furthermore, there was no change in pathology despite cognitive improvement.

**Conclusion**: MB may be a very promising drug for the treatment of AD and related tauopathies and the development of future therapeutics.

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**Abstract #**: B-84

Presented by: Mibel Pabón, MS, Graduate Student

**Fractalkine as a Neuroprotective Therapy in a Model of Parkinson’s Disease**

M.M. Pabón, J. Jernberg, C.E. Hudson, A. Bachstetter, C. Gemma, P.C. Bickford

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**Keywords**: Key words 6-OHDA- 6-Hydroxydopamine FKN- Fractalkine PD- Parkinson Disease

**Objective**: A pathological component of Parkinson’s disease (PD) is brain inflammation due to the activation of microglia. This activation causes neuronal damage through the production of cytokines and reactive oxygen species. Fractalkine (FKN) is a chemokine that regulates excessive microglia activation. With age there is a loss of FKN and this may make microglia primed for excessive activation, as seen in age-related neurodegenerative diseases. It is not known if addition of exogenous FKN beyond otherwise physiologically normal levels can be therapeutically beneficial following a neurotoxic insult.

**Methods**: To test the hypothesis we used the intrastriatal 6-OHDA model of PD. 6-OHDA (20µg/4µL) was infused into the left striatum of 3 month-old F344 male rats; followed by sustained delivery of FKN via osmotic pump at 3ng of FKN, 30ng of FKN, 90ng of FKN or 90ng of heat-inactivated fractalkine (as a control) doses for 28 days into the site of the lesion. After 28 days the rats were anesthetized, transcardially perfused. Immunohistochemistry was performed for tyrosine hydroxylase (TH) and MHC class II (OX-6) throughout the entire dorsal striatum.

**Results**: Using the cavalieri method of unbiased stereology to calculate the TH negative lesion volume revealed that exogenous soluble FKN was neuroprotective resulting in a significantly smaller lesion. Importantly, FKN produced a dose dependant decrease in activated microglia supporting the proposed mechanism of action.

**Conclusion**: FKN plays a neuroprotective role in 6-OHDA-induced dopaminergic lesion and it might be an effective therapeutic target for PD, where inflammation plays and important role.

**Research supported by**: This work was supported by the VA Medical Research Service (PCB)
Nanolipidic Particles Improve the Bioavailability and Alpha-Secretase Inducing Ability of Epigallocatechin-3-Gallate (EGCG) for the Treatment of Alzheimer’s Disease

Presented by: Adam Smith, M.S., Graduate Student

Nanolipidic Particles Improve the Bioavailability and Alpha-Secretase Inducing Ability of Epigallocatechin-3-Gallate (EGCG) for the Treatment of Alzheimer’s Disease

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Keywords: Nanoparticle, EGCG, Bioavailability, Alzheimer’s Disease, Pharmacokinetics

Objective: This study investigated the ability of nanolipidic particle complexes for increasing the oral bioavailability of EGCG.

Methods: Preparation of Nanolipidic EGCG Particles Nanolipidic particles were prepared using a proprietary co-solubilization methodology involving use of monophasic liquid preparations developed by Nature’s Defense Systems, Tampa, Florida. Neuronal sAPP-α ELISA Murine neuroblastoma cells that were stably transfected with the human APP gene were cultured, differentiated, and treated with various EGCG formulations for 18 hours. The conditioned media was collected and sAPP-α levels were quantified using a sAPP-α sandwich ELISA protocol. Pharmacokinetic Screening The EGCG formulations were delivered via oral gavage to male Sprague Dawley rats at a dosage of 100 mg EGCG/kg body weight. Blood was collected at 0, 5, 10, 30, 60, 120, 240, and 480 minutes and analyzed for EGCG content using liquid chromatography with tandem mass spectrometry.

Results: We found that forming nanolipidic EGCG particles improves the neuronal α-secretase enhancing ability in vitro by up to 91% (P<.001) and it’s oral bioavailability in vivo by more than two-fold over free EGCG.

Conclusion: This study provides important preliminary evidence that nanolipidic particles might be useful for safely translating EGCG into human clinical trials. Not only did NanoEGCG more than double the oral bioavailability of EGCG in rats but also was more effective at promoting α-secretase activity in vitro, even at reduced concentrations. Taken together, it is possible that NanoEGCG will be therapeutically effective at doses that would be considered acceptable in the clinical setting.

Research supported by: SBIR grant (R43AT004871) from the NCAAM (JT) R21 grant (R21AG031037) from the NIA (RDS).

ApoER2 Function in Retinal Synaptic Connectivity and Aging

Presented by: Justin Trotter, BS, Faculty

ApoER2 Function in Retinal Synaptic Connectivity and Aging

Trotter, J.H.*, Herz, J.**, and Weeber, E.J.*
Dept. of Molecular Pharmacology & Physiology, College of Medicine, University of South Florida **University of Texas Southwestern Medical Center

Keywords: ApoER2, Retina, Macular Degeneration, and Synaptic Development

Objective: The function of ApoER2 in the retina has not been established, although its major ligands are involved in retinal development and age-related degeneration. We determined the function of ApoER2 in the establishment and maintenance of retinal synaptic connectivity.

Methods: Changes in the connectivity of retinal circuitry in ApoER2 mutant mice were determined immunohistologically using antibodies against cell-specific markers and electrophysiologically using the full-field electroretinogram.

Results: We observed concerted changes in Reelin, Dab1, and ApoER2 expression, localization, and splicing during retinal development that paralleled morphogenesis of both rod bipolar cells (RBCs) and All amacrine cells. ApoER2 knockout (KO) mice were found to have similar rod bipolar morphogenic defects as those found in Reelin or Dab1 KO mice. In addition, aberrant rod bipolar axons were associated with ectopic All amacrine processes and abnormal localization of the gap junction protein, connexin-36. Age-related changes in All amacrine morphology in the ApoER2 KO were also noted, with an accumulation of abnormal, lobulated neurites in the OPL and surrounding loss of rod bipolar cells. Electroretinogram (ERG) revealed a significant reduction in rod transmission in ApoER2 KO. Furthermore, we found age-dependent reductions in both rod transmission and cone function in the absence of either ApoER2 or VLDLR.

Conclusion: These results suggest an important role for ApoER2 in the establishment and maintenance of normal retinal synaptic connectivity and provide a foundation upon which the function of apoE, a putative genetic risk factor for macular degeneration, can be evaluated.
**Abstract #**: B-87

**Presented by**: Stephen Aradi, Undergraduate

**Cortical Circuitry in Mild Cognitive Impairment & Alzheimer’s Disease: Changes in Dendritic Spines**

Stephen Aradi (1), Robyn A. Long (1,2), James Kotick (2,3), Adam Winkler (4), Mrunal Shah (2,5), Aaron Lozano (1), Stephen Scheff (6), Elliott Mufson (7), Ronald F. Mervis (2,8) (1)The Honors College, USF, Tampa, FL; (2)Neurostructural Research Labs, Tampa, FL; (3)Univ Miami School of Medicine, Miami, FL; (4) USF College of Medicine, Tampa, FL; (5) Univ Florida College of Medicine, Gainesville, FL; (6) Sanders Brown Center on Aging, Univ Kentucky, Lexington, KY; (7) Dept Neurological Sci, Rush Univ Med Ctr, Chicago, IL (8)Center on Aging & Brain Repair, Dept Neurosurgery, USF College of Medicine, Tampa, FL

**Keywords**: Alzheimer’s Disease, dendritic spines, mild cognitive impairment, Golgi staining

**Objective**: Synaptic loss is a primary correlate of cognitive dysfunction in Alzheimer’s disease (AD). The vast majority of synapses on cortical neurons occur on dendritic spines. Analysis of dendritic spines therefore is a highly accurate means of assessing the integrity of cortical circuitry. Mild Cognitive Impairment (MCI) is a prodromal stage of AD. The objective of this study was to determine the pattern of spine changes on neurons in cortical brain tissue harvested from non-cognitive impaired (NCI) subjects as well as from individuals who had been diagnosed with MCI or AD.

**Methods**: Formalin-fixed tissue blocks were obtained from the superior frontal cortex (SFx, Brodmann area 9), parietal cortex (Px, areas 39-40), and inferior temporal cortex (ITx, area 21). The tissue was Golgi stained, all slides coded, and layer II-III pyramids randomly selected for analysis of spines along dendritic branch segments.

**Results**: Compared to the Non-Cognitively Impaired (NCI) group, all three cortical brain regions (SFx, Px, ITx) showed significant spine loss in AD (minus 20-30%). However, these regions showed a significant difference in the MCI phase: Spines in the frontal cortex (SFx) remained essentially unaffected in MCI, whereas there was a 23% loss of spines in the Px and a 14% loss in the ITx.

**Conclusion**: The neocortex is a heterogeneous structure with regard to the effect of the progressive damage of AD on brain circuitry. In the initial prodromal MCI phase, the frontal cortex is largely resilient to spine loss whereas the parietal cortex shows susceptibility to early disruption of brain circuitry, with the Tx being intermediate in the response to MCI. These findings largely parallel results from a dendritic branching study of AD and MCI.

**Research supported by**: Supported by AG14449

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**Abstract #**: B-88

**Presented by**: Jorge Fernandez, Undergraduate

**Oral Vaccines to Protect Against Viral Infections**

Jorge Fernandez, Honors College, USF, Alberto van Olphen, Patrick Maignan Center for Biological Defense and Global Health Disease Research Program, Department of Global Health, College of Public Health, University of South Florida

**Keywords**: Adenovirus Influenza Oral Vaccine Recombinant

**Objective**: The economic impact caused by influenza due to decreased productivity and increased health care utilization is in the billions of dollars while the social impact from terminal disease is incalculable. Thus, preparedness for a coming pandemic will require development of new vaccines and antiviral therapeutics. The purpose of this research is to investigate a new vaccination strategy for the protection against viral diseases.

**Methods**: The hemagglutinin and neuraminidase from A/Cal/04/2009, and matrix (M1) genes from H3N2 A/Wyoming/03/2003 and H1N1 A/WSN/1933 influenza viruses were cloned into pDC515, a shuttle vector for the Admax adenovirus expression system. These plasmids were each recombined with an adenovirus genomic plasmid in HEK293 cells in separate calcium chloride transfections. Recombinant adenovirus was plaque-purified twice, and purified using cesium chloride banding. Immunofluorescence using rabbit anti-HA antibodies was utilized to demonstrate expression of hemagglutinin protein in MDCK and HEK293 cells.

**Results**: To date, recombinant adenovirus containing influenza genes coding for the hemagglutinin (HA), neuraminidase (NA), and matrix (M1) proteins have been created and purified, and the expression of the HA protein by mammalian cells infected with the recombinant virus has been verified with immunofluorescence.

**Conclusion**: Future experiments will further demonstrate expression in cells infected the recombinant adenovirus we have generated. Once expression is confirmed, the virus can be encapsulated in alginate microspheres and used for testing as an oral vaccine with the goal of producing an effective, egg-free oral vaccine for use in humans.

**Research supported by**: United States Department of Defense
Comparative Immune Factors and Effects of Preterm versus Term Colostrum

Nagwa El-Badri (Dajani), Department of OB/GYN, Department of Molecular Medicine, College of Medicine, University of South Florida; Terri Ashmeade, Department of Pediatrics, College of Medicine, University of South Florida; Maureen Groer, College of Nursing, University of South Florida

Keywords: Colostrum, sIgA, cytokines, lymphocyte proliferation

Objective: The first objective was to compare key immunological factors in preterm and term colostrum. These factors included a panel of cytokines, chemokines, secretory IgA, and the effects of preterm and term colostrums on lymphocyte proliferation of PBMCs collected from healthy volunteers. A second objective was to investigate changes over time in these key immunological factors in preterm milk.

Methods: Preterm and term colostrum samples were collected on day 1 postpartum and filtered. Cytokines were measured in these samples by a 26-plex Millipore kit on the Luminex 200. sIgA was analyzed through an ELISA technique (ALPCO). PBMCs were collected and then separated by density gradient centrifugation. Lymphocyte proliferation was carried out using a standard tritiated thymidine uptake assay.

Results: Significant differences were found between preterm and term colostrums for G-CSF, IL-α, IL-8, and IP-10. Preterm colostral sIgA was significantly higher than term colostrum sIgA at day 1 postpartum. Preterm milk sIgA was highest at day 1 and declined over time through 8 days post birth. Several cytokines were correlated with sIgA in preterm milk. The lymphocyte proliferation assay stimulation index (S.I.) was significantly higher in the preterm colostrums samples compared to term colostrums. The S.I. decreased over time in the preterm milk samples through 8 days post birth.

Conclusion: Preterm and term colostra are uniquely suited to the immune status of infants. Preterm infants are born immunodeficient and human milk provides them with immune boosting factors such as cytokines and secretory immunoglobulins that are both protective and immune enhancing.

Research supported by: College of Nursing and the Department of Pediatrics

The Impact of Acute Stress on Individual, Group, and Performance Outcomes

Erin Eatough (Department of Psychology, University of South Florida), Chu-Hsiang Chang (Department of Environmental & Occupational Health, University of South Florida), Nicholas Hall (College of Nursing, University of South Florida), and Danesh Jaiprashad (Department of Psychology, University of South Florida)

Keywords: Acute Stress, Performance Outcomes

Objective: This study examined how undergoing an acute stress event may have beneficial effects for employees and workgroups.

Methods: Using a within-subjects design with pre- and post- tests, seventy-eight full-time employees completed an acutely stressful ropes course challenge.

Results: Paired samples t-tests showed significant improvements of in self-efficacy (d = .38), group-efficacy (d = .89), group cohesion (d = .72), and trust in group members (d = .56) after the acute stress event; these changes were positively related to performance.

Conclusion: Consistent with sociological findings, these results suggest experiencing stressful events together may bring group members closer and improve individual- and group-level outcomes.

Research supported by: USF Sunshine Education and Research Center.
Mild Cognitive Impairment and Alzheimer's Disease: Effect on Dendritic Branching of Cortical Neurons

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Keywords: Alzheimer's disease, Mild Cognitive Impairment, Dendrites, Neocortex, Neuroplasticity

Objective: Mild cognitive impairment (MCI) is a prodromal stage of Alzheimer's disease (AD). Cognitive dysfunction in AD correlates best with loss of synapses and disruption of brain circuitry. We assessed changes in dendritic branching in cortical tissue harvested from individuals diagnosed with no cognitive impairment (NCI), MCI, or AD. We wished to determine how (and to what extent) dendritic parameters of layer II-III pyramids from 3 different neocortical regions were affected by NCI, MCI, and AD.

Methods: Formalin-fixed tissue blocks were obtained from the superior frontal cortex (SFx, Brodmann area 9), parietal cortex (PCx, areas 39-40), and inferior temporal cortex (ITx, area 21). The tissue was Golgi stained, all slides coded, and layer II-III pyramids randomly selected for dendritic analysis of the basilar dendritic arbor. Camera lucida drawings were prepared. Sholl analysis evaluated the amount and distribution of the dendritic branching.

Results: In the ITx, there was a significant (~20%) reduction in neuronal branching in MCI compared to NCI, with an additional 5% loss in AD neurons. In the PCx pyramids, there was an initial 4% loss of dendritic branching in MCI with a further -10% in AD. In the SFx, the MCI cases showed a 7% increase in dendritic branching followed by a subsequent 21% reduction in neuronal branching in AD.

Conclusion: Different regions of the human neocortex show significant heterogeneity manifesting the effects of MCI and AD on dendritic circuitry. While neurons of AD cortex show widespread dendritic atrophy of layer II-III pyramids, in MCI, SFx neurons show a neuroplastic increase in dendritic branching while the PCx neurons show minimal atrophy, and ITx neurons exhibit a widespread loss of dendritic arborization.

Research supported by: AG14449

Microglia Reduction in the Spinal Cord of ALS Mice with Optimal Dose of MNC hUCB Cells

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Keywords: ALS, microglia, activation, MNC hUCB

Objective: Amyotrophic Lateral Sclerosis (ALS) is a fatal disease characterized by motor neuron degeneration in the spinal cord and brain. Activated microglia and reactive astocytes may contribute to the damage by releasing pro-inflammatory cytokines. Current treatments for ALS are only palliative. Cell therapy might be a promising treatment for ALS. A recent study by Garbuzova-Davis et al (2008) compared three doses (10 million, 25 million, and 50 million) of mononuclear human umbilical cord blood (MNC hUCB) cells administered intravenously into G93A SOD1 mice. Results showed that a 25 million cell dose increased mouse lifespan and delayed disease progression, likely via modulation of the inflammatory response. However, MNC hUCB anti-inflammatory properties are unknown. The aim of this study was to determine effects of MNC hUCB cell doses upon spinal cord microglia in ALS mice.

Methods: Immunohistochemical staining was performed using IBA-1 antibody for ramified microglia and OX6 antibody for activated microglia. Microglia density (MD) was measured in the cervical and lumbar spinal cord for both cell types. MD was compared between cell-treated, non-treated (media), and control (cyclosporine), and C57BL/6J control mice.

Results: Current results show reduced ramified and activated microglia density among mice receiving 25 million cells. G93A mice injected with 10 million or 50 million cells show no significant differences from media injected G93A mice.

Conclusion: These results confirm that the 25 million cell dose, previously shown most beneficial, also provides the most anti-inflammatory benefit through inhibition of activated microglia.

Research supported by: NIH STTR (Phase I) grant 1R41NS46870-01A1. SGD is a consultant and PRS is a co-founder of Saneron CCEL Therapeutics, Inc.
Application of Laparo-Endoscopic Single Site (LESS) Surgery to Foregut Disorders.
Michelle Vice, Sharon Ross MD, Sujat Dahal MD, Connor Morton BS, Kenneth Luberice BS, Michael Albrink MD, Alexander Rosemurgy MD Dept. of Surgery, College of Medicine, University of South Florida

Keywords: LESS Cumulative Experience

Objective: We have pioneered transumbilical Laparo-Endoscopic Single Site (LESS) surgery to promote superior cosmetic outcomes. This study was undertaken to review our experience with LESS surgery.

Methods: Patients were prospectively followed after LESS foregut surgery. Among many outcomes, duration of operation, conversion to conventional laparoscopy or "open", length of stay, and perioperative complications were recorded.

Results: Since 2007, 365 patients have undergone LESS surgery including 213 cholecystectomies, 61 Heller myotomies, 64 fundoplications, 2 appendectomies, 6 inguinal hernia repairs, 8 umbilical hernia repairs, 2 hepatic cyst excisions, 1 adrenalectomy, 1 distal pancreatectomy, 2 intraperitoneal mass excisions, 1 salpingectomy and 1 hysterectomy. Using the same transumbilical incision, LESS cholecystectomy was concomitantly undertaken with 4 LESS fundoplications, 2 Heller myotomies, 7 appendectomies, 5 umbilical hernia repairs, 1 salpingectomy, and 1 hysterectomy. 2 LESS cholecystectomies were done during pregnancy and 8 without general anesthesia. 27 LESS operations required additional ports, 5 were converted to "open" operations. LESS cholecystectomy was complicated by 1 bile duct injury and 1 cystic duct leak. 77% of patients were discharged the day of operation. All LESS operations were undertaken without apparent scarring.

Conclusion: LESS surgery can be safely applied to a broad range of foregut operations; it offers the benefits of conventional laparoscopy with superior cosmetic results. LESS surgery provides suitable access to all quadrants of the abdomen and pelvis, allowing multiple concomitant operations. The escalating application of LESS surgery opens new horizons for a diversity of even complex abdominal disorders.

Quantitative Approach in Comparing Back to Back Games with Performance in the NHL
Austin Witnauer, Senior Athletic Training Student; Jeff Konin, PhD, ATC, PT; Associate Professor of Athletic Training Education Program; Steve Ruhmel, Senior Athletic Training Student, Orthopaedics and Sports Medicine Department, College of Medicine, University of South Florida

Keywords: NHL Performance Study

Objective: Find and further understand the correlation of back-to-back games with performance on the second game compared to the first of NHL hockey games.

Methods: Over the course of the 2008-2009 NHL season, the investigators examined statistics for back to back games for all 32 NHL teams. The investigators generated a list of controlled variables to measure player performance. The investigators then compared second game statistics to the first game of the back to back series. The controlled variables that were accounted for include: -Shots For -Shots Against -Penalty Minutes -Wins -Losses -Overtime Losses

Results: Based on evidence and data collected, there is minimal to almost no significant drop-off of player performance in the second game of a back to back series. Although, it should be noted that all variables collected do show a slight negative outcome in performance on the second day.

Conclusion: According to the data collected, league-wide statistics show that there is no evidence of back to back game days negatively effecting performance. Although slight performance drop-offs are evident, they are minimal and do not allow the researchers to claim that performance is significantly reduced on the second game.

Research supported by: USF Athletic Training Education Program
Abstract #: B-95

Presented by: Erica Anstey, MA, Graduate Student

The Impact of Maternal Overweight and Obesity on Breastfeeding Duration Erica H. Anstey, MA Department of Community & Family Health, College of Public Health, University of South Florida

Keywords: breastfeeding lactation obesity overweight BMI

Objective: Addressing barriers to breastfeeding initiation, duration, and exclusivity are important for increasing national and global rates of breastfeeding and improving children's health. One such barrier may be the worldwide rise of obesity among reproductive-aged women, which is associated with several adverse perinatal health outcomes. This review aims to determine the association between Body Mass Index (BMI) on breastfeeding duration.

Methods: This systematic review included only empirical epidemiologic studies that (1) examined an association between prepregnant BMI and breastfeeding duration, and (2) included some clear differentiation between BMI categories (underweight, normal weight, overweight, obese). Twenty articles published between 1992 and 2009 met the study criteria.

Results: Of the 20 included studies, 9 were prospective cohort, 4 were retrospective cohort, 6 were cross-sectional, and 1 was a randomized case-control. The number of participants ranged from 57 to 37,459. The combined results support the hypothesis that there exists a dose-dependent inverse relationship between maternal BMI and duration of breastfeeding among the women studied. This decrease in breastfeeding duration is evident from descriptive statistics, survival analyses and other multivariate analyses such as logistic regression. Even when adjusting for potential confounding variables, BMI remained significant in most studies.

Conclusion: The negative association between BMI and breastfeeding duration suggested by this review is an important issue with regard to improving the health of women and infants. The obesity epidemic is a challenge for health professionals attempting to meet national and global health goals to increase breastfeeding initiation and duration.

Abstract #: B-96

Presented by: Mariana Arevalo, BA, Graduate Student


Keywords: Process evaluation, data collection, medically underserved, minority, Patient Navigation.

Objective: 1. To conduct a process evaluation on self-reported data collection procedures used by the PNRP – Tampa site. 2. To identify essential components of the data collection process, and discuss the influence of contextual factors in challenges to data collection procedures. 3. To describe some challenges to collection of self-reported data in medically underserved and minority populations, discussing the contextual factors that may contribute to these challenges.

Methods: Adults with abnormal breast and colorectal cancer screenings, were recruited from community health centers in the Tampa Bay area, as part of the Patient Navigator Program (PNRP). PNRP participants were asked to take part in a 60-minute in-person interview after their cancer abnormality had been resolved. Participants were contacted over the phone, and all contacts were recorded on a Participant Contact Sheet. A process evaluation was conducted using the collected data. MS MapPoint and SPSS were used to analyze the data.

Results: Research Assistants made up to 12 attempts to contact potential survey participants. On average, RAs made calling attempts for 30 days. 65% of all contacted subjects were scheduled for an interview. Interviews were conducted across 5 counties within the Tampa Bay Area, and 81% of participants choose to be interviewed at home. Over half of all the interviews were conducted at the end of the week.

Conclusion: Contacting potential participants to conduct survey research was a challenge in terms of participant availability and time of interview. In addition, participants’ literacy level plays an important role in the collection of self-reported data. Lessons learned will be discussed.
Abstract #: B-97  Presented by: Khaliah Fleming, BA, Graduate Student

**Barriers to Disclosing HIV + Status among Black Young Women**  
Khaliah Fleming, Department of Community and Family Health, Renee Clarke, MPH, Department of Community and Family Health  
Dr. Stephanie L. Marhefka, Department of Community and Family Health, College of Public Health, University of South Florida

**Keywords:** Barriers to HIV+ Disclosure  
**Objective:** Non-disclosure of HIV+ status to sex partners is common among Black young women. To develop appropriate interventions for this population, more research is needed to understand what hinders Black HIV+ young women (BHIV+YW) from disclosing their status.

**Methods:** Twenty HIV + young women ages 16 to 24 years were recruited from an adolescent HIV clinic in Tampa, FL; only BHIV+YW were included in these analyses. Participants completed a computerized survey of their sexual behavior and disclosure experiences. Descriptive analyses were conducted using SPSS.

**Results:** Of the 17 BHIV+YW, 4 did not disclose their status to any sex partners, and 12 disclosed to > 1 partner (with 1 person not responding). Reasons for nondisclosure included: potential lack of understanding (n = 8), fear of partner rejection (n = 6), not knowing how to disclose (n = 8), lack of confidentiality (n = 9), and fear of anger (n = 7).

**Conclusion:** Numerous concerns prevent BHIV+YW from disclosing their status to sex partners. Behavioral interventions that focus on increasing disclosure-related skills may be critical. Creative community-focused interventions may also be important for reducing stigma and changing social norms for discussions with sex partners about HIV and other sexually transmitted infections.

**Research supported by:** This research was supported by grants from the Florida Center for AIDS Research (in development) and the University of South Florida Division of Sponsored Research (PI: Marhefka).

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Abstract #: B-98  Presented by: Janet Hess, MPH, Graduate Student

**Type of Provider as a Predictive Factor in Planning for Health Care Transition**  
Janet Hess, MPH, CHES, Dr. PH graduate student, College of Public Health, and Project Director, College of Medicine, Department of Pediatrics, College of Medicine, University of South Florida

**Keywords:** Children and youth with special health care needs, disabilities, adolescent medicine, health care transition  
**Objective:** To assess type of provider as a predictive factor in planning for transition from pediatric to adult health care among YSCHCN ages 12-17 in the U.S.

**Methods:** In an analysis of the 2005-2006 NS-CSHCN, interview records from parents of 18,198 YSCHCN were examined. Provider categories included type of personal doctor (pediatrician, general doctor, specialist, nurse practitioner, physician’s assistant) and having doctors who treat only children. Using the MCHB’s measurement framework, bivariate analyses were conducted to determine whether type of provider was associated with meeting a composite transition planning outcome and 3 component outcomes: YSCHCN received anticipatory guidance from providers about future health care needs and health insurance, and providers encourage YSCHCN to take responsibility for their own health care.

**Results:** Having a pediatrician as a personal doctor or having at least one provider who treats only children significantly reduced the odds of meeting component outcomes for anticipatory guidance about future health care needs (OR: 0.75 [95% CI: 0.75-0.76], and OR: 0.70 [CI: 0.69-0.70], respectively) and health insurance (OR: 0.74 [CI: 0.74-0.75], and OR: 0.77 [CI: 0.77-0.77]). However, YSCHCN with these types of providers were slightly more likely to have met the composite outcome.

**Conclusion:** This study adds to the body of knowledge concerning MCHB’s performance outcomes for transition to adulthood. It also highlights the importance of assessing strength and direction of association between type of provider and each transition service component. Further examination of these relationships will allow us to better understand and improve the degree to which health care providers assist YSCHCN and their families in transition planning.
**Assessment of Food Insecurity among Low-Income Persons Living with HIV and Receiving Food Assistance**  
Sandra Delgado, Nicole Demetriou, Rosa Joseph, Laura Merrell, Andrea Naylor  
Department of Global Health, College of Public Health, University of South Florida

**Keywords:** PLWH, Food Security, Nutrition, Low Income, Food Assistance  
**Objective:** Low-income persons living with HIV (PLWH) are at increased risk of experiencing food insecurity. Proper nutrition has been documented to be of significant importance for increased immune functioning and protection against the progression of HIV. This assessment aimed to determine if low-income PLWH who receive minor food assistance experienced food insecurity within 30 days of being surveyed. The study also sought to identify the impact of access issues (i.e. transportation) or self-efficacy of current health status on food security status.

**Methods:** A convenience sample (N=125) from the Drug Abuse Comprehensive Coordinating Office’s (DACCO) Food Bank Program completed a paper-based survey in English (n=112) or Spanish (n=13). Questions were drawn from the NHANES and complemented by questions specific to program goals and the sample population.

**Results:** Results indicate that, despite the Food Bank program, 89% of the sample met study-based criteria for food insecurity in the past 30 days. Fifty-nine percent received additional food assistance and 30% sought emergency food sources. Differences between males and females emerged, with males reporting greater rates of food insecurity and less access to reliable transportation. English-speakers were also more food insecure than Spanish-speakers. Interestingly, 70% classified their health as good to excellent, indicating an opportunity for capacity building within the population.

**Conclusion:** It was concluded that low-income PLWH experience food insecurity related to a complex interaction of economic, political and social factors. Thus, clinicians and case management agencies must address not only the clinical impacts of nutrition but the overall impact of food insecurity and issues related to access.

**HPV Vaccination Uptake and Completion: An Analysis of Data from The National Survey for Children’s Health**  
Jaime L. Myers, Russell Kirby  
Department of Community & Family Health, College of Public Health, University of South Florida

**Keywords:** HPV human papillomavirus vaccines immunizations adolescents  
**Objective:** The Advisory Committee for Vaccine Practices recommends routine human papillomavirus (HPV) vaccination for adolescent girls ages 11 and 12 and catch-up vaccination for girls ages 13 to 26 years old. This study seeks to describe the national prevalence of HPV vaccine uptake and completion among adolescent girls ages 12-17 years old and explores factors that predict vaccine uptake and completion.

**Methods:** A secondary data analysis of data from the National Survey of Children’s Health was conducted. Parental reports of adolescent females ages 12 to 17 years old (N = 16,738) were examined. Data were weighted for national estimates.

**Results:** Approximately 19% reported having at least one dose of the HPV vaccine. Among starters, 24% completed the three-dose regimen. Doctor recommendation (p < 0.001), poverty level (p < 0.05), and having received a tetanus booster shot since age 11 (p< 0.001) predicted vaccine uptake. Adequacy of insurance (p < 0.05) and having a doctor who listens carefully (p < 0.10) predicted vaccine completion.

**Conclusion:** HPV vaccination remains low despite ACIP recommendations. Higher vaccination rates among those in the lowest poverty level may speak to the successes of the Vaccines for Children program. Increasing doctor recommendation rates offers the best opportunity to increase HPV vaccination uptake rates. Evidence suggests that strategies for increasing vaccine uptake may not be unique to HPV specifically, but rather vaccines in general. However, different factors explain vaccine uptake and vaccine completion. It is important to adopt intervention strategies designed to increase uptake and completion that are tailored to each specific vaccination goal in order to reduce rates of HPV.
Abstract #: B-101

Presented by: Sarah Smith, MPH, Graduate Student

An Exploration of Oral Health Providers’ Perceived Roles, Attitudes, and Communication Behaviors Regarding Oral Cancer and HPV

Sarah Smith, MPH1, Karen Dyer, MA, MPH1, Hannah Helmy, MA, MPH1, Hollie Fuhrmann, MA1, Rita DeBate, PhD, MPH, CHES2, Ellen Daley, PhD, MPH2,3,1

(Dept. of Community & Family Health, COPH, Dept. of Anthropology, USF); 2(Dept. of Community & Family Health, COPH, USF)

Keywords: oral cancer; human papillomavirus (HPV); dentistry; doctor-patient communication; continuing education

Objective: Recent biomedical research has uncovered a link between certain oral cancers (OC) and the human papillomavirus (HPV), the sexually transmitted infection that is a necessary factor in the development of cervical cancer. This poster will present the results of a qualitative study with oral health providers (OHP) examining their perceived roles and comfort with patient/provider communication in light of this recent discovery.

Methods: Five focus groups were conducted in February 2009 with dentists (n=3 groups, 17 participants) and dental hygienists (n=2 groups, 21 participants) in Florida to explore OHP knowledge, attitudes and perceived roles regarding primary and secondary prevention of HPV-related OC.

Results: Important themes include low levels of knowledge about HPV and its link to OC; a dissonance between OHP’ perceived role and the clinical practice of OC screening; and a need for knowledge and skills related to communication strategies about HPV-related OC. Participants acknowledged the importance of universal screening and patient communication; however, they did not report consistently practicing these behaviors—citing low self-efficacy, time constraints, and lack of professional guidelines. Many discussed the disciplinary divisions between dentistry and medicine, arguing that because of the increased recognition of the importance of oral health with regards to the rest of the body, dentistry must become more holistic.

Conclusion: Findings suggest a need to shift the current dualistic paradigm between medicine and dentistry towards an interdisciplinary one that emphasizes the oral-systemic health link. Continuing professional education is critically needed to address current gaps in knowledge and understanding of HPV-related OC.

Research supported by: Moffitt/UF

Abstract #: B-102

Presented by: David Tilley, MS, Graduate Student

Acceptability and Suitability of Technology-Based HIV Prevention Interventions for Women Living with HIV (WLH): Results from Hillsborough County, Florida

David Tilley, MS1, Stephanie Marhefka, PhD1, Jamila Ealey, MPH1, Bernice Lopez, BA,3 Akilah Benton, BA1, 2Department of Community and Family Health, College of Public Health, Hollie Fuhrmann, MA,2 2Departments of Community and Family Health and Anthropology, Colleges of Public Health and Arts and Sciences, 3 Department of Global Health, College of Public Health, University of South Florida

Keywords: HIV, Women, Technology-Based Interventions

Objective: Over the past 29 years, some HIV prevention interventions have been created to address the needs of people living with HIV. However, very few have been adapted for Internet delivery using video chatting or videoconferencing. Interactive video technologies could bring interventions to many people in areas where such programs are not available.

Methods: A convenience sample of 33 women living with HIV (WLH) was recruited from two HIV/AIDS clinics in Hillsborough County, Florida, in December 2009. WLH completed an in-person interview with a female graduate student. Respondents provided information about their health and behavior, including experience with the Internet and computers. Data collection is ongoing with the goal of recruiting 100 WLH.

Results: WLH were age 44 years on average (SD = 10 years) and the majority were racial minorities (85%). Most were comfortable or very comfortable using a computer (64%); however, one-third of the WLH reported no current Internet use. Additionally, the majority of the WLH did not have a computer in their home (63%) and had never video-chatted or participated in a videoconference (87%). However, 55% said they would be willing to attend an Internet-based intervention accessed at a community organization; another 15% said they might be willing to do so.

Conclusion: In this population of WLH, providing HIV prevention interventions via video chatting or videoconferencing would necessitate providing many of them with computers and Internet access, which may not be cost-effective. Alternatively, enabling WLH to access Internet intervention programs at local community organizations is a promising strategy for intervention diffusion. Future research should explore the feasibility and effectiveness of this strategy.
**Abstract #**: B-103

**Presented by**: Christopher Wheldon, Graduate Student

**Sexual Risk Behaviors Among Men Seeking Men Online**

Christopher Wheldon, Department of Community and Family Health, College of Public Health, University of South Florida

**Keywords**: MSM Behavior Risk-taking Internet STI

**Objective**: Recent research has focused on the Internet as a venue that may facilitate sexual risk behaviors among men who have sex with men (MSM). The purpose of this study was to identify risk behaviors associated with meeting sexual partners on the Internet.

**Methods**: A cross-sectional Internet-based survey was used to collect data for this study in the spring of 2004. The sample included 1,026 sexually active MSM recruited from a variety of Internet venues. Logistic regression analysis was used to estimate odds ratios in order to identify predictors of using the Internet to meet sexual partners within the previous six months.

**Results**: Respondents ranged in age from 18 to 54 (M = 36, SD = 12). The majority of men were white (83%), college-educated (60%), and identified as homosexual or gay (81%). More than 6 out of 10 of the respondents reported having engaged in sexual activity with a partner they met online in the previous six months (n = 628; 61%). Men with Internet partners were more likely to have used Amyl Nitrate inhalants (“Poppers”) during sexual activities (OR = 1.50; 95% CI = 1.07-2.10), more likely to have had unprotected anal intercourse with more than four partners in the previous six months (OR = 5.00; 95% CI = 2.02 – 12.16), more likely to have ever met a sex partner at a bath house (OR = 1.51; 95% CI = 1.11-2.06), and to have been diagnosed with an STI in the previous six months (OR = 3.13; 95% CI = 1.34-7.32).

**Conclusion**: These findings suggest that MSM who meet sex partners online engage in a number of high-risk sexual behaviors that may lead to the transmission of STIs. Internet-based interventions tailored for MSM who meet sex partners online are needed to reduce sexual risk-taking.

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**Abstract #**: B-104

**Presented by**: Amanda Evans, BS, Graduate Student

**An Investigation of Small-Scale Spatial Variability in Aldehyde Concentrations Through Passive Sampling and Analysis**

Amanda M. Evans and Amy L. Stuart PhD, College of Public Health, Department of Environmental and Occupational Health, University of South Florida

**Keywords**: Aldehydes, Restek Allure AK HPLC column, spatial variability, passive sampling

**Objective**: Regulatory monitoring networks for air toxics are typically spatially sparse. However, to understand and mitigate exposures, it is important to quantify ambient concentration distributions at small spatial scales. Here, a pilot study was designed and performed to investigate variability in concentrations of aldehydes.

**Methods**: A literature review was first done to select and evaluate current passive sampling and analysis methods. Radiello diffusive 2,4-dinitrophenylhydrazine (DNPH) cartridges for the sampling and high performance liquid chromatography (HPLC) for the analysis were chosen. Multi-point calibration curves were established. A spatial sampling network was then designed for an urban census block group that also contained a currently operating regulatory monitor for both ozone and carbonyls. Collected samples and blanks were eluted with acetonitrile and analysis was performed with the HPLC. Concentrations of formaldehyde and acetaldehyde in air were determined using sampling rates that were corrected for temperature and ozone concentration.

**Results**: Overall sample descriptive distribution statistics and the spatial distribution pattern were investigated to characterize spatial variability within the block group. Measured concentrations were also compared with values at the nearby regulatory carbonyl monitor.

**Conclusion**: In order to minimize exposure misclassification in epidemiological studies it is important to have accurate exposure metrics. Hence, for appropriate use to determine exposures, the spatial scales that concentration measurements represent must be understood. Results of this pilot study provide data on the variability of formaldehyde and acetaldehyde concentrations over the census block group scale.

**Research supported by**: NSF Grant 0846342
Dust and Silica Exposures Resulting from Gravel Mining Operations  Laura Farina, Dr. Steven Mlynarek, PhD, CIH  Department of Environmental & Occupational Health, College of Public Health, University of South Florida  

**Keywords:** Aerosol Silica Dust Particulate Respirable  

**Objective:** Assess total particulate, respirable particulate, and the course content sampled particles.  

**Methods:** Traditional NIOSH methods for total and respirable particulate sampling. The results obtained from these methods will be compared to a new, real-time instrument known as the EPAM 5000.  

**Results:** Dust levels measured using the NIOSH methods were all below the limit of detection. There were measurable dust levels in all three sizes (PM10, PM2.5, PM1) for the EPAM 5000.  

**Conclusion:**  
- There was no visible dust in the air on any of the five sampling days.  
- Traditional NIOSH methods resulted in levels below the limit of detection for both total and respirable particulates for all five sampling days.  
- No significant silica content was found using traditional methods.  
- The EPAM 5000 has a limit of detection lower than the OSHA methods, and was able to measure particulate mass at all three size fractions.  
- Size distribution of indoor particulate matter in homes can range from 0.02 to 0.06 mg/m³. The samples obtained from the mining operation were all within these limits or lower.  
- In order to compare the traditional NIOSH dust sampling methods with the EPAM 5000 unit, more data collection is needed.

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Modeling the Environmental, Economic and Socio-Cultural Dynamics of Mercury  *Ryan Michael, Dept. of Environmental and Occupational Health, College of Public Health, University of South Florida  **Amy L. Stuart, PhD, Dept. of Environmental and Occupational Health, College of Public Health, University of South Florida*  

**Keywords:** Mercury cycling, Methylmercury, industrial ecology, systems modeling, sustainability  

**Objective:** Due to its high mobility, mercury is a global pollutant whose effects may be far removed from the source of emissions. Deleterious human health effects include kidney damages, impaired mental function, and neurological disorders, particularly to the fetus. The overarching goal of this study is to improve understanding of the social, cultural, economic, and environmental factors affecting community sustainability related to mercury use, contamination, and human exposures, with a focus on applying mathematical modeling tools.  

**Methods:** To date, our methods have included literature review and investigation of a few modeling tools. Specifically, we have applied a mass balance-based life cycle inventory approach to explore mercury sources and releases in the Tampa Bay area. We are also using receptor and trajectory modeling to understand and allocate sources. Using STELLA systems modeling software, we have begun to study the interaction of physical, chemical, and socioeconomic interactions contributing to mercury cycling in two case study areas. Finally, the Comsol Multiphysics fluid dynamics model is being applied to study mercury cycling in a model reservoir.  

**Results:** Preliminary investigations reveal that biogeochemical cycling of mercury is significantly impacted by industrial emissions in some locations, whilst others are affected by small-scale economic, socio-cultural, and religious activities.  

**Conclusion:** A sustainable solution to mercury pollution likely requires a balance between economic, cultural and social, and environmental needs  

**Research supported by:** University of South Florida, Sustainable Healthy Communities grant.
Measurement of Tensile Force in a Retractable Intramuscular Syringe: A Comparison of Two Methods
Rachel Williams, (USF College of Public Health, Department of Environmental and Occupational Health), Donna Haiduven (USF College of Public Health, Department of Global Health), Shawn Applegarth (VA Patient Safety Center), and Margaret Tenouri (USF College of Public Health, Department of Global Health)

Keywords: engineered sharps injury protection needlestick safety tensile force

Objective: One of the most widely used devices with engineered sharps injury protection(ESIP) is the retractable intramuscular syringe. The objective of this study was to measure the tensile force required to draw fluid into the barrel of a retractable intramuscular syringe, using two different testing methods.

Methods: A laboratory-based experiment was conducted on one commercially available 3cc retractable intramuscular syringe. Researchers tested devices using two different methods: 1) a manually-operated digital force gauge and 2) a computer-controlled universal testing machine, to measure the tensile force in lbs. required to draw 2cc of saline into the barrel of the syringe. A total of 200 devices were tested, 100 with each of the two methods. Data were analyzed using descriptive statistics and independent t-tests.

Results: There was a statistically significant difference (p < 0.000) between the mean tensile forces when comparing methods 1 and 2. The mean difference between the two methods was 0.116 lbs. The confidence interval was 0.077 – 0.154. The range and standard deviation of tensile force in lbs. for methods 1 and 2 was 1.10-1.84 and 0.145, and 1.14-2.03 and 0.130, respectively.

Conclusion: This study identified two methods for assessing one component of the forces involved in the injection procedure. The contribution of tensile force to the total force exerted by healthcare workers, and any potential injury risk, requires further investigation. Future evaluations of devices with ESIP should include tensile force in the multiple criteria for laboratory-based testing. This information has implications for healthcare facilities in choosing the optimal sharps devices to protect their workers.

Research supported by: Sunshine ERC

Air Pollutant Dispersion Modeling and Analysis of Equity Impacts in the Tampa Area
Haofei Yu, and Amy L. Stuart, Department of Environmental & Occupational Health, COPH, University of South Florida

Keywords: air quality, exposure, NOx, environmental equity

Objective: Air pollution is recognized as a significant problem because of its adverse health effects. Some population subgroups have been found to be disproportionately exposed to air pollution based on comparison of residential location with surrogates of pollutant concentration. One of the common air pollutants, nitrogen oxides (NOx) was chosen here for analysis. The spatial distributions of its concentrations in outdoor air in Tampa is modeled and compared to spatial distributions of population demographics for investigation of potential inequities in exposure.

Methods: The CALPUFF air pollution dispersion model is used to determine NOx concentration footprints resulting from point sources and roadways for 2002. Emissions parameters and amounts for point sources are obtained from the 2002 US National Emissions Inventory. Roadway emissions are estimated from 2002 Florida traffic data and the MOBILE 6.2 emissions factor model. Simulated pollutant concentrations are compared to spatial distributions of selected population subgroups using mapping and spatial analysis tools in ArcGIS. An equity index is calculated to quantify potential disparities.

Results: The distribution of NOx concentration follows the pattern of major roadways instead of the location of the point sources, even though the emission rate from point sources is about three times the roadway emission rate. The highest concentration can be found along the busiest roads such as I-75, I-4 and I-275.

Conclusion: Potential inequities in residential exposures to the highest modeled NOx concentrations were found for the below property group while equities issue for other subgroups are relatively complex.

Research supported by: NSF grant CBET-0846342 University Graduate Fellowship COPH Doctoral Fellowship Research Computing
**Private Plans in Medicare: Do Medicare Advantage Plans Ensure Access to Primary Care for Florida Seniors?**

Meg Comins, MPA, Barbara Langland Orban, PhD, Etienne E. Pracht, PhD
Dept. of Health Policy & Management, College of Public Health, University of South Florida

**Keywords:** Medicare Access Primary Care

**Objective:** Recent polls show that Medicare enrollees are happier with traditional Medicare than with private Medicare Advantage (MA) plans, based on perceptions of better access to care. Primary care providers are often paid less through MA plans which may delay or prevent access to care. This paper examines trends in inpatient clinical and demographic characteristics of Medicare enrollees to assess whether problems exist with access to primary care.

**Methods:** The primary source of data was 2005 and 2007, hospital discharge and financial records of Medicare and MA enrollees from the Florida Agency for Health Care Administration. ANCOVA analysis was run to test for differences between the groups and over time.

**Results:** MA patients are more likely to be admitted to an Emergency Department (p<.001), with the difference between groups decreasing over time. MA patients are more likely to spend less than 2 days, and less likely to spend 7 or more days in the hospital, p<.001. More MA patients are 79 or younger.

**Conclusion:** More admits from the emergency department point to limited access to care. Problems with access to care can be partially attributed to low payments to physicians from the MA plans. A solution may be to require that a portion of payments to MA plans be used to increase plan payments to primary care physicians; this would likely increase access to care and reduce emergency department use. What must also be considered is that MA plans are attractive to lower SES groups that are not able to afford supplemental premiums. Higher ED admissions for MA enrollees may support past research on the association between lower SES and reliance on the ED as a source for routine health care.

**Psychological Factors and Persistence of HPV in Men Participating in a Natural History Study**

Department of Community and Family Health, College of Public Health, University of South Florida

**Keywords:** HPV, persistence, psychological, men, psychoneuroimmunology

**Objective:** The field of psychoneuroimmunology focuses on the link between psychological and physiological processes and suggests that psychological variables could affect immune function. Persistent HPV infections are a necessary cause of cervical cancer and are associated with numerous other cancers including oral, penile, and vulvar cancers. Little is known about psychological factors and their association with HPV persistence.

**Methods:** Males participating in a natural history study of HPV also completed a psychosocial questionnaire which included questions on avoidance and negative emotions related to receiving an HPV test result. HPV+ participants were followed to their next study visit, approximately 6 months.

**Results:** Of the 84 HPV+ participants, 59 remained HPV+ and 25 tested HPV-. Avoidance was not associated with clearance of HPV infection, RR=0.99, 95%CI: 0.74-1.33. Compared to participants who reported experiencing at least one negative emotion, those who did not were significantly more likely to clear their HPV infection, RR=3.18, 95% CI: 1.42-7.15. There were no differences in clearance by marital status, race, ethnicity, or education. Participants who cleared their infections were younger, but this difference was not significant.

**Conclusion:** Although this analysis has several limitations – including the unique aspect of this natural history study in which males receive HPV test results – it suggests that psychological factors may be associated with HPV persistence. Future research is needed to determine the effect of other psychosocial and behavioral factors such as stress, sleep, coping, and social support which may impact HPV persistence. Determining factors associated with HPV clearance could lead to interventions among patients persistently HPV+.
Optimization of Weights in Non-Parametric ROC Comparison for Repeated Biomarkers

Ping Xu MPH, Dept. of Epidemiology and Biostatistics, Yougui Wu PhD, Dept. of Epidemiology and Biostatistics, College of Public Health, USF; Yiliang Zhu PhD, Dept. of Epidemiology and Biostatistics, College of Public Health, USF; Getachew Dagne PhD, Dept. of Epidemiology and Biostatistics, College of Public Health, USF; Jeffrey P. Krischer PhD, Pediatric Epidemiology Center; Craig Beam PhD, Pediatric Epidemiology Center

Keywords: weight optimization, ROC, repeated measures

Objective: Receiver Operating Characteristic (ROC) curves are often used to compare the performance of biomarkers, which might be used to supplement or replace standard clinical examinations. In a previous study, a non-parametric ROC approach was introduced to compare biomarkers with repeated measurements. An asymptotically normal statistic, which contains the subject-specific weights, was developed to compare the areas under ROC curve of biomarkers. Although two weighting schemes were suggested to be optimal when the correlation within is 1 or 0 by the previous study, the universal optimal weight was not determined.

Methods: We propose a solution to weight optimization in non-parametric AUCs comparison to improve the efficiency of the estimator. It is demonstrated how the Lagrange multiplier can be used as a strategy for finding the weights which minimize the variance function subject to constraints.

Results: We show substantial gains of efficiency by using the novel weighting scheme when the correlation within biomarker is high, the correlation between biomarkers is high, and/or the incidence of disease is low, which is the case for many diseases. An illustrative example is presented to apply the proposed methodology to a diabetes dataset.

Conclusion: Simulation results suggest that the optimal weight performs well with a sample size as small as 40.

A Characterization of the Cytokine Response in a Monocytic U937 Cell-line Infected with Dengue Virus Serotypes 1-4 Using a 27-plex Microsphere-based Immunoassay (MIA)

Jason H. Ambrose, MPH, (University of South Florida, College of Public Health, Tampa, FL; Florida Department of Health, Bureau of Laboratories, Tampa, FL), Lillian M. Stark, PhD, MPH, (University of South Florida, College of Public Health, Tampa, FL; Florida Department of Health, Bureau of Laboratories, Tampa, FL), Kelly Fitzpatrick, MPH, (Centers for Disease Control and Prevention, Atlanta, GA), Azliyati Azizan, PhD, (College of Public Health, Tampa, FL), University of South Florida

Keywords: dengue virus, cytokines, microsphere-based immunoassay (MIA), arbovirus, hemorrhagic fever

Objective: Dengue virus (DENV) is a major cause of morbidity worldwide and severe forms increase the risk of fatal outcomes. Cytokines are thought to play a key role in severe manifestations of dengue. Severe illness is also correlated with enhanced infection of monocytes and macrophages. Characterization of these responses may help to elucidate the immunopathogenic mechanisms responsible for these severe forms of dengue. Here, we report the characterization of the cytokine response in a human histiocytic lymphoma cell-line (U937) infected with DENV serotypes 1-4 using a 27-plex cytokine microsphere-based immunoassay (MIA).

Methods: DENV-1-4 were used to infect U937 cell cultures at a multiplicity of infection of 10. Samples from aliquots taken at 6 time points post-infection (p.i.) were assayed using a 27-plex MIA for quantification of selected cytokines. The presence of DENV RNA in samples was confirmed by RT-PCR.

Results: A number of cytokines that are thought to be related to severe DENV illness were found to be elevated in U937 cell culture supernatants, including IL-1ra, IL-8, IL-10, IL-12, TNF-α, GM-CSF, MCP-1, RANTES, PDGF-bb, and VEGF. These cytokines were seen to be most elevated at either 24h or 144h p.i. and are primarily involved with the processes of inflammation and/or angiogenesis. Interestingly, these responses were not identical across the four DENV serotypes, indicating serotype-specific responses.

Conclusion: The cytokine response observed in the U937 cell-line infected with DENV-1-4 may represent that of monocytes infected in vivo. Characterizing these responses in other models will help further this understanding of the immunopathogenesis of DENV infection.

Research supported by: Southeastern Center for Emerging Biologic Threats (SECEBT)
Core Promoter Function in Brugia malayi  

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Keywords: Filariasis, Promoter, Transfection, Trans-splicing

Objective: Previous studies have indicated that the promoters of the human filarial parasite Brugia malayi are unusual in that they do not exhibit the CAAT or TATAA sequences usually found in the core domains of promoters of most eukaryotic organisms. Analysis of the promoters of the ribosomal proteins showed that the region flanking the splice leader (SL) addition site plays an important role in transcription and may function as the core promoter domain in B. malayi. To test the hypothesis that the SL addition domain is the most important essential region of the ribosomal protein promoters, the SL addition site of the BmRPL13 gene was replaced with the SL addition domains from other ribosomal protein genes from B. malayi.

Methods: The promoter activity of the replacement constructs were tested using a transient transfection dual luciferase assay.

Results: Of the three SL domains tested in this manner, all exhibited activity that was statistically indistinguishable from that of the native promoters from which they were derived, confirming the hypothesis that the SL domain is the most important determinant of promoter activity in these genes.

Conclusion: Tests for statistical significance and principal component analysis will be applied to the data once the analysis of the promoters of the remaining genes is complete.

Understanding the Health Needs of Newly-Settled Iraqi Refugees  

Tiffany Crawford, in partnership with Gulf Coast Jewish Family Services, Inc. Dept. of Global Health, College of Public Health, University of South Florida

Keywords: Refugee health, Iraqi health needs

Objective: This study aimed to better understand the health needs of a growing refugee population in Hillsborough County as well as potential gaps in services. Iraqi refugees represented the single largest refugee group resettled in the United States in 2008 with 13,755 new arrivals and they will continue to be resettled in large numbers.

Methods: This was a qualitative study designed to describe the experiences service providers have had in working with their Iraqi refugee clients and their perceptions of their clients’ needs. A one-time interview was conducted with a non-random, convenience sample (n=4) where participants were recruited through Gulf Coast Jewish Family Services, Inc. The four case studies offer an initial investigation into the experiences of some service providers with their Iraqi clients and can serve as the basis for a larger study in the future.

Results: Challenges of limited healthcare plans, challenges of the American healthcare system and differences between Iraqi refugee clients and other refugees were described by their service providers. These insights were combined with data on the current health status of Iraqis to develop recommendations for service providers working with this population.

Conclusion: Recommendations for improved services include cultural training for case workers, more resources for healthcare navigation services, chronic disease interventions and sexual health education. CBPR should be used to inform appropriate intervention strategies. Training for healthcare providers and Public Health practitioners on the health beliefs of practicing Muslims is also needed. Public Health practitioners should be aware of common health beliefs among this group and incorporate these into any educational program or intervention.
**Abstract #**: B-115  
Presented by: Ligia Cruz, MD, MPH, Graduate Student

*Inactivation of Ascaris suum by Ammonia under Laboratory Conditions* Ligia Cruz, MD, MPH, Ricardo Izurieta, MD, Dr.PH, MPH, Department of Global Health, College of Public Health, University of South Florida

**Keywords**: Sanitation, Ascaris suum, inactivation, ammonia

**Objective**: To determine the inactivation of Ascaris suum by ammonia in a solution simulating the physical-chemical (pH =9; 28°C - 45°C) parameters achieved in the solar toilet prototype IV

**Methods**: Ascaris suum ova, distributed in permeable nylon bags (10,000 ova per bag), were placed in triplicates tubes at different ammonia concentrations (1%, 2%), pH value (7, 9) and temperature (28°C, 35°C, 40°C, and 45°C) for a period of three days. After the treatment period, pH and ammonia value were measured and Ascaris suum ova were incubated for 21 days. Ova viability was assessed at the end of incubation by microscopy. A minimum of 100 A. suum ova per sample were observed and the percentage of viable ova were calculated.

**Results**: At 45°C all A. suum ova were inactivated independently of pH and ammonia value. At 28°C the maximum inactivation reached was 73.5%. Most samples placed at a pH 9 with a 1% ammonia concentration and temperatures of 35°C and higher achieved 99% to 100% inactivation. All samples with 2% ammonia concentration, pH of 9, and temperatures of 35°C and higher, achieved 100% A. suum ova inactivation.

**Conclusion**: The critical physical and chemical parameters required for a 99%-100% A. suum inactivation found in this pilot test were: a temperature of 35°C or higher, a 1-2% ammonia concentration in water, and a pH value of 9 for a period of three days. These data will be used as a baseline to compare future studies in the laboratory and the field in an effort to evaluate parasite inactivation in dry sanitation systems.

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**Abstract #**: B-116  
Presented by: Benjamin Klekamp, BA, Graduate Student

*Development of an NS1-specific ELISA as a Drug Screening Tool Against Influenza* Benjamin Klekamp, Theresa Trindade, and Alberto van Olphen, Center for Biological Defense and Global Health Infectious Disease Research program, Department of Global Health, College of Public Health, University of South Florida.

**Keywords**: Influenza, ELISA, nonstructural protein one (NS1), drug screening

**Objective**: In today's highly mobile society, influenza is a serious threat to the global economy, healthcare system, and population. Perpetual mutation and reassortment of the influenza virus allows development of resistance to anti-influenza drugs and may nullify the effect of future vaccines due to the current outdated and lengthy manufacturing process. Cell-based screening assays used to identify antiviral compounds rely on measurement of cell viability, rather than on direct quantification of virus progeny. The purpose of this study was to develop a sensitive screening assay for measurement of virus burst which is needed to aid in the identification of anti-influenza virus compounds. Quantification of NS1 protein with an ELISA would measure production of progeny virus, without interference from input inoculums. This assay could be used in conjunction with a cell viability test to improve current antiviral screening assays.

**Methods**: The work presented here describes the expression, and purification of recombinant NS1 protein from influenza strains A/WSN/1933 and A/Wyoming/03/2003, generation of anti-NS1 polyclonal antibodies and development of NS1-specific ELISA.

**Results**: Our results indicate that this NS1-specific ELISA can accurately quantify the expression of NS1 during viral infection.  

**Conclusion**: This sensitive and specific immunoassay will serve multiple purposes in Influenza research including drug screening. This study is the foundation for development of assays that distinguish between vaccination and natural infection, a pre-requisite for the evaluation of vaccine efficacy in the general population.

**Research supported by**: Department of Defense
Verification and Evaluation of Real-time PCR for the Serotyping of Dengue Virus Serotypes 1-4
Elisabeth S.Y. Lim, BS, Jason H. Ambrose, MPH, Lillian M. Stark, PhD, MPH, (Dept. of Global Health, College of Public Health, University of South Florida, Tampa, FL; Florida Department of Health, Bureau of Laboratories, Tampa, FL, Yves Frantz Jean Louis, Jr., MPH, Azliyati Azizan, PhD)(Univ. of South Florida, College of Public Health, Tampa, FL)

Keywords: dengue serotyping, real-time RT-PCR, DENV, TaqMan, arbovirus

Objective: Currently, 50 to 100 million infections of dengue viruses (DENV) occur worldwide. A recent outbreak in the FL Keys prompted the need for a real-time RT-PCR assay for diagnostic purposes at the Florida Department of Health laboratory (FDOH-BOL). Real-time PCR offers advantages due to its sensitivity, specificity, rapidity, and ability to quantify RNA.

Methods: RNA extracts from DENV serotypes 1-4 were analyzed using group-specific primer/probe set, followed by the serotype-specific primer/probe sets using TaqMan real-time RT-PCR. Optimal concentrations of primer/probe sets, sensitivity, specificity, and efficiency of the RT-PCR assays were assessed on the ABI7500FX. Assays for cross-reactivity between DENV serotypes as well as for other arboviruses endemic to Florida were performed.

Results: Optimal concentrations and thermal cycling conditions of primer/probe sets for detection of group-specific and serotype-specific DENV were determined. The specificity of the assay was found to be 100% when tested against a variety of related arboviruses. No cross-reactivity between DENV serotypes was found for the serotype-specific primer/probe sets. The efficiency of the PCR reaction for all primers/probe sets were greater than 99%.

Conclusion: With the report in 2009 of the first cases of locally acquired dengue within the state, it now has the potential to become an urgent public health concern in FL. In this study, we verified, optimized, and validated a one-step real-time RT-PCR method for the detection of DENV serotypes using specific primer/probe sets. Incorporation of the real time RT-PCR method will improve the diagnostic capabilities at the laboratory level.

Research supported by: Southeastern Center for Emerging Biologic Threats (SECEBT)

Acinetobacter baumannii Outbreak in Primary Care and Long Term Acute Care Settings
Christen L Mayer, Department of Community & Family Health, College of Public Health, University of South Florida

Keywords: Acinetobacter, outbreak, nosocomial, infection control

Objective: This research outlines the identification and management of simultaneous nosocomial A. baumannii outbreaks in a community based primary care hospital and its affiliate long term acute care facility from April 2009 to January 2010.

Methods: A. baumannii was identified in 24 patient specimens, 9 from the primary care hospital and 15 from the long term care facility using the Vitek 2 organism identification system (bioMerieux, Durham NC). These patients had been negative for the organism prior to admittance to the hospital. The patient isolates were sent to a reference laboratory for PFGE cluster typing (Tampa General Hospital Tampa, FL). Environmental cultures were performed (Rodac media, BD-Diagnostics, Franklin Lake NJ) to identify the pathogen source.

Results: Ten isolates tested were identified as A. baumannii cluster Type A with 5 patients at the primary care hospital and 5 at the long term care facility. Seven of the isolates were identified as A. baumannii cluster Type O with 6 at the long term care facility. Environmental cultures grew A. baumannii from sinks, toilets, and bed handrails of patient rooms at the long term care facility after the rooms were sterilized and cleared for a new patient.

Conclusion: A nosocomial outbreak of A. baumannii occurred at the primary and long term care centers. Two genetic clusters were responsible for the majority of the outbreak. Both clusters appeared at both facilities suggesting transmission of pathogen between the two facilities by patients, personnel, or supplies. A. baumannii was discovered in clean patient rooms highlighting the need for proper sterilization techniques and ongoing personnel infection control education to prevent the spread of pathogenic organisms.

Research supported by: University Community Hospital
Abstract #: B-119

Presented by: Christine McGuire-Wolfe, MPH, Graduate Student

**Practices, Attitudes, and Belief of Emergency Medical Services Personnel Regarding Hand Hygiene**
Christine McGuire-Wolfe (1), C. Duncan Hitchcock (2), Donna Haiduven (1). (1) USF-COPH-Dept. of Global Health (2) Pasco County Fire Rescue, Pasco County, FL

**Keywords:** Hand hygiene; Emergency Medical Services; Fire Department; Alcohol based hand gel

**Objective:** The primary objective of this study was to document the practices, attitudes, and beliefs of personnel regarding hand hygiene in a joint EMS/fire department setting.

**Methods:** The sample consisted of paramedics and emergency medical technicians from a suburban, county fire department providing fire and EMS response in Florida. A one-page survey was delivered to 387 personnel at 21 fire stations. Participants returned the anonymous surveys through the inter-office mail system. Data were analyzed and descriptive statistics generated using EpiInfo 6.0.

**Results:** A total of 228 surveys were completed, representing a 58.9% response rate. Over 50% (131) of participants indicated they had not received any training on hand hygiene from the fire department. In various questions, 90 to 93% of respondents demonstrated a perception of personal risk due to pathogens encountered on the job, as well as the potential negative impact of poor hand hygiene. Responses to additional comments regarding frequency of hand hygiene revealed important differences between self-reported practices and observations of the practices of others.

**Conclusion:** EMS workers in this sample demonstrated an understanding of the importance of and reported consistently practicing hand hygiene, but indicated concern that co-workers were not consistently doing so. This descriptive study is an important first step in documenting the perceptions and reported practices of EMS providers regarding hand hygiene, providing a foundation for future interventions to improve methods, rates, and consistency of hand hygiene in this sample. Additional efforts should aim to understand the practices, attitudes, and beliefs of EMS personnel regarding hand hygiene in a variety of practice settings.

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Abstract #: B-120

Presented by: Janus Patel, MPH, Graduate Student

**Characterization of the Optical Properties of P. falciparum, Leishmania spp., and Other Infectious Diseases**

**Keywords:** spectrometry, malaria, leishmania, optical

**Objective:** Objective of this research is to characterize the optical properties of various pathogens for use in developing a device for rapid detection, diagnosis, and quantification of disease pathogens.

**Methods:** Pathogens were grown in a laboratory and tested with a spectrometer. Curves were analyzed and a theoretical model was created.

**Results:** P. falciparum has distinct spectral features for the different stages. The same can be said for leishmania parasites and even trypanosomes.

**Conclusion:** In conclusion, P. falciparum has spectral features that show the pigment hemozoin quite clearly in a first-derivative of the measured spectra. The nucleic acid material was also very pronounced in the spectra. These factors can all be put into a theoretical model that recreates spectra based on various parameters, such as nucleic acid material, vacuole size and volume, and hemozoin content. These factors will considerably aid in the development of a rapid diagnostic test that utilized a novel miniaturized spectrophotometer.

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Measuring Hand Sanitation in an Airport Setting  
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Keywords: Infectious Disease, Travel, Influenza, Globalization, Public Health

Objective: To determine baseline rates of hand-washing among restroom users in an airport setting.

Methods: The authors devised and conducted an observational study to determine the rates of hand-washing in an airport setting. They positioned themselves in and around the restrooms at Tampa International Airport (TIA) and Raleigh Durham International Airport (RDU) over 40 hours.

Results: The combined observed rate of hand-washing with at least water usage for both males and females was 71.7%. The rate for males using at least water was 62.8%. The rate for females using at least water was 80.2%. Of that initial 80.2% of females who used water, only 68.7% used soap and dried their hands with paper towel. This equates to 55.1% of the total of sample of women.

Conclusion: Public health practitioners should review the barriers to appropriate hand hygiene, such as the deficiency of supplies and scientific information that demonstrates the impact of enhanced hand hygiene, on the health of the traveling population. Moreover, they should factor in the environmental and time constraints of the traveling population, specifically how high-stress situations and overcrowding may reduce the likelihood of soap and water use. Subsequent research and a framework for change should consider these restrictions and any subsequent hand hygiene promotion activities, which are well warranted, should seek to directly address them.

Factors Influencing Nutritional Behaviors Among Local High School Students: A Needs Assessment  
Kansas Sparks, Christina Karges, Jennifer Peregrine, Elizabeth Helfert, Stacey Owiti, Rachel Pyngolil, Klaus Ryding, and Jaime Corvin Department of Global Health, College of Public Health University of South Florida

Keywords: Adolescent, nutrition, behavior, needs assessment

Objective: Previous studies illustrate the importance of nutrition in adolescent development. As adolescents spend the majority of their day in school, the nutritional value of meals is a public health concern. Thus, this study aimed to assess factors influencing dietary intake, nutritional attitudes and behavior among local high school students.

Methods: Self-reported questionnaires were administered to a convenience sample of 55 sophomore and junior students ranging in age from 14 to 18 years. Questionnaires contained 47 items addressing opinions of school lunch options, eating behaviors, and factors influencing adolescent food choices. Data was analyzed by standard statistical techniques including frequencies and means followed by Pearson correlation coefficients.

Results: Students reported eating 1.80±1.65 servings/day of fruits and vegetables, which is significantly lower than the FDA’s recommendations. Students also reported drinking an average 1.46±1.49 servings of non-diet soda on a weekly basis. Results showed general dissatisfaction with the lunch options provided. Students strongly agreed that the number and length of lunch lines affected their food choices, with the majority agreeing that there was not enough time to purchase and eat lunch during the lunch period.

Conclusion: Results suggest the need for further nutritional assessments in the Hillsborough County School District. Preliminary findings highlight nutritional needs expressed by students and findings may help inform school administration of potential adjustments to the current nutrition policy.
A Cross Sectional Study of Prescription Drug Abusers in Treatment Programs in Sarasota, FL
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Jaime Corvin PhD (Global Health) College of Public Health, University of South Florida

Keywords: Prescription drug abuse

Objective: The nonmedical use of prescription pain relievers is second most common form of illicit drug use in the United States. Sarasota County has one of the highest rates of unintentional poisoning deaths and hospitalizations from prescription drug overdoses in Florida. This study was designed to describe the population of prescription drug abusers currently seeking treatment in Sarasota County, the sources of their abused medication, and their perceptions that may have influenced their drug abuse.

Methods: The study was conducted using a self-administered anonymous questionnaire developed for use with an online survey software.

Results: A preliminary descriptive analysis of 27 survey respondents revealed that most of the respondents abused narcotics (96.3%) or tranquilizers (70.4%), 69% abused both drugs. Consistent with national population based studies, the most common source of medication was from friends or family for free. Purchasing the drugs from friends or dealers contributes significantly to their sources of drugs, greater than previously cited in national studies. Among the study population, receiving medication from a single doctor and/or doctor shopping is not demonstrated to be a primary source of abused medication. Additionally, this study has revealed several indications that abusers were exposed to a drug culture among friends, with greater than 80% of respondents reporting being around someone taking narcotics or tranquilizers in the past year.

Conclusion: The interpretation of this preliminary data analysis is limited due to the small sample size and specific recruitment criteria. Since the initial analysis, a total of 83 surveys have been collected and are undergoing analysis.