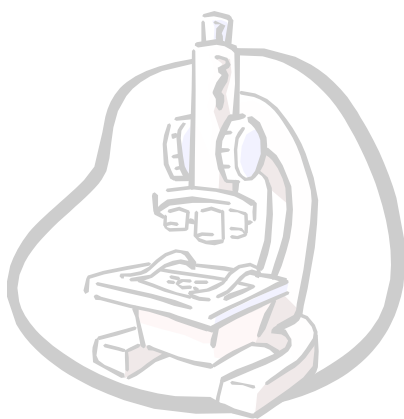


2005

Summer Research Program

STUDENT ABSTRACTS



THE UNIVERSITY *of* TEXAS

HEALTH SCIENCE CENTER AT HOUSTON

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Preface

The University of Texas-Houston Medical School (URHSC-H) Summer Research Program provides intensive, hands-on laboratory research training for MS-1 medical students and undergraduate college students under the direct supervision of experienced faculty researchers and teachers. These faculty members' enthusiasm for scientific discovery and commitment to teaching is vital for a successful training program. It is these dedicated scientists who organize the research projects to be conducted by the students.

The trainee's role in the laboratory is to participate to the fullest extent of her/his ability in the research project being performed. This involves carrying out the technical aspects of experimental analyses, interpreting data and summarizing results. The results are presented as an abstract and are written in the trainees' own words that convey an impressive degree of understanding of the complex projects in which they were involved.

To date, more than 1,400 students have gained research experience through the UTHSC-H Summer Research Program. Past trainees have advanced to pursue research careers in the biomedical sciences, as well as gain an appreciation of the relationship between basic and clinical research and clinical practice.

This year a new program was initiated for international medical students from schools with whom we have cooperative agreements. These international medical students perform research over an eight-week period and participate in all of the Program's supplemental activities. Abstracts submitted by the international medical students are in this publication (see: *International Medical Students* section.)

Student research training is supported by a grant from the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) and by financial support from the Dean and the departments and faculty of the Medical School.

Science education remains a vital and integral part of our nation's interests. The UTHSC-H Summer Research Program, and the dedication of our faculty and administration exemplify the institution's commitment to training and educating the future leaders in our scientific communities.



Gary C. Rosenfeld, Ph.D.
Director, Summer Research Program
Assistant Dean for Educational Programs

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Acknowledgements

This publication marks the completion of the twentieth year of The University of Texas-Houston Medical School (UT-HMS) Summer Research Program. The longevity and success of the program are rooted in the overwhelming support received from the deans, faculty, staff and students of UT-HMS.

Indicative of this support is the administrative assistance and financial support provided by the UT-HMS. Sincere appreciation is expressed to Stanley G. Schultz, M.D., Dean, and to Patricia M. Butler, M.D., Associate Dean, Office of Educational Programs, who continue to insure the yearly success of the Summer Research Program.

Major financial assistance for our Program has also been provided by the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) through a short-term research grant (5 T35 DK007676).

Dr. Hui-Ming Chang, Associate Vice President for International Programs and Special Advisor to the President, has negotiated cooperative agreements with the Instituto Tecnológico y de Estudios Superiores de Monterrey (Mexico) and Fu-Jen Catholic University (Taiwan) to set up tailored programs for selected international medical students. This new international initiative now provides the opportunity for our Program to expand into a new area of research education that will be expanded in years to come.

The success of our Summer Research Program depends primarily on the faculty who volunteer to mentor the trainees. These dedicated educators organize and guide the research projects that, for each student, includes data analysis, preparation on an abstract and public presentation of results. Our sincere appreciation to all faculty mentors.

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Lab Research Ownership

Publication and/or Disclosure

There are standard restrictions on student participants regarding publishing or use of their research results from laboratory research performed as summer research students through the UT at Houston Medical School Summer Research Program.

All of the student participant's laboratory research is **CONFIDENTIAL**. Although the student's abstract will be available through the Summer Research Program website and in publication form, the student

must **Not** independently disclose any research findings made during their participation in the Summer Research Program.

Also, because the student's research was generated from ideas and funds that originated through their faculty mentor,

"ownership" of any data generated by the student in their faculty mentor's laboratory belongs to the faculty mentor/Principle Investigator (PI) of the lab.

The Faculty/PI controls the data and any disclosure of that data.

The student should not communicate or present any of the research data at a meeting or conference, or to the public or scientific community, without the express prior written approval from their faculty mentor. Likewise, the student is not free to publish any of the data.

The student must contact their UT at Houston faculty mentor for written approval prior to presenting their research Abstract for any of their university's events, conferences or publications.

If the student wishes to submit their abstract to an outside entity or use it at their university, the student must first contact their faculty mentor – thru letter or email - no less than 3 weeks prior to any deadlines, and get their faculty mentor's written support.

For any questions, please contact the Summer Research Program coordinator, at 713/500-5334.

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MEDICAL STUDENTS

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ABSTRACT

Recombinant Factor VIIa Partially Reverses the Effects of Anticoagulants and Antiplatelet Agents on Thrombin Generation and Thromboelastography

MARIAM L. ABDULLATIF *The University of Texas at Houston Medical School* *Class of 2009*

Sponsored by: Miguel A. Escobar, M.D., Department of Pediatrics

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Recombinant Factor VIIa, anticoagulants, antiplatelet, thrombin generation

Anticoagulants and anti-platelet agents have the potential drawback of bleeding complications and some lack an antidote for their quick reversal. Recombinant factor VIIa (rFVIIa) has recently been suggested as a general hemostatic agent but its use has only been approved for hemophiliacs with inhibitors. The purpose of this study was to evaluate the *in-vitro* ability of rFVIIa to reverse the effects of the following anticoagulant and/or anti-platelet agents: aspirin/plavix, heparin/aspirin/plavix, and coumadin. After IRB approval and informed consent, blood was collected from patients receiving these medications and the effects of rFVIIa were assessed using the thrombin generation assay and rotational thromboelastography (RoTEG®). In platelet-poor plasma (PPP), the lagtime for thrombin generation was increased in all patients receiving anticoagulants and/or antiplatelet agents. The endogenous thrombin potential (ETP) was decreased only in patients receiving coumadin. The addition of rFVIIa (2 and 6 ug/ml) shortened the lagtime in all patients so that it was not significantly different from controls. However, rFVIIa did not increase the ETP in the coumadin group. In whole-blood thromboelastography, the clotting time (CT) in the heparin/aspirin/plavix and coumadin groups were increased compared to controls. Addition of rFVIIa (6 ug/ml) reversed the anticoagulant effect in the coumadin group but not the heparin/aspirin/plavix group. The mean clot firmness (MCF) was decreased only in the heparin/aspirin/plavix group and addition of rFVIIa (6 ug/ml) reversed this effect. This study shows that rFVIIa accelerates thrombin generation and CT, but does not completely reverse the effects of these agents on thrombin generation and thromboelastography.

ABSTRACT

Familial Thoracic Aortic Aneurysms and Dissections: Clinical Complications with Pregnancy

TRENTON D. BRYSON The University of Texas at Houston Medical School Class of 2008

Sponsored by: Dianna M. Milewicz, M.D., Ph.D., Department of Internal Medicine

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
and

Dianna M. Milewicz, M.D., Ph.D., Department of Internal Medicine

Key Words: Aortic aneurysms and dissections, pregnancy

Familial thoracic aortic aneurysms and dissections (TAAD) are inherited in an autosomal dominant manner with decreased penetrance and variable expression. It has been well documented that pregnancy can induce vascular disease in genetically predisposed individuals. To evaluate the contribution of pregnancy to the clinical risk of developing aortic disease for women with a family history of TAAD, a cohort of 174 women belonging to 155 families identified to have familial TAAD was analyzed. Women were scored as affected according to the criteria of either having had surgical repair of an aneurysm or dissection, by autopsy report, or by medical record of aortic imaging. Demographic information was collected for these women and their children to evaluate the age of onset, time to onset from last parturition, as well as the effect of multiple birth pregnancies. Although rare, several cases of aneurysm or dissection were noted during gestation or immediately post partum. For comparison, similar information was collected from affected male patients and affected women with no children. On average, disease presentation was delayed in women with larger number of children, perhaps due to anticipation. Male children of an affected parent were more likely to also develop disease than female children ($p < .0001$). Further analysis for the possibility of anticipation will better help understand these findings. Women with a family history of TAAD do not necessarily need to avoid pregnancy, but should have regular screening throughout pregnancy and during the post partum period to prevent untreated dissection or rupture during this time.

ABSTRACT

The Circadian Clock within Murine Adipose

DANA T. BUI

The University of Texas at Houston Medical School

Class of 2008

Sponsored by: Martin E. Young, D.Phil., Brown Foundation Institute of Molecular Medicine

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
Dianna M. Milewicz, M.D., Ph.D., Department of Internal Medicine

Key Words: Gene expression, diurnal variations, metabolism

Disruption of normal sleep/wake patterns is associated with development of obesity, a major health problem worldwide. Perturbations in sleep/wake cycles influence the circadian clock, an intracellular molecular mechanism that enables biological processes to occur at optional times in a day. Little is known regarding the role(s) of the circadian clock within the adipocyte. We hypothesize that the circadian clock within the adipocyte modulates gene expressions, metabolism, and function of adipose tissue over the course of a normal day, and that impairment of this mechanism precipitates metabolic maladaptation (i.e., obesity and/or diabetes). Using real time RT-PCR, we have characterized fully the circadian clock within both murine epididymal and subcutaneous adipose, as well as investigated diurnal variations in the expression of multiple genes regulating adipocyte metabolism and function. All circadian clock genes investigated (*bmal1*, *clock*, *per1*, *per2*, *per3*, *cry1*, *cry2*, *dbp*, *hlf*, *tef*, *e4bp4*, *rev-erba*, and *dec1*) exhibited significant oscillations in expression for both tissues. However, differences were observed between the two adipose depots for a subset of the genes investigated. For example, *per2* and *per3* exhibited significantly greater levels of expression in epididymal fat as compared to subcutaneous fat. Significant oscillations in expression were also observed for various adipose-derived hormones/cytokines (e.g., *leptin*), metabolic genes (e.g., *adrp*, *acs1*), and adipose-enriched transcription factors (e.g., *cebp β*). In conclusion, these data are consistent with the operation of a fully functional circadian clock within the adipocyte, which potentially regulates adipose function over the course of the day.

ABSTRACT

Anti-RNA Polymerase III Autoantibodies in Scleroderma Patients: Comparing a New ELISA Testing Method to the Currently Used Immunoprecipitation Method

RACHEL M. COHEN

The University of Texas at Houston Medical School

Class of 2008

Sponsored by: Maureen D. Mayes, MD, MPH, Division of Rheumatology

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
Maureen D. Mayes, MD, MPH, Department of Internal Medicine

Key Words: Systemic sclerosis, scleroderma, RNA Polymerase III, autoantibodies

Background: The purpose of this study was to determine the accuracy of a new ELISA testing method compared to the “gold standard,” immunoprecipitation, in identifying the Anti-RNA Pol III autoantibody. Scleroderma, also called systemic sclerosis, is an auto-immune disease characterized by specific clinical manifestations. Some of the individual disease features have been linked to the presence of a specific autoantibody. This study focused on the Anti-RNA Pol III autoantibody which has been linked to renal crisis.

Methods: 23 serum samples from the GENISOS cohort, which had already tested positive by immunoprecipitation, were tested using ELISA kits provided by Rhi Gene, Inc. Two different calibration methods were used, so each plate was run twice. Comparison of proportion was used to calculate p values.

Results: With the first method, 8 of 23 (34.78%) were positive by ELISA; whereas with the second method, 9 of 23 (39.13%) were positive. Our two ELISA methods were comparable, yielding a concordance ratio of 95.7% ($p=0.9973$). The sensitivity of the new ELISA method was only 36.96%.

Conclusion: The low concordance between the two tests suggests that the new ELISA kits are not accurate enough to implement in clinical use. Due to the surprising inaccuracy of the kits, the manufacturer was contacted. They reported other facilities had similar problems, and they were in the process of reformulating the kits. When ready, new kits will be supplied and the tests will be repeated.

ABSTRACT

Evaluation of Natural History and Treatment Benefit of Patients with Renal Cell Carcinoma Involving the Bony Pelvis

BILL R. COVINGTON The University of Texas at Houston Medical School Class of 2008

Sponsored by: Kamran Ahrar, MD, M.D. Anderson Cancer Center, Department of Diagnostic Radiology, Section of Interventional Radiology

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35 DK007676-13

Key Words: Renal Cell Carcinoma, pelvic metastasis, rate of progression

Purpose: The purpose of this study was: 1) to describe the rate of progression of metastases to the bony pelvis in patients with Renal Cell Carcinoma (RCC), 2) to study the potential benefit of aggressive treatment of pelvic bone metastases.

Materials and Methods: The medical histories of all patients diagnosed with RCC metastatic to the bony pelvis between 1998 and 2004 were reviewed. To evaluate the rate of progression of untreated pelvic metastases, three-dimensional measurements were collected. We then computed tumor volume and evaluated the progression over time. To study the benefit of aggressive treatment of pelvic metastases, the rate of progression of untreated/pre-treatment tumors was compared to the rate of progression of post-treatment tumors.

Results: Of 67 patients with RCC metastatic to bony pelvis, 11 met the criteria for the study (4 women and 7 men; mean age 58 years). Five patients received local treatment for their pelvic metastases (3 radiation, 1 radiofrequency ablation (RFA), 1 embolization with RFA). The mean follow-up period was 10.8 months (range 2-56 months) for the untreated group and 11.4 months (range 6-20 months) for the treated group. Analysis indicated a mean rate of progression of 2.24 cm³/month for untreated pelvic metastases. The rates of progression of untreated/pre-treatment metastases versus treated metastases were 2.23 and .98 cm³/month, respectively. The data suggested a 91% probability that a difference exists between the progression rates of untreated and treated tumors. However, due to small sample size, a statistically significant difference could not be demonstrated.

Conclusion: This study demonstrates a steady rate of progression for pelvic metastases from RCC. It also suggests that aggressive, local treatment can potentially reduce these tumors' rates of progression. Further studies are needed to show that aggressive, local treatment may decrease tumor-related symptoms and improve patients' quality of life.

ABSTRACT

Effects of Ketamine on GI Dysfunction in LPS Model of Sepsis

CAROLINE DAIGLE

The University of Texas at Houston Medical School

Class of 2008

Sponsored by: David Mercer, M.D., Department of Surgery

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Gastrointestinal ileus, inducible nitric oxide synthase, cyclo-oxygenase-2

Gastrointestinal (GI) ileus is a frequent occurrence in critically ill septic patients that may predispose them to the development of secondary infections such as nosocomial pneumonia via aspiration of gastric contents. GI dysfunction is further exacerbated by commonly used sedatives and anesthetics and their use is therefore a major concern in the ICU. Because the anesthetic ketamine has anti-inflammatory properties in some animal models of sepsis, this study was undertaken to examine whether ketamine improved gastric emptying or intestinal transit in rats following lipopolysaccharide (LPS) as a model of sepsis. We hypothesized it would.

Method: Fasted rats received intraperitoneal (IP) ketamine (5 mg/kg) or saline 1h before a 5h treatment with saline or LPS (20 mg/kg IP). Thirty minutes prior to sacrifice, rats were given a 1cc gastric gavage of 0.1 ml of 5 mM Rhodamine-labeled Dextran in 0.9 ml of saline followed 5 minutes later by 0.15 cc 5 mM FITC-Dextran injected into a duodenal catheter. At sacrifice, gastric fluid was collected and the small intestine was removed, clamped, and cut into 10 equal segments. Each segment was flushed with equal volume and the contents collected separately. The luminal contents were evaluated by spectrophotometer for chemiluminescent intensity of each dye at its respective wavelength to determine concentration. Stomach and intestine were also evaluated by Western immunoblot for inducible nitric oxide synthase (iNOS) and cyclo-oxygenase-2 (COX-2) as indices of gut inflammation .

Result: LPS significantly delayed gastric emptying and intestinal transit and upregulated iNOS and COX-2 in both the stomach and the intestine. Ketamine did not improve gastric emptying following LPS, but did improve intestinal transit. In addition, ketamine prevented LPS induced upregulation of COX-2 in the gut but not iNOS.

Conclusion: These data suggest that iNOS does not play a significant role in mediating the ability of LPS to impair GI transit. Moreover, the finding that ketamine improved intestinal transit after LPS may prove to be useful in the ICU setting, although comparator studies with other analgesics/sedatives are needed.

ABSTRACT

Immunolocalization of Cyclooxygenase Isoenzymes During Osteointegration and the Effects of NSAIDs on Bony Integration: Defining and Developing an Animal Model

BRIAN P. F. DEUELL The University of Texas at Houston Medical School Class of 2008

Sponsored by: Catherine G. Ambrose, PhD, Department of Orthopaedic Surgery
Dhiren S. Sheth, MD, Department of Orthopaedic Surgery

Supported by: Catherine G. Ambrose, PhD, Department of Orthopaedic Surgery *and*
Dean Stanley Schultz, MD, The University of Texas at Houston Medical School

Key Words: cyclooxygenase, osteointegration, trabecular metal

The process of bone ingrowth (osteointegration) is often characterized using the fracture repair model. This model includes the inflammation and subsequent repair processes, the role of various mediators (including cyclooxygenase), and the effects of NSAIDs on this healing. However, it is well recognized that the process of osteointegration (intramembranous ossification) differs from that of fracture healing (endochondral ossification), and many assumptions that are based on the fracture-healing model may not be applicable during osteointegration. In this project, an animal model has been developed to analyze the temporal involvement of cyclooxygenase and bony ingrowth resulting from the injury required to implant a porous metal implant. Injuries were created in twenty Sprague-Daley rats by reaming into the left femoral canal in a retrograde fashion through the knee. The animals were then sacrificed at varying timepoints: twenty-four hours, forty-eight hours, five days, seven days, and fourteen days. After sacrifice both femurs were removed and processed for standard paraffin histology and immunohistochemistry. These prepared sections are currently being analyzed for expression of COX-1, COX-2, and bony ingrowth. During the course of this project several improvements were made to the protocol. The surgical method and post-operative care that minimizes confounding factors, such as suture failure, has been improved upon as well. This model will be applied in a further studies designed to elucidate the mechanisms of osteointegration and growth factor expression in arthroplasty models.

ABSTRACT

Early Endocrine Effects on Hearing

DAVID T. DOAN

The University of Texas at Houston Medical School

Class of 2008

Sponsored by: Lincoln Gray, PhD, Department of Otolaryngology, Head and Neck Surgery
Robert Lasky, PhD, Center for Clinical Research and Evidence Based
Medicine

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: DPOAE, chickens, Endocrine disruptors

Endocrine disruptors are exogenous molecules that can influence the production, secretion, transport, binding action, metabolism and catabolism of hormones within the body. They can elicit adverse outcomes in the development of an organism. It is thought that environmental endocrine disorders impede the development of hearing, which could be quantified through distortion product otoacoustic emissions (DPOAEs). In DPOAEs, two pure tones of differing frequencies, designated as F_1 and F_2 , are played to the ear. A normal ear produces a recordable third tone with the frequency of $2F_1-F_2$, known as the distortion product. Chickens were exposed as embryos to high or low lead levels, saline or an unexposed control. Newborn chickens were sexed by feather patterns. DPOAEs recorded from the chickens showed a significant difference from DPOAEs attempted on an inanimate cavity of similar volume to the ear canal. Male and female newborn chickens also presented with distinctive DPOAEs, males having a slightly higher level than females. In humans, DPOAEs in females are higher compared to males. The disparity between the two may be due to the homozygosity of the female sex chromosome (XX) in humans and of the male sex chromosome (ZZ) in chickens. Exposure to lead further enhanced the DPOAEs in male chickens but adversely depressed those in female chickens. The study suggests that DPOAEs in hatchling chickens may be a sensitive indicator of early endocrine disruption.

ABSTRACT

Computer Tomography Analysis Model for Identifying Aorta Trauma

DAVID T. FETZER

The University of Texas at Houston Medical School

Class of 2008

Sponsored by: West, O. Clark, MD, Department of Diagnostic and Interventional Imaging

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
Susan D. John, MD, Department of Diagnostic and Interventional Imaging
O. Clark West, MD, Department of Diagnostic and Interventional Imaging

Key Words: aorta injury, computer tomography, CT, blunt thoracic trauma

Acute traumatic aorta injuries (ATAIs) and other great vessel injuries (GVIs) following blunt thoracic trauma can lead to surgically important arterial injuries. Rapid and accurate diagnosis is essential for institution of life-saving treatment. Computer tomography (CT) has become the diagnostic modality of choice in many trauma centers. We developed a model using specific CT diagnostic criteria: eleven areas of potential hematoma formation were identified in and around the mediastinum. Short and long axis of aorta cross-section at the level of greatest diameter was measured. Qualitative morphologic information (contour change, intimal flap, etc.) was recorded. Smoothness of the aorta wall was defined. These characteristics were quantified and analyzed for statistical significance. Average weighted kappa was 0.74, showing reliable agreement among the two observers utilizing this model. Marked improvement in distinguishing equivocal studies as either negative or positive injuries was observed. The ROC curve calculated from the radiologist interpretation contained 86.1% area under the curve while the curve for the new model contained 97.5%. The likelihood ratio increased from 30.06 to 48.67. The degree to which the new measure improved prediction over physician diagnosis was tested using a nested model, and yielded a reliable increment in model fit ($\Delta\chi^2(3)=19.762$, $p\leq 0.0001$). The model does not predict GVI as successfully as ATAI. However, several characteristics involving injury morphology and thoracic hematoma location showed a strong correlation with GVI, unique findings of this research. In conclusion, we have developed a diagnostic tool that may help radiologists and trauma surgeons better evaluate CT for ATAI and GVI.

ABSTRACT

The Effect of Fear on Time Perception

MATTHEW P. FIESTA *The University of Texas at Houston Medical School* *Class of 2008*

Sponsored by: David M. Eagleman, Ph.D. Department of Neurobiology and Anatomy

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
David M. Eagleman, Ph.D. Department of Neurobiology and Anatomy

Key Words: fear, brain, adrenaline, time perception, vision

Individuals in fearful situations frequently report the sensation that time seems to “slow down.” This phenomenon may be the result of the brain processing information more quickly in the fearful moment; alternatively, emotionally salient memories may be recollected in a different manner, and the event is interpreted to have gone in slow motion in retrospect – but more information was not actually absorbed during the event. In order to distinguish these two hypotheses, we have engineered a device that alternates positive and negative images of digits at specific frequencies. Each LED making up a digit alternates rapidly between the ON and OFF state. At rapid frequencies of alternation, the subject perceives the LED array to be uniformly lit. We found that each subject has a specific frequency threshold at which the alternating images can be deciphered. We asked whether an individual’s threshold frequency would increase during a fearful situation. Two members of our laboratory piloted the device by viewing a string of two randomly selected alternating numbers – presented faster than their threshold – while dangling 150 feet in the air. They were then dropped 100 feet into a net while they continued to look at the device. Under this fearful situation, one lab member reported that he was able to see digits right before the drop. The other lab member reported seeing only one digit, but could not identify the other. While these reports are promising, they are still inconclusive until we run several more subjects.

ABSTRACT

Effect of Acular LS on Macular Foveal Thickness after Phacoemulsification

STEPHANIE AMBER FINCH The University of Texas at Houston Medical School Class of 2008

Sponsored by: Richard W. Yee, MD, Department of Ophthalmology and Visual Science

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
Richard W. Yee, MD, Department of Ophthalmology and Visual Science

Key Words: Cataracts, cystoid macular edema, phacoemulsification

Phacoemulsification with implantation of a foldable intraocular lens is the most prevalent method for treatment of cataracts. There exists a 3% risk of developing cystoid macular edema (CME) after phacoemulsification. CME is an inflammation of the macula that alters its function leading to vision impairment. The purpose of this study is to determine if there is a clinical difference in macular foveal thickness in patients, who have undergone uncomplicated phacoemulsification and developed CME, using Acular LS (ketorolac tromethamine ophthalmic solution 0.4%) compared with using artificial tears. OCT measurements were performed preoperatively and postoperatively on subjects undergoing routine phacoemulsification to determine their macular foveal thickness. Subjects who developed greater than a 10% change in macular thickness were randomized to use either Acular LS or artificial tears. Of the fourteen patients in the study, three developed CME. At 1-2 weeks the average thickness change from preoperative measurements was $2.9\mu\text{m} \pm 25.2\mu\text{m}$, at 4 weeks $16.2\mu\text{m} \pm 21.1\mu\text{m}$, and at 6 weeks $11.9\mu\text{m} \pm 24.9\mu\text{m}$. In a 6-week span, preliminary results show a $14\mu\text{m}$ decrease in macular thickness from preoperative measurements in the patients using Acular LS, and an average increase of $9.5\mu\text{m}$ using artificial tears. However, the accuracy of OCT measurements has only been determined to be within $7.0\mu\text{m}$. Due to rare occurrences of CME in subjects included in the study, the clinical difference between Acular LS and artificial tears has not been conclusively determined. OCT may not be sensitive enough for detecting subtle changes in macular edema. Additional subjects are currently being recruited to participate in the study.

ABSTRACT

Composite Forearm Free Fillet Flaps to Preserve Stump Length Following Traumatic Amputations of the Upper Extremity

MITCHELL D. FLURRY

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Class of 2008

Sponsored by: Christopher Livingston, MD, Department of Surgery – Division of Plastic and Reconstructive Surgery
E. G. Melissinos, MD, Department of Surgery - Division of Plastic and Reconstructive Surgery

Supported by: Division of Plastic and Reconstructive Surgery, Department of Surgery, The University of Texas at Houston Medical School

Key Words: Composite forearm free fillet flaps, traumatic amputation of the upper extremity

Traumatic amputation of the upper extremity is a rare but devastating event. Replantation is usually not possible due to level of injury, associated life threatening injuries, patient age, comorbid diseases, ischemia time, and/or avulsion of proximal structures (1). In these unfortunate injuries, every effort should be made to preserve stump length or critical joints (2). Stable soft tissue coverage is mandatory to aide in prosthetic fitting and function which improves eventual outcome.

The use of otherwise uninjured tissue from the non-replantable or unsalvageable amputated segment can be used microsurgically to cover large and complex stump wounds which would have otherwise required revisionary amputations with stump shortening and increased patient morbidity (3). This “spare parts or free fillet flap” concept is well documented and widely utilized for lower extremity stump preservation (2,4) but only recently has been reported for upper extremity traumatic amputations or following radical upper extremity tumor resections (1-7). The benefits of this technique include avoidance of donor site morbidity, immediate resolution of the injury, potential shorter hospital stay requiring fewer operations and less patient psychological trauma, and reduced financial burden. Furthermore, sensory innervation can be utilized enhancing postoperative prosthetic fit and function while reducing pressure-related complications.

There are numerous examples reported within the literature of the use of free forearm fillet flaps based on the brachial or radial arteries to cover humeral or elbow stumps following traumatic amputations as well as following radical tumor excision (1-14). We present 8 cases of composite forearm free fillet flaps covering humeral and shoulder soft tissue defects following traumatic amputation. Stump length was preserved and all flaps remained viable with only two complications in one patient, wound infection and partial flap edge necrosis. In most cases recovery was within weeks and patients could return to work within months. Due to financial reasons, prosthetics have been difficult to obtain. However, stable soft tissue coverage has been achieved allowing successful prosthetic fit and function without wound or skin breakdown. This appears to be the largest series to date reported in the literature.

ABSTRACT

First Place Co-Winner, 2005 Frank Webber Prize for Student Research

A Retrospective Review of Fluid Resuscitation in Burn Patients

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Sponsored by: David J. Wainwright, MD, Department of Plastic Surgery

Supported by: Donald H. Parks, MD, Department of Plastic Surgery,
Daniel J. Freet, MD, Department of Plastic Surgery,
Timothy C. Hollenbeck, MD, Department of Plastic Surgery
Elisa M. Chi, PharmD, Memorial Hermann Hospital

Key Words: Burn; total body surface area; fluid resuscitation; Parkland Formula

The objective of this study was to investigate how closely fluid resuscitation of burn patients at Memorial Hermann Hospital (MHH) corresponded to the standard, accepted Parkland Formula (PF) (24 hr. total (cc)=4X%BurnXWeight(kg)). A retrospective chart study was performed of 20 patients who sustained >20% burns. Epidemiological data included age, weight, gender, % total and 3^o burn, burn etiology, presence of inhalation injury, associated and premorbid medical problems, time from the burn to burn unit admission and colloid usage. Fluid input data (type, volume) and output data (urine output (UO)) was collected at hourly intervals. The recommended input volumes (PF) and "goal" urine output volumes (.5- 1cc/kg/hr) were compared with the study population.

Results: Input volumes as a % PF were as follows: 1st 8 hrs - 114% (58- 320%); 2nd 8 hrs. - 162% (42- 349%); 3rd 8 hrs. - 137% (36- 438%); 1st 24 hrs - 132% (58- 273%). Average UO was higher than "goal" volumes in over 40% of patients at all time intervals (highest 1st 8 hrs – 60% patients) and below "goal" most often in the 2nd 8 hrs (40% patients).

Conclusions: Based on this small sample size: 1. Burn patients at MHH received 32% more fluid than calculated by the Parkland Formula. 2. This increased fluid input was most significant in the 2nd 8 hours. 3. MHH patient's UO was generally greater than "goal" volumes. 4. Higher UO was smallest in the 2nd 8 hours (when input was highest over Parkland) which may represent the need for increased volume administration in response to low UO during this time period. 5. The analysis of individual patient responses and subgroups is an ongoing project in the Division of Plastic Surgery at MHH.

ABSTRACT

Third Place Winner, 2005 Frank Webber Prize for Student Research

Abstract: Identification of Causative Bacteria in Osteomyelitis Using Rapid and Sensitive 16S rRNA Detection

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Sponsored by: Heidi B. Kaplan, PhD, Department of Microbiology

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Osteomyelitis, 16S rDNA, PCR Amplification, Identification of bacterial pathogens

Osteomyelitis can arise either hematogenously or secondary to a contiguous focus of infection. Both Gram-positive and Gram-negative bacteria cause a significant number of infections. The current gold standard for diagnosis of osteomyelitis is culture of a biopsy followed by biochemical analysis to determine the identity of the causative organism. In this study a molecular genetic approach is being developed to determine the identity of infectious agents. Using primers for the conserved regions of the 16S rRNA gene, the PCR was performed and DNA sequence analysis was used to determine the identity of the infectious agent. The sensitivity of the technique was determined using serial dilutions of pure cultures of *S. aureus* and *E. coli*; limits of detection for these two species were 70 CFU and 170 CFU respectively. In polymicrobial infections the DNA sequence is unreadable. To determine the identity of each infectious agent, the 16S rDNA fragments are cloned, PCR amplified, and sequenced. The cloning protocol was tested using *E. coli* DNA amplified with primers containing restriction sites. Cloning into the pKS vector and subsequent transformation yielded clones that were amplified with T3 and T7 primers and sequenced. Amplification was successful for eight of nine clones and sequence analysis of two inserts confirmed that the *E. coli* 16S rDNA had been cloned. Preliminary tests with three infected tissue biopsies resulted in positive PCR amplification with unreadable sequence indicating polymicrobial infections. Our goal is to standardize this protocol for detection of agents causing osteomyelitis by continuing to process biopsies from suspected osteomyelitis infections.

ABSTRACT

Management of Acute Traumatic Lower Extremity Arterial Injuries

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Sponsored by: Tam Huynh, M.D., Department of Cardiovascular and Thoracic Surgery

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Revascularization, orthopedic manipulation

The sequence of intervention in patients with acute traumatic injury of the lower extremity requiring combined revascularization and orthopedic intervention remains controversial. In this study, we review our experience in treating patients with acute traumatic femoral and popliteal arterial injuries. From January 2001 to February 2005, we performed surgical revascularization for 51 acute traumatic lower extremity arterial injuries, in level 1 trauma center. The mechanism of injury was blunt in 36 (71%) patients and penetrating in the remaining 15 (29%). The majority of patients were men 41/51 (80%). Median age was 30 years-old. Autogenous veins were used in all patients. Our limb-salvage rate was 46/51(91.2%). Thirty patients had orthopedic fixation for associated bony fractures or dislocations; this was done before revascularization in 10 (33%) and after in 20 (67%). There was no complication related to the arterial repair after orthopedic manipulation. Twenty-eight (55%) patients required calf-fasciotomy. The limb-salvage and fasciotomy rates were similar in patients whether the orthopedic intervention or revascularization was performed first. However, the average hospital length of stay was significantly longer for patients whom had fasciotomy (19 vs. 9 days; $p<0.001$). In conclusion, our study shows that revascularization prior to orthopedic manipulation is durable and should be performed first.

ABSTRACT

Annexin A1, a Potential Prognostic Marker in Brain Tumors

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Sponsored by: Constance T. Albarracin, MD, PhD, Department of Pathology, The University of Texas M.D. Anderson Cancer Center

Supported by: Janet M. Bruner, MD, Chairman of the Department of Pathology, The University of Texas M.D. Anderson Cancer Center

Key Words: Annexin A1, brain tumor, glioma

Background: Gliomas, the most common malignant primary brain tumors, are derived from neuroepithelial cells and can be divided into two principal lineages: astrocytic and oligodendrocytic. The astrocytic lineage has been shown to have a worse prognosis than the oligodendrocytic lineage. These lineages can be further classified as low grade (astrocytoma and oligodendroglioma), mid grade (anaplastic astrocytoma and anaplastic oligodendroglioma), and high grade (glioblastoma multiforme). Currently, no definitive prognostic marker exists to identify the more aggressive phenotypes. A promising candidate is annexin A1 (ANXA1), a cytoskeletal binding protein that is a major substrate for the tyrosine kinase activity of the epidermal growth factor receptor.

Objective: The purpose of this study is to determine whether ANXA1 is differentially expressed in the different types and grades of gliomas.

Methods: The tumors were diagnosed according to the WHO classification of tumors of the nervous system. A tissue microarray was assembled from over 130 surgical glioma cases at M.D. Anderson Cancer Center and ANXA1 expression was determined by immunohistochemistry.

Results: The lowest ANXA1 expression was found in oligodendrogliomas (15%). ANXA1 expression correlated with increasing levels of the astrocytic component and was expressed in 33% of low grade astrocytomas. Increasing levels of ANXA1 expression correlated with increasing tumor grade, with the highest ANXA1 levels present in gliosarcomas (75%) and glioblastomas (87.5%).

Conclusion: ANXA1 expression correlates with increasing grade of glioma, suggesting a role in glioma tumor progression. More importantly, ANXA1 may provide a potential target for therapeutic intervention.

ABSTRACT

Herpes Zoster: An Epidemiological Survey of Clinical Characteristics

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Sponsored by: Stephen Tyring, MD, PhD, MBA, Department of Dermatology

Supported by: Stephen Tyring, MD, PhD, MBA, Department of Dermatology

Key Words: Herpes Zoster, shingles, epidemiology, dermatology

Each year in the US, Herpes Zoster (HZ) affects approximately 1,000,000 people or roughly 20% of people with a history of primary Varicella Zoster Virus (VZV). However, the only reliable predictors of who will become one of the 20% are age and immunosuppression. The purpose of this study is to investigate clinical characteristics that may help determine which people are at risk for HZ. 504 HZ patients were included in this retrospective chart review and phone survey. Quantitative data was analyzed using student's t test and ANOVA with Scheffe's post hoc, while qualitative data was analyzed using Chi square. Results indicated an older age of onset in females than males ($p=0.002$). Likewise, Caucasians seemed to have an older age of onset than either Hispanics ($p=0.000008$) or African Americans ($p=0.004$). Females were more likely to see a doctor within 72 hours of rash onset ($p=0.01$), report a blood relative with history of HZ ($p=0.01$), and report a stressful prodromal event ($p=0.00002$). Incidence of HZ was not found to be equal among all possible months of onset ($p=0.02$). Age of onset seemed to be associated with presence of a stressful event ($p=0.04$) and memory of primary VZV seemed to be associated with ethnicity ($p=0.001$). 8% of patients reported recurrent HZ, 39% reported blood relatives with HZ, and 23% reported no memory of primary VZV. In light of the new VZV vaccine to prevent HZ, these epidemiological factors could help predict those people most in need of the vaccine.

ABSTRACT

Systemic Sclerosis in African Americans and Caucasians

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Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Systemic sclerosis, scleroderma, African American, autoantibody, HLA

Background: Systemic sclerosis (SSc) is a chronic, autoimmune disease characterized by organ fibrosis, vasculopathy, and immune activation. This study was designed to determine differences in HLA, autoantibodies, and clinical characteristics between African Americans and Caucasians with scleroderma.

Materials and Methods: A case control study was performed along with sub-analyses by ethnicity using three separate cohorts, including 360 Caucasians, 191 African Americans, 204 Caucasian controls and 129 African American controls. MHC Class II alleles, including DRB1, DQB1, and DQA1 were typed using genomic DNA from peripheral blood and standard oligotyping. Anti-centromere autoantibodies (ACA) were detected by indirect immunofluorescence on HEp-cells and other antibodies, including anti-topoisomerase 1 and RNP, were determined by immunodiffusion against calf thymus extract.

Results: Comparing AA controls (n=129) to AA SSc cases (n=183), it was found that SSc patients are more likely than controls to have the HLA alleles *DRB1*1302* (OR=3.69, 95% CI 1.53, 10.23) and *DRB1*08* (OR=2.15, 95% CI 1.08, 4.51). AA patients compared to Caucasian patients (n=353) were more likely to have the alleles *DRB1*08* (OR=2.46, 95% CI 1.44, 4.20), and *DQA1*0501* (OR=2.46, 95% CI 1.40, 4.32), and less likely than Caucasians with disease to have *DRB1*01* (OR=0.30, 95% CI 0.15, 0.56). Caucasians have ACA (30%) more frequently than AA patients (7%; p<.0001). African Americans more commonly have anti-topoisomerase 1 (21% vs 12%; p=0.0039), anti-fibrillarin (21% vs 7%; p<0.0001), and anti-RNP (18% vs 7%; p=0.0002) when compared to Caucasians.

Conclusion: These findings add further support to the premise that there is a strong genetic influence on scleroderma susceptibility and expression which differs between ethnic groups.

ABSTRACT

The Genetic Basis of Associative Memory

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Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Aplysia, Classical Conditioning, Gene Expression

The difference between associative and nonassociative learning has long been a source of debate. To address the possibility of a genetic difference between these two forms of learning, an *in vitro* analogue of classical (Pavlovian) conditioning of feeding in *Aplysia* was used to determine whether the expression of specific genes implicated in nonassociative learning (*ApCREB1*, *ApCREB2*, and *Ap-Uch*) were also induced by associative learning. The expression of the gene *Apdop1* was also examined for changes induced by the *in vitro* analogue because this gene encodes the dopamine D1 receptor, and dopamine is believed to mediate the unconditioned stimulus of classical conditioning of feeding in *Aplysia*. In the *in vitro* analogue, paired electrical stimulation of two selected peripheral nerves served as the conditioned and the unconditioned stimulus. The level of expression of the above mentioned genes was quantified by PCR and compared among paired and unpaired protocols, and naïve controls. The results indicate that the gene *Apdop1* exhibits a trend of decreased expression 100 minutes after paired training in both the cerebral and buccal ganglia when compared to the unpaired and naïve control groups. In addition, the genes *ApCREB1*, *ApCREB2*, and *Ap-Uch* show a trend of pairing-specific decreased expression in the buccal ganglia, and no change in expression in the cerebral ganglion. The profile of expression of *ApCREB1*, *ApCREB2*, and *Ap-Uch* exhibited as a result of classical conditioning appears to differ from that induced by nonassociative learning. These findings begin to unveil a genetic difference between associative and nonassociative learning. In addition, the pairing-specific trend of decreased expression of *Apdop1* may represent a molecular correlate of the psychological phenomenon known as US devaluation.

ABSTRACT

Outcome of Out-of-Hospital Cardiac Arrests in Houston, Texas

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Sponsored by: Richard N. Bradley, MD, Department of Emergency Medicine

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Utstein, cardiac arrests

Purpose: The purpose of this study is to determine if the outcome of out-of-hospital cardiac arrests (CA) in Houston, Texas is superior to the outcomes experienced in other major United States cities.

Methods: This study performed an Utstein template-based retrospective observational cohort analysis of all patients with out-of-hospital CAs of cardiac origin in the City of Houston during calendar year 2003. The primary outcome measure was one year survival.

Results: In the year 2003, there were 1,407 out-of-hospital CAs of cardiac origin in which resuscitation was attempted. Of those 300 (21.3%) were witnessed by a bystander and had an initial rhythm of ventricular fibrillation (VF). Among these patients, 45 (15%) survived the initial event and were discharged alive and 25 (8.3%) were still alive one year after the event. The results for survival to discharge among patients experiencing bystander witnessed VF were higher than those in Chicago (4.0%; 99% CI, 1.9% to 7.5%), Los Angeles (6.1%; 99% CI 3.3% to 11.0%), and New York City (5.3%; 99% CI, 2.9% to 8.8%).

Conclusion: The one year survival in patients experiencing witnessed VF CAs of cardiac origin in Houston is superior to that of similar patients in Chicago. We demonstrated a non-significant trend towards superior outcomes when compared to Los Angeles and New York City. One-year survival rates are still lower than desired and further research is needed to determine how to improve patient the survival rate.

ABSTRACT

Effect of Immune Enhancing Agents on MMP Production in the Jejunum Following Intestinal Ischemia/Reperfusion Injury

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Sponsored by: Emily K. Robinson, MD, Department of Surgery

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: MMP, ischemia/reperfusion, arginine, glutamine

Introduction: Matrix metalloproteinases (MMPs) are a group of endopeptidases which are induced in the stomach, in association with increases iNOS, and contribute to gastric injury in a model of endotoxemia. Previous studies in our laboratory have demonstrated that intestinal ischemia/reperfusion (I/R) induces MMP production, specifically MMP-9. Prior investigators have demonstrated that I/R induces iNOS in the intestine and that this increase can be modulated by the addition of the luminal nutrients arginine and glutamine. Additionally, glutamine attenuates I/R induced intestinal injury while arginine exacerbates this effect. The purpose of these studies was to evaluate the effects of I/R on intestinal MMP production and its modulation by luminal nutrients.

Methods: Jejunal sacs were created in Sprague-Dawley rats and filled with 60 mM glutamine or arginine, followed by 60 min of superior mesenteric artery occlusion and 6 h reperfusion and compared to shams. The rats were sacrificed, jejunal tissue harvested and MMP-9 production assessed by gelatin zymography while MMP-2, and iNOS protein production were assessed by Western analysis. (N \geq 5/group;ANOVA) **Results:** MMP-2, MMP-9 and iNOS levels were increased after I/R compared to shams. Arginine maintained while glutamine inhibited the I/R induced increase in MMP-2 and iNOS while neither arginine nor glutamine had any effect on MMP-9 production.

Conclusions: These data demonstrate a correlation between MMP-2 and iNOS production following I/R, and indicate that both jejunal iNOS and MMP-2 levels can be modified by luminal nutrients. Further studies are needed to investigate the link between increased metalloproteinase levels and intestinal injury in this model.

ABSTRACT

Does Cocaine Treatment in Adolescence Cause Cross-sensitization to Methylphenidate in Adulthood?

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Class of 2009

Sponsored by: Nachum Dafny, PhD, Department of Neurobiology and Anatomy

Supported by: Alan C. Swann, MD, Department of Psychiatry

Key Words: Sensitization, locomotion, dose response, psychostimulants

Cross-sensitization occurs when chronic treatment with one psychostimulant leads to increased sensitivity to single treatment with a different psychostimulant. This phenomenon occurs between amphetamine and cocaine and amphetamine and methylphenidate. However, there is limited knowledge whether cross-sensitization occurs between cocaine and methylphenidate (MPD). MPD, commonly known as Ritalin, is a medication frequently used to treat attention deficit/hyperactivity disorder (ADHD). This study sought to determine whether rats chronically exposed to cocaine during adolescence could exhibit cross-sensitization to MPD as adults. Adolescent female Sprague-Dawley rats were randomly divided into four equally populated groups (1) given saline in adolescence and adulthood, (2, 3, 4) given 3.0, 7.5, or 15 mg/kg cocaine intraperitoneally for six consecutive days, three days of washout, and a day of cocaine re-challenge as adolescents and similarly treated two weeks later as adults. This data was compared to four groups of rats that received the exact treatment regimen except only as adults. To test for cross-sensitization with MPD all groups received one injection of 2.5 mg/kg MPD at the end of their adulthood treatment regimen. Changes in locomotion and stereotypic behaviors were recorded using a computerized activity monitoring system. Results revealed that chronic cocaine treatment leads to sensitization to cocaine after a period of washout, especially with the 7.5 mg/kg dose. Furthermore, cross-sensitization to MPD was observed and found to be cocaine dose dependent. The occurrence of cross-sensitization between MPD and cocaine has severe implications for the abuse potential and deleterious effects of chronic treatment of children with Ritalin.

ABSTRACT

Functional MRI Comparison of Working Memory in Cocaine Users Due to Route of Administration

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Class of 2008

Sponsored by: Joel L. Steinberg, MD, Department of Psychiatry and Behavioral Sciences

Supported by: The Bernard Saltzberg Summer Research Fellowship

Key Words: Cocaine, blood oxygen level dependent functional magnetic resonance imaging (BOLD fMRI), working memory, immediate memory task/delayed memory task (IMT/DMT)

Cocaine acts as a powerful CNS stimulant by interfering with the reabsorption of dopamine and causing long-term dysfunction in the higher thought and decision-making centers of the brain. Research indicates that cortical regions required for working memory, primarily the anterior cingulate and the prefrontal cortex, are also impaired by extensive cocaine abuse. The objective of this study was to examine the extent of cerebral impairment based upon the route of cocaine administration. Cocaine users were scanned using BOLD fMRI to measure their hemodynamic response (HDR) while performing an IMT/DMT working memory task. The IMT consisted of a visual string of 3, 5 or 7 numbers flashed at 1-second intervals while the DMT had a 3-second interval with distracter numbers in between. The subject pressed a button when the displayed number (probe) matched the preceding number (target). The data was analyzed using Statistical Parametric Mapping (SPM2) software to contrast the HDR of the cocaine users according to route of administration. A group comparison of cocaine users who smoked (n=6) versus intranasally inhaled (n=4) showed significantly greater activation ($p < 0.05$) during the DMT relative to IMT in the left superior parietal lobule (BA7), cingulate gyrus (BA23,32), and prefrontal cortex, including the right inferior frontal gyrus (BA47), left middle frontal gyrus (BA9) and bilateral superior frontal gyri (BA6). The superior parietal lobule is linked to visual working memory and the cingulate gyrus with the prefrontal cortex being implicated in dopamine circuitry and further memory processing. The reduced BOLD activation in these cortical areas of cocaine snorting subjects may be associated with a more severe deficit in working memory compared to cocaine smokers. Due to fixed effects data analysis, these conclusions are limited to only the subjects in this study.

ABSTRACT

Differential Influence of Fatty Acids on Cardiac Metabolic Gene Expression

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Sponsored by: Martin E. Young, D. Phil., Institute of Molecular Medicine

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: fatty acids, cardiomyocytes, gene expression, metabolism

Diabetes mellitus is a major risk factor for the development of cardiovascular disease. A major phenotypic alteration of the heart during diabetes is increased reliance on fatty acids as a fuel. Such a metabolic perturbation is mediated, at least in part, by fatty acid-activation of the nuclear receptor peroxisome proliferator-activated receptor α (PPAR α). The latter activates the expression of genes promoting fatty acid oxidation (e.g. malonyl-CoA decarboxylase (*mcd*) and cytosolic thioesterase 1 (*cte1*)), as well as those repressing carbohydrate oxidation (e.g. pyruvate dehydrogenase kinase 4 (*pdk4*)). It is becoming increasingly clear that different fatty acid species can have distinct pathophysiological effects on the heart, including induction of arrhythmias and apoptosis. We therefore hypothesized that fatty acids of varying chain length and saturation mediate one or more of their pathophysiological effects through differential regulation of myocardial gene expression. Using a candidate gene approach, we investigated the direct effects of linoleic acid, octanoate, oleate, palmitate, and stearate on metabolic gene expression in adult rat cardiomyocytes. We report that different fatty acids have distinct time- and dose- dependent effects on specific target genes. For example, oleate induces both *mcd* and *pdk4* to a greater extent than the other fatty acids, while palmitate induces *cte1* to the greatest extent. Such observations suggest fatty acids influence myocardial gene expression through multiple mechanisms, and not just through PPAR α . Furthermore, these findings may also explain why some fatty acids (e.g. oleate) are more cardioprotective than others (e.g. palmitate).

ABSTRACT

Mutations in TGFBR2 and TGFBR1 in Familial Thoracic Aortic Aneurysms and Dissections and Related Syndromes

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Sponsored by: Dianna M. Milewicz, MD, PhD, Department of Internal Medicine, Division of Medical Genetics

Supported by: Doris Duke Charitable Trust, NIH RO1 HL62594 (D.M.M.), MO1RR02558 (University Clinical Research Center)
Dean Stanley Schultz, MD, The University of Texas at Houston Medical School

Key Words: aortic aneurysm, aortic dissection, TGFBR2, mutations

Thoracic aortic aneurysms are often asymptomatic until a life threatening event like dissection or rupture occurs. Up to 19% of thoracic aortic aneurysm and dissection (TAAD) cases have family history of the disease. Using a large family with autosomal dominant inheritance of TAAD, a locus for the disease, subsequently named TAAD2, was mapped to 3p24-25. With knowledge of Marfan syndrome, caused by a mutation in the fibrillin gene, and its indirect effect upon transforming growth factor- β (TGF- β) signaling, it is suspected that the alterations in TGF- β signaling cause the syndrome's characteristic thoracic aortic aneurysms. Therefore, it is hypothesized that mutations in the gene, transforming growth factor- β receptor type II (TGFBR2), which is within TAAD2, result in a proportion of familial cases of TAAD. DNA samples were collected from 23 families with history of TAAD. All eight exons of TGFBR2 were amplified and sequenced. Blast alignment was used for analysis of possible mutations and revealed eight persons from two different families to possess mutations in TGFBR2. Remarkably, the two different mutations affected the exact same amino acid, arginine at position 460, proposing a possible mutational hot spot at that location. Furthermore, three patients with possible Loeys-Dietz aortic aneurysm syndrome, characterized by aggressive thoracic aortic aneurysms at a very young age, were sequenced for mutations in TGFBR1, a gene in the locus TAAD1, and TGFBR2. One patient did, in fact, reveal a mutation in exon 6 of TGFBR1 supporting the evidence of TGF- β alterations leading to TAAD. Additional work will investigate the role of TGF- β in the pathogenesis of thoracic aortic aneurysms and dissections.

ABSTRACT

Tear Evaporation Rates from the Ocular Surface

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Sponsored by: Richard W. Yee, MD, Department of Ophthalmology

Supported by: Richard W. Yee, MD, Department of Ophthalmology

Key Words: Evaporimetry, Ocular Surface Disease Index (OSDI)

Evaporimetry is a sensitive method used to measure the evaporation rate of tears from the ocular surface. The instrument used in this study was the evaporimeter, which measures evaporation in $n \times 10^{-7} \text{g/cm}^2/\text{sec}$ at a relative humidity of 30%. We hypothesize that evaporation rates are repeatable and that they will increase from morning to afternoon and from Monday to Friday. We collected readings from 28 non-contact lens wearers and divided them into two groups: symptomatic and asymptomatic based on the individual's score on the Ocular Surface Disease Index (OSDI). The OSDI is a subjective questionnaire that can assess information on how often a patient experiences symptoms of ocular surface disturbances, including dry eye disease symptoms and the effect of these symptoms on various activities over the last week. Symptomatic subjects scored higher than 15 on the OSDI, and Asymptomatic (control) subjects scored 15 or lower on the questionnaire. There was a slight decrease found in evaporation rates among all subjects from morning to afternoon, but this difference was not significant. When evaporation rates were compared throughout the week, a slight increase was noted, and was significant among asymptomatic patients ($p=0.048$), but was not significant among symptomatic patients ($p=0.29$). We also found statistical significance between the evaporation rates of symptomatic vs. asymptomatic subjects ($p=0.024$) regardless of time of day or day of week taken. The mean evaporation rate among symptomatic subjects was found to be $25.94 \times 10^{-7} \text{g/cm}^2/\text{sec}$, and asymptomatic subjects had a mean evaporation rate of $20.11 \times 10^{-7} \text{g/cm}^2/\text{sec}$.

ABSTRACT

Effect of Inducible Nitric Oxide Synthase (iNOS) on Noise-Induced Hearing Loss

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Ah-Lim Tsai, PhD, Department of Internal Medicine, Hematology

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School

Key Words: Noise-induced hearing loss, iNOS

More than 30 million Americans, nearly 10% of all adults, are considered to be hearing impaired. Noise induced hearing loss (NIHL) is the second most common cause of sensory hearing loss, primarily resulting from prolonged exposure to high-intensity sounds. One of the mechanisms proposed for NIHL is reactive oxygen species (ROS) including nitric oxide. Nitric oxide is involved in macrophage cytotoxicity, vasodilation, and neuroplasticity. Three distinct isoforms of nitric oxide synthase have been identified: 1- inducible NOS, 2- neuronal NOS, and 3- endothelial NOS. There are several inhibitors of NOS, including inducible NOS (iNOS) specific s-isothiourea (SITU). To investigate whether iNOS is involved in NIHL, four groups of chickens were established: 1- not exposed to sound, 2- exposed to sound, 3- exposed to sound after 5uL normal saline placed behind tympanic membrane, and 4- exposed to sound after 5uL 1mM SITU placed behind tympanic membrane. Sound exposure designates high-intensity pure-tone sound (900Hz at 120dB) for eighteen consecutive hours. Distortion product otoacoustic emissions (DPOAEs) were measured both before and after exposure to sound in an attempt to further examine the damage caused by high-intensity sound. Electron paramagnetic resonance (EPR) studies were inconclusive to the amount of nitric oxide radical produced during exposure to high-intensity sound. Findings from this study include a significant increase in the amount of iNOS produced during exposure to high-intensity sound ($p < 0.01$) as well as a significant difference between SITU and saline or sound-exposed birds in the intensity of the DPOAEs ($p < 0.05$) around the frequency of sound exposure.

ABSTRACT

The Chemotherapeutic Effects of Ibuprofen-PC in a Murine Colon Cancer Model

EVAN G. MEEKS

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Class of 2008

Sponsored by: Lenard M. Lichtenberger, PhD, Department of Integrative Biology and Pharmacology
Elizabeth J. Dial, PhD, Department of Integrative Biology and Pharmacology

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Colon cancer, ibuprofen, phosphatidylcholine, NSAIDs

Colon cancer is too often diagnosed late and is becoming a leading cause of death in the United States. This study was conducted to examine the chemotherapeutic properties of ibuprofen against colon cancer. Previous studies show that ibuprofen has anti-proliferative properties against both human and murine colon cancer cell lines. Ibuprofen and other NSAIDs are toxic to the protective mucosal barrier in the gastrointestinal tract. By coupling NSAIDs with phosphatidylcholine(PC), the toxic side effects are alleviated and the drugs have equal or better effectiveness than NSAIDs alone. An in-vivo murine colon cancer model was developed to test the hypothesis that ibuprofen-PC would have better cancer fighting properties than ibuprofen alone with less injury to the intestinal lining. Cells from the MC-26 murine colon cancer line were injected into the spleen of male BALB/c mice. The mice were dosed with ibuprofen or ibuprofen-PC at 20mg/kg or 50mg/kg, or 90G at 100mg/kg twice a day for 14 days. At sacrifice, the spleen, liver, serum, intestinal contents, and fecal samples were collected to evaluate the growth and spread of the cancer and the effects of the treatments on the GI tract. The ibuprofen and ibuprofen-PC groups showed dose dependent decreases in cancer growth with the ibuprofen-PC 50mg/kg group having the greatest anti-proliferative properties. Also, this study revealed that ibuprofen-PC 50mg/kg caused the least intestinal damage. In conclusion, the ibuprofen-PC dose of 50mg/kg was safer and more effective as a chemotherapeutic agent than ibuprofen alone against murine colon cancer.

ABSTRACT

Use of Controlled Hyperosmolality with Hypertonic Saline Solutions in Traumatic Brain Injury: Case Study of 18 Pediatric Patients

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Sponsored by: Stephen A. Fletcher, D.O., Department of Surgery - Pediatric Neurosurgery

Supported by: Department of Surgery - Pediatrics, The University of Texas at Houston Medical School

Key Words: Hypertonic saline, traumatic brain injury, increased intracranial pressure

Current guidelines for the care of traumatic brain injury (TBI) indicate neither preference for mannitol sodium nor hypertonic saline for osmolar therapy. The benefits of using concentrated sodium chloride solutions include the ability to achieve higher, therapeutic serum osmolar targets, longer duration of action, and ease of maintenance of a normovolemic state. We report the use of controlled elevation of serum osmolality with hypertonic sodium chloride solutions at our institution with 18 pediatric patients. Data was collected by retrospective chart review from 2002-2005 at an urban Level 1 pediatric trauma center. Age, sex, injury, imaging by CT or MRI, along with surgical intervention, intracranial pressure (ICP) values, serum sodium, serum osmolality, and serum creatinine were recorded in a database for analysis. Number, use, duration, and timing of hypertonic saline were recorded to correlate to ICP. Eighteen with TBI received hypertonic saline as bolus doses of 7.3% and as a drip of 3%. Three deaths (15%) occurred. Average length of stay was 18.3 days. There were no MRI changes after being in a hyperosmolar state. Further, at 6 month follow-up, rehabilitative outcomes were equivalent to those treated with mannitol. Hypertonic sodium chloride appears to be effective for reducing intracranial pressure due to head injury related cerebral edema. This study has created the stimulus to further evaluate and compare this regimen to other modalities for the treatment of cerebral edema in head injury.

ABSTRACT

7.5% Hypertonic Saline Provides Optimal Protection Against Remote-Organ Injury following Gut Ischemia/Reperfusion

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Sponsored by: Rosemary A. Kozar, M.D., Ph.D., Department of Surgery

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Hypertonic saline, ischemia/reperfusion, multiple organ failure

Post-injury multiple organ failure (MOF) has been linked to neutrophil invasion resulting from the inflammatory response of the gut following ischemia/reperfusion (I/R) due to shock. Hypertonic saline (HS) has been shown to protect against neutrophil-mediated injury; however, the optimal dose was not known. We hypothesized that a dose-dependent effect exists related to increasing tonicity and that the optimal gut protective dose would provide better protection against remote organ injury than large volume isotonic crystalloids. **Methods:** In experiment 1 (dose response) rats were assigned to controls (sham/no resuscitation, sham/4ml/kg 7.5% HS, SMAO/no resuscitation), SMAO/equal volume (4ml/kg 0.9% NS, 4ml/kg 2.5% HS, 4ml/kg 5% HS, 4ml/kg 7.5% HS and 5 ml/kg 10% HS) or SMAO/equal sodium (3ml/kg 0.9% NS, 12ml/kg 2.5% HS, 6ml/kg 5% HS, 4ml/kg 7.5% HS and 3ml/kg 10% HS). In experiment 2 (end organ injury), rats were assigned to the same control groups, and to either SMAO/NS (33ml/kg 0.9% NS, equal salt load) or SMAO/HS (4ml/kg 7.5% HS). The SMAO was clamped for 60 minutes and boluses given 5 minutes prior to clamp removal. After 6 hours ileum and lungs were harvested for analysis of histologic injury, myeloperoxidase (MPO) as an index of neutrophil mediated injury, and serum ALT and AST drawn as a marker of liver injury. **Results:** In experiment 1, equal volume and equal sodium decreased injury and inflammation in a dose dependent fashion, with the optimal effect seen at 7.5% HS. In experiment 2, NS resuscitation resulted in minimal improvement of SMAO-induced increases in serum ALT and AST or lung injury and inflammation whereas 7.5% HS resuscitation significantly decreased these parameters. In conclusion, the protective effect of HS is due to increasing tonicity. NS had little effect on SMAO-induced remote organ injury, while optimal dose HS resuscitation was quite protective. This supports the growing evidence that HS protection may be due to its gut protective effects.

ABSTRACT

The Effects of Artificial Tears and Micro-Environment Glasses on Signs and Symptoms of Dry-Eye in Computer Users

MARTIN T. PAUKERT The University of Texas at Houston Medical School Class of 2008

Sponsored by: Richard W. Yee, MD, Department of Ophthalmology

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
Richard W. Yee, MD, Department of Ophthalmology

Key Words: Computer vision syndrome(CVS), Micro Environment Glasses(MEGs), Dry-eye, Video Display Terminal(VDT)

Research has shown both a high association between dry-eye complaints and daily computer use as well as a drastic decrease in blink rate while working on a computer. We hypothesize that this decrease in blink rate allows excessive evaporation of tears to reduce ocular surface protection, allowing environmental insult to damage the surface of the eye. In this study we evaluated the efficacy of two means of protecting the ocular surface while using the computer: artificial tears which replace lost tears, and Micro-environment Glasses(MEGs) which hyper-humidify the ocular micro-environment, resulting in decreased tear evaporation. 33 subjects were divided into symptomatic and asymptomatic groups according to OSDI classification. Each of the subjects was assessed at baseline and then after playing a high concentration computer game for 30 minutes in each of four conditions: no protection, artificial tears, MEGs, and a combination of MEGs and tears. The efficacy of the three interventions was assessed by comparing each trial to no protection. Tear breakup time(TBUT), lissamine green staining, fluorescein staining, and conjunctival injection were measured and a subjective comfort questionnaire given. In the MEGs trial, there was improvement in TBUT(from 7.57s to 10.40s, $p=0.036$), and subjective discomfort score (from 2.63 to 1.65, $p=0.0003$) among symptomatic subjects. Similar results were obtained in the MEGs+tears. Symptomatic subjects also had a reduction in lissamine green staining in the MEGs+tears trial (from 11.60 to 7.64, $p=0.0008$). We conclude that MEGs are effective at reducing dry-eye complaints in symptomatic subjects, and that artificial tears offer additional protection when used in conjunction with MEGs.

ABSTRACT

Aortic Baroreceptors and MAP Following Aortic Arch Repair

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Class of 2008

Sponsored by: Charles C. Miller III, PhD, Department of Cardiothoracic and Vascular Surgery

Supported by: Department of Cardiothoracic and Vascular Surgery

Key Words: Aortic baroreceptor, blood pressure, aortic aneurysm

Introduction: Blood pressure in humans is regulated in part by baroreceptors and chemoreceptors in the carotid bodies and the aortic arch. Large aneurysms arising from the aortic arch are life threatening and require surgical repair to prevent rupture. Extensive repair may require resection of the aortic receptors. We studied the change in mean arterial blood pressure (MAP) in patients following different degrees of aortic arch resection, to assess the relationship of aortic body resection with MAP control.

Methods: We reviewed blood pressure records pre-operatively and for forty-eight hours post operatively for fifty ascending arch patients selected by random number algorithm from our database. Polynomial statistical models were constructed to evaluate change in blood pressure as a function of aortic segment removed controlling for inotropic and vasoactive drug use.

Results: Resection and graft replacement of the greater curvature of the aortic arch is associated with significant alterations in blood pressure in the first forty-eight hours postoperatively, independently of administration of inotropic and vasoactive medications. The effect appears to be most pronounced in the first twelve hours.

Conclusion: Location of resection is correlated with postoperative blood pressure control, which may be related to loss of aortic baroreceptor function. Future studies should address the role of carotid receptors and long term blood pressure control.

ABSTRACT

The Effectiveness of a Resorbable Plates in Nasal Reconstructive Surgery

STACI D. PEREZ The University of Texas at Houston Medical School Class of 2008

Sponsored by: John F. Teichgraeber, MD, FACS., Department of Surgery, Division of Pediatric Surgery *and*
James J. Xia, MD, PhD, Department of Oral & Maxillofacial Surgery

Supported by: Department of Surgery, Division of Pediatric Surgery

Key Words: cheiloplasty, resorbable plate, nasal asymmetry

Purpose: To evaluate the surgical outcomes after primary placement of resorbable internal plates in the nasal tip in patients with cleft lip and palate.

Materials and Methods: Pre- and post-operative photos of nine primary and secondary cheiloplasty patients were used. Photogrammetric nasal analysis was performed using a custom computer program. The following measurements were made on both the right (cleft side) and left side (non-cleft side): nasal tip projection, alar tangent angle, total length of nostril, ala contour, alar width and height, and alar base width. The columella deviation was also measured. The difference between the right and left alar contour was calculated. The ratios were calculated for the other right and left measurements. Means and standard deviations were calculated. Finally, paired t-tests were performed.

Results: There were statistically significant differences between all the pre- and post-operative measurements. The nasal tip projection and columella deviation were corrected significantly towards the symmetry. The alar width on the cleft side was significantly reduced towards the normal, while the alar height was significantly increased towards the normal. The length of the alar base and the total length of the nostril on the cleft side were significantly reduced. The alar tangent angle on the cleft was significantly increased towards the non-cleft side after the surgery. Finally, the alar contour difference showed a significant improvement of the alar contour on the cleft side.

Conclusion: The effectiveness of the nasal reconstructive surgery using a resorbable internal plates has been proven.

ABSTRACT

Teleconsultation Utilizing a Digital Camera Phone for Plastic Surgery Hand Emergencies

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Sponsored by: Erik S. Marques, MD, Department of Surgery, Division of Plastic Surgery

Supported by: The Dr. Thomas D. Cronin Chair in Plastic Surgery, The University of Texas at Houston Medical School

Key Words: Telemedicine, plastic surgery, hand emergencies

Purpose: Ideally, clinical evaluation of a patient with a hand emergency should be performed immediately by an attending surgeon with expertise in hand and microsurgery. At most academic medical centers, surgeons with hand experience are on-call 24 hours per day, 7 days per week; however, they are not in-house. Therefore, initial evaluation of these patients is frequently carried out by physicians with less training and less experience than the on-call plastic surgeon. Management decisions are guided by the on-call resident's verbal description provided to the attending. There are few instances in which patient care is compromised. However, there have been occasions when a patient's injury has been underestimated by the resident and the appropriate care delayed as a result. Hand injuries mandating emergent operative intervention have gone unrecognized with resultant patient morbidity (infection, functional compromise, digital loss). The purpose of this study is to investigate whether the use of a digital camera phone will help minimize errant management decisions.

Methods: This is a 3 month prospective study conducted at Memorial Hermann Hospital. Plastic surgery hand consults while Dr. Marques is on-call are included in the study. The mobile phone used in the study is a Verizon LG-VX-8000 with a color display and built-in 1.3 megapixel high resolution digital camera. The resident performs a focused history and physical examination. Images of the injury and any pertinent x-rays are taken with the mobile camera phone. Resident findings and recommendations are then discussed with Dr. Marques by telephone. The digital images are then transmitted via resident camera phone to attending (ESM) e-mail or camera phone for immediate viewing. Management decisions may be modified after attending review of the digital images.

Results: During the 3 month study period, 18 days of plastic surgery hand call were taken (6 calls / month). 22 patients that required immediate evaluation were included in the study (16 males and 6 females). Of the 22 patients, 19 patients had traumatic injuries (86%) and 3 patients had non-traumatic hand emergencies (14%). 73 digital images were transmitted using the camera phone (mean 3.3 images / case). After case presentation and review of the digital images by the attending, the resident's plan was modified 2 times (9%). The attending agreed with the plan outlined by the resident 91% of the time (20/22 patients). Both patients in which the resident plan was modified underwent immediate procedures. One patient required bedside drainage of a hand abscess. The other patient was taken emergently to the operating room for exploration of a forearm laceration with a transected radial artery who otherwise would have been discharged home from the ER.

Conclusion: We have found that teleconsultation with the digital camera phone is a user-friendly and cost-effective way to minimize potential errors in management. We plan to continue utilizing the digital camera phone in evaluating hand emergencies.

ABSTRACT

T SPOT-TB, a New Tuberculosis (TB) Diagnostic Test: Sensitivity and Specificity in Children

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Sponsored by: Kimberly C. Smith, MD, MPH, Department of Pediatrics

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
Kimberly C. Smith, MD, MPH, Department of Pediatrics

Key Words: Tuberculosis, enzyme-linked immunospot assay, diagnosis

The Mantoux tuberculin skin test (TST) is one of the only available diagnostic tools for latent TB infection, but is known to have impaired sensitivity/specificity, because it can cross react with BCG vaccine and *non-TB Mycobacterium*. The objective of this study was to determine the sensitivity and specificity of T spot-TB, a new enzyme-linked immunospot assay for interferon gamma (IFN- γ) using two antigens highly specific for *M. tuberculosis*. Lack of tools to accurately diagnose latent tuberculosis infection (LTBI) and lack of a gold standard test with which to compare T spot-TB, makes assessing its sensitivity/specificity challenging. We present degree of agreement between Mantoux TST and T spot-TB and a comparison of test performance between TST and T spot-TB. Individuals with and without TB participated in the study by giving a 4 mL blood sample and having a TST placed. A total of 78 participants were included in the data analysis, median age 9.5 yrs. T spot-TB agreed with TST 62% of the time. For participants without LTBI (n=26), both T spot-TB and TST were negative 100% of the time. For participants with culture confirmed TB (n=4), both T spot-TB and TST were positive 75% of the time in the same patients. For participants with *clinically* diagnosed TB (n=11) T spot-TB was positive 82% of the time compared to 91% for TST. For LTBI (n=37) T spot-TB was positive 48% of the time. In conclusion, T spot-TB demonstrated at least equal performance as TST in cases where diagnosis was certain. In cases where there was more diagnostic uncertainty, the results are more difficult to interpret especially since the TST is used as part of the diagnosis and may be inaccurate. More research will need to be done to establish accurate measures of sensitivity and specificity of T spot-TB in cases of diagnostic uncertainty. However, preliminary results are promising.

ABSTRACT

Pathophysiologic Mechanisms in Acute Bacterial Diarrhea

JOSEPH J. SCHLESINGER The University of Texas at Houston Medical School Class of 2008

Sponsored by: Herbert L. DuPont, MD; Department of Internal Medicine

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Bacterial diarrhea, breath hydrogen yesting, Irritable Bowel Syndrome (IBS)

Studies have shown that enteric bacterial infection predisposes to persistent intestinal symptoms and post-infectious IBS. In our study, 52 U.S. college students studying for the summer in Mexico 2005 were classified as asymptomatic (AS) (n=28), or with acute (A) (n=13) or persistent (P) (n=11) diarrhea. We employed the non-absorbed lactulose in a breath test to determine bacterial overgrowth or oro-cecal transit time. Water load tests documented changes in gastric accommodation and visceral hypersensitivity. A test of abnormal intestinal permeability seen in IBS was executed with lactulose, sucrose, sucralose, and mannitol dissolved in water to ascertain the ratio of the aforementioned sugars in the urine. Finally the HADS and SF-36 psychological tests were administered. A and P patients were matched with AS patients based on chronological enrollment. Breath samples were analyzed with the Quintron Microlyzer. Treatment for travelers' diarrhea was with known effective antibacterial drugs. Concentration of urine lactulose varied greatly; however, A patients exhibited 0.02 +/- 0.005mg/ml. A patients exhibited the lowest concentration of urine sucrose at ND-0.0076mg/ml. P patients exhibited the highest concentration of urine mannitol at 0.9922-3.3230mg/ml suggesting existence of a permeability defect. Our studies showed positive glucose results in 67% (4/6), 100% (3/3), and 50% (1/2) for AS, A and P diarrhea subjects, respectively. Lactulose breath tests showed positive results in 63% (5/8), 63% (5/8), and 50% (3/6) for AS, A, and P cases, respectively. Glucose/lactulose combination showed 79% (11/14), 100% (2/2), and 100% (3/3) for AS, A and P cases, respectively. Of all subjects studied, there were positive results in 73% (8/11), 59% (13/22), and 84% (16/19) for glucose, lactulose, and combination, respectively. The frequency of positive results was higher than would be expected for healthy persons in the United States. American students in Mexico may experience alterations of oro-cecal intestinal transit time; however, this would not explain increased breath hydrogen levels with ingestion of glucose. Small bowel bacterial overgrowth occurring shortly after arrival in Mexico is the likely explanation for our findings. The changes appear to occur early after arrival to Mexico. It is desirable in future studies to obtain baseline data before leaving the U.S., or on the day of arrival in Mexico.

ABSTRACT

Isolation and Characterization of *Bacillus anthracis* Mutants Altered for Protective Antigen Synthesis

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Sponsored by: Theresa M. Koehler, PhD, Department of Microbiology and Molecular Genetics

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35 DK007676-13

Key Words: *Bacillus anthracis* toxin, protective antigen, transposon

The toxin produced by *Bacillus anthracis* consists of three proteins: lethal factor, edema factor, and protective antigen (PA). PA is required for entry of the toxin into eukaryotic cells and is thus a central component of the toxin. The PA gene *pagA* is controlled by multiple *trans*-acting regulators, but none have been shown to act directly on the *pagA* promoter sequence. To find additional *pagA* regulators, I screened a *himar1* transposon insertion library of *B. anthracis* for mutants exhibiting altered PA synthesis. Clones from the insertion library were cultured in conditions that favor *pagA* expression. Culture supernates were spotted onto membranes and probed with polyclonal anti-PA serum. Of 2304 mutants screened, eight appeared to be affected for PA synthesis. One of these consistently showed a PA-negative phenotype in repeated assays. SDS-PAGE and Western Blot analysis confirmed the mutant phenotype. In a parallel effort, I created a recombinant plasmid that will facilitate replacement of gene BA5151 in *B. anthracis*. The BA5151 locus was identified previously in a similar immunoscreen for PA-deficient mutants. I cloned a spectinomycin-resistance gene flanked by sequences upstream and downstream of BA5151 into a plasmid vector. The construct was electroporated into the parent strain. Recombination between the vector and the BA5151 locus will result in a BA5151-null mutant. The isolation of a new PA-deficient mutant and the steps toward creation of a null-mutation in a locus associated with PA synthesis have advanced efforts to reveal the molecular basis for control of an important anthrax toxin gene.

ABSTRACT

Evaluation of Antibiotic Impregnated Microspheres

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Sponsored by: Catherine Ambrose, PhD, Department of Orthopaedic Surgery

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
and

Zimmer grant to Catherine Ambrose, Ph.D and Terry Clyburn, MD, for
*Evaluation of Trabecular Metal Implants and Antibiotic Impregnated Microspheres in
Osteomyelitis*

Key Words: Microsphere, tobramycin, osteomyelitis

Biodegradable microspheres capable of being impregnated with antibiotics have been developed as a drug delivery system. This study fabricated microspheres with various drug-formulations in order to assess which has the most effective elution profile for treatment and prophylaxis of osteomyelitis. Five formulations were assessed: 10% chemical grade tobramycin; 10% medical grade tobramycin; 25% medical grade tobramycin; 10% recombinant human lactoferrin (rhLTF); and 10% medical grade tobramycin with 10% rhLTF. The entrapment efficiency of each formula was evaluated by dissolving samples of the microspheres in dichloromethane. Using the entrapment efficiency and the amounts of tobramycin that released into PBS over four weeks, individual elution profiles were calculated. 10% tobramycin formulas possessed the greatest entrapment efficiencies, near 50%. The 25% tobramycin formula had the lowest entrapment efficiency, 19.2%; however, its elution profile indicated the greatest initial and cumulative release of drug, eluting nearly 70% in five days. The elution profiles of the microspheres containing 20-25% drug display one burst that releases most of the tobramycin and slowly levels. The 10% drug formulas eluted with an initial burst, a plateau, and a second, slower burst. These data indicate that although the 25% tobramycin microspheres have the lowest entrapment efficiency, they deliver the greatest dose at the fastest rate, which may be predictive of the greatest treatment efficacy. Further studies are needed to investigate the therapeutic efficacy of the various formulas in vivo, the impact of rhLTF on osteomyelitis treatment, and determine the elution profiles for rhLTF release.

ABSTRACT

Diffusely Adhering *Escherichia Coli* as a Cause for Diarrhea in Travelers to Mexico

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Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: DAEC, afa/Dr+, adherence

Diffusely Adhering *E. Coli* (DAEC) are a common cause of urinary tract infections. Their role as agents of diarrhea is controversial. DAEC are defined by their specific adherence to intestinal cells and by the presence of specific genes. We investigated the prevalence of DAEC in North American adults that acquired diarrhea while traveling to Mexico during the summer of 2004. Eligible patients had acute diarrhea of less than 72 hours duration with at least 3 unformed stools in 24 hours plus at least one symptom of gastrointestinal infection such as nausea, vomiting, cramping, excessive gas, urgency, frequency or tenesmus and who had not received prior antibiotic therapy. Five colonies of *E. coli* isolated from each diarrheal stool were tested for the presence of the DAEC gene afa/Dr+ by colony polymerase chain reaction (PCR). 96 subjects with diarrhea provided a stool specimen and an average of five *E. coli* colonies per patient were tested by PCR analysis using a nonspecific primer to find the afa operon which codes for the afimbrial attachment sheath that is found in DAEC and then by a specific primer to find the afa/Dr+ gene. We found that only one subject harbored DAEC as determined by PCR. We conclude that DAEC is not a significant cause of diarrhea in North American travelers to Mexico.

ABSTRACT

Synthesis of Fluorescent Cardiolipin

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Sponsored by: Dr. Diane Bick Ph.D. Department of Pathology

Supported by: Dr. Diane Bick, Department of Pathology, National Institutes of Health grant
5RO1HL72770-2

Key Words: cardiolipin, apoptosis, phospholipids, Barth syndrome, lipids, phospholipids

Cardiolipin, an inner mitochondrial membrane polyglycerophospholipid, has been shown to be important in both eukaryotic oxidative phosphorylation and apoptosis. Cardiolipin anchors cytochrome C in the inner mitochondrial membrane. Upon disassociation, cytochrome C leakage into the cytosol can lead to activation of the caspase cascade, and ultimately apoptosis. A deficiency in cardiolipin is also present in Barth Syndrome, a rare X-linked disorder leading to cardioskeletal myopathy and neutropenia.

Much of the work done today relies on ^{32}P , ^{14}C or ^3H labeled phospholipids to study the synthesis, remodeling and trafficking of cardiolipin. It was our aim to devise a non-radioactive assay for cardiolipin (CL) synthase, the rate-limiting enzyme in mammalian cardiolipin (CL) synthesis. Cardiolipin (CL) synthase catalyzes the condensation of phosphatidylglycerol (PG) and cytidine 5'-diphosphate phosphatidylglycerol (CDP-PG) to cardiolipin (CL) with cytidine monophosphate (CMP) as a by-product. We hypothesized that we could use a fluorescent phosphatidylglycerol, NBD-PG as a substrate, which would result in a fluorescent cardiolipin molecule, when paired with the proper assay. The advantages of this procedure are two-fold. The first being it is more cost effective. Second, it is more environmentally sensitive.

We needed a way to effectively separate cardiolipin (CL) from other phospholipids that are produced in our synthesis. These include phosphatidic acid (PA) and phosphatidylglycerol (PG). Also, we needed to ensure there would not be excessive steric hindrance that could prohibit the enzyme, cardiolipin synthase, from properly interacting with our substrates NBD-phosphatidylglycerol (NBD-PG) substrate and cytidine 5'-diphosphate phosphatidylglycerol (CDP-PG).

Using thin layer chromatography (TLC) and an appropriate solvent system, we have shown that it is possible to synthesize fluorescently labeled cardiolipin (NBD-CL) *in vitro*, using crude preparations of cardiolipin synthase (CL synthase) extracted and isolated from neonatal rat livers. This assay should aid in future research into the role of cardiolipin (CL) and cardiolipin (CL) remodeling in apoptosis, and in diseases such as Barth Syndrome, by allowing researchers to study cardiolipin (CL) synthesis and biophysical properties using non-radioactive but extremely sensitive fluorescent techniques.

ABSTRACT

Survey of Genotype-Phenotype Correlation in Patients Previously Diagnosed with Reis-Buckler Corneal Dystrophy

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Sponsored by: Richard W. Yee, MD, Department of Ophthalmology and Visual Sciences
Xinping Zhao, PhD, Department of Ophthalmology and Visual Sciences

Supported by: Richard W. Yee, MD, Department of Ophthalmology and Visual Sciences

Key Words: Corneal dystrophy, Reis-Buckler, anterior basement

Reis Buckler Corneal Dystrophy (RBCD) is an autosomal dominant anterior basement corneal dystrophy characterized by hazy opacities in a geographic pattern. Clinically the distinction of RBCD from other anterior corneal dystrophies is a diagnostic challenge. Therefore emphasis on genetic analysis has become an important diagnostic tool. The mutation responsible for RBCD has been identified on the *BIGH3* gene, mapped to the 5q31 chromosome locus. The purpose of this study was to confirm or revise the initial clinical diagnosis of RBCD in eleven families using molecular diagnosis and to search for novel mutations. Corneal phenotypes were assessed by biomicroscopy, slit lamp and clinical history. Blood samples were collected from the affected and unaffected family members. After extraction and purification of DNA, polymerase chain reaction was used to amplify exons 4 and 12 of the gene, followed by single strand conformation polymorphism and bidirectional sequencing. Molecular analysis resulted in eight families with R555Q mutation and one family with R124L, both previously associated with RBCD. Another family had R124C, a mutation that has been established as a Lattice Type I/IIIA dystrophy despite the clinical diagnosis of RBCD. In addition, one family had a novel mutation H626P. This study confirms that molecular analysis can provide a more accurate way to diagnosis corneal dystrophies. It also reveals a novel mutation previously not associated with RBCD. Further assessment of clinical data and images will provide information about the role specific mutations play in clinical presentation.

ABSTRACT

First Place Co-Winner, 2005 Frank Webber Prize for Student Research

Metabolic Dysregulation as a Cause of Insulin Resistance in Skeletal Muscle in a Rat Model of Diet Induced Obesity

MAI K. TRAN

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Sponsored by: Heinrich Taegtmeyer, MD, DPhil, Department of Internal Medicine

Supported by: National Institute of Diabetes and Digestive and Kidney Diseases, T35
DK007676-13

Key Words: Obesity, Insulin Resistance, Metabolism

Obesity is a risk factor for many diseases including diabetes. Having previously studied the effects of obesity on the heart, we now investigated the metabolic changes that occur in skeletal muscle as a result of diet-induced obesity. We wanted to determine whether skeletal muscle of rats fed western and high fat diets would become insulin resistant.

Male Wistar rats (2 months of age) were fed control, western, and high fat (10%, 45%, and 60 calories from fat) diets for 1, 4, and 8 months. Insulin sensitivity was determined using the isolated soleus muscle incubation (n=8/group). Glucose oxidation, oleate oxidation, and glycogen production were measured after stimulation with a logarithmic range of insulin. After eight months, rats fed western and high fat diets weighed more than rats fed control diet (986±51 and 925±16 vs. 842±34 g, respectively; p<0.01). Glucose oxidation was decreased at all time points in the high fat (-50%) and western (-10%) groups compared to control (p<0.05). There was a trend for increasing oleate oxidation in the western (+23%) and high fat (+31%) fed groups. Glycogen synthesis increased in western and high fat groups compared to control at one month (+47% and +61%, respectively, p<0.05) but was not increased at 4 and 8 months.

In conclusion, skeletal muscle of rats fed western or high fat diets exhibits decreased response to insulin. There appears to be a discordance in glucose uptake and oxidation in the early stages of diet induced obesity that is absent with continued feeding. The mechanisms and consequences of obesity on skeletal muscle insulin resistance requires further study.

ABSTRACT

Cytokine Regulation of the Peptidase CD26: Implications for Hematopoietic Stem Cell Transplantation

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Class of 2008

Sponsored by: Dr. Kent Christopherson II, PhD, Institute of Molecular Medicine

Supported by: Dean Stanley Schultz, MD, The University of Texas at Houston Medical School
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Key Words: Peptidase CD26, hematopoietic stem cell transplantation, umbilical cord blood

Hematopoietic stem cell transplantation (HSCT) serves as a successful treatment option for patients suffering from severe hematologic diseases. However, patient survival is compromised when umbilical cord blood (CB) is used as a source for transplant into adult patients. This results from a discrepancy between the need for large numbers of transplantable Hematopoietic Stem Cells (HSC) and the limited quantity available in a single CB collection. We have recently delineated a novel method by which inhibition of the peptidase CD26 on donor HSC increases the number of donor HSC trafficking to recipient's bone marrow. The purpose of this study was to examine the effect of cytokines on CD26. CD34⁺CD38⁻ defined HSC were isolated from CB and treated with cytokines commonly utilized in the clinical setting to help patients' hematopoietic systems recover from stresses such as chemotherapy or anemia. Cells were treated with Granulocyte-Colony Stimulating Factor (G-CSF), Granulocyte Macrophage-Colony Stimulating Factor (GM-CSF), Erythropoietin (EPO), Thrombopoietin (TPO), or Stem Cell Factor (SCF) in IMDM media, 20% FBS, 37°C, 5%CO₂ for 18 hours. Cells were then analyzed for changes in CD26 expression in the context of the chemokine receptor CXCR4 by multi-variant flow cytometric analysis. G-CSF, GM-CSF, and SCF treatment resulted in increases in the percentage of cells expressing CD26 as well as the amount expressed per cell. Treatment with EPO and TPO showed modest changes in CD26 activity. Differential expression of CXCR4 was also noted. These observations have critical clinical implications which could intimately affect the treatment protocol established for adult post-HSCT patients.

ABSTRACT

Calpain 1 Gene Expression and Calpain-Like Protease Activity Increase with Atrophic Remodeling of the Heart

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Key Words: Calpain, atrophy, cardiomyocyte, gene expression

Background: Cardiac hypertrophy is an independent risk factor for the development of heart failure. Regulators of cardiomyocyte atrophy are potential targets for reversing cardiac hypertrophy. Calpain 1 and calpain 2 are essential regulators of skeletal muscle atrophy. Their role in heart muscle is undefined.

Hypothesis: Calpain 1 and 2 regulate cardiomyocyte atrophy.

Methods: We used two models of unloading-induced and one model of starvation-induced cardiomyocyte atrophy. Failing human hearts of 10 patients (8 men, age: 40.4 ± 5.7) were unloaded with a left ventricular assist device (mean duration was 200 ± 38 days, range: 64-437 days). Normal rat hearts were unloaded by heterotopic transplantation for 7 days. Neonatal rat ventricular cardiomyocytes (NRVM) were starved of amino acids for 18 hours. Using quantitative RT-PCR we measured transcript levels of calpain 1 and calpain 2 at baseline and after treatment. In the unloaded rat hearts we also measured calpain-like protease activity.

Results: Mechanical unloading increased transcript levels of calpain 1 in 8 out of 10 patients and did not significantly change calpain 2 gene expression. The change in calpain 1 gene expression correlated negatively with duration of unloading ($r^2=-0.73$, $p<0.01$). Unloading of the heterotopically transplanted rat heart significantly increased calpain 1 transcript levels and calpain-like protease activity. Starvation (no amino acids) increased calpain 1 gene expression in NRVM.

Conclusions: Calpain 1 gene expression and activity increases during cardiomyocyte atrophy suggesting that calpain 1 may be a potential target for reversing hypertrophy. Future studies will examine the trophic effect of calpain 1 inhibitors in models of atrophy and calpain 1 activators in models of hypertrophy.

INTERNATIONAL MEDICAL STUDENTS

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ABSTRACT

Analysis of the Phospholipid Components at the Cell Poles

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Supported by: Fu-Jen Catholic University, Taiwan

Key Words: Minicell,,cardiolipin,pgsA

To date, at least thirteen proteins have been demonstrated to be required in *E. coli* for division site selection and septum formation; however the specific involvement of phospholipids in these membrane-associated processes has not been investigated. Determining the roles of phospholipids and phospholipid-protein interactions during cell division will be necessary to understand this complex process. Heterogeneous distribution of cardiolipin (CL), along bacterial membrane, is demonstrated by using of the cardiolipin-specific fluorescent dye, 10-*N*-nonyl acridine orange, which shows CL-enrichment at the cell poles and cell center. The lipid composition of minicells, dividing from the cell poles in Δ min mutants, reflects that of the cell poles. It is demonstrated that CL is fourfold increase in minicells compared with normal *E. coli* cells, which is consistent with the enrichment of CL at the cell poles. We hypothesize that the anionic domains appear to participate in the binding of amphitropic proteins (MinD, DnaA and FtsA) and they are responsible for correct selection of cell division site. To further test whether anionic lipids segregate into specific domains at the cell poles of *E. coli*, the phospholipid composition of the poles of the triple mutant UE54 (*pgsA lpp2 rcsF*) will be analyzed. This mutant completely lacks of phosphatidylglycerol and CL but accumulates their precursors, phosphatidic acid and CDP-diacylglycerol, which are also anionic lipids. If anionic lipids are concentrated at the cell poles of UE54 strain, then enrichment of anionic lipids will be observed in minicells. The Δ min CDE mutation is transferred into the UE54 strain and minicells produced by resulting strain are analysed. Minicells are successfully isolated after six sucrose gradient centrifugations. Further analysis and comparison of the phospholipid components in minicells and parental cells, from which minicells are derived, help to define the roles of anionic phospholipids domains in the view of cell division.

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ABSTRACT

Knockdown of RGS2 by RNAi

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Supported by: Fu-Jen Catholic University, Taiwan

Key Words: G protein signaling, RNAi, adenylyl cyclase

Regulators of G protein signaling (RGS) proteins accelerate the GTPase activity of G protein subunits in vitro, negatively regulating G protein-receptor signaling. An RGS family member RGS2 has been shown to decrease cAMP accumulation by inhibition of the enzyme, adenylyl cyclase. Previous work also showed that RGS2 mediates vascular smooth muscle relaxation and blood pressure via regulation of Gq-signaling. However, the true physiological role of RGS2 is not fully understood. Here we used RNAi to knockdown RGS2 in order to understand the regulation of cAMP production in cardiac myocytes by RGS proteins.

Method: We designed four 19-nucleotide oligonucleotides (306, 341, 540, and 545) based upon the rat RGS2 sequence, and cloned these into the pSIREN-DNR-DsRed RNAi vector. The resulting plasmids were transfected into PC12 cells in order to initially characterize these RNAi sequences. After 36-hours of culture, the cells were harvested and Western blots were performed with antibody against RGS2.

Results: Based on the DsRed expression, the transfection rate of these RNAi plasmids was 30-40%. In the Western blots, we observed a 30% decrease in RGS2 expression using RGS2-545i compared with the negative control luciferase RNAi.

Conclusion: RGS2-545i decreased RGS2 expression. This construct will be used to make an adenoviral RNAi against RGS2 and further research on the physiological role of RGS2.

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UNDERGRADUATES

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ABSTRACT

Invitro Stress Resistance in Pathogenic Candida Species

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Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: Candida, fungal infection

The yeast genus *Candida* contains several pathogenic strains. Through patient studies we have observed that there might be a connection with the growth environment and how pathogenic a species is. In this study we focused on determining any correlation between stress resistance and the strains' pathogenicity. Using 10 related species (*C. albicans*, *S. cerevisiae*, *C. glabrata*, *C. lusitaniae*, *C. guilliermondii*, *C. dubliniensis*, *C. parapsilosis*, *C. tropicalis*, *D. hansenii*, and *L. elongisporus*), we grew the strains at 30°C by streaking them on various media including serum, acetate, ethanol, oleate, SLAD, various percentages of ethanol on YPD, various molarities of calcofluor on YPD, various pH levels, oxidative stress, and salt concentrations. We also tested the strains on YPD at various temperatures, ranging from 30°C to 46°C, and performed heat shock tests. As well as streaking these plates, we also plated dilutions (78,000-fold) of the strains on the same media. Through these tests, we concluded there was a correlation with extreme pH and high temperatures with virulence potential. The other stresses did not appear to be correlated to the pathogenicity, therefore their results are not relevant to this study. Recognizing these connections will later aid in patient studies. More studies focusing on how extreme pH and high temperatures affect the genome of these pathogens will be needed in order to truly benefit patient study outcome.

ABSTRACT

Obese Mice and Pregnancy

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Key Words: Obesity, exercise, pregnancy, mice

As mice age, they tend to become obese when given unlimited food supply. Studies have shown that in the past, obese mice have had difficulties with conception. Our hypothesis was that if given a chance to exercise, the mice would lose weight and have more pups. In this study, obese ICR mice from a previous food experiment were selected and placed as breeder pairs in different cages. Three breeding pairs were put in standard mouse cages measuring 6X11X5 inches. The other three pairs were put in larger rat cages measuring 8X18X9 inches and supplied with a running wheel. Weekly weights of all mice were obtained for one month. The weights were difficult to utilize because all of the females became pregnant over the course of the study, causing them to increase in mass. All six of the females gave birth to numerous pups. After the pups were a week old, they were counted. The mice in the regular cages averaged fourteen pups. In the running wheel cages, the mice averaged ten pups. Although the results did not confirm our hypothesis, more data with additional mice would be needed to further come to significant conclusions. The mice did, however, seem to use and enjoy the running wheel.

ABSTRACT

Bond Strength of belleGlass Indirect Composite to Enamel and Dentin

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Supported by: The University of Texas at Houston Dental School

Key Words: Indirect Composite, Resin Cements, BelleGlass

Purpose: To compare and contrast the strength of bond of belleGlass Indirect Composite to both enamel and dentin using 4 resin cements with bonding agent (Multilink, Nexus, Panavia, RelyX ARC), 1 self adhesive resin cement without bonding agent (RelyX Unicem), and 1 resin modified glass ionomer luting cement (RelyX Luting Plus).

Methods and Materials: 96 specimens of buccal and lingual surfaces of freshly extracted teeth were mounted in resin and ground to expose 48 enamel and 48 superficial dentin surfaces. The surface was polished with 600 grit SiC discs. Each adhesive system was used according to manufacturer's directions. Prior to bonding, the belleGlass Indirect Composite material was molded and cured in the shape of inverted truncated cones with a diameter of 3 mm at the bonding surface and 5 mm at the top of the cone, then microetched. 8 specimens were used for each condition. After bonding, specimens were kept in water in an incubator at 37°C for 24 hours. They were then debonded in tension using an instron testing machine at across-head speed of 0.5 mm/min.

Results: RelyX ARC to enamel and dentin and Nexus to enamel displayed the highest bond strengths.

Conclusion: When bonding indirect composites to enamel and/ or dentin a total etch/ bonded cement should be used for the best bond strengths.

ABSTRACT

Residential Pool Awareness

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Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: drowning, prevention, education, awareness

Background: Drowning in Houston, Texas has risen dramatically in the past year. Beginning in June 2005, there has been an average of one pediatric drowning per week. In response, the Houston Police Department (HPD), Houston Trauma Link, and the City of Houston Health and Human Services (HHHS) have united to spread awareness and aid in the prevention of residential drowning. HPD has a mandate to inspect community and residential pools, using law to enforce improvements, while Trauma Link is using this opportunity to educate.

Objective: The objective was two-fold: 1) To create a tool, i.e. a police checklist, that targets pool safety violations 2) To create a door-hanger that identifies violations, educates through text and images, and gives resource information.

Methods/Design: A literature search was done, the state and city codes and ordinances were reviewed, and P.I (MG) shadowed pool inspectors. The information from these sources was compiled into the checklist and door-hanger. A pilot is being conducted using these tools.

Results: Violations are based on law: fencing, drains, and water clarity; while educational intervention targeted parental supervision and the ideal pool environment.

Conclusions: The door-hanger and checklist will be applied to areas of Houston (separated by zip code) where there is a high concentration of pools. Based on the findings from our checklist, we will target residential pool violations and launch an educational campaign in those areas.

ABSTRACT

Passive Exposure Induces Persistent Learning Effects

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Key Words: 'exposed' location, 'unexposed' location, d'

The visual system is constantly bombarded with both relevant and irrelevant stimuli. Attentional mechanisms allow us to attend to relevant stimuli while filtering out irrelevant stimuli, and yet whether this filtering process effectively eliminates irrelevant information is poorly understood. Do irrelevant stimuli have the capacity to influence our behavior? To answer these questions we examined the discrimination of stimulus orientation when subjects were passively exposed to patterned stimulation. Each session consisted of two training phases in which subjects were instructed to maintain fixation to a central dot while counting how many times a letter appeared at an above location. This procedure allowed us to control spatial attention, and, simultaneously, present oriented stimuli at an 'exposed' location and unoriented stimuli at an 'unexposed' location. To be confident that attention was directed to the letter identification task we required an accuracy of 90%. The exposed region alternated between sequences of sine-wave gratings at 60° followed by 150° , while the unexposed location was stimulated by a gray circle. During the test phase, 100 trials were presented to each region, and subjects indicated if the test (presented at $150^\circ \pm 4^\circ$) was similar to the sample (which was always 150°). The analysis consisted of determining the difference between z-scores for the hit rate (HR) and the false alarm rate (FAR): $d' = z(\text{HR}) - z(\text{FAR})$. We plotted d' for exposed and unexposed locations as a function of time and observed a steady increase at the exposed region and minimal changes at the unexposed region. Importantly, exposure-based perceptual learning was most prominent when the exposure stimuli were alternating orthogonal gratings. We conclude that passive exposure has a significant effect on visual behavior. This indicates that theories of perceptual learning assuming that attention should always be directed to the trained location need to be revised.

ABSTRACT

Psychophysical Eye Tracking

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Key Words: Eye tracking, psychophysics, double-step, saccade, face,

During rapid eye movements known as saccades, the brain suppresses the visual system so that the world does not appear to go streaking by during this high-velocity motion. However, it is unclear how the visual system deals with this small 'gap' in time (about 80 milliseconds) while the eyes are in flight. Previous studies have suggested that the gap is filled in retrospectively by whatever images the eyes land on. To study this hypothesis, we asked participants to view a face on the computer screen. At the beginning of each trial, the subject was instructed to fixate on the eyes of the face, which return their gaze, and then to execute a quick saccade to a 'name tag' (containing 4 random letters) as quickly as possible after the face's eyes look away. Additionally, the subject was told to return as quickly as possible to the center of the screen after viewing the stimulus, as the computer face's eyes will return in order to 'catch' him. The subject then provided two reports: 1) the 'name' on the tag, and 2) whether they were caught by the face on the computer. Eye movements were monitored at 30 Hz with a video-based eye-tracker. We found that when the subject's eyes and the computer's eyes returned to the center at roughly the same time, subjects reported being caught 20% more often than the camera reported. In other words, subjects' assessments of their eye positions lagged their true eye movements. This simple yet novel finding contributes to a growing body of data on timing judgments around the time of saccades.

ABSTRACT

Effects of Chemical Chaperones and Temperature on Expression of Prostaglandin H Synthase in a Baculovirus Expression System

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Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: chemical chaperones, prostaglandin, PGHS, ibuprofen, glycerol

Establishing an efficient overexpression system is a major challenge in elucidating the reaction mechanism of prostaglandin H synthase (PGHS), the key enzyme of eicosanoid biosynthesis and the target of aspirin and many other non-steroidal anti-inflammatory agents. The baculovirus expression system commonly used gives low yield, thus limiting the availability of mutant enzyme for mechanistic studies. Previous work using small molecules to facilitate the folding and processing of proteins suggested potential alternatives to increase the yield of the post-translationally modified PGHS-2 [Bernier, V.; et al. (2004) *TRENDS in Endocrinology and Metabolism* 15, 222-228]. Ibuprofen, a cyclooxygenase inhibitor that could bind to the nascent protein, aiding in folding and structural stability and prevent degradation in the proteasome pathway, was added during expression at four concentrations (1 nM, 1 uM, 10 uM, 100 uM). Glycerol, which stabilizes and increases the maturation level of protein through an unknown mechanism, was also tested at four concentrations. Lowering the expression temperature, which may slow down the folding process, allowing a greater proportion of proteins to reach native conformation was also tried. Western blot analysis shows decreased protein yield at 25°C and in the presence of glycerol. It increased at 1 nM ibuprofen concentration but decreased at all higher concentrations. Results have indicated that ibuprofen and glycerol have a limited effect on protein yield and enzyme activity. Additional work on other chaperones may hold promise in the future for both isoforms of PGHS.

ABSTRACT

Bond Strength of Resin Composite to Desensitized Dentin

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Sponsored by: Magda S. Eldiwany, DDS, Restorative Dentistry

Supported by: The University of Texas at Houston Dental School

Key Words: Desensitizers, total-etch, superficial dentin, bond strength

Purpose: The movement of fluid in the dentinal tubules is the basic cause of tooth sensitivity. Desensitizers affect the tubular fluid movement thus eliminating pain. The purpose of this study was to investigate the effect of desensitizers on the bond strength of direct composite restoration after using three total-etch bonding agents.

Materials and Method: Freshly extracted teeth were mounted in resin and ground to expose superficial dentin. Specimens (n=8, 96 total) were polished to 600 grit. Each adhesive/composite system was bonded according to manufacturers' instructions. The composite took the shape of an inverted, truncated cone and was stored in water at 37° for 24 hours. The specimens were debonded using an Instron testing machine.

Results: The results are shown in the table. The manufacturer for BisBlock cautioned against using Prime and Bond NT because of its acidic primer.

Table 1. Bond strengths (MPa)

	Control	Hemacil & Cide	D/Sense Crystals	BisBlock Oxalate
Single Bond Plus/Filtex Z250	21.4 (3.7)	18.1 (5.6)	5.4 (1.6)	5.9 (2.1)
Prime & Bond NT/TPH	18.0 (4.9)	23.4 (4.7)	5.0 (1.3)	6.1 (3.6)
One-Step Plus/AELite	27.8 (8.3)	14.4 (3.3)	7.6 (2.3)	15.8 (4.3)

Conclusions: In general, desensitizers reduced the bond strength of resin composite to dentin. For desensitizer Hemacil and Cide, the bond strength of Prime and Bond NT increased. Products were provided by the manufacturers.

ABSTRACT

Tissue-Specific Expression of a Ubiquitin-Conjugating Enzyme (GTAP) in Mice Prone to Atherosclerosis and Cardiomyopathy

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Supported by: The University of Texas at Houston Medical School - Summer Research
Program

Key Words: Gene expression, heart disease, protein ubiquitination, atherosclerosis

Ubiquitination represents a primary mechanism for protein degradation by the 26S proteasome complex. The novel ubiquitin-conjugating enzyme GTAP has been shown to regulate ubiquitination and protein homeostasis during early development. This study tested if expression of GTAP is altered in tissues containing progenitor cells in the murine models of atherosclerosis and hypertrophic cardiomyopathy. Total proteins were extracted from different tissues (eg, testis, kidney, lung, fat, spleen, heart, aorta, liver, skeletal muscle, and thymus) of three murine strains, wild-type, apolipoprotein-E deficient, and GTP-binding protein Gs \square transgenic. Proteins were fractionated on SDS-PAGE and then subjected to Western blot analysis with antibodies specific for the COOH-terminus of GTAP to determine relative abundance of GTAP to GAPDH, a housekeeping protein. Among all tissues tested, only aorta, skeletal muscle, and spleen showed significant GTAP expression. Although no significant difference in relative abundance of GTAP existed in the Gs \square mice, the GTAP levels in the aortas of apolipoprotein-E-null mice differed from wild type controls. Densitometry for GTAP levels showed that the aortic GTAP levels were significantly ($p < 0.05$, t -test) lower in apolipoprotein-E null, atherosclerosis-prone mice (0.37 ± 0.07) than those of wild type controls (0.86 ± 0.10). Immunocytochemistry with anti-GTAP antibodies illustrated positive GTAP signals in the aorta, heart and skeletal muscle of wild type mice, particularly in the endothelium of the aorta and the coronary vessels. The data indicate the downregulation of GTAP in atherosclerotic aorta but no change in other tissues, suggesting the potential involvement of protein ubiquitination in atherosclerosis.

ABSTRACT

Estimation of Volumetrics from Magnetic Resonance Brain Images of Children with Traumatic Brain Injury, Spina Bifida / Hydrocephalus, or Autism

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Key Words: Brain MRI, Brain Volumetrics, MR image analysis, Cerebrospinal fluid, Gray/White Matter, Normal Brain Development, Traumatic Brain Injury, Autism, Spina Bifida

We use a novel fuzzy-clustering algorithm to estimate the relative proportion of gray and white matter in relation to cerebrospinal fluid in magnetic resonance (MR) images of the brain across child populations diagnosed as having 1) Traumatic Brain Injury, 2) Spina Bifida / Hydrocephalus, 3) Autism, or 4) normal. MR images consisted of T1 and T2 weighted images, which are pre-processed, aligned/co-registered, masked, and clustered. We estimate volumetrics data within specific regions of interest (ROI), specified by the mask volume, and record these quantities in a comprehensive neuro-behavioral database on the children. In these studies, the ROI was limited to the cerebrum. We verified the correctness of clustering solutions by generating digital phantom scans, corresponding to a 3D image of the clustering solutions as they would appear distributed over the mask volume. The overall goal of the project is to identify any correlations between the volumetric data with behavioral abnormalities resulting from injury, spina bifida or autism in these child populations. The specific goals of our lab group include performing the image analysis component of the broader studies.

ABSTRACT

Reaction Mechanism of Endothelial Nitric Oxide Synthase: Redox Recycling of Tetrahydrobiopterin Radical

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Sponsored by: Ah-Lim Tsai, PhD, Department of Internal Medicine

Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: nitric oxide synthase, BH₄

Endothelial nitric-oxide synthase (eNOS), which catalyzes the conversion of L-arginine to nitric oxide (NO), plays a significant role in vascular function. NOS contains two major functional domains. The oxygenase domain binds the heme prosthetic group and the tetrahydrobiopterin (BH₄), while the reductase domain holds binding sites for FMN, FAD, and NADPH. Transient kinetic studies indicate that one electron from the flavin reductase domain reduces the heme to ferrous form to enable oxygen binding. The iron bound dioxygen is further reduced by an electron from BH₄ resulting in a tetrahydrobiopterin radical. However, the steady state reaction following the addition of both BH₄ and L-arginine offers little insight into how BH₄ partakes in the intricate reaction. To better examine redox communication among FMN, biopterin and the heme, specifically the recycling of the BH₄ radical during catalysis, we decided to overexpress protein in a simplified system containing a plasmid construct of eNOS oxygenase domain (eNOS_{ox}) and a single FMN domain. To accomplish this, overexpression of His-tagged eNOS_{ox}+FMN in BL21 E. coli cells followed PCR amplification of the cDNA construct, cloning, subcloning, coexpression with PCW vector, and DNA sequencing for the final DNA product. PCR and restriction enzyme mapping verified the successful cDNA subcloning. Protein purification of wild type human eNOS_{ox} was performed utilizing nickel column chromatography to simulate future eNOS_{ox}+FMN purification. After characterization and activity analysis of the recombinant protein, stopped-flow and rapid freeze quench EPR measurements will be conducted to resolve the mechanism of BH₄ radical recycling and its redox communication with heme and FMN.

ABSTRACT

Investigation into Multiple Signaling of Prostacyclin Receptor

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Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: Prostacyclin IP receptor, Calcium Mobilization, G-Protein coupled receptor, Iloprost

It is known that the prostacyclin receptor (IP) mainly couples to Gs protein increasing intracellular cAMP concentration, which mediates vasodilation and anti-platelet aggregation functions. However, many experimental data have also indicated that the receptor can couple to Gq to increase intracellular calcium concentrations. Little information is available regarding the molecular mechanisms for the different signaling. One hypothesis is that different signaling pathways are based on different G protein coupling sites in the intracellular domains of the receptor. To test this hypothesis, the seven residues (Ala42-Ala48) in the first intracellular domain (iLP1) of the IP receptor important to the Gs signaling previously identified by our lab were investigated for their effects on Gq coupling using a calcium mobilization assay. Seven of the site-directed mutants corresponding to the Ala42-Ala48 with a Gly replacement were constructed by PCR approach, and then expressed in COS-7 cells using Lipofectamine 2000. The transfected cells were incubated with FURA2/AM fluorescent dye for the detection of calcium mobilization. The Gq coupling-mediated calcium mobilization activity of each mutant was compared with the IP wild type under dose-dependent stimulation using increasing amounts of iloprost (the receptor agonist). Preliminary results showed a persistent activity of calcium mobilization in the mutants. However, when compared to wild type, the mutants showed significantly decreased sensitivity to the iloprost stimulation. Further studies are required to investigate to what degree and how the mechanism of Gs coupling-mediating signaling may interfere with Gq coupling-mediating signaling to decrease the sensitivity of the IP receptor to its agonist stimulation.

ABSTRACT

Analysis of IGF-1R Levels in the Progression from Well-Differentiated to Undifferentiated and Invasive Endometrial Adenocarcinoma Type 1

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Sponsored by: Peter J. Davies, MD, PhD, Department of Integrated Biology and Pharmacology

Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: Endometrial adenocarcinoma, IGF-1R, transcript expression level

Endometrial adenocarcinoma Type 1 (EC1) is known to progress from a well differentiated state in grades 1 (ECg1) and 2 (ECg2) to an undifferentiated state with invasion of the myometrium by grade 3 (ECg3). In normal cells, the insulin-like growth factor 1 (IGF-1) pathway is known to play a role in regulating cellular proliferation as well as embryonic development. Several types of cancer have increased expression of the IGF-1 receptor (IGF-1R), a key component in the pathway. It has been suggested that increased expression of the receptor may contribute to increased rates of proliferation and cell invasion. We have previously shown IGF-1R is over-expressed and hyper-activated in ECg1 as compared to endometria with complex atypical hyperplasia (CAH) or normal endometrium. In this study, we evaluated the expression of IGF-1R in ECg2 and ECg3. The level of IGF-1R transcripts were quantified, using real-time Q-PCR, in RNA extracted from formalin-fixed paraffin embedded human endometrial tissue (n= 23 ECg1, n=14 ECg2 and n=6 ECg3). The transcript levels obtained were normalized for RNA loading by comparison with 18S ribosomal RNA. A significant increase ($p<0.05$) in ECg3 over ECg2 and ECg1 was found, with transcript levels 5-fold higher in ECg3 than in the well-differentiated cancers: ECg2 levels were comparable to ECg1. The increased level of IGF-1R expression in Grade 3 endometrial cancer suggests that the IGF-1R may cause increased proliferation in the endometrium and may contribute to the invasiveness of advanced endometrial cancer.

ABSTRACT

A Qualitative Comparison of Gutta Percha versus Resilon

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Class of 2006

Sponsored by: Roberta Pileggi, DDS, MS, Department of Endodontics

Supported by: The University of Texas at Houston Dental School

Key Words: Gutta percha, Resilon, obturation

A significant step in the root canal procedure is proper obturation of the root canal system. The customary obturation material is gutta percha, an isomer of rubber. Gutta percha cannot completely prevent microbial leakage, and thus long-term success of root canals is highly dependent upon high-quality coronal restorations. Recent research has yielded a new obturation material, Resilon, which claims to provide a better seal than gutta percha and possibly strengthen treated teeth. To compare the root adaptation of gutta percha to that of Resilon, sixty molar canals were cleaned, shaped and randomly divided into two groups of thirty. Group I was obturated with gutta percha and Group II was obturated with Resilon. An additional ten canals were used as positive control and another additional ten canals were used as negative controls. Teeth were tested for dye leakage using Methylene blue dye and cleared following decalcification. Areas of leakage were measured using the Image Pro Plus computer software, and the data was statistically analyzed using a t- test ($p < 0.05$). Teeth obturated with Resilon showed less dye leakage than those obturated with gutta percha, but there was no statistical significant difference between the two groups ($p = 0.10$). Nine canals in Group I (gutta percha) showed leakage, while three canals in Group II (Resilon) .

ABSTRACT

Orientation Discrimination is Consistent with an Internal Model of Gravity

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Class of 2006

Sponsored by: Valentin Dragoi, Ph.D., Department of Neurobiology and Anatomy

Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: gravity, bias, orientation, discrimination, horizontal

Gravitational laws fundamentally shape the physical environment surrounding us. Is it possible that evolution of the brain has captured Newtonian laws in a way that would influence the way we see the world? To examine this issue, we successively flashed two oriented gratings and asked subjects to determine whether the orientation of the second grating was different from the first. The first grating was always presented at 45 degrees, and the second grating could either go toward horizontal, vertical, or stay the same with respect to the first. Sensitivity to motion in the horizontal direction would be expected if visual perception were consistent with an internal model of gravity. We use a staircase threshold procedure to assess the change in orientation at which subjects could discriminate between the gratings. Separate thresholds for downward (toward horizontal) and upward (toward vertical) moving stimuli were compared. We found that the horizontal threshold was significantly lower than the vertical threshold, indicating that subjects could better discriminate gratings moving toward horizontal. Additional experiments tested whether varying the time delay between gratings would still reveal the bias. Preliminary results show a bias toward horizontal, indicating that the visual system's ability to discriminate between successive stimuli not only depends on the physical differences between them, but also the temporal order in which they are presented. Further, these results support the hypothesis that the human visual system has adapted during its evolution to be more sensitive and attuned to those visual stimuli that follow the laws of gravity.

ABSTRACT

Evaluation of a Treatment Readiness Assessment Tool for HIV Patients

JULIANNE A. SEFKO

Cedar Crest College, Pennsylvania

Class of 2006

Sponsored by: Philip C. Johnson M.D. Department of Internal Medicine

Supported by: The University of Texas at Houston Medical School - Summer Research Program *and the*
T. Ragan Ryan Memorial Summer Research Student Fund

Key Words: ARV therapy, HIV, readiness assessment

Antiretroviral (ARV) therapy effectively treats Human Immunodeficiency Virus (HIV). Patient adherence to ARV is critical to their ability to remain healthy with HIV infection. Patient readiness to begin ARV therapy is believed to be a factor associated with their likelihood to adhere to their ARV regimen, but this has not been proven. Treatment Readiness Assessment Protocol (TRAP) is a required assessment form for all clinics providing care under Ryan White Titles from HIV Services of Harris County. The HIV Wellness Program of Christiana Care Health Services at the Porter State Service Center, developed the initial TRAP, which includes five dimensions that help determine a patient's readiness to begin ARV therapy. These dimensions (mental health, substance use, environmental, cognitive and attitudinal) are expected to demonstrate a patient's positive or negative readiness. Contacts were made with professionals who developed TRAP to determine proper scoring. They reported that the TRAP was a tool which led to subjective decision making only. This led to the formation of a research committee to develop a format for scoring the tool. At St. Hope Foundation, a committee composed of a Ph.D./nurse practitioner, a physician assistant, two social workers, a college student, and a Ph.D. AIDS consultant was formed to determine a more quantitative assessment instrument. This assessment will be evaluated in a prospective study.

ABSTRACT

Effects of Low-Dose PEG-ADA on ADA Deficient Mice

BENJAMIN SHIELDS

Rice University

Class of 2007

Sponsored by: Rodney E. Kellems, MD, PhD, Department of Biochemistry and Molecular Biology

Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: Adenosine deaminase, transgenic mice, severe combined immunodeficiency

Adenosine deaminase (ADA) deficiency creates severe combined immunodeficiency (SCID) in both humans and mice. Until recently, opportunistic pulmonary infections were thought to be responsible for premature deaths in ADA deficient mice; however, recent data suggests an intrinsic pulmonary deficiency due to elevated adenosine levels. Providing mice with low-dose of ADA has been shown to relieve the pulmonary deficiency in mice while retaining at least a partially compromised immune system. It is, therefore, of interest to find a level of dosage such that mice can survive to 8 weeks but still have a compromised immune system. Transgenic ADA $-/-$ mice were used to test three different levels of dosage with PEG-ADA: 2 μ L twice a week, 10 μ L twice a week, and 20 μ L once a week. At 8 weeks, the spleen and thymus were harvested and cells were stained with CD4, CD8, and B220. Flow cytometry was then used to analyze the data and showed that the thymus cell count of mutant mice was approximately ten percent of a wild type mouse, and the spleen cell count was between thirty and fifty percent of a wild type mouse. These results support the idea that low dose PEG-ADA treatment can overcome the pulmonary deficiency while still retaining a compromised immune system; however, they also suggest that another lower dosage is probably possible that would allow mice to survive till 8 weeks of age or longer and retain a more compromised immune system.

ABSTRACT

Designing a DNA Microarray to Detect Large Deletion/Duplication Mutation for Tuberous Sclerosis Complex Patients

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Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: Tuberous sclerosis complex, deletions, duplications, DNA microarrays

Tuberous sclerosis complex (TSC) is a genetic disorder caused by mutations in either TSC1 or TSC2 gene and is characterized by benign tumors growing in various organs of the human body. Direct sequencing of the TSC1 and TSC2 genes can detect 80% of mutations (including missense, nonsense, small deletions or duplications). The remaining 20% of mutations are known to include large deletions/duplications and somatic mosaicism. We aim to develop better methodologies to detect these latter two classes of mutations. Specifically, for the large deletions and duplications, we are attempting to produce a DNA microarray that includes all coding exons of the TSC1 and TSC2 genes, exons of other human genes as positive controls and non-human genes as negative controls. Using PCR, I had amplified 54 of the 64 exons for TSC1 and TSC2 genes, 80 exons of other human positive controls from 9 autosomes and the X and Y chromosomes (for gender identification), and 4 from non-human plasmid and viral genes. I made 100 ul PCR for each exon enough to provide sufficient DNA to make 100 microarrays. These PCR products are purified by G-50 Sephadex minicolumns. A concentration of 1 ug/ul of each exon in spotting solution will be spotted onto glass slides by a microarray spotter with 5 replicates per exon in each array. Patient and control DNA will be labeled with fluorescent dyes Cy3 and Cy5 respectively, and hybridized to exons spotted on an array to detect deletion/duplications shown by an increase/decrease of Cy3 signal on patient DNA.

ABSTRACT

Statistical Analysis of Length of Stay Comparisons for Palliative Patients vs. Non-Palliative Patients at Memorial Hermann Hospital

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Sponsored by: Philip C. Johnson, MD, Department of Internal Medicine

Supported by: Thomas Ragan Memorial Summer Research Fund

Key Words: symptom management, diagnosis related groups, outliers

Palliative care is medical care that focuses on symptom management rather than curative treatment. The goal is to relieve suffering and improve the quality of life for patients with advanced illness. An interdisciplinary team, including physicians, nurses, social workers, chaplains, counselors and other healthcare professionals, often provides this medical care. This study was undertaken to analyze statistical outcomes of a newly established palliative care program at Memorial Hermann Hospital. We searched data for all expired patients from October 2004-May 2005. We recorded dates of admission, dates of first palliative care visit and discharge or expired dates. Finally, lengths of stay comparisons were made. Patients staying in the hospital <48 hours and >90 days were excluded. Results showed total of 125 palliative care consults. 37 patients expired and the average amount of time from date of admission to a palliative care consult was 10.7 days. After consultation, expired patients were followed for 1.6 days on average. 88 palliative patients survived and the average amount of time from date of admission to a palliative care consult was 8.3 days. After consultation survivors were followed for 4.4 days on average. The average length of stay of expired palliative care patients (n=37, 12.31 days) was not statistically different from expired non-palliative care patients (n=475, 9.89 days). We conclude there appears to be a significant delay in physicians calling for a palliative care consults at Memorial Hermann Hospital.

ABSTRACT

Optimizing Co-expression of COX-2 and mPGES in HEK-293 Cell Line

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Sponsored by: Ke-He Ruan, MD, PhD, Department of Internal Medicine

Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: Prostaglandin E₂, HPLC Assay, COX-2, HEK-293

Biosynthesis of prostanoids involves two-step reactions. The first step is conversion of arachidonic acid (AA) to prostaglandin H₂ (PGH₂) by cyclooxygenase-1 (COX-1) or cyclooxygenase-2 (COX-2), and the second step is isomerization of PGH₂ into prostaglandin (PG) E₂ (PGE₂), PGD₂, PGF₂, PGI₂ and thromboxane A₂ by individual synthases. Over-synthesis of PGE₂ by inducible expressions of COX-2 and microsomal PGE₂ synthase (mPGES), mediated inflammation and cancer formation have been reported. Thus, co-expression of the recombinant COX-2 and mPGES proteins in vitro could be an approach to up-regulated PGE₂ production and used for anti-inflammatory and cancer drug screenings. In the current studies, the recombinant COX-2 and mPGES protein were co-expressed in human embryonic kidney 293 (HEK-293) cells by a transient protein expression approach using Lipofectamine™2000. The highly co-expressed COX-2 and mPGES in the cells were confirmed by Western blot. The activities of the recombinant proteins converting AA into PGE₂ was determined by a High Performance Liquid Chromatography assay using ¹⁴C-AA and ³H-PGH₂ as substrates for the cells over-expressing the recombinant proteins. The optimal co-expression and HPLC assay conditions for the recombinant COX-2 and PGES were tested. As a conclusion, co-expression of COX-2 and mPGES in HEK-293 cells is an effective approach to specifically synthesize PGE₂ from AA in vitro. The system can be used for anti-inflammatory and anti-cancer drug screening.

ABSTRACT

Salivary Assays as an Evaluation of Physiological Stress in Humans

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Sponsored by: Anil D. Kulkarni, PhD, Department of Surgery
Lalita Sunderesan, PhD, NASA and Department of Surgery
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Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: saliva, cortisol, DHEA, SlgA

The extended duration of space missions, especially future Moon and Mars missions, presents issues that are serious risk factors to crew health and space missions. It is imperative that these issues be addressed in preparation for long term missions. Group dynamics and crew relationships during these extended periods of confined isolation are such issues. While the psychological dimension of stress experienced by these individuals has been studied in the past, there has not been a well-documented physiological study. Stress levels of individuals training in groups under isolation for extended periods of time were measured using the stress hormones cortisol, dehydroepiandrosterone (DHEA), and secretory immunoglobulin A (slgA). Salivary specimens with these hormones were measured by ELISA assays. Sensitivity of saliva to changes in these hormones, and the non-invasive method of collection made this a suitable choice. Assays for cortisol showed samples to vary greatly in concentration based upon time of day, with morning samples being ten times more concentrated than evening samples. A correlation of DHEA levels with age and time of day was also observed. For these reasons, it was determined that accurate monitoring of stress needs to be assessed on an individual rather than comparative basis, and furthermore that samples taken from an individual must be at a consistent time of day. Both these factors must be taken into account in future sample collection.

ABSTRACT

Dissociation of Calmodulin from Ca^{2+} /Calmodulin-dependent protein kinase II at different $[\text{Ca}^{2+}]$

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Key Words: Calmodulin, CaMKII, calcium

Calmodulin (CaM) is a ubiquitous, calcium-binding protein that acts as a calcium sensor and signal transducer in activating a multitude of target proteins. The four Ca^{2+} binding-sites on CaM provide interesting, but kinetically complex, possibilities for decoding intracellular Ca^{2+} changes and the binding of Ca^{2+} is tuned when CaM interacts with targets. These properties enable CaM to bind specific proteins and elicit a range of responses. One major target for CaM is Ca^{2+} /calmodulin-dependent kinase II (CaMKII); a Ser/Thr-specific, calcium-sensitive kinase, which has been implicated in various neuronal functions, including synaptic plasticity. Although the kinetic rates of the reciprocal interactions between these two proteins are well established, these rates have not been used to observe the affinity of CaM for CaMKII at different $[\text{Ca}^{2+}]$. Since it is not yet possible to experimentally discriminate between conformations of CaM, this study employed computer simulated EGTA-induced dissociations of CaM from CaMKII at different $[\text{Ca}^{2+}]$ using a limited number of measured experimental parameters. Analysis of the dissociation curves revealed that CaM developed a double exponential dissociation from CaMKII at lower $[\text{Ca}^{2+}]$. At a closer look, between 100nM (basal) and 1 μM $[\text{Ca}^{2+}]$ (during synaptic stimulation), a shift in the dissociation pathway was detected. Furthermore, when other CaM target proteins (RC3 and PEP19) were included, the CaM bound to CaMKII shifted to the fully calcium saturated functionally active form. Experimental data with a stopped-flow fluorimeter confirmed this finding, but due to experimental limitations data at high $[\text{Ca}^{2+}]$ was not collected, which leaves room for future experiments.

ABSTRACT

Serum Free Fatty Acids and Cardiac Function in Clinically Severe Obesity

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Class of 2008

Sponsored by: Heinrich Taegtmeyer, MD DPhil, Cardiology

Supported by: The University of Texas at Houston Medical School - Summer Research Program

Key Words: Obesity, free fatty acids, diabetes

The purpose of this study was to analyze baseline left ventricular and metabolic data for clinically severe obese patients undergoing bariatric surgery. Our hypothesis is that increased substrate availability negatively influences cardiac function – “the heart fails in the midst of plenty.” This project is part of a prospective longitudinal cohort evaluating the effects of weight loss on metabolic parameters and cardiac function. We analyzed baseline parameters of 58 patients (BMI > 35kg/m²), including 2-D complete echocardiogram with M-Mode and tissue Doppler imaging, anthropometric measurements, and analysis of blood chemistries. All results are reported using median values [25th, 75th percentiles] and were analyzed using Pearson Correlation coefficients. The median age and BMI were, 46 years [37,53], and 49.4 kg/m² [42.7,56.2], respectively. The prevalence of diabetes, hypertension and insulin resistance were, 29%, 54%, and 75% respectively. Levels of fasting insulin and free fatty acids (FFA) were significantly greater than normal throughout the cohort. Global cardiac function was preserved (EF = 63% [59, 68]). However, 40% of the cohort demonstrated age adjusted diastolic impairment measured by Tissue Doppler imaging (TDI). An age adjusted, statistically significant association was found between the level of FFA and diastolic function ($r = 0.30$, $p = 0.0297$), as measured by TDI, that remained significant even when diabetes was excluded. The negative association between FFA and diastolic function, in the setting of insulin resistance, suggests that excess FFA may exert a lipotoxic effect on cardiac diastolic function, independent of diabetes.

ABSTRACT

Neurophysiological Effects of Acute Cocaine Administration

TED H. YAMAMOTO

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Class of 2007

Sponsored by: Nachum Dafny, PhD, Department of Neurobiology and Anatomy *and*
Alan C. Swann, MD, Department of Psychiatry

Supported by: The University of Texas at Houston Medical School - Summer Research
Program

Key Words: Cocaine, drug-seeking, electrophysiology, mesolimbic

Acute effects of cocaine can induce neuroadaptations that lead to further drug-seeking behavior. These neuroadaptations are thought to be localized in the "motive circuit," a subcircuit of the mesolimbic dopamine system which is comprised of the ventral tegmental area (VTA), prefrontal cortex (PFC), nucleus accumbens (NAc), ventral pallidum (VP), and amygdala. Previous studies have examined spontaneous electrophysiological changes of mesolimbic neurons to acute cocaine treatment. These studies fail to describe cocaine's ability to modulate sensation, a relevant notion considering relapse potential is closely associated with encounters of drug-related cues. In the present study, freely behaving rats previously implanted with permanent electrodes in motive circuit sites were used to assess cocaine dose response effects on sensory evoked potentials (SEPs). Animals were connected to a neurophysiological recording system and SEPs were recorded before and after 3.0mg/kg, 7.5mg/kg, and 15.0mg/kg intraperitoneally administered cocaine. Amplitudes of P2, N2, and P3 components of SEPs were evaluated. In a dose dependent manner, cocaine attenuated SEPs in the VTA, NAc, and PFC by 16%, 29%, and 35% for doses 3.0mg/kg, 7.5mg/kg, and 15.0mg/kg, respectively. Similar dose dependent responses were observed for N2 and P3. These data suggest that cocaine plays a crucial neuroadaptive role in mesolimbic structures by modulating sensation.

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2005 SUMMER RESEARCH PROGRAM UNDERGRADUATE PARTICIPANTS

