51st Annual AOA Research Conference—Abstracts, 2007

This issue of JAOA—The Journal of the American Osteopathic Association *(August 2007) contains abstracts for poster presentations that will be given at the 51st Annual American Osteopathic Association (AOA) Research Conference. Poster presentations will be provided by AOA research fellows (abstract series F), as well as osteopathic physicians, medical students and educators, clinicians, and researchers on the following general topics: osteopathic manipulative medicine and osteopathic principles and practice (series P), clinical studies (series C), basic sciences (series B), and medical education (series M).*

The 2007 AOA Research Conference will take place from Sunday, September 30, to Tuesday, October 2, during the American Osteopathic Association's (AOA) 112th Annual Convention and Scientific Seminar in San Diego, Calif. The theme of this year's conference is "Chronic Disorders of Aging in the 21st Century: New Insight and Approaches." For more information on the Annual Convention and Scientific Seminar, please see the DO-Online Web site (http://www.do-online.org).

Abstracts are reproduced as submitted. The AOA and The Journal assume no responsibility for the content of abstracts presented herein.

AOA Research Fellowships

♦F1

Comparing the Effects of Cranial Manipulation on Heart Rate Variability and Traube-Hering-Mayer Waves

T. Pexton, DO, K. Nelson, DO, T. Glonek, PhD, K. Heinking, DO, R. Kappler, DO, N. Serfueef, DO (France), Dept. of OMM, Midwest University, Chicago College of Osteopathic Medicine, Downers Grove, IL

Hypothesis: It has been shown that the cranial rhythmic impulse (CRI) is simultaneous with a measurable oscillation in blood flow velocity known as the Traube-Hering-Mayer (THM) wave. It has also been shown that the THM wave can be enhanced with cranial manipulation. Based on physiological principles, it is reasonable to expect that heart rate variability (HRV) will similarly be enhanced by cranial manipulation.

Materials and Methods: A heart rate monitor and a laser Doppler flowmeter were used to measure HRV and the THM wave simultaneously. The protocol consisted of a pre-treatment phase, a CV-4 treatment phase and a post-treatment phase. Each phase lasted approximately 5-10 minutes. Subjects remained quietly in the supine position for the duration of the protocol. The following standard HRV measurements were generated: RMSSD; pNN50; Total Power (0-0.4Hz); High Frequency, HF (0.15-0.4Hz); Low Frequency, LF (0.04-0.15 Hz); Very Low Frequency, VLF (0-0.04 Hz); LF/HF; and the ratio of the powers of various frequency ranges to the Total Power. THM wave power spectra were generated with WinDaq Software.

Results: 20 healthy subjects were evaluated. All signed an institutionally approved IRB consent form. Subjects were divided among 3 physicians (N of 3, 4 and 13). For the entire group (N=20), there was no significant change in pre- to post-treatment measurements for any of the standard HRV parameters. For one physician, a significant increase in the Square Root of the Mean of the Squared Differences Between Adjacent Normal RR Intervals (RMSSD) was found pre- to post-treatment [N=4; (mean±SD) pre 51.85±24.33, post 60.7±23.42; P=0.050]. Simultaneous THM wave measurements did not show any significant changes for these subjects [N=4; (mean±SD) pre 1093.3±143.9, post 1079.6±159.1; P=0.601].

Conclusion: An increase in the THM wave following a CV-4 treatment was not demonstrated. However, a significant post-treatment increase in one of the standard HRV measurements was demonstrated. A decreased HRV has been shown to be one of the best predictors of mortality in CHF and post-MI patients. Given the prevalence and mortality associated with heart disease, further research is warranted.

♦F2

Measuring pressures used by physicians and students for cervical diagnosis of segmental somatic dysfunction using the Iso-TOUCH® Palpation Monitor System

N. T. Jean OMS-III[1], M. L. Kuchera, DO, FAAO[1], B. Schoenfeldt, OMS-III[1], R. T. Dombroski, DO[1], T. Vardy, DO[2], L. Williams, OMS-III[1], [1]Center of Chronic Disorders of Aging, [1]Philadelphia College of Osteopathic Medicine, Philadelphia, PA, [2]Neuro-

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

muscular Engineering, Tweed Heads, North South Wales, Australia

For manual medicine (MM) physicians, developing palpatory skills needed to diagnose somatic dysfunction prior to treatment and assess subsequent change is crucial in determining safety and efficacy. Several scientific recording methods have attempted to quantify palpation demonstrated technological limitations.

Hypothesis: It was hypothesized that MM students would use more pressure during cervical spine diagnosis than experienced MM practitioners.

Methods: 10 osteopathic students and 10 osteopathic physicians palpated the cervical spines of volunteers (n=28, 12 by students, 16 by physicians). At each session a student or physician palpated two volunteers and then reported the side-bending/rotational motion characteristics of the perceived "worst segment."

A prototypic Iso-TOUCH® Palpation Monitor System (Neuromuscular Technologies) was developed to provide instrumentation to quantify pressure ranges used by successful practitioners to accurately diagnose (and later, successfully treat) somatic dysfunction. This technology utilizes finger, thumb and palm sensors to measure contact pressures during palpation of a given body region.

Finger pressures over the cervical articular pillars during side-bending and rotation were recorded and graphed by customized Iso-TOUCH® software. They were analyzed using a one-factor analysis of variance test performed by the PCOM statistician. The PCOM Institutional Review Board approved this study.

Results: In comparing all students to physicians, students used an average of 0.82 Lbs more palpatory pressure ($p<0.01$) than physicians (1.22-2.88 versus 1.03-1.30 Lbs). Additionally, physicians were found to compress the soft tissues prior to motion testing using 0.39 Lbs while students did not compress the tissues at all ($p<0.01$). Of the 10 students, 7 used more force than any physician palpator. The remaining 3 students used more force than most (7 of 10) of the physicians.

Conclusion: This study tends to confirm the hypothesis that as palpatory skills develop, less pressure is required to accurately diagnose somatic dysfunction. Furthermore, experienced clinicians incorporated loading pressures omitted by students. Based upon these findings, future studies are warranted with this technology to document palpation characteristics. Studies in educational settings are also planned to determine if this real-time feedback system might improve palpatory skills and confidence of future practitioners.

Acknowledgment: This project was partially funded by an AOA Research Fellowship Grant.

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

♦F3

Matrix Metalloproteinase-9 Processes Chemokines and Cytokines During Chlamydia Muridarum Urogenital Tract Infection of Mice

J. T. Distelhorst BS[1], M. T. Imtiaz, MD[1], S. R. Lacy, DO[1], J. S. Schripsema, BS[1], I. M. Sigar, PhD[1], J. N. Kasimos, DO[2], K. H. Ramsey, PhD[1], [1]Microbiology and Immunology Department, [2]Pathology Department, Midwestern University-Chicago College of Osteopathic Medicine, Downers Grove, IL

Hypothesis: Matrix metalloproteinase-9 is involved in the pathogenesis of chlamydial infection in a mouse model.

Methods: Mice with a genetic deletion of matrix metalloproteinase-9 (MMP-9 KO) or strain-matched wild type (WT) mice were infected intravaginally with *Chlamydia muridarum*, mouse pneumonitis biovar (MoPn). Following infection, cervical-vaginal swab samples were collected at 0, 4, 7, 10, 14, 28, 35, and 56 days post-infection for culture confirmation and quantitation of infection. Chlamydial inclusions were enumerated by indirect fluorescence microscopy. At intervals, a subset of mice was euthanized, necropsied, and their urogenital tracts were excised. Some tissues were homogenized in buffer for the assessment of chemokine and cytokine production profiles by antigen capture enzyme-linked absorbent assay (ELISA) as well as antigen capture microarray. Tissue homogenates were also used to assess processing of chemokines by Western blot and to isolate MoPn in the upper genital tract. Lastly, tissues were fixed in buffered formalin, paraffin embedded, sectioned, stained with hematoxylin and eosin and blindly assessed for histopathological parameters. **Results**: In the lower tract, the infection course remained unchanged as a result of the MMP-9 KO whereas in the upper genital tract, there was a slight reduction in MoPn isolated from the MMP-9 KO mice. Pathological parameters, particularly acute inflammation and hydrosalpinx formation, were significantly reduced in the MMP-9 KO mice when compared to WT controls. Antigen capture ELISA and microarrays yielded quantitative differences in certain chemokines and cytokines and Western blot analysis indicated MMP-9 is involved in the proteolytic cleavage of specific chemokines and cytokines.

Conclusions: MMP-9 (gelatinase B) is a mediator of pathological host responses during *Chlamydia muridarum* infection in a mouse model. A likely explanation for this is observation is the proteolytic processing of chemokines and cytokines to modulate their activity.

Acknowledgement: Supported by N.I.H. 1 R01 AI49354-05.

♦F4

Six Etiologies of Low Back Pain Found in the US Army

M. M. Braun, DO, M. L. Kuchera, DO, T. E. Borden, DO, N. Ondrasik, DO, D. E. Jones, DO, G. V. Manco, PhD, M. V. Krueger, DO, Philadelphia College of Osteopathic Medicine, Philadelphia, PA

Objective: To determine the prevalence of six somatic dysfunction diagnoses in the presence or absence of low back

pain. These somatic dysfunctions, closely related to six causes of chronic low back pain labeled by Greenman as the "Dirty Half Dozen," include: short-leg syndrome, psoas syndrome, non-neutral sacrum, sacral shear, pubic shear, and non-neutral lumbar dysfunction[1].
Setting: Womack Army Medical Center, Fort Bragg, NC
Methods: Subjects were randomly selected. Subject eligibility was determined by answers to written and oral interview questions. The subjects were evaluated clinically using a standardized osteopathic structural examination for various abnormalities involving the pelvis, sacrum, lumbar spine, and lower extremities. The examining team member was unaware of the patient's low back pain status. Statistical analysis included one-sided T-test for difference in proportions and nonparametric "exact" tests.
Results: 265 subjects were enrolled. Of these, 20 were not eligible to participate based on the initial screening. Of the remaining 245 subjects, 150 (61.1%) interviewed claimed to have back pain issues. 95 patients denied presence of back pain (control group). Of interest, 75% of the subset consisting of all airborne soldiers (n=39) reported back pain. Subjects showing two or more findings exhibited a greater amount of back pain compared to those subjects with 1 or less findings (p=0.046, LCB 95%). Of the "Dirty Half Dozen" diagnoses, subjects exhibiting the following had significant findings with back pain compared to controls: psoas syndrome (p=0.0000009, LCB 95%); non-neutral lumbar dysfunction (p=0.0308, LCB 95%); pubic shear (p=0.00000005, LCB 95%); and non-neutral sacrum (p=0.0185, LCB 95%).
Conclusions: Our study indicates that there is an association between back pain and presence of the "dirty half dozen" physical findings in the U.S. Army. Further study is required to determine if Osteopathic Manipulative Treatment of these dysfunctions can resolve patients' pain and expediently return these soldiers to duty.
References:
[1]Kuchera, WA. Lumbar Region. Foundations for Osteopathic Medicine 2nd Ed. 2003; 727-750.

Osteopathic Manipulative Medicine/Osteopathic Principles and Practice P1

A PILOT CLINICAL TRIAL OF OSTEOPATHIC MANIPULATIVE TREATMENT DURING THIRD TRIMESTER PREGNANCY

J. C. Licciardone DO, MS, MBA[1], S. Buchanan, DO[2], K. Fulda, DrPH[2], K. Hensel, DO[3], H. King, DO, PhD[1], S. T. Stoll, DO, PhD[1], [1]National Osteopathic Research Center, [2]Obstetrics and Gynecology, University of North Texas Health Science Center, [3]Tarrant County Public Health Department, Ft Worth, TX
Purpose: The primary purpose of this study was to explore the potential therapeutic effects of osteopathic manipulative treatment (OMT) during the third trimester of pregnancy.
Methods: All study procedures were approved by IRB and the study was registered with ClinicalTrials.gov (NCT00298935). Exclusion criteria included either of the following: (1) intent to deliver at a non-designated hospital or (2) high risk pregnancy as determined by the attending obstetrician. Each subject was randomized to one of three treatment groups: (1) usual obstetrical care and OMT (UOBC+OMT); (2) usual obstetrical care and sham ultrasound treatment (UOBC+SUT); or (3) usual obstetrical care only (UOBC only). The UOBC+OMT and UOBC+SUT groups were scheduled to receive treatments at the 30th week (visit 1), 32nd week (visit 2), 34th week (visit 3), 36th week (visit 4), 37th week (visit 5), 38th week (visit 6), and 39th week (visit 7). The primary outcome measures included: (1) an 11-point scale for the typical or average level of back pain; (2) the Roland-Morris Disability Questionnaire; and (3) the SF-12 Version 2 Health Survey (SF-12) scale score for bodily pain and the summary scores for physical health and mental health. All analyses were based on the intention-to-treat principle. Missing data for all primary outcomes were imputed using the carry-forward method. Repeated measures analysis of covariance (ANCOVA) was used to test the hypotheses.
Results: A total of 49, 48, and 49 subjects were randomized to the UOBC+OMT, UOBC+SUT, and UOBC only groups, respectively. Typical or average back pain levels decreased over time in the UOBC+OMT group, remained essentially unchanged in the UOBC+SUT group, and increased in the UOBC only group (ANCOVA treatment group x time interaction, P=.02). The Roland-Morris disability scores increased over time in all three treatment groups; however, the rate of increase was significantly different among the groups (ANCOVA treatment group x time interaction, P<.001). Disability progressed less rapidly in the UOBC+OMT group than in the UOBC only group (P<.001), and there was also a trend toward less rapid progression in the UOBC+OMT group in comparison with the UOBC+SUT group (P=.08).
Conclusion: OMT was associated with favorable disability and pain outcomes during the third trimester of pregnancy.
Supported by a grant from the Osteopathic Heritage Foundations.

♦P2

The Burden of Musculoskeletal Conditions in Ecuador

E. M. Timoshkin BA[1], A. J. Caban-Martinez, MPH[1], E. M. Wallace, DO, MSc[1], G. P. Suciu, PhD, MSPH[2], [1]Osteopathic Principles and Practice, [2]Masters of Public Health Program, Nova Southeastern University-COM, Fort Lauderdale, Fl.
Objective: Understanding and elucidating acute and chronic musculoskeletal somatic dysfunction in regional areas of Ecuador has yet to be ascertained and remains a priority for the World Health Organization. Anecdotal data indicates that developing countries in South America are experiencing increases in work related musculoskeletal disorders. The lim-

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

ited body of literature on musculoskeletal conditions, specific to body region and type of pain are poorly characterized in Ecuador. In the present study, using a musculoskeletal survey, we assessed the proportion of musculoskeletal complaints by body region, at rural and urban Ecuadorian sites.

Methods: A modified 7-item version of a validated musculoskeletal survey was randomly administered to 116 adult patients during the NSU-DOCARE 2007 Ecuador Medical Mission. Descriptive analyses of the 7 selected variables from 100 completed surveys were tabulated using SPSS. Participants were consented prior to participation in the survey and the Nova Southeastern University's institutional review board approved the study.

Results: Among the 100 completed musculoskeletal surveys, adult participant ages ranged from 18-80 years old (mean±SE; 44.1±1.7), height from 48-69 inches (60.3±0.4), weight from 70-206 pounds (137.4±2.3), and BMI from 15.2-46.2 lb/in2 (26.8±0.5). Survey participants most frequently self-reported discomfort of the upper back (65%), knee (63%), and neck (62%) in the past 12-months; and upper back (61%), neck (58%) and low back (58%) body regions with the highest frequency in the past week. Fisher's test performed for site versus musculoskeletal pain location, demonstrated a significant difference for male gender between Baeza and Quito regions of Ecuador for the period of the past week, with a higher proportion of knee pain within Baeza (p=0.024).

Conclusion: Findings from the present study indicate there is a considerable prevalence of musculoskeletal conditions in specific Ecuadorian communities. Individuals residing in more labor intensive communities (Baeza) tend to self report higher levels of knee pain as compared to their urban counterparts. Focused strategies and interventions to ameliorate the current burden among this population are highly warranted.

P3

Student Achievement in a Virtual Reality Test of Palpatory Diagnosis

J. N. Howell, PhD[1], J. M. Burns, DO[2], D. C. Eland, DO[2], R. L. Williams, PhD[3], R. R. Conatser, MS[1], [1]Dept. of Biomedical Sciences, [2]Dept. of Family Medicine, [3]Dept. of Mechanical Engineering, Ohio University College of Osteopathic Medicine, Athens, OH

Evaluation of tissue textures is a major part of the palpatory diagnosis of somatic dysfunction. Assessment of tissue textures requires detection of differences in tissue compliance (1/stiffness). In this study the Virtual Haptic Back (VHB) was used to determine the smallest differences in compliance osteopathic medical students could detect. The VHB is a virtual reality, force-feedback simulation of the contour and compliance of the human back, developed as a teaching/learning aid in palpatory diagnosis (*JAOA*, in press).

Hypothesis: By palpation medical students can detect differences in compliance on the VHB comparable to the 8 to 12% obtained by other investigators, utilizing simpler test algorithms.

Methods: In the fall of 2006, 89 first-year medical students were required, as part of their manipulative skills training, to perform a pre-test, 6 practice sessions, and a post-test on the VHB. The VHB simulation was based on the contours and compliance of the back of a 51 year old female. Programmed into the back were 2.5 X 3 cm regions of abnormally high paravertebral stiffness, which varied randomly from trial to trial as to side (L/R) and vertebral level. The difficulty level was progressively increased by having the difference in compliance between background and abnormal region progressively decrease. The task for the user was to find these abnormal regions by palpation.

Results: As a group, students improved during the practice sessions from being unable to detect differences of less than 28% to detecting differences as little as 14%. Ten students were able to detect differences as little as 7%, which represented achievement ⩾ 1 standard deviation (SD) above the mean. The performance of these students in the pre-test was not significantly better than the students whose highest achievement during subsequent practice sessions was ⩽1 SD below the mean, but was better (P⩽.05; t-test) in the first practice session. Improvement rates in the remaining sessions did not differ.

Conclusion: Despite the more complex context of the discrimination task, the highest achievers in our sample performed at least as well the 6 subjects tested by De Gersem et al. (2005), although the average for the entire class, 14%, was somewhat poorer. The performance difference between high and low achievers was not apparent in the pre-test, but appeared during first practice session following the pre-test.

Supported by the Osteopathic Heritage Foundation, Columbus, OH.

♦P4

Inter-rater Reliability of Rib Strain-Counterstrain Tenderpoints

J. Hartinger OMS-IV, N. Cloar, OMS-IV, M. Seffinger, DO, Western University of Health Sciences/COMP, Pomona, CA

Objective: The purpose of the study was to assess the reliability between two Osteopathic Manipulative Medicine student teaching fellows palpating a designated set of rib counterstrain tenderpoints.

Methods: After IRB approval, eighty volunteer subjects were recruited from the staff and student body at Western University of Health Sciences. Of the 80, 33 (41%) were randomly selected for the reliability study using an uncharted wrist tenderpoint between the distal radial-ulnar bones. Subjects were included if they were 18 or over, had no current open wounds or fractures in the regions of palpation, and had no tenderness at the uncharted wrist tenderpoints. A total of 14 points were assessed for each subject. Rib tenderpoints, which are on the

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

bony surface of the ribs, consisted of anterior ribs 2-6 and posterior rib 1 bilaterally. The omohyoid tenderpoints (located at the inferior attachment of that muscle on each scapula), were used as non-bony reference points. A 0-10 point verbal pain scale was used upon digital provocation. The pressure applied for palpating the tenderpoints was pre-determined between the examiners to blanch no greater than the first 1/3 of the fingernail bed. Examiner 1 entered the room with the subject supine on a treatment table, assessed all 14 tenderpoints and then left the room. Examiner 2 entered the room within 1 minute, blinded to the results of examiner 1, and instructed the subject to give responses on the 0-10 verbal pain scale independent to the responses given to examiner 1.

Results: Pain scores within a 1-point difference or within a 2-point difference at each tenderpoint were considered as "agreement". Rib scores within 1-point difference: 71% agreement; κ=0.61; within 2-point difference: 88% agreement; κ=0.84. For the omohyoid tenderpoint, within1-point difference: 85% agreement; κ=0.76; within 2-point difference: 92% agreement; κ=0.88. There were no significant differences between kappa scores for the rib and muscle tenderpoints ($p>.05$)

Conclusion: There is good inter-examiner reliability between OMM student teaching fellows at assessing standard rib counterstrain tenderpoints using agreed upon amount of digital pressure. Inter-examiner reliability is comparable between the rib and reference muscle tenderpoints.

♦P5

The Effect of Cranial Osteopathic Treatment on Visual Function

A. Caban-Martinez, MPH[1], E. Timoshkin, BA[1], M. E. Sandhouse, DO[1], D. Shechtman, OD, FAAO[2], G. Fecho, OD, FAAO[2], A. Snyder, DO[1], M. Patterson, PhD[1], J. Shallo-Hoffmann, PhD[2], P. Hardigan, PhD[3], [1]Dept. of Osteopathic Principles and Practice, Nova Southeastern University COM, [2]College of Optometry, Nova Southeastern University COO, [3]Statistical Support Center, Nova Southeastern University Health Prof Division, Fort Lauderdale, FL.

Objective: The effects of cranial osteopathy on visual function, particularly on changes of the visual field, and the binocular alignment of the eyes, are limited in the literature. Anecdotal evidence indicates that patients who undergo cranial osteopathic manipulation claim to experience an improvement in visual performance. A pilot study recently conducted by the authors demonstrated that a single treatment using cranial osteopathic manipulation produced statistically significant differences ($p<0.05$) within groups for multiple optometric parameters. In the present full study three important issues were addressed and included in the design of this study: increasing sample size, measuring the cumulative effects of multiple treatments, and measuring the temporal permanence of these effects.

Methods: Using a randomized, double-blind, placebo-controlled protocol, we assessed for a change in visual function following cranial manipulation among a sample of healthy adults with cranial asymmetry. 113 adults (18-35 years old), who were free of active ocular or systemic disease, were consented prior to and entered into the study. The Nova Southeastern University's institutional review board approved the study. Following a battery of optometric examinations, all participants were evaluated for cranial dysfunction. The treatment group received cranial osteopathic manipulation to correct cranial dysfunctions, while the control group had a light pressure of a few ounces of force applied to their cranium without the use of osteopathic manipulation. All subjects were re-evaluated for cranial dysfunction after the intervention, and then all optometric parameters were re-measured. This was repeated for a total of 8 visits, followed by another 8 visits during which only the optometric measurements were performed.

Results: 89 subjects completed the trial, 42 in the control group and 47 in the treatment group. A two-way ANOVA revealed statistically significant differences within groups in seven of the twelve parameters measured, and between groups in Pupillary size measured under bright illumination OS ($p<0.05$) and Near Point of Convergence break measurement ($p<0.04$).

Conclusion: The changes in both groups suggest that active motion testing of the sphenobasilar synchondrosis (SBS) may alter the cranial system to a sufficient extent as to alter visual function.

♦P6

Evaluation of the Effects of Heel and Forefoot Lifts on Body Weight Distribution, Center of Mass, and Muscle Activity in the Lumbar Region

K. Quinones BS[1], E. Shamus, PhD[1], M. Fernandez, PhD[2], C. Sheehan, MS[3], M. Akin, BS[2], [1]College of Osteopathic Medicine, [2]Department of Public Health, [3]Center for Psychological Studies, [1]Nova Southeastern University, Fort Lauderdale, FL

The ability to symmetrically distribute body weight has been reported to be essential to normal balance and to the ability to perform daily activities. Therefore, decreasing asymmetry and enhancing dynamic stability are important therapeutic goals. Postural instability is a growing problem in the older population, leading to falls that may result in major health problems. Even in the younger population, a number of physical problems have been associated with unbalanced weight distribution. In this study we are using the quadruple postural scale to measure body weight distribution and the Surface Electromyogram (SEMG) to measure muscle activity while wearing different heel and forefoot lifts. Results from this study may lead towards understanding if certain footwear can be helpful in treating postural asymmetries and preventing associated pathologies.

Hypothesis: Varying the heights of heel and forefoot lifts will have an effect on anterior/posterior weight distribution and

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

muscle activity in the lumbar spine.
Materials and Methods: Fifty female NSU students ages 18 to 35 are being recruited to participate in this study. The quadruple postural scale measures the participants' weight distribution under five conditions: 1)barefoot, 2)1 inch heel lifts, 3)2 inch heel lifts, 4)[1½] inch forefoot lift, and 5)1 inch forefoot lifts. While the SEMG records muscle activity in the lumbar spine while under the five conditions. To control for order effects, the order of presentation of the conditions is determined randomly. Procedures have been approved by the HPD Research Committee and the NSU IRB.
Results and Conclusion: Preliminary (N=9) results have shown a significant movement towards balance of weight distribution in participants with an initially posterior stance when wearing 2 inch heel lifts (p=0.013). SEMG results have shown significant decrease in muscle activity in both the right and left lumbar region when wearing 1 inch heel (left: p=0.027; right: p=0.045), 2 inch heel (left: p=0.029), and 1 inch forefoot lifts (left: p=0.022; right: p=0.029) when compared to baseline (barefoot). The results from the 50 participants will be presented at the meeting.

♦P7

Reproducibility Assessment of the Response to CV4 as Measured by the Laser Doppler Flowmeter and Electrocardiogram

K. K. Guinn, OMS-III[1], C. Rodriguez, BS[2], D. Bucheli, BS[2], R. L. Alsip, OMS-II[2], M. A. Seffinger, DO[2], M. L. Smith, PhD[1], [1]UNTHSC, Fort Worth, TX, [2]Western University Health Science Center COMP, Pomona, CA

Background: Osteopathic manipulation techniques, such as Compression of Fourth Ventricle (CV4), have been shown to influence autonomic balance as measured by Laser Doppler Flowmeter (LDF) and electrocardiogram (EKG). In this study, authors assess reproducibility of a subject's response to CV4, measured by LDF and EKG.
Objective: Assess reproducibility of subject's response (change in autonomic variability) to CV4 by an alternate operator as compared to response elicited by reference standard operator.
Methods: 22 healthy volunteers were enrolled in IRB approved project. Simultaneous LDF and EKG readings were obtained before, during, after CV4 in 7min segments. Five minutes of data with least amount of noise were selected from each segment for analysis. Each subject underwent this protocol on 2 different occasions, once each with reference standard operator and an alternate operator. All operators were trained in CV4 technique. Timed periods were compared using WINDAQ and BIOPAC software for LDF and Heart Rate Variability (HRV) software used for EKG. Spectral analyses were performed using Discrete Fourier Transformation and areas under the curve of the low frequency Traube-Hering wave (TH) (0.8-.15 Hz) were measured. Results were analyzed using a paired samples t-test in SPSS. Duration of change in area under curve of TH wave during CV4 performed by each operator was compared using a paired samples correlation in SPSS.
Results: Paired samples t-test (SPSS) demonstrated significant ($p<.05$) decrease in autonomic variability from baseline for all subjects measured by LDF and EKG, regardless of the operator. Results of alternate operator were compared with results of primary investigator for each subject. Overall, there was no significant difference ($p>.2$) in the subject's response to CV4. Comparison of duration (seconds) of change, determined by a 40% decrease from baseline in area under curve (TH) during CV4 revealed correlation of .777 p=.001.
Conclusion: Decreased autonomic variability and length of its duration during CV4 are reproducible in same subject by an alternate operator as compared to response elicited by reference operator. This response to CV4 as measured by LDF was mirrored in changes seen in HRV, as obtained from EKG. Any potential therapeutic effects of CV4 are undetermined at this time, it is recommended that LDF or EKG be used in clinical outcomes studies to investigate efficacy of CV4 in select patient populations.

P8

Prolonged Effects of Maximal Effort Exercise (with Valsalva) and Osteopathic Manipulative Treatment in Women with Multiple Sclerosis

M. L. Kuchera DO, FAAO[1], T. C. Vardy, DO[2], H. Yates, DO[3], B. Stouch, PhD[1], J. C. Johnson, MA[3], CCDA, [1]Human Performance & Biomechanics Lab, Philadelphia College of Osteopathic Medicine, Philadelphia, PA, [2]Neuromuscular Clinic, Tweed Heads, North South Wales, Australia, [3]Kirksville College of Osteopathic Medicine, Kirksville, MO

Benefits gained through most exercise programs typically diminish shortly after discontinuing the program. This study investigates the effect over time of discontinuing a uniquely structured machine-assisted exercise and manual medicine protocol that previously demonstrated short-term benefits (*JAOA*, May 2002). The exercise previously used employed isometric and eccentric vertical leg presses as well as isometric semi-erect whole body lunges. Each exercise had been performed for an average of 6 seconds duration with Valsalva and 3-5 repetitions of each exercise was performed on specialized equipment (IsoPUMP®, Neuromuscular Technologies). OMT had been integrated to reduce coexisting somatic dysfunction each session and to help prepare the subject for exercise.
Hypothesis: We hypothesized that previously reported improvement in strength, endurance, fatigue, coordination, and ambulation in females with Multiple Sclerosis (MS) as a result of a twelve-week physical intervention protocol combining machine-assisted MEE and OMT would be maintained for at least nine months after discontinuing the protocol.
Materials and Methods: Strength (isometric lunge, isometric and eccentric leg press), ambulatory ability (Timed 25-foot

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

Walk) and coordination (Block-and-Box Test, BBT) were measured at 3 and 6 months post-intervention (n=7) and at 9 months (n=6). Repeated-measures ANOVAs were used to determine whether outcomes previously reported as a consequence of the MEE/OMT protocol were maintained after discontinuing the treatment. This study was approved by the KCOM Institutional Review Board where all data had been gathered.
Results: Significant increases in strength during isometric lunge ($P<0.001$) and BBT ($P<0.001$) as well as reductions in Timed 25-foot Walk ($P=0.007$) due to the MEE/OMT protocol were maintained at 3-, 6-, and 9-month follow-up. Gains in strength during isometric leg press were lower at six months than immediately post-intervention, but still retained significance above baseline at all three follow-up times ($P<0.001$).
Conclusion: Prolonged positive effects, including increased strength, ambulatory ability, and coordination, results from a combined MEE/OMT protocol in women with mild-moderate MS impairment. Measurable benefits in walking and strength still existed nine months after discontinuing the twelve-week protocol.
Acknowledgment: Supported by the KCOM Strategic Research Initiative.

♦P9

Foot Ankle Biomechanics: Effects of Manipulative Intervention on Plantar Fasciitis Subjects

A. L. Morton BS[1], A. McGrew, BS[1], J. M. Burns, DO[1], E. Karodogan, MS[2], R. Conatser, MS[1], J. N. Howell, PhD[1], [1]Biomedical Science, [2]Engineering, OUCOM-IINR, Athens, OH

Counterstrain (CS), an Osteopathic Manipulative Medicine (OMM) technique that involves passively shortening painful tissues, significantly reduced plantar fasciitis (PF) pain in a prior OUCOM study (Wynne, et. al., *JAOA* Sep 2006; 106:547-556). Triceps surae peak force (Torque) production as well as time needed to reach peak force increased in both PF and control subjects without altered muscle spindle gain (H-reflex). This lack of a neuroreflexive explanation prompts possible involvement of passive mechanical tissue properties of the foot/ankle complex.
In order to isolate these properties, force production was measured on the same apparatus, and the ankle was dorsiflexed the same number of repetitions and degrees as the former study; but slow enough to prevent eliciting an Achilles tendon reflex. Non-therapeutic passive 90 second shortening represented a sham for CS.
Hypotheses: Alteration of passive mechanical tissue properties of the foot/ankle complex by stretching or shortening the plantar surface of the foot will result in: 1. Increased force production and time to reach peak force, 2. Decreased PF pain.
Methods: Case-control study following informed consent per OU IRB.
Cohort: 12 subjects with plantar fasciitis, 5 controls. Force against the foot plate of a motor driven apparatus was recorded in supine subjects while the ankle was dorsiflexed (5° over 500 msec) 10 times followed by a 30 second rest then repeated 10 times. This was done at baseline and after two manipulative interventions: 1) a 90 second hold of the plantar fascia in a shortened position and 2) a 90 second stretch of the plantar fascia. Subjects were randomized to receive either stretching or shortening first.
Results: RMANOVA statistical analysis showed no difference, following either intervention, in peak force production or time needed to reach peak force ($p \geq 0.05$). Plantar fasciitis subjects reported no change in foot pain following either or both interventions.
Conclusion: If the decrease in pain or increases in peak force production and in time needed to reach peak force seen in the previous experiment resulted primarily from an alteration in the passive biomechanical components of the foot-ankle complex, similar results would be expected in this study. This was not the case. Force generation under the ball of the foot may also be influenced by reflex activity of intrinsic foot muscles and is the subject of future study. Further research into the mechanisms of CS is warranted.

P10

Developing technology and protocols to measure pressure characteristics used by physicians and students for diagnosis and treatment of cervical somatic dysfunction

M. L. Kuchera, DO, FAAO[1], N. T. Jean, OMS-III[2], B. Schoenfeldt, OMS-III[2], L. Williams, OMS-III[2], T. Vardi, DO[3], R. T. Dombroski, DO[2], [1]CCDA, Human Performance & Biomechanics Lab, [2]Philadelphia College of Osteopathic Medicine, Philadelphia, PA, [3]Neuromuscular Engineering, Tweed Heads, North South Wales

Risk-to-benefit data concerning cervical spinal manipulation (CSM) provokes great interest among manual medicine (MM) practitioners whose concern extends to safety-efficacy issues performing and teaching CSM.
Hypotheses: A protocol for a prototypic Iso-TOUCH® palpation system (Iso Technologies) was developed to quantify and provide feedback concerning cervical pressure used in diagnosis and CSM. It is hypothesized that this protocol and equipment would be sensitive enough to differentiate student from physician pressures used to palpate and perform CSM. Also that data gathered would demonstrate intra-examiner consistency and suggest adequate sample sizes for future studies.
Methods: Osteopathic physicians (n=10) and students (n=10) wore pressure sensors over thumbs, index fingertips, and thenar eminences. The system was tared to compensate for finger size. Customized software converted electrical sensor signals into pounds using pre-determined calibration curves. Palpators applied articular pillar pressure to diagnose cervical somatic dysfunction. Each provided self-selected thrust, muscle

♦ Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

energy, or balanced tension CSM to the "worst" dysfunction. Individually "stacking" each barrier (side-bending, rotation, and flexion/extension) prior to applying activating forces helped measure individual and total barrier engagement and isolated CSM pressure and duration. Between trials, taring limited sensor deformation drift.
Results: Individual palpators' pressures used in diagnosis were internally consistent from cervical segment-to-segment. Sampling rates captured beginning and end treatment forces even in thrust activations of short duration. Data documented that students use more pressure than physicians in both diagnosis (1.22-2.88 versus 1.03-1.30 Lbs) and CSM (1.87-11.61 lbs versus 0.87-11.42 lbs). The data suggests future cervical trials should use 0.5 -15.0 pound calibrations (creating a sensitivity range of +/- 0.1-0.2 pounds). Protocols to detect physician-student differences should involve each palpating the same 16 subjects or 29 unique subjects (n=58).
Conclusions: Carefully described protocols using calibrated digital and manual sensors could provide reliable, reproducible data needed to quantify pressure characteristics used by MM physicians and students in cervical diagnosis and CSM. The protocol could be further enhanced by using a scale to create customized sensor calibration curves for each palpator.

Clinical Studies

C1

Free Serum Acrolein Levels in Several Disease States Associated with Oxidative Stress or Inflammation

A. Gugliucci MD, PhD[1], C. Hathuc, OMS-III[2], J. Schulze, BS[2], S. Kimura, MD[3], [1]Dept. of Biochemistry, Touro University-CA, [2]Glycation, Dept. of Oxidation and Disease Laboratory, Touro University-CA, Vallejo, CA, [3]Dept. of Laboratory Medicine, Showa University, Northern Yokohama Hospital, Yokohama, Japan

We tested the hypothesis that free serum acrolein levels are increased in conditions associated with increased oxidative stress and inflammation: renal failure, stroke, infection and diabetes, as well as in liver disease by impaired metabolism. We performed a nested case-control study with 35 chronic liver disease patients; 40 chronic renal failure (CRF) patients undergoing dialysis ; 22 type 2 diabetic patients (mean HbA1c 10%); 15 stroke (CVA) patients; 10 acute pneumonia patients and 40 age-matched controls. Fasting blood samples were analyzed for acrolein using a fluorometric method. Serum acrolein levels were 312±76 nmol/l for control subjects; 914±210 nmol/l for CRF patients; 404±96 for diabetic patients; 300±89 for chronic liver disease patients; 330±91 for CVA patients and 1212±210 for pneumonia patients. The differences between controls and diabetes, CRF, and pneumonia patients are significant with a $p<0.01$; 0.001 and 0.001, respectively. In diabetes patients, acrolein correlates with fasting glycemia and HbA1c (r=0.5 and 0.41 respectively, $p<0.05$). We failed to see a difference in acrolein levels in liver disease, suggesting that liver metabolism is not a major player in the circulating acrolein pool. Acrolein levels are extremely high in pneumonia, suggesting it as a potential marker of infection and/or damage by inflammation. This new finding also lends mechanistic support to the synthesis of acrolein from threonine by phagocytes MPO as a major source of acrolein in vivo.

C2

PROVIDER OPINIONS AND PRACTICES IN MANAGING POSTPARTUM DEPRESSION

J. L. Drowos DO, MPH[1], A. Alonso, MD, MPH[2], G. P. Suciu, MSPH, PhD[3], R. D. Lovinger, MD[4], [1]Dept. of Family Practice Residency, Broward General Medical Center, PBCHD, NSU-COM, [3]Public Health Program, NSU-COM, Davie FL, [2]Palm Beach County Health Department, [4]Preventive Medicine Residency Palm Beach County Health Department, West Palm Beach, FL

Purpose: Despite the availability of effective screening tools for postpartum depression this condition remains largely underdiagnosed in the United States. We performed a pilot study to survey attitudes and practices among healthcare providers in diagnosing and treating postpartum depression.
Methods: A cross-sectional analysis was performed based on a 21 item questionnaire. Questions were based on prior screening studies for evaluating attitudes and practices for post partum depression. All healthcare providers serving new mothers and newborns at the Palm Beach County Health Department clinic locations received copies of the survey for participation. This included family physicians, pediatricians, obstetricians, physician assistants and nurse practitioners. Parameters were evaluated independently using the SAS statistical computer package for contingency table analysis, using Chi-square test of independence and the corresponding p-values to determine the significance between particular variables.
Results: 24 providers (50% response rate) completed questionnaires for inclusion in the data analysis. Trends were apparent including a number of providers (7 or 29.2%) who reported they had never received training pertaining to postpartum depression. Significant relationships were found between variables including provider gender and general beliefs about postpartum depression, as well as between specialty practiced and personal attitudes toward postpartum depression.
Conclusions: Trends in managing postpartum depression related to provider characteristics can be understood. Surveys can uncover providers who have not received training on the subject, and to identify those who feel uncomfortable with their knowledge base. Identifying those specific providers who are most likely to care for affected patients and who are unfamiliar with screening and treatment measures can facilitate designing educational interventions and policies for Palm Beach County Health Department clinicians.

♦C3

Withdrawn at the Author's Request

♦C4

BREAST CANCER AND MAMMOGRAPHY: HOW KNOWLEDGEABLE ARE WOMEN OF THE RECOMMENDATIONS?

S. K. Desai, BS, BA[1], J. V. Davis III, DO[2], A. Gnanadev, MD[3], D. T. Wong, MD[3], [1]WesternU-COMP, [2]Dept. of Surgery, WesternU-COMP/ARMC/OPTI, Pomona, CA, [3]Dept. of Surgery, Arrowhead Regional Medical Center, Colton, CA

Background: Several randomized trials have shown that screening mammography decreases Breast Cancer mortality specifically, in women 50 and older. Significant protection has also been demonstrated in women who begin screening at 40 years of age. The estimate for reduction of Breast Cancer mortality attributable to mammogram screening in the U.S. is approximately 10%. The American Cancer Society, American College of Radiology, National Cancer Institute, and the American College of Obstetrics and Gynecology all recommend routine screening at age 40.

Purpose: To determine how frequently women diagnosed with Breast Cancer at Arrowhead Regional Medical Center (ARMC) obtained a mammogram the year before their most recent mammogram leading to their diagnosis.

Methods: We conducted a retrospective study with 87 female patients diagnosed with Breast Cancer between Jan 2005-March 2006. We reviewed ARMC Tumor Registry charts to obtain demographic, staging, and diagnostic information.

Results: 70 out of 87 (80.5%) Breast Cancer patients did not complete a mammogram one year prior to their diagnosis. Of these 70 patients, 16 were categorized with Stage I, 22 with Stage II, 16 with Stage III, and 15 with Stage IV Breast Cancer. The average age of patients was 51.7±10.6. Of the 70 females who did not have a mammogram one year prior to their diagnosis, 59 (84%) were age 40 and above.

Conclusion: The recommendation of routine screening in women over 50 years of age is well established. Though it is controversial between 40 and 50 years of age. Our data indicates that the vast majority of women at our county facility have not received timely and appropriate mammographic evaluations prior to their diagnosis. This is unfortunate because it has been shown that women who have had one screening mammogram in the previous 2 years were less likely to be stage II B or higher. There may be a lack of understanding, or a failure to follow through even when knowledgeable. Also, it may be an inability to access resources that prevents them from obtaining care. Among our patients, it may be a communication or cultural issue as well. Better cultural and language instruction for non-English speaking patients may improve compliance.

C5

The Safety and Efficacy of Duloxetine Hydrochloride for the Treatment of Fibromyalgia: Results from a 6-month Randomized, Double-Blind, Placebo-Controlled, Fixed-Dosed Trial

M. Wohlreich, MD[1], J. Russell, MD, PhD[2], P. Mease, MD[3], T. Smith, MD[4], D. Kajdasz, PhD[1], D. Walker, PhD[1], F. Lewis, DO[5], A. Chappell, MD[1], [1]Eli Lilly and Company, Indianapolis, IN, [2]University of Texas Health Science Center, San Antonio, TX, [3]University of Washington School of Medicine, Seattle, WA, [4]Mercy Health Research, Chesterfield, MO, [5]Global Therapeutic Development in Neurology and Ps, Weston, FL

Objectives: The primary objective was to determine whether treatment with duloxetine [DLX] 120 mg/d for 3 months was effective in reducing pain severity in patients with fibromyalgia syndrome [FMS].

Methods: DLX60 mg/d, DLX120 mg/d, and placebo [PBO] were compared during 6 months of treatment in adults with American College of Rheumatology-defined primary FMS. Coprimary efficacy measures included the Brief Pain Inventory Average Pain Score [APS], and the Patient's Global Impressions of Improvement [PGI-I] questionnaire. Safety and tolerability were assessed.

Results: At 3 months, patients treated with DLX120 mg/d showed greater improvement in change from baseline in APS score [-2.31 vs. -1.38, $P<0.001$] and endpoint PGI-I score [2.89 vs. 3.39, $P=0.004$] vs. PBO-treated patients. At 6 months, the DLX120 mg/d group still exhibited greater improvement in APS change [-2.25 vs. -1.42, $P=0.003$] and PGI-I [2.93 vs. 3.37, $P=0.012$]. The DLX60 mg/d group showed significant improvement compared with PBO on both measures at 3 months and on APS change at 6 months. At 6 months, response, defined as $\geq$50% reduction from baseline in APS, was greater with DLX120 mg/d [35.9%, $P\leq0.01$] and DLX60 mg/d [32.6%, $P\leq0.05$] vs. PBO [21.6%]. DLX was similarly efficacious in patients with [N=122] or without [N=375] major depressive disorder [MDD] with regard to both co-primaries. Discontinuation rates over 6 months were similar among the groups (DLX60 mg/d 45.3%, DLX120 mg/d 46.3%, PBO 50.0%). Adverse event-related discontinuation was significantly higher in the DLX120 mg/d [25.9%, P=.009] but not in the DLX60 mg/d [15.3%, P=0.400] group compared with the PBO (11.8%) group.

Conclusions: DLX60 and DLX120 mg/d are efficacious and safe treatment options for pain associated with FMS, whether or not MDD is present

♦C6

Minimally Invasive Posterior Lumbar Interbody Fusion after Treatment with Recombinant Human Bone Morphogenic Protein-2 Added to Bioresorbable Implants: Sur-

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

gical Technique, Clinical Results, and Review of the Literature

A. J. Scumpia, OMS-IV[1], K. O. Colle, DO[2], A. C. Mapley, OMS-IV[1], M. C. Overby, MD[2], M. B. Eisenberg, MD[2], M. E. Levine, MD[2], P. H. Hollis, MD[2], [1]Dept. of Neuroscience, New York College of Osteopathic Medicine/NYIT, Old Westbury, NY, [2]Dept. of Neurosurgery, North Shore University Hospital-LIJ Health System, Manhasset, NY

Hypothesis: The purpose of this study is to evaluate the clinical suitability of combining recombinant human bone morphogenic protein-2 (rhBMP-2) to bioresorbable implants as interbody spacers in spinal lumbar fusion surgery, via the minimally invasive posterior lumbar interbody fusion (PLIF) technique.

Materials and Methods: This is a retrospective study of 40 consecutive patients who underwent the minimally invasive PLIF procedure after treatment with rhBMP-2 added to bioresorbable implants as interbody spacers in single and multi-level applications for various lumbar spine pathologies. This study assessed the use of combining rhBMP-2 to bioresorbable implants in forty consecutive patients with at least two years of follow-up who underwent the minimally invasive PLIF for various spinal pathologies, particularly spondylolisthesis, herniated nucleus pulposis, and degenerative disc disease.

Results: At the nine month post-operative follow-up, 38 (95%) were found to have solid fusions. Independent neuroradialogists' evaluations of CT scans with reconstructions were obtained at 3, 6, 9, 12, and 24 months respectively.

Conclusions: The minimally invasive PLIF procedure utilizing bioresorbable implants in conjunction with rhBMP-2 can achieve successful lumbar fusion safely and effectively, for a variety of common lumbar spine pathologies.

C7

Dyslipidemia among Stroke Patients in a Large, Urban Tertiary Care Center

A. N. Russman DO, D. R. Nerenz, PhD, Henry Ford Hospital, Detroit, Michigan

Hypothesis: Dyslipidemia is an important modifiable risk factor in patients with ischemic stroke. Prior to a first stroke, the current NCEP III guidelines suggest maintenance of low-density lipoprotein (LDL) less than 130 and high-density lipoprotein (HDL) greater than 40. Clinicians will need to address dyslipidemic management during the initial admission.

Methods: Following IRB approval, we queried the Henry Ford Hospital database for single-payer HMO patients with an admission and an ICD-9 code for ischemic stroke during the period from 01/01/2001 to 06/30/2006. Medical records were reviewed and data collected on demographics, lipid studies, and length of stay (LOS).

Results: We identified 944 patients, 51% women, mean age of 66 years (range 27-97). Average fasting lipid values were cholesterol 183, triglycerides 143, LDL 110, and HDL 46. The average LOS was 5.4 days. Using LOS as a proxy for disease severity, patients with LDL greater than 130 tended to have a lower average LOS (5.0 versus 5.5 days, p=0.24) compared to those with levels less than 130. In addition, patients with HDL less than 40 tended to have a higher average LOS (5.7 versus 5.1 days, p=0.14) compared to those with an HDL greater than 40. Women were more likely to have a higher LDL (115 versus 104, $p<0.001$), and HDL (50 versus 42, $p<0.001$) than men. Patients over the age of 65 were more likely to have LDL less than 130 (34% versus 26%, p=0.01) and HDL levels greater than 40 (43% versus 35%, p=0.02). However, LOS was higher for patients over the age of 65 (5.8 versus 4.8 days, p=0.006) compared to those under 65.

Conclusion: Among our urban population, younger patients tended to have a less favorable profile of high LDL and low HDL. Women were more likely to have an intermediate risk profile with higher serum LDL and HDL. Low serum HDL may be a risk factor for longer length of stay and possibly disease severity. Our data suggest that studies are needed to better characterize the relationship between ischemic stroke severity and dyslipidemia.

♦C8

Influence of Customized Proprioceptive Foot Orthotics in Posture, Balance and Pain

V. A. Cohen MS, OMS-I, M. L. Kuchera, DO, FAAO, P. E. Stewart, PhD, CCDA, Human Performance & Biomechanics Laboratory, Philadelphia College of Osteopathic Medicine, Philadelphia, PA

Background: The adult body constantly receives information from proprioceptors throughout the body, including the soles of the feet. The CNS integrates this proprioceptive information with visual and vestibular information. The input integrated from these 3 systems is important in homeostatic maintenance of balance and posture from moment to moment as the body ages.

Hypothesis: Custom-fitted foot orthotics working through the proprioceptive system will provide a greater level of pain relief, range-of-motion (ROM), and postural balance than sham foot orthotics.

Methods: 22 subjects with low back pain were randomized into treatment (n=11) or placebo/sham (n=11) insole groups, with 13 subjects (7 treatment, 6 placebo/sham) completing the entire study. All subjects completed an Oswestry Disability Questionnaire (ODQ), pain VAS, balance test, and ROM evaluation during the 1st visit. Additionally, each participant received a "body balance check" (according to Vabena protocol) that dictated the specific prescription for customized insoles. Upon receiving their insoles, balance and ROM tests were rechecked. During the 3rd and 4th visit, all surveys and tests were repeated. Additionally, at the 4th and final visit, a post treatment body balance check was conducted and randomization was revealed.

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

Results: A clinical importance measure was demonstrated in this study. The treatment group achieved the ODQ mean clinically important difference (MCID ?6%) at both 3rd and 4th (11.04%, 10.86%) visits. Additionally, a 2-tailed fisher's exact test detected a significant difference (p=0.0344) in the proportion of treatment to placebo subjects clinically improving between first and last visits. Aside from a suggested significant baseline difference in ODQ (p=0.063) between the treatment and placebo groups, ANOVA and univariate statistics showed no statistically significant pain, balance, or postural differences between or within treatment and placebo groups. However, upon reviewing subject compliance, those who most regularly wore their insoles demonstrated the greatest decrease in their ODQ score.

Discussion: This study demonstrated that systematic proprioceptive stimulation using neuromuscular proceptor soles (Vabena protocol) may be clinically beneficial in decreasing patient pain perception. Larger trials, controlled for compliance, however are warranted to fully understand the impact of these insoles in treating patient posture, balance, and chronic back pain.

C9

Ropinirole CR Extended Release Reduces Mood Disturbance in Patients with Moderate-to-Severe Primary Restless Legs Syndrome (RLS): Results from a 12-Week Pivotal Trial

R. B. Ziman MD, FACP[1], J. S. Gitt, DO[2], M. Castiglia, PharmD[3], N. Williams, MSc[4], [1]Northridge Neurological Center, Norridge, CA, [2]Hope Research Institute, Phoenix, AZ, [3]GlaxoSmithKline, [4]Research Triangle Park, NC, GlaxoSmithKline, Harlow, Essex

HYPOTHESIS: Patients with Restless Legs Syndrome (RLS) may report comorbid symptoms of depression and/or anxiety. A 14-hour extended-release formulation - ropinirole CR - was developed to provide symptom coverage in patients with primary RLS who experience symptoms that occur in the late afternoon/early evening and during the night. The effect of treatment with ropinirole CR on mood in patients with moderate-to-severe RLS was assessed.

MATERIALS AND METHODS: In a 12-week pivotal study (protocol 101468/205), patients with moderate-to-severe primary RLS experiencing symptoms in the evening and at nighttime were randomized to ropinirole CR (n=189), 0.5-6.0 mg/day, or placebo (n=195), titrated as needed and tolerated, taken once daily (4pm or later) 1-2 hours before the usual onset of RLS symptoms. Effect on mood was assessed by mean changes from baseline in the Hospital Anxiety and Depression (HADS) subscale scores (in symptomatic subpopulations defined by a baseline score ⩾8) and in the Profile of Mood State (POMS) scale six domain scores and total mood disturbance score at Week 12 last observation carried forward (LOCF). The primary endpoint for the study was mean change from baseline in International Restless Legs Scale (IRLS) total score at Week 12 LOCF.

RESULTS: Greater improvements from baseline (reductions) in mean HADS anxiety and HADS depression subscale scores were observed at Week 12 LOCF for ropinirole CR vs. placebo, with a statistically significantly greater improvement seen for the HADS depression subscale (adjusted mean treatment difference [AMTD]: -2.3; $p<0.01$). The AMTD for the HADS anxiety subscale was -1.3 ($p=0.13$). In addition, a greater improvement in each domain and the total mood disturbance score of the POMS scale was observed with ropinirole CR vs. placebo at Week 12 LOCF (not tested). For the primary endpoint, a significantly greater improvement (reduction) in IRLS total score was seen for ropinirole CR vs. placebo (AMTD: -5.9; $p<0.001$).

CONCLUSIONS: Ropinirole CR reduces RLS severity and depressive mood symptoms associated with RLS in patients with moderate-to-severe primary RLS.

SUPPORTED BY: GlaxoSmithKline Research and Development.

C10

Ropinirole CR Extended-Release Treatment Improves Both the Symptoms and the Impact of Restless Legs Syndrome

R. B. Ziman MD, FACP[1], J. S. Gitt, DO[2], N. Williams, MSc[3], [1]Northridge Neurological Center, Norridge, CA, [2]Hope Research Institute, Phoenix, AZ, [3]GlaxoSmithKline, Harlow, Essex, UK

HYPOTHESIS: Restless Legs Syndrome (RLS) is a chronic neurological disorder. The International Restless Legs Scale (IRLS) is a validated, disease-specific, 10-item scale for assessing RLS symptoms, from which two subscales can be derived: the Symptoms subscale (items 1, 2, 4, 6, 7, and 8, which relate to the severity/frequency of core symptoms and sleep disturbance [the primary morbidity of RLS]), and the Symptoms Impact subscale (items 5, 9, and 10, relating to the impact of core symptoms on daytime sleepiness, daily functioning, and mood). Ropinirole CR, a novel 14-hour extended-release formulation, was developed to provide extended coverage of RLS symptoms. The effect of ropinirole CR treatment on the core RLS symptoms and impact of symptoms was assessed using IRLS subscale data from a pivotal study in patients with moderate-to-severe primary RLS.

MATERIALS AND METHODS: In this 12-week, double-blind, placebo-controlled study (protocol 101468/205), patients with moderate-to-severe primary RLS were randomized to ropinirole CR (n=189), 0.5-6.0 mg/day, or placebo (n=195), titrated as needed and tolerated, taken once daily (⩾4pm), 1-2 hours before usual onset of RLS symptoms. The effect of treatment on the core RLS symptoms and impact of symptoms was evaluated by *post-hoc* analysis of mean changes from baseline in IRLS Symptoms and Symptoms Impact subscale scores (ranges 0-24 and 0-12, respectively) at Week 12 last observation carried forward (LOCF). The primary endpoint was mean change from baseline in IRLS total score (range 0-40) at Week 12 LOCF.

RESULTS: At baseline, mean IRLS subscale and total scores were similar between treatment groups. At Week 12 LOCF, statistically significant treatment benefits were seen for ropinirole CR, compared with placebo, for improvements (decreases) in the IRLS Symptoms and Symptoms Impact subscale mean scores (adjusted mean treatment difference [AMTD]: -3.8 and -1.6, respectively; both $p<0.001$). Ropinirole CR also significantly improved (decreased from baseline) mean IRLS total score at Week 12 LOCF compared with placebo (AMTD: -5.9; $p<0.001$).
CONCLUSIONS: Ropinirole CR extended release improves the core symptoms of RLS and the impact of these symptoms in patients with moderate-to-severe primary RLS.
SUPPORTED BY: GlaxoSmithKline Research and Development.

C11

Efficacy Comparison of Albendazol and Ivermectin in Treatment of Ascariasis in Children

A. Moradi M.D., MPH & TM, P. Mortazavi, PharmD, R. Nassiri, DSc, Department of Pharmacology and Public Health LECOM College of Osteopathic Medicine, Bradenton, FL

Abstract: Ascariasis is an intestinal infection caused by the parasitic roundworm *Ascaris lumbricoides*. It is the most common of intestinal helminthes infection which is limited to humans. The infection can be seen in all age groups but the highest morbidity appears to be in children.
Objective: To evaluate the high efficacy of albendazole in comparison to ivermectin for the treatment of intestinal infection caused by parasitic roundworm ascaris.
Material and Method: The treatments used in this study were a single dose of oral Albendazole 400mg and a single dose of oral Ivermectin 200mcg/kg. We tested the efficacy of both medications in two control trial groups of 105 school children during a light infection stage (early stage) with ascaris in Bahamas. This study clearly demonstrated that the treatment efficacy in children with a single dose of albendazoloe considerably differs than a second group with a single dose of ivermectin.
Result: The treatment with a single dose of albendazole in first group of children produced cure rates of over 93%. On the other hand a single dose of ivermectine in the second group of children had only produced a 75% cure rate initially and needed an additional dose given at a 7 days interval to archive a 95% cure rate.
Conclusion: Our result suggests that single dose oral albendazole has higher efficacy against light ascaris infection than ivermectin in a similar condition. These findings are of major public health relevance in cost-effectiveness in treating school age children with very effective therapy and no need for further doses to be given at interval time.

C12

Tolerability with No Loss of Efficacy after Overnight Conversion to a Novel Extended-Release Formulation, Ropinirole CR, in Primary Restless Legs Syndrome (RLS)

A. L. Ellenbogan DO[1], MPH, M. Castiglia, PharmD[2], R. A. Hodge, MSc[3], [1]Quest Research Institute, Bingham Farms, MI, [2]GlaxoSmithKline, Research Triangle Park, NC, [3]GlaxoSmithKline, Harlow, Essex, UK

HYPOTHESIS: Many patients with moderate-to-severe primary Restless Legs Syndrome (RLS) experience symptoms that occur in the late afternoon/early evening and at night, and may benefit from an extended-duration treatment. Safety and tolerability were assessed after overnight conversion from the current formulation of ropinirole (ropinirole IR) to a new, once-daily, 14-hour, and extended-release formulation - ropinirole CR - in patients with primary RLS.
MATERIALS AND METHODS: In a randomized, double-blind, three-cohort study (protocol 101468/805), patients currently treated with ropinirole IR for RLS entered one of three cohorts (A, B, or C) based on their stable dose of ropinirole IR (1mg, 2mg, or 4mg, respectively). Patients were then randomized (1:1) to one of two dosing groups within each cohort and entered a 4-week treatment phase. At one of two pre-identified clinic visits, patients were converted overnight to ropinirole CR for 1 week (2mg, 3mg, and 6mg in cohorts A, B, and C, respectively). To allow for comparison between groups in each cohort and to maintain the blind, at each conversion time-point one group underwent an actual conversion to ropinirole CR, while the second group underwent a "dummy" conversion. Primary endpoint was incidence of post-conversion-emergent adverse events (AEs), defined as first occurrence, new episode, or increase in severity of an ongoing AE. Efficacy was assessed by pre- to post-conversion change in International Restless Legs Scale (IRLS) total score.
RESULTS: Baseline demographics and characteristics were similar between groups. Emergent AEs (any) following actual vs. dummy conversion occurred in 37% vs. 43%, 33% vs. 37%, and 46% vs. 36% of patients in cohorts A, B, and C, respectively. The most common AE following actual conversion in each cohort was nausea (A and B) and somnolence (C). No patients discontinued due to AEs in cohorts A or C; 2 patients (1 IR, 1 CR) discontinued in cohort B. Overall, conversion to ropinirole CR did not lead to a loss of efficacy based on mean change from pre- to post-conversion in IRLS total score.
CONCLUSIONS: Overnight conversion from ropinirole IR to the longer-acting ropinirole CR extended release was generally well tolerated, with no loss of efficacy.
SUPPORTED BY: GlaxoSmithKline Research and Development.

C13

Bipolar Depression: Diagnosis, Recognition, and Treatment Experience with Quetiapine

S. D. Levy DO, West Chester Psychiatric Associates, West Chester, PA

Hypothesis: Bipolar disorder is a complex illness with episodes

of mania, hypomania, depression, or mixed states alternating with euthymia. Bipolar depression is now recognized to be an extremely common and disabling part of the illness and is often misdiagnosed as major depression[1]. For appropriate management, it is important that primary care physicians also be involved in the accurate diagnosis and recognition of bipolar disorder. Currently available atypical antipsychotics (olanzapine, quetiapine, risperidone, aripiprazole and ziprasidone) have demonstrated efficacy in treating bipolar mania as monotherapy but only quetiapine monotherapy has received approval in the US for treating bipolar depression[2,3].

Materials and Methods: A post hoc evaluation of outpatients with DSM-IV bipolar disorder experiencing depressive episodes combined from two similarly designed, 8-week, double-blind, randomized, placebo-controlled trials of quetiapine monotherapy (fixed doses, 300 or 600 mg/d once daily; BOLDER I & II).The primary efficacy endpoint was mean change from baseline to Week 8 in MADRS total score (ANCOVA/LOCF analysis).

Results: Combined data from these trials (n=1045) showed that quetiapine monotherapy (300 or 600 mg/d) significantly ($P<0.001$) improved MADRS total score from Day 7 to Day 56 (effect size: 300 mg/d 0.65; 600 mg/d 0.69), and significantly improved HAM-D, HAM-A, CGI-S, and YMRS scores, compared with placebo. All individual MADRS items were significantly ($P<0.001$) improved, by week 8 of treatment compared with placebo, with 7 of the items being significant from Week 1 through Week 8.Quetiapine monotherapy also significantly ($P<0.001$) improved patients health-related quality of life. Quetiapine was generally well tolerated in both studies, with low and similar incidences of treatment-emergent mania in quetiapine and placebo groups.

Conclusions: Quetiapine is the first of the currently available treatments to demonstrate substantial clinical efficacy as monotherapy (likely optimal dose: 300 mg/d) for treating depressive episodes for both bipolar and bipolar II disorder. Moreover, unlike antidepressants, quetiapine monotherapy did not appear to increase the risk for switching into hypomanic or manic states.

References

[1]Post RM. J Clin Psychiatry. 2005;66 Suppl 5:5-10.

[2]Calabrese JR, et al. Am J Psychiatry 2005; 162:1351-60.

[3]Thase ME, et al. J Clin Psychopharmacol 2006; 26:600-9.

Supported by funding from AstraZeneca Pharmaceuticals LP.

C14

Higher Levels of Ischemia-Modified Albumin in Acute Cerebrovascular Accidents: A Pilot Study

A. Gugliucci MD, PhD[1], S. Kimura, MD[2], T. Menini, MD, MS[3], [1]Dept. of Biochemistry, Touro University-CA, Vallejo, CA, [2]Laboratory Medicine, Showa University, Yokohama, Yokohama, Japan, [3]Dept. of Basic Sciences, Touro University-CA, Vallejo, CA

Ischemia modified albumin (IMA) could become a triage tool in acute coronary syndrome patients. It does not appear to be very tissue or clinically specific and might also find utility in stroke. In this pilot study we set out to confirm this contention and to test the hypothesis that IMA levels raise even more in subsequent days after the acute episode.

We studied 12 consecutive patients presenting within 6 h of the onset of an acute neurological deficit as well as 40 age-matched controls. Serum samples were obtained for all patients at initial presentation and repeated only in patients with stroke at 2, 4 and 6 days. IMA was measured by the albumin-cobalt-binding test and expressed as absorbance arbitrary units. The intra-assay CV was 5%. The initial IMA was 0.57 ± 0.08 AU for stroke patients vs. 0.55 ± 0.09 AU for control subjects, showing no significant differences ($p<0.2$). At 2-4 days IMA levels raised to 0.69 ± 0.1 AU ($p<0.001$ vs. day 0). IMA levels not only do not return to control levels in 2 or 4 days but continue to rise and, in some cases, remain elevated for more than a week (0.7 ± 0.03, n=3). Even with the limitations of small n, this is the first study to serially measure IMA levels in stroke patients after 24h post-episode, and the data are consistent with a persistent elevation of IMA levels beyond the first day, suggesting this marker is sensitive to changes in brain tissue resulting from ischemic injury and its oxidative stress consequences.

C15

Diabetes Self Care: Unrealistic Expectations?

J. Shubrook DO[1], G. Klein, OMS 1[2], F. L. Schwartz, MD[3], A. Wapner, DO[4], [1] Dept. of Family Medicine, [2]Dept. of Student Services, [3]Dept. of Specialty Services, [4]Dept. of Pediatrics, OUCOM, Athens, OH

Background: It has been estimated that 90% of diabetes self-care falls on the patient. Diabetes educators are critical providers to help patients learn the skills and habits necessary to successfully manage their diabetes. A previous study by Russell, Suh, and Safford showed in focus groups with diabetes educators that time demands for diabetes self-management were substantial if patients followed educators' recommendations. A pilot study by the lead author found that self-care for people with type 2 diabetes (DM) can require up to 3 hours a day. In this study, the authors quantified the necessary time needed to complete all of the tasks for self-management in a national sample of diabetes educators.

Methods: The prototypical patient for this study was an adult with chronic type 2 diabetes on oral medications alone. Questionnaires were administered to a national list of diabetes educators. The questionnaire addressed the estimates of time demands for self-care as directed by the ADA guidelines. This study was approved by the Ohio University Institutional Review Board.

Results: At the time of submission 674 questionnaires have been analyzed. The time needed for the required elements for a person with type 2 DM was 234 minutes (3 hours and 54 minutes). There was substantial variation in the estimates of each

component of self-care. In addition, recommendations varied by the training of the educator and their previous experience with diabetes.

Conclusions: The recommendations for DM self-care are likely too time intensive for most people to successfully complete. Despite national guidelines explaining the components for self-care the actual recommendations vary greatly. These differences should be evaluated and addressed.

C16

KCUMB DOCARE: A Service Oriented, Educational, and Research Outreach Program in Guatemala

G. J. Desai DO[1], D. L. McCandless, PharmD[2], A. G. Glaros, PhD[3], D. Dyck, DO[3], R. Magie, DO[5], R. Ogden, DO[6], [1]Dept. of Medical Affairs, Family Medicine, [2]Dept. of Pharmacology, [3]Dept. of Basic Medical Sciences, [4]Dept. of Family Medicine, [5]Dept. of Pediatrics, [6]Dept. of Family Medicine, KCUMB, Kansas City, MO

Hypothesis The investigators conducted a medical mission in February 2007, in conjunction with DOCARE International, in Guatemala, C.A. Prior missions resulted in clinical impressions that the most common diagnoses were gastroesophageal reflux disease (GERD), headache, cough and upper respiratory symptoms, musculoskeletal pain, and gastroenteritis (GE). Based on past experience, the investigators also felt it likely that most of the patients would be women and children. Our goal in this year's project was to continue to provide screening and educational services and to assess the accuracy of prior clinical impressions using a structured, data collection form.

Materials and Methods With IRB approval, patients seen during the 2-week mission had their non-identifiable data gathered in a computer-readable sheet that was used as the encounter form. All individuals screened during this visit lived in rural areas several hours distant from the capital, Guatemala City.

Results Proportions of valid, non-missing data include 1477 patients, 70% female, and 28% male. Age ranges most represented were 1-4 (10% of total pts), 5-11 (24%), 18-39 (29%), and 40-64 (19%), Diagnoses most often made were GERD (17%), headache (17%), rhinitis (8%), GE (8%), and arthropathy (8%). The most commonly dispensed medications were acetaminophen (24% of visits), children's multivitamins (23%), adult multivitamins (19%), H2 - blockers (17%), and ibuprofen (16%).

Conclusions The most commonly seen diagnoses correlated to an earlier study, as well as previous missions. The majority of patients were women and children, confirming earlier clinical impressions. We don't have data on why the proportion of men was so low. We hope to increase the number of men seen in the future. Infectious diseases often affect the health of rural Guatemalans, and development of public health programs emphasizing general hygiene may have a positive impact. Some data may be skewed as medication supplies affect dispensing habits, and we often deplete the entire stock of medication. The international rotation served to improve health of Guatemalans, and served as an eye-opening experience for medical students, faculty and staff, with extremely positive subjective feedback from participants and patients. The data will be used to better understand the population served and provide an empirical basis for planning for future missions.

♦C17

Barriers to Effective Chronic Pain Management: Perceptions of Prescribers in Rural Appalachia

E. N. Remster OMS-IV, T. L. Marx, DO, CMD, Department of Family Medicine
OUCOM, Athens, OH

Context: Chronic pain is a significant cause of morbidity among adults worldwide. Previous research indicates "prescriber reluctance to prescribe Opioid" and "inadequate knowledge of appropriate pain management" as major barriers to effective management of chronic pain. Few studies have evaluated the opinions of prescribers working with the unique patient population in rural Appalachia regarding pain management.

Hypothesis: Prescribers in rural Appalachian Ohio encounter the same barriers as prescribers elsewhere in providing effective management of chronic pain.

Materials and Methods: This IRB-approved prospective study was designed to evaluate prescriber opinions of chronic pain management in rural Appalachian Ohio. The investigators developed a 30-item questionnaire and distributed it to physicians and nurse practitioners attending a CME conference on pain management. Demographics and aspects of pain management were assessed and participants were asked to rank a list of twelve perceived barriers to effective chronic pain management.

Results: Thirty-six questionnaires were distributed, 22 were returned (response rate of 61.1%). All participants reported practicing in rural Appalachian Ohio; fifteen practice primary care (family medicine, internal medicine and geriatrics) and 7 reported other disciplines. Overall, participants were male physicians with a mean age of 50. Most participants were involved with chronic pain management "several times each week" or more; fifteen participants reported that they wrote prescriptions for Opioid medications "several times each week" or more. A calculation of mean was used to identify whether participants ranked barriers as "more significant" or "less significant". The most significant barriers were 1) prescriber reluctance to prescribe Opioid; 2) inadequate access to health care due to financial burden; and 3) inadequate access to pain specialists.

Conclusions: Prescribers in rural Appalachian Ohio did identify the same top barrier to effective chronic pain management as prescribers elsewhere, but also identified issues of

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

health care accessibility as significant barriers. The researchers intend to use these preliminary study results to direct a larger, more comprehensive study involving all physicians in Appalachian Ohio.

C18

Patient Reported Effectiveness and Tolerability of Frovatriptan in a Large Population Previously Taking Other Triptans for Acute Migraine Treatment: An Open-Label, German Postmarketing Surveillance Study

B. L. Peterlin DO[1], S. Barlas, PhD[2], B. A. Jones, PhD[2], J. Campbell, BSc[2], [1]Department of Neurology Drexel University College of Medicine, Philadelphia, PA, [2]Medical Affairs, Endo Pharmaceuticals Inc., Chadds Ford, PA

Hypothesis: Migraineurs who are not effectively treated with one triptan may respond to another (Dodick D. *Headache*. 2005;45(2):156-162). In this retrospective sub-analysis of a large migraine postmarketing surveillance study, we examined the hypothesis that patients would report frovatriptan to be an effective and well-tolerated acute migraine treatment in migraineurs who had previously taken another triptan.

Materials and Methods: Among 7107 German patients initiated on frovatriptan in a post marketing observational study for acute treatment of migraine, 2437 were identified as previous triptan (other than frovatriptan) users. Baseline data included previous migraine characteristics, and effectiveness and tolerability with previous therapies which were assessed categorically as Poor, Satisfactory, Good, or Very Good. Ordinal data were described as both the absolute number and as the percentage of the overall cohort. Intrapatient change in ratings was evaluated by McNemar's test of agreement.

Results: The mean±SD age was 44 ±11 years, and 85% were women. Fifty-four percent of triptan-experienced patients had ⩾3 attacks per month; 34% reported attacks lasting >24 hours when untreated. Patients rating their treatment effectiveness as Good or Very Good improved from 39% with their previous triptan to 84% with frovatriptan. Similarly, 60% rated the tolerability of their previous triptan Good to Very Good compared with 93% with frovatriptan. Intrapatient comparison showed that 51% rated their previous triptan as Satisfactory to Poor and improved to Very Good or Good on frovatriptan ($P<0.001$). After switching to frovatriptan, patients' ratings of efficacy and tolerability were 8.2 times and 15.8 times more likely to improve to Good or Very Good than to decrease to Satisfactory or Poor ($P<0.001$). At the conclusion of the study, 85% of patients indicated they would continue treatment with frovatriptan.

Conclusion: Patients who reported less than Good efficacy and/or Good tolerability with their previous triptan reported clinically meaningful improvements in acute migraine treatment after switching to frovatriptan.

C19

Immediate and Prolonged Effects of a 10-Week Maximal Effort Exercise (with Valsalva) Protocol on Cognition in Men and Women with Multiple Sclerosis: A Multi-Center Study

M. L. Kuchera, DO, FAAO[1], R. T. Dombroski, DO[2], T. Vardy, DO[3], F. Thomas, MD, PhD[4], P. Yagnik, MD, PhD[5], K. Wenzel, MA[6], S. Stoll, DO[7], PhD, B. Stouch, PhD[2], M. T. Wallin, MD, PhD[8], [1]CCDA, Human Performance & Biomechanics Lab, [2]PCOM, [3]Dept. of Neuromuscular Engineering, Tweed Heads, North South Wales, Australia, [4]Saint Louis VAMC, St. Louis, MI, [5]Philadelphia VAMC, Philadelphia, PA, [6]Rocky Mountain Multiple Sclerosis Center, Denver, Colorado, [7]UNTHSCFW-Texas College of Osteopathic Medicine, Fort Worth, TX, [8]Washington VAMC, Washington D.C.

Half of all Multiple Sclerosis (MS) patients develop cognitive dysfunction (CogD) with slowed ability to reason, concentrate, or remember. While 5-10% develop problems interfering with activities-of-daily living, mild CogD decreases quality-of-life and contributes to depression. CogD, 1of 3 MS Functional Composite measures, can be seen early but is more common in long-term MS.

Hypothesis: We hypothesized that adding a 10-week protocol of twice-weekly progressive maximal effort exercise (MEE) would improve scores on a standardized CogD test, the Paced Auditory Serial Addition Test (PASAT). We also hypothesized that benefits would be retained when rechecked at 6 and 12 weeks.

Methods: 68 subjects (60% female; median age 50 years) with mild-to-moderate MS participated in an adjunctive, standardized, multicenter 10-week twice-weekly non-fatiguing progressive MEE protocol. During MEE, isometric and eccentric vertical leg presses and semi-erect whole body (lunge) exercises (4 seconds each with Valsalva) were repeated 3-5X on specialized exercise and strength-sensing equipment (IsoP-UMP®, Neuromuscular Engineering).

PASAT-3, a 3-minute, validated instrument requiring concentration, short-term memory, and simple addition skills (numbers electronically spoken every 3 seconds) was performed at baseline; twice during the 10-week intervention; and at 6- and 12-week rechecks. One research coordinator scored all subjects.

The Institutional Review Boards (IRB) of Philadelphia College of Osteopathic Medicine (lead site), Texas College of Osteopathic Medicine and Veterans Administration Medical Centers of Philadelphia, St. Louis, Denver, and Washington all approved this study.

Results: Where perfect=60, PASAT-3 correct scores at baseline averaged 41.16 (range 12-60; 39.62-44.70 [95%CI]); 48.00 after 10-weeks MEE (range 18-60; 45.02-50.98 [95%CI]); and 49.06 three-months after discontinuing exercise (range 20-60; 46.08-52.03 [95%CI]). Significant change (~12%) from baseline correct was noted during the exercise period (+3.44 at week 6, $p<0.001$) and increased thereafter: end-MEE (+5.04, $p<0.0001$) and 12-week post-MEE (+6.64, $p<0.0001$).

Conclusions: Integration of a progressive, non-fatiguing IsoP-

UMP® MEE protocol twice weekly for ten-weeks results in improved cognition lasting at least 3 months post-MEE ($p<0.0001$).

Acknowledgment: Supported by joint civilian - Veterans Affair Project

C20

Immediate and Prolonged Effects of a 10-Week Non-Fatiguing Maximal Effort Exercise Protocol on Strength in Deconditioned Men and Women with Multiple Sclerosis: A Multi-Center Study

M. L. Kuchera, DO, FAAO[1], R. T. Dombroski, DO[2], T. Vardy, DO[3], F. Thomas, MD, PhD[4], S. O'Brien, MD, PT[9], P. Yagnik, MD, PhD[5], K. Wenzel, MA[6], S. Stoll, DO[7], PhD, B. Stouch, PhD[2], M. T. Wallin, MD, PhD[8], [1]CCDA, Human Performance & Biomechanics Lab, [2]PCOM, [3]Dept. of Neuromuscular Engineering, Tweed Heads, North South Wales, Australia, [4]Saint Louis VAMC, St. Louis, MI, [5]Philadelphia VAMC, Philadelphia, PA, [6]Rocky Mountain Multiple Sclerosis Center, Denver, Colorado, [7]UNTHSCFW-Texas College of Osteopathic Medicine, Fort Worth, TX, [8]Washington VAMC, Washington D.C., [9]Colorado Multiple Institutional Review Board, Denver, CO

Multiple Sclerosis (MS) patients become deconditioned and weak; poorly tolerating aerobic exercise or fatigue. Pilot data (*JAOA* 102(5):267-275; 2002) showed significant strength improvement after a 12-week program combining maximal effort exercise (MEE) with osteopathic manipulative treatment (OMT).

Hypothesis: In MS subjects, 20 2x-weekly MEE sessions will result in proximate and prolonged increases in isometric and eccentric strength measures above baseline.

Methods: 68 MS subjects (60% female; ageave=50 yrs) participated in a multicenter 10-week 2x-weekly progressive MEE protocol (without OMT) with rechecks at 6 & 12 weeks. MEE consisted of isometric & eccentric leg presses (IsoLP, EccLP) and total body lunges (IsoTBL, EccTBL) performed maximally (4 secs, 3-5 reps). MS Functional Composite, fatigability, and quality-of-life measures were followed longitudinally along with IsoPUMP® (Neuromuscular Engineering) strength measures. All college and VA IRBs approved the protocol.

Results: IsoLPave strengths maximally improved by week 6 ($p<0.0041$) and most retained increases 3-months post-protocol. Immediate post-MEE strengths increased 67 Lbs (range IsoLPave=21-113, $p=0.005$), a 13% increase. (Ave IsoLPmax=492 Lbs). At both post-exercise points, IsoLPave remained +52 Lbs (+8%) overall; some retaining +100 Lbs improvement (6-weeks post-MEE, $p=0.024$; 12-weeks post-MEE, $p=0.045$). EccLPave strength +46 Lbs (range=16-77, $p=0.004$) during exercise becoming insignificant by 6-weeks post-MEE.

IsoTBL strength was compared for 3 matched leg positions. Subjects gained considerable ($p<0.01$) strength in all positions at 6-weeks MEE, end-MEE, 6- and 12-week post-MEE (each IsoTBL gain averaged 51-63 Lbs; range=29-85), a 30% IsoTBLave strength increase. (IsoTBLmax averaged 174 Lbs).

Of 3 matched leg positions, EccTBL strength showed gain throughout the MEE protocol ($p<0.032$) for right foot posterior, however these gains were not maintained 6 & 12 weeks post-MEE.

Twelve standardized questions assessed fatigue. Reduction was seen at 7-weeks exercise as well as 1 and 6 weeks post-MEE (10/12, 11/12, 12/12 respectively).

Conclusions: Integration of a progressive, non-fatiguing IsoPUMP® MEE protocol twice-weekly for ten-weeks results in significantly increased isometric strength gains lasting three months post-MEE.

Acknowledgment: Supported by a joint civilian-Veterans Affairs Project.

C21

Withdrawn at the Author's Request

♦C22

Development of an Objective Instrument and Protocol to Document Two- and One-leg Balance with and without Visual Input

T. R. Drames, OMS-II[1], M. L. Kuchera, DO[1], M. Tratenberg, OMS-III[1], L. Yoo, BS, P. E. Stewart, PhD, T. C. Vardy, DO[2], R. T. Dombroski, DO[1], [1]Center for Chronic Disorders of Aging, Philadelphia College of Osteopathic Medicine, Philadelphia, PA, [2]Dept. of Neuro Muscular Engineering and Technology, Tweed Heads, Australia

Standing balance tests can be used to diagnose and assess neurological and musculoskeletal abnormalities that affect the lower extremity and central nervous system. The IsoBALANCE (*Neuromuscular Technologies*) force plate system tracks postural sway in the coronal and sagittal planes and quantifies total distance traveled as well as the percentage of time a subject remains within a 0.5, 1.0, and 1.5 inch radius from his/her mean center of gravity (MCOG).

Hypothesis: Standing balance tests performed using the IsoBALANCE system can provide an effective method in evaluating center of gravity (COG) and distance traveled in normal, healthy individuals.

Methods: Three standing balance test positions where performed in socks, with eyes open and eyes closed: two-leg stance (TLS), left and right one-leg stance with knee flexed 90° (OLS 90°), and with foot brought forward (OLS forward). All test positions were repeated and measured twice. OLS 90° and OLS forward were compared. Study subjects consisted of 18 male and 22 female healthy individuals, between ages 18 and 55, who reported no history of ankle instability or proprioceptive/vestibular defects. Subjects were required to hold each position for 30 seconds with the exception of eyes closed OLS, where tests lasting at least 10 seconds would be considered valid. All OLS trials were fully randomized in order to prevent adaptation.

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

Results: Statistical analysis using the Student-Newman-Keuls Test showed that OLS performance was better with eyes open than closed ($p<0.05$) relative to distance traveled and all MCOG radii tested. However, TLS with eyes open was only significantly different than TLS with eyes closed using the subject's 0.5 inch radius MCOG. There was no significant difference between OLS 90° and OLS forward trials. Furthermore, in OLS eyes closed trials, females performed significantly better than males ($p<0.05$).

Conclusions: These results demonstrate that the equipment and protocol can serve to effectively evaluate a number of balance-related parameters (including COGs, percent time in various COG ranges using MCOG, time in a given position, and total distance traveled over time), thereby providing a standard of normality from which comparisons may be made for interpretation of balance performance deficits.

C23

Positional differences in Paraspinal Soft Tissue Compliance as measured with the "Back Poker 7.0"

B. J. Rockey, BS[1], M. Streng, BS[1], W. Ji, BS[1], R. R. Conatser, MS[2], D. Noyes, PhD[1], J. N. Howell, PhD[2], J. M. Burns, DO[3], [2]Dept. of Biomedical Science, [3]Dept. of Family Medicine, [1]IINR at Ohio University, Athens, OH

Osteopathic physicians (D.O.'s) rely upon the ability to discriminate changes in tissue compliance to diagnose and treat neuromusculoskeletal disorders.

Clinically, D.O.'s are able to sense postural differences in regional compliance between standing, seated, and prone positions. In order to objectively measure such changes (i.e., pre- and posttreatment, etc.) a reliable instrument as sensitive as a D.O. is needed.

The Back Poker 7.0, under development at Ohio University, employs a haptic device, the PHANToM Premium 3.0 (SensAble Technologies, Inc.), to calculate compliance by measuring displacement per unit force applied perpendicularly to tissue. This study measured paravertebral compliance changes between subjects in commonly used diagnostic positions: seated and prone.

HYPOTHESES

1. The Back Poker is a reliable measuring instrument.
2. Objective compliance measurements of the paraspinal tissues taken in the seated position are significantly different from those in the prone position.

METHODS

Cohort: n=12 (6 male, 6 female); avg. age=32.5 yr; avg. BMI=24.89

Informed consent obtained per Ohio University IRB. For both seated and prone positions, 4 nonconsecutive measurement trials were taken 2 cm lateral to the right of spinous processes T3, T7, and L4, a total of 12 measurements per position. Subjects were randomized as to whether seated or prone tests were obtained first.

RESULTS

Effects of trial were minimal. Intraclass correlation coefficients demonstrated a high degree of reliability (1.0=perfect correlation). Seated: T3=0.98, T7=0.959, L4=0.976; Prone: T3=0.987, T7=0.981, L4=0.984. Repeated Measures ANOVA (RMANOVA) revealed statistically significant ($p<.05$) differences in compliance between seated and prone groups at both the T3 ($p=0.003$) and L4 ($p=0.041$) paraspinal levels, but not between seated and prone positions at the T7 level ($p=0.877$). RMANOVA showed no significant order (seated first/prone first) or sex effects.

CONCLUSIONS

The Back Poker is a reliable device for the objective measurement of tissue compliance as evidenced by the high intraclass correlation coefficients. Our results, which show significant differences in compliance at both the T3 and L4 levels between seated and prone positions, are comparable to a previous study that showed differences between standing and prone postions with a hand-held device (Waldorf et al., 1991).

C24

Clinical and Ultrasonic Comparisons in Acute Cervical Radiculopathy

I. Sharapov MD, PhD, V. V. Belyakov, MD, PhD, N. P. Yeliseev, MD, PhD, The Manual Therapy Center of Kaluga Region, Obninsk, Russia

Objective: To study radiographic, ultrasonographic, and color doppler features of acute cervical compression radicular syndromes.

Methods: We examined 20 patients (12M, 8F; average age 39.9±12.4 years) with cervical compression radicular syndrome. Twenty patients with reflex pain syndromes served as a comparison group, while another 20 patients served as control group. Investigations included clinico-neurological examinations, X-ray studies of the cervical spine, ultrasound studies of cervical intervertebral discs in B-mode, and color doppler energy imaging.

Results: Clinico-neurological examination diagnosed compression syndrome of the C5 root in 2 patients (10%), C6 root in 11 patients (55%), and C7 root in 7 patients (35%). According to data, spinal root compression occurred at the level of a corresponding disc, and the pain syndrome lateralization coincided with the side of the disc's largest herniation. Ultrasonographic study showed disk herniation size at the C5-C6 level was 3.01±0.81 mm for compression radiculopathy, 2.19±0.66 mm for reflex syndromes, and 0.15±0.46 mm for control group (<0.001). Herniation size at the C6-C7 level was 2.70±0.91 mm for compression radiculopathy, 2.15±0.54 mm for reflex syndromes, and 0.05±0.22 mm for control group. Sagittal diameter of the spinal canal at the C5-C6 and C6-C7 level was 11.31±1.52 mm and 12.38±1.53 mm respectively for compression radiculopathy, 12.61±1.27 mm and 13.30±1.43 mm respectively for reflex syndromes, and 13.26±0.87 mm and 13.73±1.17 mm respectively for control group. Using doppler energy imaging signs of cessation, epidural blood flow was

revealed in 12 patients with the compression radicular syndrome at the level of an injured intervertebral disc. This showed disturbance of venous circulation, stasis and epidural edema. Eight patients also had signs of disturbed venous circulation at the level of the underlying intervertebral disc (extended epidural edema).
Conclusions: The leading causes of acute cervical compression radicular syndromes are intervertebral disc hernias (55%) and protrusions (45%). Compression syndrome is developed when herniation size is 3.01 ± 0.81 mm ($p<0.001$) at the C5-C6 disc level and 2.70 ± 0.91 mm ($p<0.001$) at the C6-C7 disc level. Important pathogenetic factors of the compression radicular syndrome are epidural edema and stasis in veins of the epidural space accompanied with aseptic inflammation.

C25

DIFFERENTIAL DIAGNOSIS IN SPONDYLOGENIC DISEASES, Moscow, Russia

S. Anatoly, MD, FIMM Educational and Scientific Congress

AIM: The Center of Manual Therapy postulated that applying the following standard differential diagnostic principles in patients referred for manual therapy would identify a significant number of neoplasms.
1. Discrepancy of subjective and objective clinical symptoms.
2. Presence of vegetative component that cannot be successfully treated with medication or physical methods.
3. Bilateral pain radiation, failure of sensibility, or paresis of lower extremities if vegetation.
4. Combination of distinct vegetation with paresis of the proximal and distal sections of the lower extremity.
MATERIALS & METHODS: The Center of Manual Therapy analyzed 300,000 referral patients classified as "back pains" who were sent from other medical institutions to receive manual treatment. Personnel used the above principles to differentially diagnose diseases of the spine and joints versus other nosological forms.
Every manual therapeutist of the Center was recommended to thoroughly examine all referral patients not relying on X-ray information alone. X-ray examinations that were not of perfect quality or older than one month were repeated. Radiographic reports without films were not accepted. In the case of suspected metastasis, MRI was also obtained at the Center. Early recognition of mammary gland cancer with metastases into bones including the spine was determined by radionuclide scintigraphy. Detectable tumors affecting the spine were divided into primary vertebral tumors (benign and malignant) and secondary bone changes connected with metastasis. Additional diagnostic techniques were obtained as needed to reach the correct diagnosis.
RESULTS: According to the data collected at The Center of Manual Therapy, reliance on roentgenographic examination provides sufficient imaging to diagnose primary tumors and metastases of the spine precisely. However, in many cases of suspected neoplasia, the standard examination is not enough, even in possession of a good quality of roentgenogram and additional tests must be ordered.
The established scheme of examining patients in the Center resulted in 24% primary diagnoses. Differential clinical and instrumental diagnosis showed 12% neoplasms (primary and secondary), 3.6% polyneuropathies, 5.4% demyelenating diseases, and 3% mental disorders. Manual therapy was not applied to those patients and they were directed to other specialists with the primary diagnosis.

C26

Natal Craniocervical Trauma Affect on Psychoneurological Development of Children

I. Li, MD, FSA-Russian Federation Presidential Medical Clinic

Objective: To compare clinico-instrumental and manual examinations to psychoneurological deficiencies found in children with Minimal Cerebral Dysfunction (MCD) and craniocervical trauma (CCT) history and the effect of adding adaptive manual therapeutics to standard care.
Materials and Methods: MCD children aged 3-6 years with historic CCT but without congenital/organic developmental defects: Group1 (n=75) received adaptive manual therapy (craniosacral and neuromuscular techniques); Group2 (n=35) had standard medical care.
Results: Retrospective history noted abnormal pregnancies (50.9%) and complicated birth deliveries (79.15%). X-ray confirmed natal CCT including C1 rotation/subluxation, ?2-?7 vertebrae disfixation, and increased C1-C2 arch-spine distance. Cranial ultrasound doppler revealed vertebrobasilar deficiency (third part).
Neuropsychological (NP) testing showed a mosaic inlaid and mainly dynamic character of mental dysfunctions from low psychic tonic. Complex motions and dynamic praxis examination showed problems automizing different motions (73.6%). General speech underdevelopments (50%) pronouncing difficult words, mechanical word memory difficulty, and unstable attention were noted. By neurologic disorder syndrome criteria, nearly all had peripheral cervical deficiency syndrome; 21.8% had shoulder girdle amyotrophia.
Manual testing revealed craniosacral asymmetries (91.8%), functional blockades at spinal transition zones, and mid-cervical/lumbar hypermobility. Vertebral syndrome was observed (90%) with cervical restriction, scoliosis, weight-bearing abnormalities, and muscle pain syndromes (esp. cervical, shoulder girdle).
Complex rehabilitation therapy effectiveness criteria included EEG and NP testing. Only Group1 showed progress ($p<0.001$) in NP testing, the majority of psychological indicators, and EEG (more regular increased alpha rhythm; reduction of theta- and delta-waves). 75% of Group1 showed accelerated development of communicative behavior, speech and motor function formation, increased attention stability, and improved perceptive-productivity processes of cognitive activity.
Conclusions: Managing CCT effect on natal structure and

function in cranial and cervical areas has benefit. Psychoneurological development dysfunction compensation was 4 times faster when based on complex rehabilitation that includes manual therapy for cranial-cervical distortion.

C27

The Results of Duplex Doppler Sonography of the Extracranial Vertebral Arteries in Patients with Spondylogenic Vertebrobasilar Insufficiency

M. Bakhatadze MD, PhD, V. N. Galaguza, MD, N. V. Sidorskaja, MD, Centre for Manual Therapy, Moscow, Russia

INTRODUCTION: One of the causes of vertebrobasilar insufficiency (VBI) can be compression of the vertebral artery (VA) before it enters transverse foramen of the sixth cervical vertebrae (V1 segment of the VA). The vessel can be compressed between scalene and long cervical muscles. This is of potential interest to those treating these muscles with post-isometric relaxation manual therapy.

GOAL: To compare peak systolic blood flow velocity (Vps) in patients with diagnosed spondylogenic VBI with that in healthy subjects, in the neutral spinal position.

METHODS: From 09/2005 to 09/2006, two groups of patients diagnosed with spondylogenic VBI were independently formed by two neurologists (M.A. and V.N.): Group1 (n=38; age 40.6±10 years;16 male; 21 female); Group2 (n=38; age 41.5±11 years; 13 male; 25 female). **Inclusion criteria** were the complex of spondylogenic VBI symptoms: neck pain, vertigo/dizziness up on cervical rotation, occipital headache, visual disturbances, and buzzing/tinnitus in the ears. **Exclusion criteria** were arteriosclerosis, arterial hypertension and arterial diseases. Thirty-seven healthy subjects (age 44.2±12.4 years; 16 male; 21 female) formed the control group.

Vps was measured with Doppler duplex sonography (N.V.) in the V1 segment of both VAs. Data in both groups of VBI patients were compared (t-test) to healthy controls.

RESULTS: In most of the VBI patients, statistically significant Doppler differences between healthy subjects and VBI patients were found for the right VA. The mean Vps in V1 segments in the control group was 46±5.87 cm/sec. VBI group mean Vps measures (Group1=41.35±6.91 cm/sec; Group2=42.45±5.75 cm/sec) were both significantly lower than control (t=3.14; p=0.002 and t=2.45; p=0.01 respectively).

CONCLUSION: Tension and compression by contraction of impinging scalene and long cervical muscles can be the reason for VA compression in the V1 segment. Such compression can be the cause of blood flow reduction in the vessel, resulting in the symptoms seen in our patients diagnosed with spondylogenic VBI. [Scalene muscles were tender to palpation]

C28

Effectiveness of Steroid Epidural Blockades in Discogenic Compression Syndromes: A Double Blinded Randomized Study

N. Sergei, MD, PhD, A. B. Sitel, MD, PhD, K. O. Kuzminov, MD, S. P. Kanaev, MD, Russian League of MM Professionals, Moscow, Russia

OBJECTIVE: To evaluate the pain syndrome in compression syndrome patients with lumbar discogenic dorsopathy, supervising them dynamically and conducting a course of Manual Therapy (MT) along with Medication Epidural Blockades (MEB).

METHODS: To investigate pain compression syndrome with different clinical symptoms, forty patients were enrolled in a double-blinded, randomized study. The study was comprised of identification, treatment (14 days) and evaluation stages. Computerized randomization split the patients who began the primary MEB+MT treatment into two groups: Group 1=MEB (including steroid) + MT; Group 2=MEB (without steroid) + MT. Every patient received MEB aseptically (lidocaine 0.5% 20.0 - 40.0) weekly (up to 2-3 medication blockades) along with 10 MT sessions that were differentially delivered depending on intervertebral disc hernia location.

Assessment incorporated a Visual Analogue Scale (VAS) with data evaluated on the 1st, 4th, 8th and 14th days. Quality of the pain syndrome was estimated according to McGill's questionnaire. The effectiveness rate of influence on continued pain sensations during the period of treatment was assessed according to AUC (area-under-curve) data with covariate analysis and the original parameter of VAS as a covariate. Mann-Whitney's U-test was used to evaluate the effectiveness of the treatment and the follow-up period. Comparable and variable quantities were evaluated using non-parameter methods.

RESULTS: Of 58 patients included in the study, 28 patients (48%) made up group 1 and 30 patients (52%) made up group 2. No great difference was noted in age between the groups ($p>0.05$).

At all times, pain syndrome intensity was statistically decreased in both groups ($p<0.05$) compared to initial levels. The data were credibly distinguished however in 2-hour and 6-hour times ($p<0.001$), then on the 4th, 7th and 8th days ($p<0.01$). According to AUC, the steroid MEB applied to group 1 in conjunction with MT were more effective compared to group 2 treatments for the first 8-9 hours ($p<0.05$) and during 7-day period of treatment ($p<0.01$).

CONCLUSION: The therapeutic effect of the steroid epidural blockade in conjunction with MT sets in gradually and is preserved for two weeks (average two blockades/course).

C29

Diagnostics and Manual Therapy of Minor Pectoral Muscle Syndrome

N. Yeliseev, MD, PhD, I. N. Sharapov, MD, PhD, V. V. Belyakov, MD, PhD, The Manual Therapy Center of Kaluga Region, Obninsk, Russia

Hypothesis: Clinically-diagnosed Minor Pectoral Muscle Syndrome (MPMS) patients have abnormalities in Ultrasound Dopplerographic (UD) and Electroneuromyographic (ENMG)

parameters that improve with Manual Therapy (MT).
Methods: Upper extremity ENMG and UD parameters were followed in 54 clinically-diagnosed MPMS patients complaining of constant anterior thorax pain worse with arm/body movement, palpatory tenderness at origin/insertion and/or trigger point sites of the minor pectoral muscle (MPM), and MPM hypertonus. MPM shortening/tension tests created clinically-apparent brachial plexus, clavicular artery/vein compression.
All received MT consisting of 6-8 sessions (over 1-3 days) of spinopelvic mobilization-manipulation; postisometric relaxation muscle-energy and counterstrain to anterior chest wall (serratus anterior, pectoralis major and minor) muscles; and visceral (pericardiac, cervical pleura, and thoracoabdominopelvic diaphragmatic) techniques.
Results: Considerable clinical improvement (decreased pain zone intensity/area, increased shoulder range-of-motion, decreased vegetative disorders in the injured arm) was documented during MT care of 50/54 MPMS patients (92.6%).
UD demonstrated ($p<0.05$) arteriolar increase of linear blood flow velocity (LBFV) on the symptomatic side (3.3±1.58 cm/sec ⧫ 5.2±1.4 cm/sec). Also, radial artery LBFV (at rest and in shoulder adduction) increased (9.4±3.3 cm/sec ⧫ 12.2±2.4 cm/sec and 8.1±2.5 cm/sec ⧫ 11.7±3.1 cm/sec respectively; $p<0.05$).
On ENMG, bilateral amplitude decreases and latent period increases (especially on ipsilateral pain side) were documented; conduction block (CB) at Erb's point was also seen related to vegetative asymmetry ($p<0.05$). Comparing patients with asymmetric generated skin-sympathetic amplitudes to those without conduction asymmetry in postganglionic sympathetic fibers, CB-related measures were 58.0⧫1.24% vs. 76.65⧫1.04% (median nerve) and 60.37⧫1.06 vs. 71.77⧫7.05% (ulnar nerve).
Discussion and Conclusion: ENMG and UD evidence support compression of the axillary artery, proximal segments of the median and ulnar nerves, and/or postganglionic sympathetic fibers of the dysfunctional MPM as a significant pathogenetic mechanism causing clinical polymorphism in MPMS. Differentiated, pathogenetically-substantiated MT delivered to restore musculo-neuro-vascular relationships and reduce neurovascular bundle compression under the MPM was objectively reflected in positive UD, ENMG, and clinical results.

Basic Sciences

B1

Promotion of Lipoxidation of Human Serum Albumin by Solvent Hydrophobicty

H. D. Craig, BA, N. W. Seidler, PhD, Department of Biochemistry, Kansas City University of Medicine and Biosciences, Kansas, MO

Acrolein-dependent protein modification is implicated in the etiology of atherosclerosis. Serum components, such as low density lipoprotein and albumin, are modified by acrolein, recognized by the scavenger receptor on macrophages and detected in atherosclerotic lesions. The factors that control deleterious modification of albumin by acrolein are not well defined. We tested whether trifluoroethanol (TFE), which alters proximal solvent hydrophobicity, affects albumin's susceptibility to acrolein modification using human serum albumin (HSA). HSA (15-100 μM) was incubated in a 5.0 mM sodium phosphate (pH 7.4) buffer containing various concentrations of ACR and TFE at room temperature unless otherwise indicated. Sample agitation included continuous vortexing (speed: approx. 300 rpm), platform rocking (speed: approx. 80% max.) or constant stirring with magnetic stir bars (speed: approx. 60% max.). Following incubation samples were analyzed using size exclusion chromatography, SDS-PAGE, tryptophan anisotropy and intensity readings. Addition of acrolein brought about the formation of chemical adducts referred to as advanced lipoxidation endproducts (ALEs). TFE as low as 10% increased the formation of acrolein-induced fluorescence adducts on HSA. With 40% TFE we observed a 5-fold increase in ALEs compared to the effects of acrolein alone. Acrolein also increased tryptophan anisotropy in HSA, suggesting that adduct formation affects protein mobility. TFE (40%) further increased HSA anisotropy. These observations suggest that solvent hydrophobicity of TFE promotes lipooxidation of HSA which may play a role in the pathogenesis of atherosclerosis.

⧫B2

Morphological Features of the Nuclei of the Trapezoid Body in Human

Randy J Kulesza Ph.D., B. R. Trale, BS, Lake Erie College of Osteopathic Medicine, Erie, PA

The superior olive is a group of nuclei within the auditory pathway that functions in multiple aspects of hearing. The SOC displays interspecies variability, but is composed of 2 principal nuclei and multiple periolivary nuclei. Most ascending axons reaching the SOC arrive via the trapezoid body (tb), a bundle of axons within the pons. Within the tb, nuclei are situated along the anterior border of the SOC and these are collectively termed the nuclei of the trapezoid body (NTB). The NTB have been studied in lab animals and include 3 cell groups: medial, ventral and lateral, that functions in sound localization and modulation of the cochlea. Despite these important functions, the NTB have yet to be studied in human. The **hypothesis** is that because of the reduced hearing range in humans, the NTB will exhibit a significant reduction in volume and neuronal number as compared to lab animals.
Methods: This investigation is based on the study of 8 brainstems (55 to 90 years of age). For cytoarchitecture, brains were cryoprotected, sectioned at a thickness of 40 um on a freezing microtome and stained for Nissl substance. For silver impregnation, 3 mm thick blocks were incubated in mordant, washed and incubated in silver nitrate under a

⧫Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

vacuum of 5 inches of Hg. Neuronal profiles were traced using a camera lucida and analyzed using ImageJ. Statistical analyses were made using Microsoft Excel.
Results: Examination of tissue from all specimens reveals the presence of 3 distinct cell groups. Notably, the medial nucleus is much reduced compared to other species and in human includes only 3,600 neurons. The lateral nucleus is by far the largest and includes 7,500 neurons. The ventral group is the smallest of the NTB and includes 1,400 neurons.
Conclusions: This work provides evidence supporting the existence of NTB within the human SOC. Notably, the number of neurons is in stark contrast to what has been reported in rodents. More in-depth studies are required before functional homology between the SOC in human and laboratory animals can be can be stated.

B3

Thymidylate Synthase Expression affects Global DNA Methylation

G. Siu, PhD, OMSIV, [1] J. Alisangco, OMSIII[2], [1]UMDNJ-SOM, Stratford, NJ, [2]Philadelphia College of Osteopathic Medicine, Philadelphia, PA

Background: DNA methylation plays an important role in epigenetic modification after replication in humans, in which the methylation helps regulate gene transcription. However, DNA methylation changes (hypo- or hypermethylation) can induce genetic instability and eventually lead to tumorigenesis. These methylation changes are frequently found in cancer cells and correlate with specific stages of tumor progression. Global DNA hypomethylation could affect growth-promoting proto-oncogenes, induction of regional alterations in DNA conformation and chromatin structure, and accessibility to DNA-damaging agents such as oxidants and endonucleases. All of these effects would lead to tumor progression and ultimately carcinogenesis. Many enzymes and substrates affect the status of DNA methylation, including those in the methionine regeneration and folic acid metabolism pathways. One of the enzymes that may play a role in DNA methylation is thymidylate synthase (TS). TS is a key enzyme involved in the nucleotide biosynthetic pathway, yielding the only source of *de novo* thymidylate production in the cell. Since TS requires a methyl donor co-factor, its activity may indirectly influence the availability of methyl groups that are required for DNA methylation reactions. Therefore, differences in TS expression may help to maintain DNA methylation and high TS expression could lead to DNA hypomethylation.
Objective: To investigate the role of TS expression on DNA methylation.
Methods: DNA methylation was determined in cell lines with either low or high TS expression over a two-week period by using a tetracycline inducible gene expression system. At different time intervals, genomic DNA was isolated and a radioactive methyl acceptance assay was used to determine DNA methylation.
Results and Conclusions: The results showed that cell lines with high TS expression lead to a global DNA hypomethylation, while methylation status was unchanged in low TS expressed cell lines. Here, we demonstrated that high TS expression can lead to global DNA hypomethylation in TS-inducible cell lines, suggesting that individuals with high TS expression may be more prone to tumorigenesis.

♦B4

The Effect of the Marine Toxin Domoic Acid on Hippocampal c-FOS Expression and Microglia Accumulation in Rates

M. T. Braddock OMS-2[1], L. A. Carlson, MS[2], M. L. Hall, MS[2], P. F. McCulloch[3], A. M. Mayer, PhD[2], [1]Midwestern University-CCOM, [2]Pharmacology, Midwestern University-CCOM, [3]Physiology, Midwestern University-CCOM, Downers Grove, IL

Introduction: Domoic acid (DA) is a marine neurotoxin that may cause Amnesic Shellfish Poisoning in humans by binding to ionotropic glutamate receptors in the hippocampus, an area responsible for learning and memory processing. We investigated neuronal c-FOS protein expression and macrophages/microglia (BMΦ) distribution in the hippocampus of rats treated with DA.
Hypothesis: We hypothesized that differential behavioral responses, c-FOS expression and BMΦ distribution in the CA 3 region of the hippocampus would be observed in rats injected with either DA or saline.
Methods: Twelve rats were injected with either 2.2, 3.3 mg/kg DA or saline (control) intraperitoneally and behavioral responses were observed over a 3 hour period. A DA specific ELISA was used to quantify DA in rat serum. Neuronal c-FOS protein was determined by immunohistochemistry and BMΦ CD11b by immunofluorescence.
Results: Rats receiving either 2.2 or 3.3 mg/kg DA showed significantly different behavioral responses when compared to controls which correlated with serum levels of DA: 2.2 mg/kg DA and 3.3 mg/kg DA, >10,000 pg/ml (n=5). A statistically significant increase in the number of CA 3 hippocampal c-FOS positive neurons per coronal section was observed in DA-treated rats when compared to controls: Saline, 2.9±1.3 (n=4); 2.2 mg/kg DA, 101.5±50 (n=4, $p<0.05$); 3.3 mg/kg DA, 220.8±88.6 (n=4, $p<0.05$). In contrast, no difference in BMΦ distribution was observed: mean BMΦ per 1,000 DAPI-stained nuclei; Saline, 23.7±4.2 BMΦ (n=3); 2.2 mg/kg DA, 23.2±1.1 BMΦ (n=3); 3.3 mg/kg DA, 26.6±7.1 BMΦ (n=3).
Conclusions: Our data provides partial support to our working hypothesis by demonstrating that during a 3 hour observation period; DA significantly affected rat behavioral responses and hippocampal c-FOS protein in a concentration- and time-dependent manner, but did not result in changes in BMΦ distribution. Supported by NIH (AMSM) and the Biomedical

♦ Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

Sciences Program, College of Health Sciences, Midwestern University.

♦B5

Escherichia coli Chemotaxis and Adherence: Response Modulation by Insulin and Carbohydrates

R. F. Martin, BA, K. Balla, MS, B. J. Plotkin, PhD., Microbiology and Immunology, Midwestern University/CCOM, Downers Grove, IL

Hypothesis: *Escherichia coli* interact with both cellular and inanimate surfaces. Human insulin modulates the response of *Escherichia coli* to glucose. We hypothesize that human insulin acts as a signal surrogate in the bacterial decision making response to various carbohydrates, thus regulating the transition from planktonic to sessile states. To test this hypothesis we measured the effect of insulin on chemotaxis (planktonic state) and adherence (sessile state).

Materials and Methods: *E. coli* ATCC 25923 was grown in peptone (1%) yeast nitrogen base broth. For chemotaxis, cells in buffer containing insulin (2, 4, 20 and 200 mU mL$^{\varnothing 1}$) with and without maltose (10^{-3}M), mannose (10^{-3}M), arabinose (10^{-1}M), lactose (10^{-3}M), galactose (10^{-3}M), or fructose (10^{-2}M) were tested. Adherence to plastic was measured in microtiter plates after growth in homologous media and detected by measuring crystal violet absorbance (595nm). Adherence to latex (10^3 CFU/ml; 30min; 37°C) was determined using 7mm latex squares and quantified by the press plate method. Controls were media or buffer alone. Glucose (1%) served as the positive control.

Results: The response to lactose was reversed in the presence of 20 and 200mU mL^{-1} insulin. Response to the other sugars was unaffected by insulin. Insulin's effect on adherence to plastic and latex was both sugar and insulin concentration dependent. Insulin (20 and 200mU mL^{-1}) at most of the sugar concentrations tested (8 concentrations representing 2 fold serial dilutions of peak chemoattractant concentration) inhibited adherence to plastic with the exception of mannose that, like glucose, enhanced adherence as compared to the sugar-only control.

Conclusion: These findings show that human insulin can modulate *E. coli*'s response to sugars in addition to glucose in a manner that affects expression of factors associated with pathogenesis, i.e., motility and adherence.

♦B6

Expression Profile of Bag-1

C. Curcio, BS, B. H. Hallas, PhD, G. Torres, PhD, Department of Neuroscience, New York College of Osteopathic Medicine/NYIT, Old Westbury, NY

Hypothesis: Bag 1 is a protein intimately involved in signaling pathways that regulate cell survival and cell death. Bag 1 associates with heat shock 70 (Hsp70), presumably to assist molecular chaperones with the removal of aberrant proteins. There is currently much interest in psychiatric research involving Bag 1, as this protein is a long term target for the actions of mood stabilizers in bipolar depression. Accordingly, it can be valuable to determine effects of postmortem factors on Bag 1 if autopsy-derived human specimens are used in psychiatric research. We examined the expression profile of Bag 1 in the brain to consider issues associated with the sampling of anti-apoptotic proteins in a rat model of the human postmortem process.

Materials & Methods: Adult male Long-Evans rats were used in this study. All animal procedures were carried out in accordance with the NIH Guide for the Care and Use of Laboratory Animals, and with approval from the NYCOM/NYIT IACUC. Rats were injected with a lethal dose of choral hydrate and left undisturbed at storage temperatures of either 4 degrees C or 24 degrees C. Following a 4h postmortem interval, brains were removed and immediately placed in fixative solution. We analyzed coronal sections via immunocytochemistry with Bag 1 antibodies. For Western blotting, hippocampal samples were homogenized, electrophoretically transferred to nitrocellulose membranes and incubated with Bag 1 antibodies.

Results & Conclusion: Postmortem tissue (up to 4h) showed a significant and prominent up-regulation of Bag 1 in CA1 and CA3 subfields of the hippocampal formation. Over-expression of Bag 1, however, could only be traced down to a storage temperature of 24 degrees C. These data suggest that storage temperatures, but not postmortem intervals, significantly affect the expression profile and cellular stability of Bag 1 proteins. The cause of this differential sensitivity to storage temperatures is unknown, but may stem from the fact that postmortem phenomena trigger the expression of anti-apoptotic proteins to maintain cell viability. Thus it is imperative to control for storage temperatures in postmortem brain studies.

♦B7

The Antioxidant Enzyme Paraoxonase 1 (PON-1) is Inhibited by Acrolein, a Major Component of Cigarette Smoke

L. Tran, MD, PhD[1], A. Gugliucci, PhD[2], J. Schulze, BS[2], [1]Dept. of Biochemistry, TUCOM-CA, [2]Dept. of Basic Sciences, TUCOM-CA Vallejo, CA

Background: Paraoxonase 1 is an esterase carried by HDL particles with antiatherogenic functions. Acrolein is a very reactive dicarbonyl that is a terminal product of lipoperoxidation and one of the most concentrated substances in tobacco smoke. Acrolein readily forms adducts with lysine and oxidizes cysteine.

Hypothesis: We tested the hypothesis that acrolein acts on HDL to decrease PON-1 activity in a time and concentration dependent fashion.

Design and Methods: Fresh human serum HDL (1 mg/ml) was incubated under sterile conditions in PBS, pH 7.4, containing increasing concentrations of acrolein (0-10 mM), at 37°

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

C for 0-4 h. Some of the incubations included acrolein at 0.5 mM and carnosine or aminoguanidine (glycation inhibitors) or cysteine (all at 0.25-1 mM). PON-1 activity toward paraoxon was measured after the reaction of paraoxon hydrolysis into p-nitrophenol and diethylphosphate. Samples were also screened for fragmentation and polymerization by SDS-PAGE.
Results: Acrolein produces a time and concentration dependent decrease in PON-1 activity in HDL, (IC 50=1 mM) at only 2 h of incubation. At 0.5 mM, an activity loss of 40% is already accompanied by detectable dimerization of apoA-I. At the same concentration carnosine and aminoguanidine do not significantly protect the activity whereas cysteine affords 95% protection ($p<0.001$).
Conclusions: The results suggest that in conditions where circulating acrolein levels are high, or where local acrolein concentrations are high (atheroma plaque, sites of lipoperoxidation), acrolein-mediated loss of PON-1 activity could be a plausible phenomenon.

♦B8

A Method for the Evaluation of Site-Specific Nephrotoxic Injury in the Intact Rat Kidney Using Ethidium Homodimer

M. V. Kowal, BS, OMS-1, P. C. Lamar, BS, W.C. Prozialeck, PhD, J. R. Edwards, PhD, E. A. Diamantakos, MBS, OMS-1. Department of Pharmacology, Chicago College of Osteopathic Medicine, Downers Grove, IL

Background: As part of our ongoing work to identify mechanisms of cadmium (Cd)-induced renal toxicity, we have developed a novel method for assessing the viability of tubular epithelial cells in the intact kidney. This assay, which involves the *in situ* perfusion of the kidney with ethidium homodimer, has been used to label necrotic cells in the proximal tubule.
Objective: The objective of the present study was to determine if this method could be used to identify necrotic cells in other segments of the nephron following toxic injury.
Methods: Adult male Sprague-Dawley rats were treated with the site-specific nephrotoxicants, gentamicin (proximal tubule; 100 mg/kg i.p. per day for 8 consecutive days), amphotericin B (distal tubule; 15 mg/Kg i.p. for 5 consecutive days), and indomethacin (papillus; 20 mg/kg oral gavage, single dose). At the end of the respective treatment periods, ethidium homodimer (5 μM) was infused (3 ml/min for 10 min) into the left intact kidney while the animal was anesthetized. The kidney was then removed, placed in embedding medium, frozen, cryosectioned at a thickness of 5μm and ethidium-labeled cells were quantified, using fluorescent microscopy.
Results: Gentamicin treatment resulted in the death of cells in the proximal tubule. Renal cell death resulting from indomethacin treatment was localized primarily in the papillus. However, the kidneys of the indomethacin-treated animals also showed modest evidence of cell death in the cortex and medulla. Finally, amphotericin B caused necrosis of distal tubule epithelial cells.
Conclusion: These results show that this *in situ* viability technique is a sensitive and accurate assay to identify cellular necrosis in different segments of the nephron following exposure to nephrotoxic substances. Supported by Grant R01 ES006478 from the National Institute of Environmental Health Sciences.

♦B9

Mechanism of Action of N-Terminal Domain of Beta-Catenin on Optic Axon Branching

M. Mora, BS, A. Wiley, BS, MPH, T. Elul, PhD, Touro University College of Osteopathic Medicine, Vallejo, CA

Cadherin and Wnt signaling are key molecular pathways involved in the establishment of axonal connectivity in the developing visual system. β-catenin, a cytoplasmic adaptor protein, is a component of both Cadherin and Wnt signaling pathways. The N-terminal domain of β-catenin contains interaction sites for α-catenin (required for Cad adhesion) and GSK3β (required for Wnt signaling). We previously showed that overexpression of NTERM in optic axons in live *Xenopus* tadpoles reduces the number of terminal arbor branches. Here we address the molecular and cellular mechanisms of action of NTERM on branching in optic axons. First, we show that overexpression of a truncated NTERM that contains only the GSK3β binding domain does not reduce branch number in optic axonal arbors. Second, with timelapse confocal imaging, we show that NTERM reduces branch number in optic axonal arbors by inhibiting dynamic extension of new branches. These data suggest that the α-catenin binding domain of β-catenin normally promotes branching in optic axonal arbors by controlling the extension of new branches.

♦B10

Does Suboptimal Activation of System B0,+ Activity Influence Placentation and Long-Term Development Through Adulthood in Mice?

C. Yuen To, OMS, A. Campione, MS, K Cordero, B.S, BA, L. J. Van Winkle, PhD, Dept. of Biochemistry, Midwestern University, Downers Grove, IL

Hypothesis: System B0,+ is thought to play a major role in the penetration stage of implantation. Less than optimal activity of this amino acid transport system may result in suboptimal implantation, placentation and birth weight. Previous studies involved two groups of ICR mice; one group (E mice) received isoleucine in their drinking water in order to attempt to inhibit system B0,+ during the peri-implantation period of development, while the other served as a control (C mice) and received regular water. Offspring born to E mice on Day 19 of pregnancy were larger than pups born to C mice, whereas the reverse was true on Day 20. Fetuses apparently continued to grow between Day 19 and Day 20 in C mice but not in E mice. Thus, off-

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

spring born to C mice on Day 20 had greater birth weights than any other group of offspring born on either Day 19 or Day 20. To investigate further the underlying mechanism of these findings, the following study was performed.

M & M: Altered placental morphology is thought to be among the possible causes of low birth weight in E mice. We studied placental weight on both Day 15 and Day 18 of pregnancy (Day 1=Day of copulation plug detection) in 2 groups of mice: Ile treated drinking water (from Day 2-10, E mice) vs. regular drinking water (C mice). Daily food and water intake in both groups of mice were recorded and analyzed for possible correlation with data collected during delivery. A total of about 40 pregnant mice were studied (about 10 mice per group). The pregnant mice were delivered by caesarian section on Day 15 or Day 18 and the weights of whole conceptuses, placentas and fetuses were determined.

Results: Analysis of the data collected leads us into two principle findings. Firstly, fetal weight was positively correlated with placental weight in both C and E mice on Day 15 ($p<0.001$), but this correlation was lost selectively in E mice by Day 18. Placentas in E mice were smaller and fetuses were larger than normal on Day 18 of pregnancy, but the reverse was true on Day 15 ($p<0.01$). Secondly, conceptuses ruptured more easily than normal in E mice on day 18 of pregnancy possibly owing to greater fragility of fetal membrane ($p<0.01$).

Conclusion: These results support the theory that the placenta and other extraembryonic tissues are altered, but the placenta appears to adapt in E mice to support a more or less normal fetal growth rate at least until day 18 of pregnancy.

♦B11

Protein Kinase C (PKC) Isoform Peptide Activators (+)/Inhibitors (-) Attenuates Polymorphonuclear Leukocyte (PMN)-induced Myocardial Ischemia/Reperfusion (MI/R) Injury

H. Y. Kay MS, OMS III, J. Teng, MS, OMS II, J. Adams, MS, OMS III, N. Atkinson, MS, C Grilli, OMS IV, G. Guglielmello, OMS IV, R. Brue, MS, DO, A. Bell, OMS-IV, M. Dawley, MS, D. Omiyi, MS, P. Taormina II, DO, Q. Chen, PhD, L. H. Young, PhD, Philadelphia College of Osteopathic Medicine, Philadelphia, PA

MI/R in the presence of PMNs results in sustained cardiac contractile dysfunction. Attenuating PMN superoxide (SO) release or enhancing endothelial nitric oxide (NO) reduces endothelial dysfunction and inhibits PMN infiltration during reperfusion. PKC regulates PMN SO release and endothelial NO release/intracellular adhesion molecule expression (ICAM-1). Selective PKC isoforms mediating these responses are not well understood.

Hypothesis: Selective PKC isoforms will attenuate PMN-induced MI/R injury when given at reperfusion in isolated perfused rat hearts via attenuating PMN SO release/infiltration or increasing NO bioavailability.

Methods: Rat hearts were subjected to I/R and reperfused with PMNs in the presence/absence of PKC peptides (delta (δ+/-), epsilon (ϵ+/-), and beta II+zeta (βII-/ζ-), Genemed Synthesis, San Francisco, CA) during the first 5 min of reperfusion. Cardiac function/histology, PMN SO release, and endothelial NO/ICAM-1 expression were evaluated.

Results: PKC βII-/ζ-, δ+/-, and ϵ- significantly restored post-reperfusion cardiac function when given at reperfusion; however, PKC ϵ+ was cardioprotective only when given prior to ischemia (preconditioning/pretreatment). Significant restoration of post-reperfusion cardiac function was associated with decreased PMN infiltration in all studies ($p<0.05$). PKC ϵ- significantly attenuated post-reperfusion ICAM-1 expression ($p<0.01$). PKC βII-/ζ-, δ-, and ϵ+ significantly increased endothelial NO release ($p<0.05$), while PKC βII-/ζ- and δ+ significantly decreased PMN SO release ($p<0.05$). PKC ϵ+/- had no effect on PMN SO release due to its absence in PMNs.

Conclusion: These data indicate that PKC ϵ plays a dual role in attenuating MI/R injury. PKC ϵ+ is cardioprotective in preconditioning I/R, which may be related to increased endothelial NO release prior to ischemia. PKC ϵ- is cardioprotective during reperfusion following acute ischemia, which may be related to decreased ICAM-1 expression. Although PKC δ+ restores cardiac function via inhibited PMN SO release, PKC δ- does so via increased endothelial NO release. PKC βII-/ζ- restores cardiac function by NO and SO mechanisms.

Acknowledgment: Supported by PCOM CCDA and NIH/NHLBI Grant 1R15HL-76235-01 to LHY.

♦B12

Chondrocyte-Produced TGFb Modulates Early Osteoblast Differentiation at the Chondro-Osseous Border

V. L. Scheinfeld, OMS-III,[1] S. E. Day, OMS-II,[1] B. N. Dragann, OMS-I,[1] E. L. Bray, OMS-III,[1] P. V. Bodine, PhD,[2] M. D'Angelo, PhD,[1] [1]Center for Chronic Disorders of Aging, Philadelphia College of Osteopathic Medicine Philadelphia, PA, [2]Women's Health Research Institute, Wyeth Research, Collegeville, PA

We have shown that hypertrophic chondrocytes produce large quantities of activated TGFβ; however, the purpose of this activated TGFβ remains elusive. During endochondral ossification, it is possible that this TGFβ regulates the differentiation of osteoblasts that come into contact with the soluble factor at the chondro-osseous border. We isolated early (EH) and late (LH) hypertrophic chondrocytes from the avian sterna and placed them in serum free alginate culture. After five days in culture, conditioned media (CM) was removed and placed over early mature osteoblasts (HOB-03-CE6; EMOB) for 6 hours. Total RNA was isolated, reverse-transcription PCR performed and PCR product expression normalized to 18s rRNA. Osteocalcin (OC), a noncollagenous bone matrix protein, was examined as a selective marker of osteoblast differentiation. OC expression was significantly inhibited after treat-

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

ment with EH CM and inclusion of a pan-specific neutralizing antibody to TGFβ resulted in a return of OC expression to control levels. These data suggest that TGFβ present in the EH CM is inhibiting osteoblast differentiation. Since neutralizing antibody activity can be transient we depleted the CM of TGFβ by passage over a strep-avidin micro-bead column containing biotin-labeled polyclonal antibody to TGFβ. Following TGFβ depletion of EH CM, OC expression again returned to control levels. In contrast, CM overlay of EMOBs with LH CM had no effect on OC expression. When neutralizing antibody to TGFβ or TGFβ depleted LH CM was added to the EMOB cultures, a statistically significant inhibition of OC expression was seen. These data indicate that chondrocyte-produced TGFβ has some role in inhibiting osteoblast differentiation, though it may also be involved in regulating other factors present in the CM. Since large amounts of activated TGFβ are present in LH CM as compared to EH CM, the TGFβ modulation of other soluble factors could potentially be dose related.

♦B13

Role of Cutaneous Chlamydia pneumoniae Infection in the Pathogenesis of Mycosis Fungoides

M. S. Lewars, MS[1], C J Hammond, MS[1], C. Angel, DO[2], V. Nozad, DO[2], G. Severs, DO[2], T. Griffin, MD[3], B. J. Balin, PhD[1], [1]Dept. of Pathology/Microbiology & Immunology-CCDA, Philadelphia College of Osteopathic Medicine, [2]Dept. of Dermatology, Frankford Hospital/PCOM, [3]Dept. of Dermatology, Drexel University, Philadelphia, PA

Background: Mycosis fungoides (MF) represents the most common type of cutaneous T-cell lymphoma (CTCL) accounting for ~50% of all primary cutaneous lymphomas with an incidence of 1 in 300,000 per year. MF is a chronic, slowly progressive disorder that typically begins as patches and may transform into plaques and/or tumors. Persistent or chronic infection has been proposed as a chronic stimulator promoting clonal expansion of epidermotropic T-cells. A previous study has implicated *Chlamydia pneumoniae* (Cpn), a common respiratory pathogen, as a chronic pathogenic stimulus in CTCL (Abrams et al, 1999). Further, Cpn has been implicated in other chronic disorders including reactive arthritis, meningoencephalitis, atherosclerosis, and Alzheimer's disease.

Hypothesis: Cpn infection in the skin provides a chronic stimulus that promotes epidermotropic lymphoproliferation, which results in mycosis fungoides.

Materials and methods: Archival paraffin-embedded skin specimens obtained from punch biopsy of lesional sites from 17 MF patients and comparable sites from age-matched controls were sectioned and immunolabeled for Chlamydia to determine the presence or absence of Cpn antigens.

Results: Typical punctate Chlamydial immunolabeling was found in the dermis of 10/17 (59%) MF skin samples. In addition, there was labeling observed in monocytes and macrophages as well as perivascular and periadnexal labeling. Interestingly, the extent of anti-Chlamydial labeling was inversely proportional to the extent of the dermal cellular inflammatory response. Minimal immunoreactivity was observed in the most highly proliferative cases as compared to the mild to moderate cases.

Conclusion: Our data suggest that the presence of Cpn in the skin may act as a chronic stimulus promoting lymphoproliferation leading to MF. Thus, linkage of Cpn with the pathogenesis of mycosis fungoides will have major implications with respect to therapeutic options.

B14

An Evaluation of New Integration Algorithms for High-Throughput Chromatography

J. A. Price PhD[1], C. Sanny, PhD[2], [1]Dept. of Pathology, OSU-COM, [2]Dept. of Biochemistry & Microbiology, OSU-COM, Tulsa, OK

Hypothesis: The new integration algorithms may produce the accurate automatic peak integration needed for high throughput chromatography, avoiding slow manual *ad hoc* analysis, and incidentally may accurately reveal the quantitative loss of peaks with higher wavelength detection, previously an evaluation by subjective manual integration editing.

Materials and Methods: We used the newest chromatographic software for analyzing peak capacity to ascertain the actual loss of detection of peaks as a function of wavelength of detection with data from a complex RP-HPLC separation of proteins and small peptides. PDA data was extracted and analyzed using default settings for each algorithm and/or product.

Results and Discussion: Poor results were obtained from a traditional integration (Waters), or default assisted curve fitting by PeakFit in any mode. An improvement was seen with Galaxie Chromatography software. The best result was by Innovations Chromperfect® showing 186 peaks at 214nm, and 79 peaks at 280nm in default mode. By comparison, the Waters Corp. Traditional algorithm has revealed at 214nm 42% of the peaks found by ApexTrack, and 31% with shoulder detection engaged. 58 of 169 peaks were lost from 214nm to 230nm, the detection limit for some mobile phases. Removing small peaks below 3 sigma of baseline had little effect on results.

In summary, for this type sample and separation, the ApexTrack and particularly Chromperfect® integration technologies not only proved substantial enhancements over the traditional integration algorithms but can be used on this type data with little or no modification.

♦B15

Mdm2 Gene Regulation by Vitamin D in Osteoblasts

R. C. Cruz OMS-2, K. Cordero, MS, N. Chandar, PhD, Dept. of Biochemistry, Midwestern University-CCOM, Downers Grove, IL

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

Hypothesis: In previous studies our laboratory has demonstrated a role for p53 tumor suppressor gene in osteoblast differentiation. Mdm2 protooncogene is a p53 regulated gene, whose targeted reduction in bone was found to result in bone pathology. In this study we attempted to determine a role for mdm2 in osteoblast differentiation independent of p53 by studying its promoter activation by bone anabolic agent vitamin D3 in osteoblasts of differing p53 status.
Materials and Methods: Using transient transfections and luciferase assays we studied the effect of vitamin D3 on the two mdm2 promoters in osteoblast cells with differing p53 status.
Results and Conclusion: Mdm2 promoters P1 (p53 independent) and T2 (p53 dependent) were found to be active in osteoblasts under normal conditions. A time dependent increase in both promoter activities was seen with vitamin D treatment. Maximal activity occurred after two hours in both promoters. Exogenous wild type p53 activated mdm2-T2 promoter and addition of vitaminD3 caused a synergistic increase in activity. This effect was seen in both p53 positive and negative cells. These studies suggest that mdm2 gene may be regulated in osteoblasts under physiological conditions by vitamin D in a p53 dependent and independent manner.

B16

The Effects of Tetrahydrobiopterin (BH4) and Dihydrobiopterin (BH2) on Nitric Oxide (NO) and Hydrogen Peroxide (H2O2) Release during Femoral Vein Ischemia/Reperfusion (I/R) by Real-time Measurement

Q. Chen PhD[1], J. J. Wood, LATg [2], L. H. Young, PhD[1], [1]Dept. of Pathology, Microbiology and Immunology, PCOM, [2]Laboratory Animal Resources, Philadelphia College of Osteopathic Medicine, Philadelphia, PA

Background: The oxidative stress (i.e., NO decreases and superoxide (SO) increases) following I/R is a major cause of endothelial dysfunction during reperfusion. It is well-known that BH_4 is a required co-factor for the synthesis of NO from L-arginine by endothelial NO synthase (eNOS). However, when BH_4 is oxidized to BH_2, under oxidative stress, such as reperfusion injury, the product profile of eNOS can shift from producing NO to SO. SO is further converted to H_2O_2 by superoxide dismutase. The effects of BH_4 and BH_2 on NO and H_2O_2 release have not been directly estimated by real-time measuring in blood vessels during I/R in vivo.
Hypothesis: BH_4 may increase NO release whereas BH_2 may increase H_2O_2 release in blood vessels following I/R compared to the sham control.
Methods: NO and H_2O_2 were directly measured by inserting either NO or H_2O_2 microsensors (100 μm diameter) into both femoral veins in the anesthetized rat. One femoral vein was subjected to I/R, which was induced by clamping both femoral artery and vein for 20 min ischemia followed by removing the clamp for 45 min reperfusion. The other femoral vein served as sham control not subjected to I/R in the same animal.
Results: NO release increased briefly in the femoral vein subjected to I/R at the beginning of reperfusion, but quickly came back to the sham control level throughout the rest reperfusion. By contrast, BH_4 (200 μM) given i.v. through the tail vein at the beginning of reperfusion showed an increase in NO release throughout the entire reperfusion. Moreover, H_2O_2 release increased throughout the first 20 min of reperfusion in the femoral vein subjected to I/R compared to the sham control. Whereas, BH_2 (100 μM) given i.v. at the beginning of reperfusion showed an increase in H_2O_2 release throughout the entire reperfusion.
Conclusion: The preliminary data thus far support our hypothesis that BH_4 can increase NO release and BH_2 can increase H_2O_2 release during reperfusion in femoral veins.
Acknowledgment: Supported by PCOM CCDA and NIH/NHLBI Grant 1R15HL-76235-01 to LHY.

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

♦B17

P53 and p300 during Osteoblast Differentiation and Regulation of the Osteocalcin Gene

S. Chaudhari, DO, K. Codero, BS, N. Chandar, PhD, Dept. of Biochemistry, Midwestern University-CCOM, Downers Grove, IL

We have previously shown p53 to have a role in osteoblast differentiation. P300 is a coactivator that regulates p53 activity. While p300 is known to be important for tissue differentiation, its mechanism of action is not well understood. In this study we followed p300 expression during osteoblast differentiation and determined its effect on the regulation of a bone specific, differentiation related gene osteocalcin.
We used an invitro model of osteoblast differentiation where ROS17/2.8 osteosarcoma cells were exposed to a differentiation promoting media for different lengths of time from 0-8 days. Western blots were used to determine levels of p300 and p53 produced at different stages of osteoblast differentiation. Using transient transfections, we studied the effect of p300 on the osteocalcin promoter using reporter assays.
Results and Conclusion: We were able to see a time dependent increase in osteocalcin promoter activity during differentiation. On long term treatment, these cells mineralized in culture and calcified matrix was quantitated using Alizarin Red S staining. There was an increase in p300 levels during differentiation and is followed by an increase in p53 levels. In transient transfections with the 882bp of the osteocalcin promoter, the presence of p53 increased the promoter activity. Addition of p300 with p53 resulted in a synergistic increase in activity. Our results demonstrate a role for p300 in osteoblast differentiation and in the regulation of the osteocalcin gene.

♦B18

Regulation of the Osteocalcin Promoter by p53 and mdm2

J. Liboon BS, BA, K. Cordero, BS, K. Kolman, BS, N. Chandar,

PhD, Department of Biochemistry, Midwestern University-CCOM, Downers Grove, IL

Hypothesis: We have previously shown p53 gene to play a role in osteoblast differentiation. Mdm2 is a p53 regulated protein and is known to be important in the regulation of p53 stability. In this study we worked on the hypothesis that p53 directly regulates the osteocalcin gene and that mdm2 has a p53 independent role in this regulation.

Materials and Methods: We used osteoblast cell culture and transient transfections with an 882bp osteocalcin promoter region to show the role of p53 and mdm2 in the regulation of the osteocalcin gene. In addition, we employed the DNA affinity immunoblotting (DAI) assay to demonstrate direct binding of p53 to the osteocalcin promoter region.

Results and Conclusion: We found p53 to increase osteocalcin promoter activity several fold by itself. P53 and mdm2 resulted in a synergistic increase in osteocalcin activity when compared to either protein alone. We were able to show the binding of p53 on the osteocalcin promoter using DAI assay. This complex also consisted of mdm2 and coactivator protein p300. The activation of osteocalcin gene by mdm2 was unique to this gene and contradictory to its role in the regulation of the p53 gene. We therefore determined the effect of mdm2 alone on the osteocalcin promoter and found it to regulate this gene independent of p53. Addition of vitamin D to the transient transfections increased the promoter activity several fold. We believe that the osteocalcin gene is regulated by a multiprotein complex that is made up of several proteins some of which include p53, mdm2, p300, vitamin D receptor and Cbfa1.

♦B19

Variation in Appearance of Laminin Isoforms in Developing Skeletal Muscle Grown on a Synthetic Matrix

S. C. Richard BA, OMS-I, P. Mattioli, BA, E. O. Adah, BS, OMS-II, M. P. Hammock, BS, C. DiLullo, Ph.D., Department of Anatomy, Philadelphia College of Osteopathic Medicine, Philadelphia, PA

Alterations in laminin binding have been shown to be a causative factor in some forms of muscular dystrophy. Laminin, an extracellular matrix (ECM) component of the basement membrane in skeletal muscle, can act as a ligand for specific integrins which are heterodimeric, transmembrane proteins. The alpha 1/beta 1 integrin heterodimer can bind to laminin. Previous analysis of the alpha 1 subunit of integrin has indicated that it spatially reorganizes early in the development of cultured primary skeletal myocytes. Alpha 1 integrin reorganizes from a punctate distribution to one with a sarcomeric periodicity in young myocytes with sarcomeres already aligned into myofibrils. To determine if the reorganization of alpha 1 integrin could be correlated to the temporal and spatial distribution of laminin in skeletal muscle, various laminin isoforms were analyzed in developing skeletal muscle. Primary skeletal muscle from 10 day chick embryos was obtained according to an approved IACUC protocol. The muscle was cultured and plated into dishes containing a synthetic matrix insert which eliminated the need to coat the surface of the dish with an exogenous extracellular matrix. Cultures were fixed on days 3, 5, & 7 and immunofluorescently labeled with antibodies to alpha 1 integrin, several laminin isoforms and a number of muscle specific proteins. Data indicates that the different laminin isoforms were localized in distinct regions of the myocyte. A limited number of these isoforms were detected with a sarcomeric periodicity. A subset of the laminin isoforms that demonstrated a periodic distribution appeared to be co-localized with the periodic alpha 1 integrin. This correlation suggests a possible link between this specific cell membrane integrin and the basement membrane laminin. Further examination of these two proteins will be necessary to determine their potential role(s) in the developmental program of skeletal muscle and/or establishment of the skeletal muscle basement membrane.

♦B20

Effects of Bilobalide on the Conformation of Aβ

B. R. Odum, BA[1], G. J. Desai, DO[2], N. W. Seidler, PhD[3], [1]Kansas City University of Medicine and Biosciences, [2]Dept. of Family Medicine, KCUMB, [3]Dept. of Biochemistry, KCUMB, Kansas City, MO

Hypothesis: Alzheimer's disease (AD) involves the abnormal processing of the amyloid precursor protein (APP), a neuronal transmembrane protein. Proteolysis of APP yields a large extracellular and a small intracellular domain. The remaining middle peptide of 37-42 amino acids is the amyloid β peptide (Aβ). Conformational transitions form soluble neurotoxic oligomers. Most therapies inhibit the production of Aβ but with limited success. Complementary therapies that target other areas are desirable. Extracts of *Gingko biloba* show clinical utility; however, the mechanism is unclear. We looked at bilobalide, a major terpenoid, and its interaction with Aβ.

Methods: Conformation of Aβ was examined using fluorometric properties of fragment 1-42 with and without bilobalide. Fluorescence anisotropy of Tyr-10 was used to assess peptide mobility. Aggregate size was measured using right-angle light scattering (RALS). Center of spectral mass (CSM) was calculated to compare the hydration properties of the Aβ oligomers.

Results: When bilobalide was incubated with Aβ under continuous stirring, RALS was greater than control at 6 days. At a 100:1 molar ratio, bilobalide progressively increased RALS over 8 weeks. Bilobalide also increased Tyr anisotropy and decreased CSM.

Conclusions: These observations suggest that bilobalide increased Aβ aggregation, while decreasing peptide mobility and dehydration. Bilobalide's effect is likely due to its chemical properties: a tri-methyl group as well as multiple sites for

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

hydrogen-bonding. Features of bilobalide-Aβ interaction may lead to development of novel therapeutics for AD.

Acknowledgement: The project was supported by Grant Number 5 R25 AT000529-03 from the National Center for Complementary and Alternative Medicine.

B21

Low Dose Antibiotic Treatment of Lyme Disease in Young versus Old Mice

C. S. Pavia, PhD, N. L. Johansson, OMS, Biomedical Sciences, New York College of Osteopathic Medicine of NYIT, Old Westbury, NY

Hypothesis: It is still unclear what the minimally effective dosage is for the antibiotics recommended for use in curing Lyme disease (LD), and whether these dosages are age dependent. In recent related studies of ours, we investigated the efficacy of short course treatment with the antibiotic ceftriaxone (CTX) in vivo in a mouse infectivity model for LD. These results showed that 5 equally spaced doses of CTX, given over a 24-hr period, were as effective as the more standard 5-day treatment regimen with a single daily dose of CTX in eliminating viable *Borrelia burgdorferi* (Bb) from young mice infected with Bb. We hypothesize that by modifying this animal model and using elderly mice, it will be possible to determine minimally effective treatment regimens that could be applicable to geriatric patients who have contracted LD.

Methods: C3H female mice were assigned to a senescent group of 12 to 18 months of age or to a young group of 5 to 12 weeks of age. Four separate groups of C3H mice (5 per group) were infected intradermally with 100,000 culture-grown, low passage strain BL206 of Bb which had been previously isolated from the blood of a LD patient having early erythema migrans. Two to 4 weeks after infecting the mice, they received 2 intramuscular injections (given 18-24 hours apart) of either saline or CTX (50mg/kg). One to 2 weeks after treatment, cultures of the urinary bladder were established in BSK media in order to determine the presence or absence of Bb.

Results: It was found that the two-dosage regimen of CTX was 100% and equally effective in sterilizing the urinary bladders from the Bb-infected mice, irrespective of their ages.

Conclusions: These experiments suggest that shorter courses of antibiotics than those currently recommended should be considered for study in patients with early uncomplicated LD. Such prospective studies should also include test subjects in all age groups in order to fully evaluate any possible age-related response differences to antibiotic treatment regimens.

♦B22

Herpes Simplex Virus 1 and Chlamydophila (Chlamydia) pneumoniae Promote AB 1-42 Amyloid Processing in Murine Astrocytes Linking an Infectious Process to Alzheimer's Disease

L. R. Triplett MS[1], A. L. Dore, MSII[2], K. S. Kralik, BS[1], B. J. Balin, PhD[1], S. T. Hingley, PhD[1], D. M. Appelt, PhD[2], [1]Pathology, Microbiology, Immunology and Forensic, PCOM, [2]Neuroscience, Pharmacology, and Physiology, PCOM, Philadelphia, PA

Background: Several studies have suggested an infectious etiology for Alzheimer's disease (AD). Previously, our laboratory identified *Chlamydia pneumoniae* (Cpn) from autopsied sporadic AD brains, as well as developed a BALB/c mouse model that demonstrated infection-induced amyloid plaques similar to those found in AD.

Hypothesis: We propose that an additional pathogen such as herpes simplex virus type 1 (HSV1), also may be a factor in the pathology seen in AD. HSV1, in addition to Cpn, may be triggering abnormal cleavage of the beta amyloid precursor protein (BAPP) into AB1-42, thereby contributing to amyloid plaque formation. Our current study examines amyloid processing following infection of primary and C8-DIA murine astrocytes with Cpn and HSV1.

Materials and Methods: Immunocytochemistry was used to analyze the outcome of infection by these two pathogens.

Results: Cpn infection resulted in an increase in cytoplasmic labeling of AB 1-42 relative to uninfected cells, while increased nuclear labeling of AB 1-42 was observed following HSV1 infection. Co-infections with Cpn and HSV1 resulted in amyloid labeling resembling that of HSV1 infection alone, though AB 1-42 labeling appeared decreased specifically in Cpn-infected cells of the co-infected monolayers.

Conclusions: These data suggest that infection of astrocytic cells by Herpes Simplex Virus 1 and *Chlamydophila (Chlamydia) pneumoniae* alter the processing of BAPP, thereby producing AB1-42. Therefore, these studies, in addition to the previous research reported by our laboratory, support an emerging linkage of the infectious process to the neuropathology characteristic of Alzheimer's disease.

♦B23

Prevalence of borrelia burgdorferi in Black-Legged Ticks Inhabiting Presque Isle State Park

D. M. Brown BS, C. C. Keller, PhD, Lake Erie College of Osteopathic Medicine Erie, PA

Hypothesis: *Borrelia burgdorferi*, the causative agent of Lyme disease, is present in *Ixodes scapularis* ticks which inhabit Presque Isle State Park (Erie, PA).

Materials and Methods: Adult *Ixodes scapularis* ticks (n=69) were obtained from Presque Isle State Park in Erie, Pennsylvania in fall of 2006. Ticks were collected from suspected habitats using a flagging method. Ticks were stored in 75% ethanol until processed for DNA isolation. DNA was isolated by crushing ticks in a 5% Chelex solution followed by centrifuging for 7 min at 12,000g. The supernatant, which contained nucleic acids, was aspirated and transferred to a sterile microfuge tube. The presence of DNA in each sample was determined by amplifying *I. scapularis* 16S ribosomal genomic DNA by PCR.

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

Resulting fragments were separated by gel electrophoresis and visualized on 1% agarose gels containing ethidium bromide. All ticks which tested positive for 16S ribosomal tick DNA were examined for the presence of *B. burgdorferi*. Bacterial 16S ribosomal DNA was amplified using a *B. burgdorferi*-specific PCR. DNA fragments were analyzed by gel electrophoresis.

Results: DNA was successfully isolated from all ticks (n=69) as evidenced by specific amplification of *I. scapularis* 16S ribosomal DNA. PCR analysis demonstrated that 53.62% (37 of 69) of the ticks were positive for *B. burgdorferi* DNA. There were no significant differences in the *B. burgdorferi* infection rate between males and females, location of collection, or date of collection.

Conclusions: The results of the present study indicate that *B. burgdorferi*-infected adult *I. scapularis* ticks are present on Presque Isle, indicating that there is a potential risk of human *B. burgdorferi* infection for the estimated 4 million annual visitors to the park.

B24

Abdominal Lymphatic Pump Treatment Increases Leukocyte Count and Flux in Thoracic Duct Lymph

L. M. Hodge PhD[1], M. K. Bearden, BS[1], H. H. King, DO, PhD[2], A. G. Williams, Jr., BS[3], J. W. Simecka, PhD[1], S. T. Stoll, DO, PhD[2], H. F. Downey, PhD[3], [1]Molecular Biology and Immunology, UNTHSC at Fort Worth, [2]National Osteopathic Research Center, UNTHSC at Fort Worth, [3]Integrative Physiology, UNTHSC at Fort Worth

The lymphatic pump technique (LPT) was designed to improve lymph flow, reduce edema, and improve the body's immunological defenses to combat infections. Studies suggest that LPT enhance immunity and resistance to infection, but direct evidence of this has not been documented. To test the hypothesis that LPT enhances lymph flow and leukocyte concentrations in lymph, the immediate effect of LPT on lymph flow and leukocyte flux and in the canine thoracic lymph duct was measured. Lymph flow data and lymph samples were collected at baseline and at 2-3 min intervals during LPT. The baseline leukocyte count was 4.8±1.7 million cells/ml, and LPT increased leukocytes to 11.8±3.6 million cells/ml ($P<0.05$). Whereas numbers of macrophages, neutrophils, total lymphocytes, T cells, B cells, and IgG forming B cells increased similarly during LPT, their relative percentage in lymph was unaltered by LPT; however, IgA antibody forming B cells increased from 5.8% at baseline to 17% during LPT. This data suggest that LPT acts preferentially on mucosal tissues, a potential source of these mobilized leukocytes. Furthermore, LPT enhanced lymph flow approximately 4-fold. Leukocyte flux was computed from the product of lymph flow and cell count, and LPT enhanced leukocyte flux from 8.2±4.1 million leukocytes per min to 60±25 million leukocytes per min. The most important new finding of this investigation was that LPT produced large increases of leukocytes in lymph. Since LPT also increased lymph flow in the thoracic duct, the flux of leukocytes transported to the circulation through the thoracic duct was greatly increased. Furthermore, LPT increased IgA antibody forming B cells, suggesting LPT enhances the mobilization and lymphatic transport of mucosal immune cells during abdominal compression. It is likely that this mobilization of leukocytes is an important mechanism responsible for the increased immune responses of patients treated with LPT. Thus, the results of this investigation may provide a rational basis for the use of LPT to enhance immunological function and treat infection.

Funding for this project was provided by the National Institutes of Health (Grant No. P01 AT 2023).

B25

Lymphatic Pump Treatment Enhances Survival and Reduces Pulmonary Bacteria during Experimental Pneumonia Infection

L. M. Hodge PhD[1], M. D. Bellard, BS[1], M. K. Bearden, BS[1], J. W. Simecka, PhD[1], S. T. Stoll, DO, PhD[2], H. F. Downey, PhD[3], H. H. King, DO, PhD[2], [1]Molecular Biology and Immunology, UNTHSC at Fort Worth, [2]National Osteopathic Research Center, UNTHSC at Fort Worth, [3]Integrative Physiology, UNTHSC at Fort Worth

The purpose of this study was to develop an animal model to examine the mechanisms by which lymphatic pump treatment (LPT) enhances immunity and clears pulmonary infection. Clinical studies have shown that patients given LPT have enhanced clearance of the tracheobronchial tissues, greater production of sputum, shorter duration of cough, and shorter duration of total and intravenous antibiotic treatment and hospitalization. These studies suggest that LPT may reduce pulmonary infection by enhancing immunity and/or facilitating antibiotic delivery to pathogens; however, the exact mechanisms responsible for the clinical benefits of LPT have not been identified. To determine if LPT enhances survival and reduces pulmonary bacteria during acute infection, rats were divided into sham treatment (manual control, MC) or LPT groups and infected nasally with $1x10^8$ bacterial colony forming units (CFU). Approximately 2 hours following infection (day 0), and on days 1 and 2 and post-infection, rats received 8 minutes of either MC or LPT under anesthesia. Weights and survival were measured on days 0, 1, 2 and 3 post-infection. On day 3 post-infection, rats were euthanized and pulmonary bacteria were enumerated. LPT enhanced survival approximately 20% by day 3 post-infection compared to MC. The benefit of LPT on delaying weight loss during infection was only observed on day 2 post-infection. Furthermore, LPT reduced pulmonary bacteria approximately 30%, suggesting that innate immune mechanisms are enhanced by LPT and contribute to clearance of pulmonary infection. This study is the first to demonstrate that LPT can enhance survival and reduce pulmonary bacteria during acute pneumonia infection. Importantly, the use of animal models allows for collection of samples that cannot be obtained using human sub-

jects. These data are essential to identifying the mechanisms by which LPT enhances immunity and clearance of pneumonia. Understanding of these mechanisms will provide scientific support for the clinical use of LPT. Funding for this project was provided by the American Osteopathic Association (Grant No. 06-11-547).

♦B26

Effects of Oxidized Low Density Lipoprotein On Hsp70 Expression In The Cerebral Cortex Of Huntington's Disease Transgenic Mice

D. H. Lee OMS III[1], T. D. Hulbert, OMS III[1], J. Schulze, BS[3], K. D. Klapstein, PhD[2], T. Menini, MS, MD[2], G. J. Klapstein, PhD[2], [1]Touro University-California, [2]Basic Sciences
Touro University - California, [3]Health Sciences, Touro University - California

Background and Hypothesis: Huntington's Disease (HD) is a progressive neurodegenerative disorder characterized by changes in movement, behavior and cognition. One hallmarks of HD cellular pathology is huntingtin protein aggregates inclusions, intracellular and intranuclear. These inclusions co-localize with heat shock proteins (HSP), which are molecular chaperones expressed during a stress response. To test our hypothesis that HSP production and transport across cortical cell membranes is altered in HD, we measured intracellular Hsc70/Hsp70 (iHSP) and extracellular Hsp70 (eHSP) in response to an oxidative stressor in R6/2 transgenic HD mice at asymptomatic (18-25 days, young) and symptomatic (>80 days, old) ages and compared them with age matched littermate controls (WT).

Methods: Cerebral cortex from young and old HD and WT were microdissected and incubated in artificial cerebrospinal fluid with low-density lipoprotein (LDL, control) or oxidized-LDL (ox-LDL, stressor) for 24h. Samples were analyzed for iHSP (immunoblots) and eHSP (ELISA).

Results: Compared to control, ox-LDL decreased iHSP and increased eHSP in young WT. In young HD, ox-LDL had no effect on iHSP but decreased eHSP. Thus, ox-LDL treated HD tissue retained relatively more iHSP. In old WT and old HD mice, ox-LDL decreased iHSP and increased eHSP; however, there was no significant difference between old HD and old WT. As the WT mice age, there was no significant difference in ox-LDL effect on iHSP or eHSP. However, as HD mice age, ox-LDL significantly decreased iHSP and increased eHSP when compared to WT controls.

Conclusion: Young HD cortical cells may have decreased ability to transport HSP following exposure to an oxidative stress but were able to transport more HSP as they aged. HSP production is thought to be cytoprotective. Therefore, increased iHSP in young HD mice may indicate an early compensatory mechanism. Furthermore, the abnormal response to stress at asymptomatic ages may contribute to the aggregation of inclusions and the pathogenesis of HD.

Acknowledgment: AAN Medical Summer Student fellowship and AMA Seed Grant (DHL).

♦B27

Differential Host-Responses to RSV Infection in Neonatal and Adult Mice

R. Mishra, OMS-III, R. Merhi, MS, OMS-III, K. Durre, BSc, W. Yang, MD, PhD, M. Qureshi, MD, PhD, Touro University-Nevada, Henderson, NV

Patients suffering from RSV broncheolitis and pneumonia during infancy develop airway hyper-responsiveness (AHR) and wheezing in later life. A predominant Th2 response prevails in the respiratory tract of these individuals. Development of Th1 vs. Th2 responses largely depends on the nature of antigen and the type of antigen presenting cell (APC) involved. Dendritic cells (DC) are the major APC in the respiratory tract. In the present studies, using a BALB/C mouse model, we examined whether young infants respond differently to RSV infection than adults, which may explain their post-RSV -infection susceptibility to asthma. Flow cytometry revealed that the proportions of myeloid DC (mDCs; $CD11b^{lo}$ $CD11c^{hi/med}$ $CD45R^{-}$) to plasmacytoid DCs (pDCs; $CD11b^{lo}$ $CD11c^{lo}$ $CD45R^{+}$) were different in the pup and adult lungs. Using an in vitro system, we also determined the differential responses of neonatal and adult DCs to RSV infection in regard to expression of MHC and co-stimulatory (CD80, CD86) molecules. In addition, neonatal lung cells produced significantly lower amount of Th1 cytokines (INF-γ and TNF-α) and higher amounts of Th2 cytokines (IL-4, IL-10 and TGF-β) compared to that of adult lung cells in response to RSV infection. Thus, different proportions of mDCs to pDCs in the neonatal lungs and inefficient maturation of neonatal lung DCs (as observed by decreased expression of MHC and CD80 and CD86 in response to RSV), may underlie the mechanism of differential responses of neonates and adults to RSV as well as development of AHR and susceptibility to asthma.

This was sponsored by Touro University Nevada Research Grant Award #05-06/104.

B28

GROWTH INHIBITION OF RENAL CELL CARCINOMA BY CARBONIC ANHYDRASE INHIBITORS

W. R. Chegwidden PhD, FRSC, D. G. Linville, PhD, LECOM, Erie, PA

Renal cell carcinoma accounts for up to 3% of all adult malignancies, and in the US about 30,000 new cases are diagnosed each year. It tends to metastasize widely before producing local signs or symptoms, and is notable resistant to both radiotherapy and currently available chemotherapy.

Carbonic anhydrase (CA) isozymes have been implicated in the growth and invasion of human cancer cells. This may be through the provision of bicarbonate, which is necessary for the

♦ Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

synthesis of nucleotides and other cell components, and/or through enhancing elimination of the acid load generated by hypoxia in cancer cells. The CA IX isozyme is virtually specific to cancer cells and is considered to be a marker for renal cell carcinoma.

We have previously demonstrated inhibition of both growth and invasion of renal carcinoma cells in culture. We have now examined the effect of methazolamide (NEPTAZANE), a specific sulfonamide inhibitor of CA, on the growth of human renal carcinoma cells transplanted into immunodeficient ('nude') mice. Preliminary data demonstrated more than 80% inhibition of tumor growth rate by methazolamide (20 mg/kg injected I.P. on alternate days). These data suggest that carbonic anhydrase is a potential new target for cancer therapy.

The support of the Hess Roth Kaminsky Foundation is gratefully acknowledged.

♦B29

PUTATIVE MECHANISM OF STRESS-INDUCED NPY RELEASE: CATECHOLAMINERGIC-NEUROPEPTIDE Y (NPY)-IMMUNOREACTIVE ASSOCIATIONS IN THE HUMAN HYPOTHALAMUS

L. Ko OMS-III, B. Dudas, MD, PhD, Lake Erie College of Osteopathic Medicine, Erie, PA

Physical and emotional stress involves profound neuroendocrinological changes in the hypothalamus of several species including human. One of the major mediators of stress is believed to be a 36 amino acid peptide, neuropeptide-Y (NPY). Indeed, previous studies confirmed that NPY release is increased by stress in several species. However, the exact mechanism of the stress-induced NPY release is not known.

In our study, we utilized double label immunohistochemistry in order to examine our hypothesis, that catecholamines directly influence NPY release in the human hypothalamus via juxtapositions that may represent functional synapses. The present study is the first that revealed these juxtapositions between the catecholaminergic and NPY-immunoreactive (IR) neurons in human. The majority of these associations were *en passant* type, where catecholaminergic fibers labeled with tyrosine hydroxylase antiserum abutted on NPY-IR perikarya. These juxtapositions were most numerous in the infundibular and periventricular areas of the human diencephalon, where NPY-IR neurons often formed 4-5 contacts with catecholaminergic fiber varicosities.

Since close examination of the hypothalamic sections with oil immersion revealed no gap between these contacting elements, the juxtapositions described in the present study may represent functional synapses between the catecholaminergic and NPY-IR elements. Since catecholamines are known to be the crucial component of the stress response, the possibility of direct catecholaminergic-NPY-IR synapses may explain the stress-induced NPY release in human. The released NPY in turn is believed to play a major role in the stress response.

B30

In vitro Human Fibroblast Model of Repetitive Motion Strain (RMS) and Direct OMT (DOMT): Roles for Proliferation and Apoptosis

K. R. Meltzer MS[1], J. F. Schad, MS[2], H. King, DO, PhD[3], S. T. Stoll, DO, PhD[3], P. R. Standley, PhD[1], [1]Basic Medical Sciences, University of Arizona, Phoenix, AZ, [2]Biomedical Sciences, Midwestern University-College of Health Sciences, [3]Osteopathic Research Center, UNTHSC at Fort Worth, Fort Worth, TX

Background: Despite clinical efficacy, the cellular basis for osteopathic manipulative therapies (OMTs) is not well understood. Previously, we described an in vitro cellular strain model useful for exploring potential cellular mediators of OMT and modeled a repetitive motion strain injury and indirect OMT. In the current study, we further this work by investigating the effects of direct OMT (DOMT) with this same modeled injury using human fibroblasts (HF).

Hypothesis: We hypothesized that HF respond to RMS by manifesting cellular apoptosis, loss of intercellular matrix, and reduced proliferative capacity. Further, we hypothesize that modeled DOMT would reverse these effects.

Methods: HF was exposed to 1) 8 hrs RMS, 2) 60 seconds DOMT, or 3) both profiles RMS+DOMT. Samplings (microscopy, apoptosis, proliferation, protein microarrays) occurred immediately and 24 hr post treatment. A total of 5 experiments were performed.

Results: RMS resulted in elongation of lamellopodia, reduced intercellular contact area, reduced cell-matrix attachment points and disrupted actin architecture. Control and DOMT groups did not display this profile. RMS resulted in a 35% increase in apoptosis vs. control ($p \leq 0.05$), whereas the RMS+DOMT group displayed no apoptotic changes vs. control. BadS91 (serine 91-phosphorylated Bcl2-antagonist of cell death protein) expression was reduced by 45% in RMS vs. control HF, but no differences in BadS91 were noted when comparing RMS and RMS+DOMT groups. Similarly DAPK2 (death-associated protein kinase 2) was elevated in RMS vs. control, but no such changes were seen when comparing RMS and RMS+DOMT. While no differences in proliferative responses were seen among the control, 24 RMS, and 24 DOMT groups, the 24 RMS+DOMT group displayed an 18% increase in proliferation ($p \leq 0.05$).

Conclusion: Our modeled injury (RMS) appropriately displayed enhanced apoptosis activity (consistent with pro-apoptotic BadS91 and DAPK2 signaling) and loss of intercellular integrity. Added DOMT to this group resulted in normalization in apoptotic rate and enhanced proliferative capacity, albeit in a BadS91 and DAPK2-independent fashion. Future studies are ongoing to identify cellular signaling pathways responsible for the observed OMT-induced reversal of the

♦ Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

injurious phenotype. These in vitro studies build on the cellular evidence base needed to fully explain clinical efficacy of OMTs post-injury. Funding: NIH P-01 AT2023 (NCCAM)

♦B31

RSV-Infection Induced Differential Polarization of Pulmonary Dendritic Cells Contributes to Susceptibility to Asthma

R. Merhi MS, OMS III, R. Mishra, OMS III, K. Durre, BSc, W. Yang, MD, PhD, M. Qureshi, MD, PhD, Touro University-Nevada, Henderson, NV

RSV infection accounts for significant morbidity in young infants. The survivors become susceptible to develop asthma later in life. Studies have shown increased airway responsiveness in these individuals and a concomitant Th2 response in their lungs. However, the underlying mechanism (s) responsible for this pulmonary Th2 response in post- RSV infection period is not well understood. In this study, using a mouse model, we examined whether differentiation of lung dendritic cells (DCs) was skewed due to RSV infection and whether a Th2 cytokine response was associated. Neonatal mice were sensitized with intraperitoneal ovalabumin (ova) and infected with intranasal RSV followed by subsequent challenge with intranasal ova. Flowcytometric analysis of lung parenchymal cells determined the proportions of $CD11b^{lo}CD11c^{hi}CD45R^{-}$ myeloid DCs (mDCs) and $CD11b^{lo}$ $CD11c^{hi}$ $CD45R^{hi}$ plasmacytoid DCs (pDCs). An increase in the numbers of pDCs was observed in the lungs of RSV-infected as well as RSV-infected and ova-exposed mice. To mention, pDCs are reported to downregulate Th1 responses. Consistent to this, augmentation of Th2 cytokine responses was observed in the RSV-infected and ova-exposed mice. Thus, polarization of lung DCs towards pDC due to RSV infection may modulate lung cytokine responses leading to susceptibility to asthma in later life. Reversal or inhibition of this skewed polarization may be considered as a clinical strategy in the post-RSV infection asthma. *This work was sponsored by Touro University Nevada Research Grant Award #05-06/104.*

♦B32

Assessing the Effects of Opioid Inverse Agonists and Neutral Antagonists In Vivo

John J Lowery OMSIII, T J Raymond, OMSI, H N Martinez, BS, E J Bilsky, PhD
UNECOM, Biddeford, ME

Context: We have characterized the classic Opioid antagonist's naloxone and naltrexone as inverse agonists in the Opioid dependent state, whereas structurally similar analogs, such as 6β-naltrexol, exhibit neutral antagonist properties in both naïve and Opioid dependent states. To further evaluate the pharmacology of these antagonists we (a) performed a more complete in vivo pA2 analysis to determine the relative potencies of naltrexone, naloxone, 6β-naltrexol, and naloxone methiodide and (b) completed respiration studies that assessed the effects of Opioid agonists and antagonists on respiration under Opioid dependent conditions.

Hypothesis: It was hypothesized that neutral antagonists (e.g., 6β-naltrexol) would differentiate themselves from inverse agonists (e.g. naltrexone) in terms of antagonist potencies to reverse agonist effects and precipitate withdrawal.

Methods: Male ICR mice were used to estimate the antagonist potencies of naltrexone, naloxone, 6β-naltrexol and naloxone methiodide. Dose-response curves for morphine-induced locomotor activity were rightward shifted with increasing doses of each antagonist using an in vivo pA2 approach. In addition, each antagonist was tested for precipitation of Opioid withdrawal using respiratory parameters as an endpoint. Lastly, the contributions of both central and peripheral Opioid receptors in Opioid-mediated respiratory depression were evaluated.

Results: All antagonists produced a dose-dependant antagonism of morphine-induced hyperlocomotion. 6β-naltrexol was approximately 12-fold less potent than the classic Opioid antagonist naltrexone. In Opioid dependant mice, naltrexone, naloxone and 6β-naltrexol produced dose-dependent increases in respiratory rate and minute ventilation with 6β-naltrexol being approximately 70-fold less potent than naltrexone in this assay. In addition, the peripherally selective Opioid antagonist naloxone methiodide dose-dependently increased these respiratory parameters suggesting that there is a peripheral component to Opioid withdrawal that impacts measures of respiration.

Conclusion: The data further supports the hypothesis that neutral antagonists may offer a greater therapeutic index (e.g. a less severe withdrawal) compared to naloxone and naltrexone in procedures involving reversal of Opioid agonist effects, especially in Opioid dependent patients.

♦B33

Pharmacological Characterization of the Novel Phenylethylmorphan ACH.1.132: A Putative Opioid Mu Agonist/Delta Antagonist

T. J. Raymond OMS-I[1], D. N. Giuvelis, BS[1], J. J. Lowery, OMS-III[1], A. C. Hiebel, PhD[2], K Cheng, PhD[2], A. E. Jacobson, PhD[2], K. C. Rice, PhD[2], E. J. Bilsky, PhD[1], [1]UNECOM, Biddeford, Maine, [2]National Institutes of Health, Bethesda, MD

Context: Current Opioid therapy for the treatment of moderate to severe pain is associated with significant side effects that limit their clinical utility. These side effects include the development of tolerance, physical dependence and addiction liability. It has been proposed that inhibition of δ Opioid receptor function during μ agonist stimulation may attenuate the development of these adverse effects while maintaining desired analgesic effects. The focus of our research has been to synthesize a single chemical entity that has both mu agonist and delta antagonist properties (e.g., ACH.1.132).

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

Hypothesis: Our working hypothesis is that compounds that simultaneously stimulate μ receptors and antagonize δ receptors will produce antinociception with fewer side effects compared to morphine.
Methods: Male ICR mice (25-35 g) were used to test the antinociceptive effects of ACH.1.132 and morphine in the 55℃ tail flick test and the acetic acid abdominal constriction assay. In vivo receptor selectivity was determined by pre-treating mice with μ (β-FNA), δ (naltrindole), and κ (nor-BNI) selective antagonists. Mice were also evaluated for acute and chronic physical dependence using naloxone to precipitate withdrawal. Lastly, the development of tolerance was examined using repeated injections of the respective agonists for three days and determining potencies of the agonists in the 55℃ tail flick test.
Results: ACH.1.132 was found to be approximately 50-times more potent than morphine in the 55℃ tail flick test and 10-times more potent in the abdominal constriction assay. The primary site of action for these effects was μ Opioid receptors in the CNS. Similar levels of acute and chronic dependence were found between ACH.1.132 and morphine; however ACH.1.132 produced significantly less tolerance.
Conclusion: ACH.1.132 produced dose-related antinociceptive effects, being more potent than morphine with similar efficacy. ACH.1.132 produced equivalent levels of physical dependence compared to morphine and less tolerance. The results are discussed in the context of developing an ideal ratio of μ agonist/δ antagonist activity and the dissociation of Opioid tolerance from physical dependence.

♦B34

Assignment of Copyright Permissions Pending

♦B35

Analysis of Chlamydophila pneumoniae and AD-like Pathology in the Brains of BALB/c Mice Following Intranasal Infection

T. Joyce BS, C. J. Hammond, MS, D. M. Appelt, PhD, B. J. Balin, PhD, C. S. Little, PhD, Pathology, Microbiology, & Immunology/CCDA, PCOM, Philadelphia, PA

Background and Hypothesis: This work continues previous studies examining the role of Chlamydophila pneumoniae (Cpn) in the induction/progression of Alzheimer's disease (AD)-like pathology in a non-transgenic mouse. BALB/c mice were inoculated with Cpn to model AD-like pathology, and presents an exemplary mode of pathogenesis possibly implicated in AD. The objective of this work is to identify Cpn antigens in the brains of BALB/c mice infected with Cpn, and to determine if Cpn localizes near the hippocampus, amygdala, and entorhinal cortex, regions of the brain that display AD-like pathology. These areas are frequently involved with AD pathology in humans and our current working hypothesis is that Cpn antigens will be present in or near regions that display AD-like pathology.
Materials and Methods: BALB/c mice were infected, intranasally, with a respiratory isolate of Cpn (AR-39) and brain and olfactory bulbs were isolated following perfusion at 1, 2, 3, or 4 months post-infection. Serial sections from brains of experimentally infected or mock-infected age and sex matched mice were analyzed via immuno-histochemistry using antibodies (monoclonal IgG) specific for amyloid or the Cpn epitopes: LPS, major OMP, and purified elementary bodies.
Results and Conclusions: Chlamydia-specific labeling was noted, in the brains of AR-39 infected mice, and was most prominent at 1 month p.i., which was prior to substantial amyloid deposition. Amyloid-specific labeling was prominent at and after 2 months p.i., but did not consistently co-localize with Chlamydia antigen. Our initial data suggest that Chlamydia infection, although initially established in the brains of BALB/c mice, is limited, and may serve as a chronic stimulus for inflammation in the brain.

♦B36

IL-8 Production by Cytokine-Stimulated Human Iris Stromal Cells

G. L. Searle BS[1], B. W. Zanotti, BS[1], M. D. Silverman, PhD[2], M. V. Volin, PhD[1], [1]Microbiology and Immunology, Midwest University-CCOM, Downers Grove, IL, [2]Internal Medicine University of Michigan Medical School, Ann Arbor, MI

Hypothesis: Acute anterior uveitis is distinguished by the presence of leukocytes and inflammatory exudates in the anterior chamber of the eye. Several chemokines have been identified in the aqueous humor of anterior uveitis patients. One of these chemokines, IL-8, plays an important role in neutrophil recruitment during inflammatory processes. We hypothesize that human iris stromal cells are impart responsible for this production of IL-8 in the anterior chamber of eyes with uveitis. To test this hypothesis we performed in vitro studies in order to determine the production of IL-8 by human iris stromal cells stimulated with cytokines present in anterior uveitis.
Materials and Methods: Human iris stromal cells were isolated from donor eyes and grown in culture. These cells were stimulated with TNFα and IFNγ (10ng/ml) for 3 or 24 hours. Antibody arrays were used to detect production of 37 different chemokines including IL-8. Additionally, a specific IL-8 ELISA was used to quantify the IL-8 levels in conditioned media from cytokine-stimulated human iris stromal cells.
Results: Antibody arrays showed increased levels of several chemokines, including IL-8, in the cytokine-stimulated human iris stromal cell media. A specific IL-8 ELISA confirmed these findings, showing significantly increased release of IL-8 in the TNFα-stimulated human iris stromal cells stimulated for 24 hours compared to non-stimulated cells (4.4 ng/ml compared to 0.03 ng/ml, $p<0.05$, n=3). However, human iris stromal

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

cells stimulated with IFNγ showed no significant change. Combined stimulation with both INFγ and TNFα also showed significant increases in IL-8 production at both 3 and 24 hours (0.9 and 6.8 ng/ml compared to 0.03 and 0.03 ng/ml respectively, $p<0.05$, n=3).

Conclusion: These results show that cytokine-stimulated human iris stromal cells produce IL-8 and therefore may potentially play a role in neutrophil recruitment into the anterior segment of the eye in anterior uveitis.

B37

Uterine Artery Vasoconstriction During the Onset of Exercise in Gravid and Nongravid Uterine Horns in Rabbit Pregnancy

J. Schriefer, AB, R. Murphy, MS, K O'Hagan, PhD, Midwestern University-CCOM, Downers Grove, IL

The normal physiological response to steady-state exercise in non-pregnant rabbits includes lowered blood flow and uterine artery conductance through an alpha-adrenergic mediated vasoconstriction of the uterine arteries. During pregnancy, this response to exercise is attenuated. Recently, we have shown that this attenuation is largely dependent on the presence of fetoplacental units, rather than circulating factors associated with the pregnant state. However, the response to the initiation of exercise had not been examined. Our hypothesis was that an initial vasoconstrictor response is neurally-mediated and will be blocked by an alpha-adrenoreceptor antagonist. Uterine artery conductance and blood flow (transit-time ultrasound flowprobe) and arterial blood pressure were measured in four female rabbits during the non-pregnant state and at days 10, 20 and 28 (term) of pregnancy. Measurements were made in both the gravid and non gravid uterine horn at rest, during exercise, and with a pharmacologic alpha-adrenoreceptor blockade. The first minute of exercise was analyzed using a general linear model with repeated measures. The analysis showed that the vasoconstrictor response to the first minute of exercise was attenuated in the gravid uterine horn at early, middle, and term pregnancy. This suggests the presence of fetoplacental units affects the constrictor response. Also, during mid and late gestation, the dynamic response to exercise was altered in both uterine horns with the peak response occurring earlier. This effect was the most pronounced in the gravid horn. Lastly, the alpha-adrenoreceptor block reduced the vasoconstrictor response at all stages of gestation and in both uterine horns. This suggests that alpha-mediated tone persists throughout pregnancy in the gravid horn. These results suggest that the vasoconstrictor response to the initiation of exercise exhibits subtle changes with advancing gestation that may be related to the presence of the fetoplacental unit.

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

B38

Tamm Horsfall Protein (Uromodulin) is Resistant to Glycation and Nitration

A. Gugliucci, MD, PhD[1], M. Rose, OMS-III[2], L. Tran, OMS-III[2], [1]Dept. of Biochemistry, [2]Dept. of Basic Sciences, Touro University-CA, Vallejo, CA

Background: Tamm Horsfall protein (THP) or uromodulin is the most abundant urinary protein. As an extracellular protein it could be submitted to carbonyl or oxidative attack, especially in conditions such as diabetes.

Hypothesis: We tested the hypothesis that THP evolved to become resistant to carbonyl and nitrosative modifications as compared to serum proteins.

Design and Methods: THP was isolated from human urine by salting out, extensively dialyzed against water and its purity confirmed by SDS-PAGE and silver staining. THP (.2 mg/ml), HDL (.2 mg/ml) and albumin (.2 mg/ml) were incubated under sterile conditions in PBS, pH, 7.4, containing increasing concentrations of acrolein (0-10 mM), methylglyoxal (MG, 0-10 mM) and SIN-1 (0-100 mM), at 37° C for 0-24 h. Acrolein is a carbonyl compound produced in lipid peroxidation and pollution smoke, MG is a key dicarbonyl in Maillard chemistry in humans and SIN-1 is a peroxynitrite generator. After extensive dialysis, the samples were screened for fragmentation and polymerization by SDS-PAGE

Results: At carbonyl concentrations of 1 mM HDL and albumin exhibit dimerization and polymerization. SIN-1 produces extensive fragmentation of HDL apolipoproteins and albumin at 10 mM whereas these changes are minimal for THP.

Conclusions: This is the first study to focus on in vitro Maillard and nitrosative modifications of THP. We show that THP is more resistant to these modifications, even under very stringent conditions, than 2 abundant serum proteins. Its high glycan content (more than 30%) as well as its particular primary and spatial structure should play a role in this resistance.

♦B39

Up-regulation of Arginase II mRNA levels in Human Gingival Fibroblasts

E. Maksimak, OMS-I, F. Daghigh, PhD, Dept. of Biochemistry, PCOM, Phildelphia, Pa

Background Availability of arginine during periodontitis may be regulated by arginase activity. Arginase expression appears to be essential in regulation of cell immune responses and inflammatory processes during periodontitis, a disease of the gingival tissue. Arginase may also regulate arginine availability for nitric oxide (NO) synthesis. Excessive NO is associated with tissue destruction and inflammation. Arginine is a common substrate of both iNOS and arginase. In addition to urea production, the role of arginase includes production of ornithine for synthesis of polyamines and proline required for cell proliferation and collagen formation. Upregulation of arginase is speculated to decrease arginine availability for iNOS and it may downregulate NO overproduction. Proin-

flammatory cytokines have been found to up-regulate iNOS in human gingival fibroblasts (HGF) but regulation of arginase in this tissue type has yet to be determined. Mammals express two isoforms of arginase, arginase I (ARGI) and arginase II (ARGII). ARGI is a component of the urea cycle while the role of ARGII is not completely understood.

Hypothesis Presence of ARGII in HGF may provide a source of polyamines for collagen synthesis and cell proliferation. It may also modulate the inflammatory response in periodontitis, limiting NO production by iNOS and exerting an antiinflammatory effect.

Materials/Methods HGF were collected from patients undergoing periodontal surgery; cells underwent 3-5 passages. HGF incubated with cytokines mimicking proinflammatory and antiinflammatory conditions: IFNγ, LPS, cAMP, and IL4 and combinations of IFNγ+LPS, IFNγ+cAMP, cAMP+LPS and IL4+cAMP over time points of 6, 12, 18, 24, and 36 hr. Real Time-PCR was used to measure RNA levels.

Results ARGII was constitutively expressed in HGF. cAMP upregulated the mRNA levels of ARGII significantly at 12 and 36 hr of incubation. Proinflammatory cytokine, IFNγ, was previously found to upregulate iNOS in HGF, it inhibited the ARGII basal mRNA levels highly significantly at 12 and 36 hr. IFNγ abolished upregulation of ARGII mRNA by cAMP after 36hr incubation. LPS attenuated the basal ARGII mRNA and blocked the effects of cAMP highly significantly at 12hr, but not at 36hr.

Conclusions ARGII and iNOS are regulated differentially and may implicate a role for ARGII in collagen synthesis and cell regeneration.

Medical Education

♦M1

Taxation and Obesity Policy: Implementation of a Novel Tax on Foods Based on Nutrition Content

B. E. Goldstein MPH, OMS-III, A. Moradi, MD, MPH, Departments of Family Medicine and Public Health, Lake Erie College of Osteopathic Medicine, Bradenton, FL

Obesity is a growing public health problem that has left legislators perplexed over how to deal with this problem adequately and cost-effectively from a policy-making standpoint. One debated approach is the use of a "snack tax." Previous snack tax efforts, or efforts to increase the price of unhealthy foods with the hope of decreasing demand, have been largely unsuccessful due to complexity in implementation, lack of evidence-based data, and backlash from lobbies and interest groups. In short, it has been difficult for legislators to decipher which foods should be subjected to these taxes. This study attempted to alleviate some of these concerns by gathering data on the efficacy of a novel taxation formula based on nutrition content of foods rather than using a flat-rate approach. Protein, carbohydrate, and fat content of twenty-five popular food items were gathered and converted into a nutrition index and taxation scheme such that foods with higher fat and carbohydrate content, as well as lower protein content, yielded higher taxation amounts. Five-hundred individuals were recruited for the study. Participants were given a questionnaire that analyzed demographic variables, stages of behavior change, and food choices given three levels of taxation. Results showed that the use of a nutrition-based taxation scheme effectively altered consumer choices. One-way ANOVA analyses showed that carbohydrate and fat content of food choices decreased ($p<0.05$) while protein content of food choices increased ($p<0.05$). Thus, this study demonstrates that the use of an equitable and objective food taxation scheme can effectively alter consumer choices, and it can simultaneously reduce lobbyist backlash and legislative pitfalls associated with flat-rate discriminatory approaches. This research will provide a steady backbone for future research on snack tax legislation.

♦M2

The Value of Positron Emission Tomography in Biopsy Site Selection

Briana Beauchene Medical Student, J Neil, MD, R Newbold, MD, R Korn, MD, A Irving, BS, Scottsdale Medical Imaging, Scottsdale, AZ

Objective: The objective of this retrospective study was to determine the added value of PET/CT upon the decision making process of the interventional radiologist when determining the most appropriate site and/or lesion to biopsy using minimally invasive techniques.

Materials and Methods: Fifty patients were selected who had a tissue biopsy performed between 01/01/2003 and 12/31/2005 and who had a PET/CT and diagnostic CT performed within 30 days of each other. The CT, PET, and PET/CT fused images for each patient were interpreted by two interventional radiologists. Data recorded for each lesion included location, shape, composition, size, and SUV (for PET and PET/CT exams). Of the recorded lesions, the interventionalists were asked to select one lesion for biopsy, and explain the reasons for their selection.

Results: Radiologist A and radiologist B identified a total of 132 and 122 lesions on PET/CT, 116 and 109 lesions on PET, and 116 and 113 lesions on CT respectively. The results showed that in the cases where more than one lesion was seen, radiologist A and B chose a different lesion to biopsy based on the PETCT versus the PET 24.2% and 32.35% of the time respectively. They also chose a different lesion to biopsy based on the PET versus CT 33.33% (A) and 42.42% (B) of the time. When the radiologists looked at the PET/CT versus the CT their decision of what lesion to biopsy changed 40.63% (A) and 44.12% (B) of the time.

Conclusion: In summary PET/CT rather than CT does change the decision making process of the interventional radiologist

♦ Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

when determining the most appropriate site and/or lesion to biopsy. The addition of PET/CT gives a radiologist a more precise visual of where each lesion is located, which lesion is the most metabolically active, easiest to access, and safest to biopsy.

M3

Applicant Selection Criteria for Osteopathic Residency Programs

G. O. Ballam, PhD[1], V. J. Guillory, DO, MPH, FACPM[2], [1]Dept. of Psychology, [2]Division of Research, Kansas City University of Medicine & Biosciences, Kansas City, MO

The number of osteopathic residency program openings in the US is competitive and somewhat limited. It is therefore to the benefit of residency program applicants to be aware of the absolute and relative importance of the criteria used to evaluate them in order to maximize their probability of a successful match. To provide information regarding these criteria and their importance, a survey was developed and distributed to all osteopathic residency program directors in the specialties of family medicine, internal medicine, pediatrics, obstetrics/gynecology, orthopedics and surgery. Of 157 surveys distributed, 98 (62%) were completed and returned. Questions on the survey were divided into the areas of academic accomplishment, personal/professional characteristics and professional recommendations. Respondents were asked to rate each item as to its importance in considering an application using a 5 point scale from unimportant to critical. Every survey response in the personal characteristic category such as importance of empathy/compassion, with the exception of a letter of recommendation from a clinical faculty within the hospital and specialty to which the applicant is applying, ranked higher than all other responses in both other categories. All scores for the personal characteristics category ranked between the categories of very important and critical. The top ranked attributes were compatibility with the program followed by commitment to hard work and personal interview. Professional recommendations were the second most important category. The importance of academic criteria followed that of professional recommendations with the most important being required clerkship grades the highest and rated just under very important. In summary, it appears personal attributes, which are best demonstrated by direct contact with those involved in the residency program to which the graduate is applying, are the most important for consideration into a residency program. Although also very relevant, professional recommendations and academic criteria are of lesser importance.

M4

Factors Contributing to Osteopathic Manipulation Usage at Army Family Medicine Residency Programs

K. E. Platz DO, C. A. Asplund, MD, Department of Family and Community Medicine
Eisenhower Army Medical Center, Fort Gordon, GA

Hypothesis: Army Family Medicine Residency Programs with a philosophy conducive to Osteopathic Manipulative Treatment (OMT) have less barriers to performing OMT and increased usage.

Material and Methods: After IRB approval, each osteopathic resident currently enrolled in one of the seven Army Family Medicine Residency Programs was sent an email requesting voluntary participation in an online survey. Programs were divided into two groups based on resident perception of whether their program had a philosophical environment conducive to OMT, as measured by a five-point Likert scale.

Results: 70% (36/53) of the residents completed the survey. When comparing programs reported as OMT-conducive residency programs to less conducive programs, 56% (10/18) versus 21% (4/15) [p=0.031] had adequate access to designated treatment tables, 72% (13/18) versus 33% (6/18) [p=0.019] participated in monthly OMT clinics, and 100% (14/14) versus 14% (2/14) [$p \leq 0.01$] had multiple osteopathic lectures and labs in the last academic year. 89% (16/18) of the OMT promoting programs' residents reported using manipulation at least monthly versus 58% (11/19) [p=0.034] at other programs. 72% (13/18) of the residents in the programs that support OMT consider manipulation usage as a determining factor in their match ranking versus 26% (5/19) [p=0.05] at non-OMT supporting residencies. Residents at non-conducive programs stated they would increase OMT usage if there were more OMT tables, clinics, and lectures/labs (74% [14/19], 79% [15/19], and 89% [17/19] respectively). 100% of residents stated they would like to improve their manipulation skills.

Conclusions: There is an association between programs whose residents report a philosophy conducive to OMT and the usage of osteopathic manipulation. Programs with a less conducive environment could easily remove the identified barriers to increase OMT usage. Residency programs, both allopathic and osteopathic, have an incentive to increase OMT use to assist with recruitment of osteopathic medical students into Family Medicine Residencies.

♦M5

Awareness of Health Disparities in Organ Donation among Osteopathic Medical Students

D. M. Burch OMS-IV, A. R. Gordon, OMS III, P. A. Burnett, Ph.D., Center of Excellence for Multicultural Medicine, Ohio University College of Osteopathic Medicine, Toledo, OH

Introduction: Close to 100,000 Americans are on waiting lists for an organ transplant; nearly half are from minority populations. The shortage of transplantable organs has a significant effect on the mortality of African Americans. Despite sponsored campaigns to recruit minority organ donors; donation rate increases have been marginal. Physicians can play a key role in educating African Americans about organ donation. We hypothesize that osteopathic medical students are not

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

aware of the cultural barriers African Americans face regarding organ donation and transplantation.
Methodology: This IRB-approved, prospective study targeted first- and second-year medical students at Ohio University College of Osteopathic Medicine. All registered students received a 22-question survey. Due to small class size, consent was limited to self-selection. The survey contained 4 subscales: awareness of organ donation barriers; perceived attitudes to organ donation; knowledge of needs and demands for organ donation; and, sensitivity to cultural/racial differences regarding organ donation. Chi square analysis was performed; significance was set at the 5% level.
Results: The survey was completed by 156 students (66 male - 43%;90 female - 57%): African Americans 24 (15%), Caucasians 111 (71%), Asians 11 (7%), Hispanics 5 (3%), Native Americans 1 (<1%), Other 4 (3%). Of the participants, 65% believe racial/ethnic differences were not barriers to successful organ transplantation (p=0.0001); 69% fear the medical community may take their organs before offering life saving treatment (p=0.0001); 78% mistrust the medical community (p=0.0001);67% fear unequal distributions of organs to non-minorities (p=0.0001).Students identified these reasons why African Americans may be less likely to become organ donors; however, some students (63%) believe disruption of the body's integrity (p=0.0001) and superstitions/religious beliefs (59%) were not reasons. Only 10% of participants scored 80% or above on the total knowledge of the needs and demands for organ donation among African Americans (p=0.0001).
Conclusion: Osteopathic medical students in this study are not fully aware of cultural barriers faced by African Americans regarding organ donation and transplantation. If students become aware of this dilemma, they will be better equipped to educate these patients and, thus, help to reduce health disparities that African Americans encounter in organ donation and transplantation.

♦M6

Attitudes towards Osteopathic Manipulation in Career Choices (ATOMICC)

Brett D Dunbar BS, MS4, G J Desai, DO, Dept. of Family Medicine, Dept. of Family, Medicine & Medical Affairs, Kansas City University of Medicine and Biosciences, Kansas City, MO

Hypothesis: The investigators surmise that osteopathic medical students at Kansas City University of Medicine and Biosciences (KCUMB) will be more likely to use Osteopathic Manipulative Treatment (OMT) in the future if: their 1st choice of medical school was osteopathic, they practice OMT outside of class/clinical, and they plan on entering primary care. The investigators also hypothesize those students who observe preceptors using OMT will be more likely to use OMT themselves.
Materials and Methods: With IRB approval, all KCUMB students (approx 950) were e-mailed requesting their participation in a 17 question survey.
Results 271 responded, with 63% reporting D.O. school as their 1st choice. Prior to medical school, 27% had OMT performed on them and 44% had observed OMT being performed. 49% want to enter primary care, 23% a medical specialty, 19% a surgical specialty, and 9% "other". 41% reported their OMT skills are not adequate to treat patients currently. 41% reported adequate skills, and 18% were unsure. Almost half (49%) felt instruction received to date has prepared them to use OMT. 30% feel unprepared and 21% are unsure. 38% never practice OMT outside the lab/clinic, 53% practice 1-2 hours/week and 9% practice 3 or more hrs/week. 22% were very likely to use OMT in their practice and 39% somewhat likely. 24% are not likely to use OMT. 100% of students felt OMT was *at least* sometimes efficacious for musculoskeletal disorders, and 79% of students for non-musculoskeletal disorders. Of students who have had rotations, 77% worked with at least 1physician who uses OMT.
Conclusions: Students have a positive attitude towards OMT, and find it at least sometimes efficacious. 63% chose osteopathic medical school as 1st choice, 49% plan on entering primary care and 61% practice OMT on their own. 60% of students are somewhat likely to use OMT in their practice. Positive behavior modeling by preceptors performing OMT, and continued emphasis on OMT in education may continue to encourage students to use OMT as physicians, keeping alive a distinguishing characteristic between osteopathic and allopathic physicians.

♦M7

Three Shortcomings of Current Osteopathic Medical Education Curricula, a Student Perspective

R. A. Alvarez, MS, OMS-II[1], S. Feitell, OMS-II [2], A. C. Jones, OMS-III[3], B. W. Landry, MSPT, OMS-III[4], A. C. Mapley, OMS-IV[4], C. R. Mason, MMS, OMS-II[4], P. A. Sarmiento, OMS-III[3], S. Schrick-Senasac, OMS-IV[5], [1]Nova Southeastern University-COM, Davie, FL, [2]PCOM, Philadelphia, PA, [3]UMDNJ-SOM, Stratford, NJ, [4]NYCOM/NYIT, Old Westbury, NY, [5]UNTHSC, Fort Worth, TX

Background: Curricular content and methodologies exhibit great variation amongst the existing COMs. Osteopathic medical students have expressed a sentiment of inconsistency regarding the educational experience between the COMs and a concern of insufficient preparation for their future careers as osteopathic physicians. The three major shortcomings identified by the 2005-2006 COSGP were the business of medicine, professional education (in the form of teaching and learning) and research. To further quantify and address this perception, the authors utilized a survey of the 23 COMs.
Methods: Written surveys were sent to the current student government presidents at the 23 campuses of the COMs. These surveys consisted of nine questions regarding the presence, con-

♦ Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.

tent, and delivery of education on the business of medicine, professional education and research at their campuses.
Hypothesis: That osteopathic medical student would identify areas of perceived deficiency in the current curricula used by COMs.
Results: Twenty one of the 23 surveys were completed by student government presidents of the COMs to achieve a 91% participation level. There was a variety of coverage of the topic areas, ranging from 95% perceived coverage of business of medicine and research to 52.6% coverage of professional education. There was a greater variety amongst some specific subtopics such as grant-writing under research, demonstrating 5% perceived coverage and other areas, such as HIPAA training under business of medicine, demonstrating 89% perceived coverage. The reported methods by which the content was delivered also varied greatly.
Conclusions: Most areas that have been deemed core competencies by the AACOM and the AOA are perceived by osteopathic medical students to be well-covered by current curricula. However, the important subject matters of the business of medicine, professional education, and research are perceived by most student governments as lacking. While not considered core competencies, these areas of deficiency are of significant importance to the future success of osteopathic students, residents and physicians alike. The identification of the aforementioned deficiencies warrants further investigation by the osteopathic profession. Potential solutions should be explored and implemented in an effort to strengthen the current system of osteopathic medical education delivery and the profession as a whole.

♦M8

Teaching Palpatory Force to Osteopathic Medical Students

R. M. Marchioni BS, A. H. Badoolah, BS, A. J. Caban-Martinez, BS, MPH, M. M. Patterson, Ph.D., College of Osteopathic Medicine, Nova Southeastern University, Ft. Lauderdale, FL

Purpose: To determine the effect of providing palpatory pressure feedback training to 1st year osteopathic medical students (OMS 1) on their ability to subsequently and repeatedly replicates a given palpatory pressure.
Methods: Twenty-one OMS 1 students were recruited and randomly assigned to control or experimental groups. Both groups palpated a standard subject over the T4 and L4 spinous processes after being told to "Palpate the area with enough pressure to spring the spinous process". Each subject applied the pressure he/she thought appropriate 4 times rhythmically. Applied pressure was monitored by a Tekscan transducer placed over the spinous process. Experimental subjects were then trained by watching the pressure readout and instructed to apply 10N of force. They practiced this task until they felt able to replicate 10 N force (about 1-2 minutes), then were immediately retested. All subjects were retested each week for 4 weeks and neither group was allowed to see the pressure readout. The NSU IRB approved the study.
Results: There was no difference in average pressure applied during the initial session between the 2 groups. Most students started with pressures of about 2 N or less, although 2 in the control and 2 in the experimental group began with T4 pressures of above 6N (4 Ss in Experimental Group for L4). Students in the control group who began above 6N dropped their pressures over the testing to 1N or less, while the others remained at about 1N. All students in the experimental group palpated at 5N or above and all but 2 palpated at pressures of 10N or above immediately after training. In subsequent sessions, most showed some decrease in palpatory pressures, but only 1 dropped to 1N or less.
Comment: One session of feedback training resulted in an immediate but variable change in palpatory behavior. The students in the experimental group verbalized an excitement in having the feedback training to guide their palpatory ability. It is evident that more training is necessary to accurately reproduce a specified pressure, but that even a short training session may provide a long-lasting change in palpatory behavior for beginning students. Further studies are underway to quantify the training necessary for students to retain the ability to accurately replicate given palpatory pressures.

♦Indicates posters entered in the Student Prize Competition, a judged event that takes place during the Poster Session at the Research Conference.