Student Involvement in Research
E. Marty Knott, OMS IV

The Student Osteopathic Medical Association (SOMA) has made a commitment to promoting student involvement in research at all the colleges of osteopathic medicine. In November 2004, SOMA hosted its 8th Annual Research Symposium at the 109th Annual AOA Convention and Scientific Seminar in San Francisco. SOMA combined efforts with the AOA to bring together 46 osteopathic medical students from around the United States, allowing them to proudly present their research findings.

Thanks to a generous $1000 gift from Nazzareno E. Liegghio, DO; a matching donation from Pfizer Inc; and money from the AOA Council on Research, cash awards were given to five osteopathic medical students, two of whom had their abstracts previously published. Two of the previously unpublished award-winning poster presentations appear here.

- **Jon C. Uggen** from Midwestern University’s Chicago College of Osteopathic Medicine won the inaugural 2004 Nazzareno E. Liegghio, DO, SOMA Student Research Award for his project titled “Tendon Gene Therapy Modulates the Local Repair Environment in the Shoulder.”

- Awards for best poster presentations were also given to:
  - **Zachary Nye** for his presentation “Morphometric Variation in Human and Chimpanzee Lumbar Vertebrae” (*J Am Osteopath Assoc*. 2004;104:387);
  - **Gina Benaquista and Jodi Speth** for their presentation “Modification of Superficial Scar Tissue Following Skeletal Muscle Laceration” (*J Am Osteopath Assoc*. 2004;104:347); and
  - **Que Huong T. Nguyen, BS**, for his presentation “A Theoretical Study of Polarization Effects on a Model Peptide Bond.”

SOMA hopes to continue its collaboration with the AOA at its 110th Annual Convention and Scientific Seminar in Orlando, Florida, to further improve the student portion of the poster session. Seventeen of the 19 abstracts that were submitted for the SOMA poster session are published here in the Student Contributions section of the *JAOA—The Journal of the American Osteopathic Association*.

Currently, SOMA is working on the SOMA Summer Research Fellowship Award. Through financial gifts from the Osteopathic Research Center (ORC) and Osteopathic Heritage Foundation (OHF), SOMA will award up to five $2000 fellowships annually to students interested in completing an osteopathic-related research project. The fellowship award will fund students for their time as well as a trip to the AOA convention to present their findings. SOMA is grateful to the ORC and OHF for their support on this project. This type of funding will no doubt lead to many significant findings in the coming years. Applications have been submitted and the selection process has begun. Final announcements will be sent out in March. More information for this year’s application can be found at the SOMA Web site (www.studentdo.com).

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Editor’s Note
Unlike the Research Conference abstracts published in the August and September 2004 issues of *JAOA*, the students’ poster presentations appearing here did not undergo review and approval by the Council on Research (COR) for presentation or publication here.

SOMA has exhibited posters at the AOA Research Conference poster presentation for several years, but always as a separate exhibit. In 2004, the COR approved SOMA’s proposal for a joint exhibit; however, that decision came after the deadline for submission. Therefore the following SOMA abstracts were not published with the COR-reviewed and approved abstracts in the August or September 2004 issues of the *JAOA*.

The following SOMA research poster presentations are published as submitted with minimal editing of text by *JAOA* staff to provide consistent style, type, and format; to identify authors by their full names rather than initials when known; and to add authors’ institutional affiliations when omitted in the original abstract.
Introduction: Rotator cuff tears are a common soft tissue injury of the musculoskeletal system. These tears heal by formation of inferior repair tissue, which may lead to severe joint dysfunction. A controversy exists over whether rotator cuff tendons heal after surgery. The endogenous healing is poor or insufficient in most rotator cuff tears and especially in large tears. A method for augmenting the endogenous healing process could be of significant clinical value. The specific aim of this study was to determine if local delivery of an anabolic growth factor using a novel, combined tissue engineering gene therapy approach results in improved healing of rotator cuff tendon defects compared to suture repaired controls.

This study contained two phases: The in vitro phase demonstrated the feasibility of using a tissue engineered gene therapy platform for tendon repair. Rat tendon fibroblasts (RTFs) were transduced with either platelet-derived growth factor-β (PDGF-β) or insulin-like growth factor-1 (IGF-1). Confirmation of active peptide was assessed, and then ability of the peptide to upregulate metabolism in an adjacent, local environment was demonstrated. The in vivo phase evaluated a new cell-polymer construct with RTFs transduced with PDGF-β or IGF-1 genes for its ability to augment tendon repair in an RTF model of rotator cuff injury.

Materials and Methods: Phase I, in vitro model: Adult male Sprague-Dawley rats were used to isolate tendons from the shoulder complex, and tendon fibroblasts were initiated in culture by explant outgrowth. Explants were fed Dulbecco’s modified eagle medium (DMEM) supplemented with 10% fetal calf serum. After RTFs were serially cultured and expanded, they were transduced with the genes for either PDGF-β or IGF-1 by retroviral vectors. The cells containing the active genes were selected by incorporation of a neomycin resistance gene added to the construct. Reverse transcriptase-polymerase chain reaction (RT-PCR) and enzyme-linked immunosorbant assay (ELISA) were used for positive confirmation of gene expression. To test whether IGF-1 gene transduced RTF cells modulate the metabolism of surrounding tissue, the following was performed: RTF cells were seeded onto a bioabsorbable polymer scaffold composed of polyglycolic acid and cultured for 5 days to assemble a tissue-engineered tendon construct. This technique was then repeated to assemble constructs containing the PDGF-β gene-transduced RTFs. The two different constructs were cultured in the same tissue culture well but separated by a membrane that allowed diffusion of soluble factors. The tested group configurations included single construct (RTF/0; Control 1), Nontransduced RTF (RTF/RTF; Control 2), and RTF/IGF/RTF (experimental). Constructs were then allowed to incubate in culture for 24 or 48 hours and then pulse labeled with tritiated proline (H3-Pro) and tritiated thymidine (H3-Thy) to assess collagen and DNA synthesis. The constructs were harvested individually and scintillation counted to determine the rate of synthesis in each RTF construct.

Phase II, in vivo model: Adult male Sprague-Dawley RTFs were isolated, cultured, and transduced with genes for either IGF-1 or PDGF-β by retroviral vectors. After selection and expansion, the transduced RTFs were seeded onto a polymer scaffold and further cultured. Rotator cuffs of rats were transected surgically and allowed to undergo an inflammatory phase for 2 weeks, at which time they then underwent repeated operation to repair the original tear. Repair included standard suture realignment as a control or suture repair with the addition of a gene-modified tendon tissue construct (experimental group). Tissue was harvested at 6 weeks postrepair and prepared for histological examination.

Results: Phase I, in vitro model: Rat tendon fibroblasts were easily cultured, readily transduced, and selected with the IGF-1 and PDGF-β genes. They demonstrated expression of the gene and active peptide via ELISA and RT-PCR analysis. RTF cells rapidly attached to polymer scaffolds and formed highly cellular tissue constructs within the polyglycolic acid (PGA)
scaffolds by 5 days postseeding. RTF constructs incubated alone exhibited a baseline level of collagen synthesis (control 1) and were not significantly stimulated by placement of a similar RTF construct (control 2). By 24 hours, PDGF-β transduced cells stimulated adjacent RTF cells to increase collagen synthesis by 300%; however, DNA synthesis was not significantly increased. IGF-1 increased collagen synthesis by 28% and DNA synthesis by almost 100% by 24-hours exposure. At 48 hours, the trends for increased collagen synthesis for PDGF-β and IGF-1 continued. In addition, PDGF-β transduced tendon cells resulted in a 300% increase in DNA synthesis, and IGF-1 maintained a 28% increase in DNA synthesis.

**Phase II, in vivo model:** Rotator cuffs receiving standard suture repair (control) healed with a range from no repair to incomplete restoration. Histological examination corroborated these findings. The experimental group receiving tissue construct grafts containing cells expressing PDGF-β plus reconstruction with suture showed a strong tendency for repair with near complete to full restoration of the torn tendon. Observation using polarized light microscopy demonstrated a restoration of normal crimp patterning and collagen bundle longitudinal alignment in experimentally treated animals compared to controls.

**Conclusions:** The in vitro study demonstrates tendon fibroblasts can be tissue engineered to deliver therapeutic peptides to local environments to stimulate a repair response. The in vivo study demonstrates the efficacy of a new type of bioactive implant for repair of rotator cuff injuries. The current in vivo model closely mimics the clinical sequelae of tear, inflammation, and repair. The goal of this work is to develop a bioactive patch capable of accelerating rotator cuff repair and modulating the quality of the repair tissue. Initial work has focused on IGF-1 and PDGF-β; however, other growth factors with different actions on tendon cells are being explored.

**A Best Poster Presentation Award Recipient**

**A Theoretical Study of Polarization Effects on a Model Peptide Bond**

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Of the many aspects that govern the structure, stability, and functionality of proteins, interactions involving peptide and hydrogen bonds often play a very important role. Peptide bonds are present in proteins between amino acid residues, covalently linking them together, and hydrogen bonds are present at, among other locations, the substrate binding sites of enzymes, as well as the structures such as the alpha helixes and beta confirmations. In order to understand the fundamental aspects of peptide and hydrogen bonds in proteins, three systems of N-methylacetamide (NMA) have been established and studied from a theoretical perspective. Through the use of *ab initio* molecular dynamics simulations, the structure of NMA in solution, as well as in the gas and crystal phases, was computed and analyzed. From this, the structural changes of NMA going from the gas to the crystal phase along with the changes of the NMA vibrational spectra in differing chemical environments were discussed and compared to available experimental data. In addition, the dipole moment of water as induced by the presence of NMA was calculated in order to quantify polarization effects present in NMA solution. Overall, the results of this study coincide those of available experimental data and can be used as the foundation for studying more complex systems involving proteins.

**Demonstration of Endopeptidase 24.15 in Rabbit Heart: Evidence of Bradykinin’s Role in Cardioprotection**

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Bradykinin (BK) plays an important role as an endogenous mediator resulting in acute protection from myocardial ischemia reperfusion injury. Shielding BK from metabolism prolongs its activity and increases protection during reperfusion following an ischemic event. EC 3.4.24.15 (EP24.15) is one of the enzymes responsible for the metabolism of BK. Previous work in this lab has shown inhibitors of EP24.15 increased BK levels in the heart and decreased reperfusion injury, but the actual enzyme’s presence has not been validated in heart tissue. The goal of this study was to demonstrate that EP24.15 is present in the heart.

Partially purified rabbit heart homogenates were separated by 10% SDS–12% polyacrylamide gel electrophoresis and underwent Western Blot analysis. The nitrocellulose membranes were incubated with anti-EP24.15 antibody (1:1500 dilution) overnight. The secondary antibody (goat anti–mouse IgG HRP in a 1:2000 dilution) was incubated for 1 hour at room temperature. The membranes underwent chemiluminescence and were exposed to x-ray film.

The analysis showed bands of protein which correspond to authentic EP24.15 in the heart tissue samples. These results demonstrate EP24.15’s presence in rabbit heart tissue, thus providing support for the idea that inhibitors of this enzyme work by inhibiting the actual EP24.15 enzyme and not by other nonspecific mechanisms.

This research was supported by a WV SOM Intramural Research Grant.
Hysteresis as a Measure of Ankle Dysfunction

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Introduction: By quantifying hysteresis (the retained energy of deformation), scientists and engineers better understand physical properties of many materials. For this study, a new instrument, the Ankle Torsion Monitor (AnTM), was designed to measure ankle hysteresis loops (AHL) during the application of inversion and eversion forces. A similar premise was successfully used to study low back rotational hysteresis. The ability to objectively document an evolving AHL spectrum during the sprain healing process and to demonstrate interventional impact on AHL would benefit treatment design choices.

Design: This was a methodological study designed to test the ability of an AnTM protocol to document AHL changes over time and in response to different therapies using intra-subject paired ankle observations (sprained vs. nonsprained repeated over time) or immediate comparison of the same ankle (pre- to post-bracing). The effect of osteopathic manipulative treatment (OMT) was also compared using intra-subject post- and pre-treatment values.

Methods: A single investigator (MLK) palpated the lower extremities of subjects presenting with grade 1-2 supination ankle sprains. If present, articular dysfunctions (cuneiform, talocalcaneal, talocrural, fibular head, and femorotibial) were graded 1 (mild), 2 (moderate), or 3 (severe) according to AOA standards. AnTM measures (using 0-, 2-, 4-, and 6-ounce weights) were made pre- and post-application of a brace and longitudinally (every 2-3 days) in a number of cases to determine its variability on the same day and over time. AHL measurements were also made pre- and post-OMT of the sprained side and compared to the untreated AHL.

Intervention: A single operator (MLK) performed muscle energy OMT for plantar flexion of the talocrural joint in addition to indirect joint techniques of all five joints examined. Total treatment time averaged 5 minutes. An ankle stabilizing brace (Mueller®) was employed for a portion of the study.

Participants: Subjects were men and women aged 23-57 years. Those with acute ankle sprains (n=17) were included for the OMT portion of this study; additional normal subjects (n=5) allowed AHL comparison of braced and unbraced ankles.

Outcome Measures: Each AHL was repeated three times with area within the loop measured by planimeter and loop extremes noting inversion-eversion range-of-motion (ROM). A cumulative somatic dysfunction load (CSDL) was determined for each lower extremity by adding all ipsilateral severity scores.

Results: Bracing statistically tightened AHLs and reduced ROM. Same-day nontreated ankles (allotted 5-10 minutes rest) demonstrated no difference in ROM or in area within the AHL. On the side of the sprain, AHL measurements showed a trend (P=.137) for ROM to change between visit 1-2, but no change on the nonsprained side.

Conclusions: AHL measurements generated by the AnTM protocol applied were shown to provide consistent, reproducible data in both normal and sprained ankles. While hysteresis measurements in this small study did not differentiate between sprained and nonsprained ankles or between those with high and low CSDL, it did demonstrate a statistical trend for AHL change over time during the healing process of a sprained ankle with a high CSDL.

Credit: A portion of this study was funded by the Commonwealth of Pennsylvania.

Reference
improved by 33 minutes. The most important contributing factor to minimizing the office time was attributed to the medical assistance personnel that took patients vital signs soon after registration. Another contributing factor was an efficient communication between the medical assistance personnel and the nursing station, which necessitated a series of staff meetings to improve upon their skills in dealing with patient’s needs in a swift and productive manner.

**Conclusion:** This study should be used as a model for learning how to improve the time patients spend in a clinic.

**Acknowledgment:** The principal author is thankful to Dr Reza Nassiri, Professor of Clinical Pharmacology and Medical Virology at LECOM for the helpful discussions on this project.

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**Prion: Gene, Structure, and Species Barrier**

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**Hypothesis:** Prion species barrier may be due to genetic variation and protein structural differences among different species.

**Methods:** Sequence analyses of open reading frames of the prion gene from different species, including human, deer, sheep, cattle, mouse, and rat were conducted. Synthetic prion-derived peptides with strategic point mutations were manufactured and studied to investigate potential conformational changes.

**Results:** Codon 129 is a polymorphism position where, in the human population, 50% is Met (Methionine)/Val (Valine), 40% Met/Met, 10% Val/Val. Other species: sheep, cattle, rat, and mouse have a conserved codon 129 with Met/Met. It is observed that with Met at codon 129, transmission of prion disorder is relatively consistent regarding incubation time. However, if codon 129 is other than Met, eg, Valine in rat, Leucine (Leu) in Wapiti deer, incubation time of prion disorder is inconsistent or prolonged. Furthermore, there are four putative alpha-helical regions forming four-helix bundles in the prion protein which are proposed to be critical in the stability of the protein. If these alpha-helical regions convert to beta-pleated sheets, the altered prion protein accumulates in the brain tissues and degenerative changes occur. Prion-derived peptide 121-231, containing Met at codon 129, demonstrated alpha-helical conformation under nuclear magnetic resonance (NMR). However, if Val or Leu is at codon 129, prion-derived peptide 121-231 demonstrated beta-pleated sheet conformation under NMR analysis.

**Conclusions:** Prion species barrier may be, at least in part, explained by differences in genetic and protein structural variations among different species.

**Sponsors:** Patricia Hentosh, ScD, and Paul Dew, MD, MPH, at Kansas City (Mo) University of Medicine and Biosciences; Stanley Prusiner, MD, at University of California, San Francisco.

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**Utilizing the Dementia Rating Scale-2 Alternate Form to Differentiate Independent and Assisted Living in a Continuing Care Retirement Community**

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**Background and Hypothesis:** At this time, there are no objective measures to help determine which level of care in the continuing care retirement communities (CCRCs) is most appropriate for an elderly individual. The goal of this project was to evaluate the usefulness of the Dementia Rating Scale-2: Alternate Form (DRS-2: AF) in differentiating appropriate placements within CCRCs. How well do DRS-2: AF scores differentiate between residents in independent living (IL) and assisted living (AS) settings? We hypothesized that individuals living independently would score significantly better than those placed in an assisted living environment.

**Materials and Methods:** Participants included 64 individuals residing at the Cadbury CCRC, Cherry Hill, NJ (27 in IL and 37 in AL). We administered the DRS-2: AF to all 64 participants. All DRS-2: AF protocols were scored, and results were entered into an SPSS database.

Living placement (IL or AL) was based on an interdisciplinary team assessment, including a geriatric physician, clinical social worker, physical therapist, and nurse.

We ran descriptive statistics, including mean and standard deviations for total score and subscales, and Student’s t-tests to examine differences between means for IL and AL participants.

**Results:** In terms of the descriptive statistics for each group (IL and AL) for total score and subscale scores, the IL group received higher scores on the DRS-2: AF total score (m = 130.52, SD = 8.10) in comparison to the AL group (m = 114.14, SD = 20.92). A similar pattern was seen across the subscale scores as well.
Conclusion: Our findings suggest the DRS-2: AF total score as well as the three subscales may be useful in differentiating residents residing in IL settings from residents in AL settings. Individuals with lower scores should be considered for an AL placement, as opposed to IL placement. The DRS-2: AF may be a useful tool in making placement decisions for potential residents within CCRCs.

Sponsors: The project was sponsored by a Pfizer grant and a Summer Research Student Fellowship program at UMDNJ-SOM.

Reference

Outpatient Prescribing Practices for Nonapproved Drugs in Children

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Objective: To determine the use of nonapproved medications (safety and effectiveness in pediatric patients have not been established) for children prescribed at outpatient physician visits.

Background: The National Ambulatory Medical Care Survey (NAMCS) is a nationwide periodic survey of non–federally employed office-based physicians primarily engaged in patient care activities. The NAMCS provides information on selected patient visits including data on patient demographics, reasons for visit, physician diagnoses, clinical services provided, and medications ordered. Up to 6 medications could be recorded for each visit. The number of pediatric outpatient visits sampled between 1993 and 2000 ranged from 3000 to 6000 each year.

Methods: NAMCS data were analyzed for the period 1993–2000 for all children <18 years of age. Medications were abstracted from the drug codes listed for each visit. For the top 200 drugs recorded, we examined the indications and usage of the drug for pediatric patients according to the 2001 Physicians' Desk Reference. The rate of nonapproved medication use was estimated according to the top 200 medications recorded. Information on the use on nonapproved medications according to medical disease and physician specialty was also examined. Weighted data were used for all analysis.

Results: For the years 1993–2000, there were an estimated 158 million pediatric outpatient visits each year to physicians in the United States. Medication was prescribed at 64.2% of pediatric visits. Of the top 200 medications prescribed to children during this time period, 8% (16/200) of these medications had not been established as safe and effective in pediatric patients. The most common prescribed nonapproved medications included Paxil®, Wellbutrin®, Vicodin®, Benzac®, and Risperdal®. The most common disorders for which nonapproved medications were prescribed included dermatological and psychiatric disorders. Dermatologists, family practitioners, psychiatrists, and pediatricians prescribed the most nonapproved medications as compared to other specialties.

Conclusion: Of the top medications prescribed to children on an outpatient basis in the United States, 8% have not been shown to be safe and effective in pediatric patients. The majority of these medications are prescribed for dermatological and psychiatric disorders and likely represents the limited availability of effective medical therapies for children with these disorders.

Protein-Protein Interactions in Vasopressin Signal Transduction: New Evidence for Cytoskeletal Dynamics

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The [Arg8]vasopressin (AVP)–induced signal transduction pathway that stimulates oscillations (spikes) in cytosolic free Ca2+ concentrations ([Ca2+]i) was examined in confluent cultures of rat A7r5 vascular smooth muscle (VSM) cells. Recent studies have suggested that AVP signal transduction may involve dynamic protein-protein interactions, ultimately leading to the phosphorylation and inhibition of delayed rectifier potassium channels ( Kv1.2). Several likely protein candidates involved in this signaling cascade have been identified, including proline-rich tyrosine kinase 2 (PYK2), which may phosphorylate the Kv1.2 channel protein and protein kinase C (PKC) family members, which may phosphorylate PYK2. In the present study, A7r5 VSM cells were treated for varying times (0–30 min) with and without physiological vasocostrictor concentrations of AVP (100 pM) or the PKC activator 4β-phorbol 12-myristate 12-acetate (PMA). Following these treatments, protein-protein interactions were examined by immunoprecipitation of PYK2 and by Western blot analysis to identify proteins that become associated with PYK2 during the time course of the treatments. Western blot analysis of untreated A7r5 VSM cells confirmed previous studies, which found that...
PYK2 is constitutively bound to the Kv1.2 channel. Western blot analysis of the AVP time-course treatment of A7r5 VSM cells also revealed that PKC ε and PKC δ transiently associate with PYK2. PMA treatment induced a similar PKC translocation. These results extend previous findings that suggested that PKC plays an active role in the signal transduction pathway leading to Ca2+ spiking. Additional new evidence of protein-protein interactions was observed in AVP treated A7r5 cells where an unknown protein band was visible on a Ponceau red stain of a Western blot membrane. This unknown protein in the PYK2 immunoprecipitate was identified as actin based on the apparent molecular weight (43 kDa), and subsequently confirmed by Western blot analysis using an antibody specific for smooth muscle α-actin. The transient association of actin with PYK2 was maximal at 10 min of AVP treatment and significantly decreased by 30 min. This novel result suggests a dynamic role of actin in vasopressin signal transduction and emphasizes the importance of the cell cytoskeleton in signaling cascades.

Is Equity Being Sacrificed? The Effect of User Fees on the Willingness and ability to pay for Schistosomiasis Control in the Lake Victoria Region of Tanzania

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This study asks: In an effort to finance schistosomiasis prevention and treatment activities, have countries such as Tanzania lost sight of those most in need? With the new policy of user-fee charges in place, is effective treatment still a priority for those in heavily endemic regions? The impact of charges on health-seeking behavior is crucial to disease control in Tanzania. The decentralization of the country’s healthcare system has led to an increase in the need for income generation at all operational levels. In general, people have accepted the costs of curative services. However, the impact of user-fee charges on health-seeking behavior has yet to be assessed. This is a cross-sectional study of willingness and ability to pay for schistosomiasis control. Questionnaires were administered to household heads of six representative fishing and farming communities in the Lake Victoria Region of Tanzania, East Africa. A total of 103 questionnaires were analyzed. The yearly mean household expenditure on schistosomiasis diagnosis and treatment was 7,000Ts ± 60,000Ts (US$1 = 1,000Ts), accounting for 1.54% of the total yearly household income. Most household heads (85 [83%]) thought schistosomiasis was the greatest health threat in their communities, but only 27 (26%) of them were willing to pay for screening, diagnosis, and treatment of the infection. On the other hand, 72 (70%) were willing to do volunteer work in exchange for schistosomiasis control. Exposure and livelihood cannot be separated; the future of disease control in this endemic region of Tanzania is about access, not prevention.

Patency and Obliteration of the Cranial Sutures: Is There a Clinical Significance?

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Previous research on cranial sutures has placed great emphasis on correlating age with timing of suture fusion and little dedication to evaluating the relation of sutural patency with clinical relevance. The external morphology of the coronal, sagittal and occipital sutures was evaluated in thirty-six human cadaver skulls. Each suture was described using a modified grading scale to quantify the extent of sutural patency and obliteration. Two significant findings were observed: (1) the lambdoid suture was overall most likely to be patent and least likely to be obliterated and (2) the sagittal suture was most likely to be obliterated and least likely to be patent when compared to the other sutures. These findings provide an insight into understanding what external forces cause cranial sutures to be more patent than others. In turn, it can help determine if they have a clinical significance beyond the estimation of age.

Effect of Bovine Lactoferrin on the MICs of Ampicillin/Sulbactam (UNASYN), Amoxicillin/Clavulanic Acid (Augmentin), Ticarcillin Clavulanate (Timentin), and Piperacillin-Tazobactam (ZOSYN), Against Clinical and Environmental Isolates of Motile Aeromonas

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Aeromonas are gram-negative rods transmitted through drinking of contaminated water whose presence poses a risk to public health. Aeromonas have shown multiple resistance to...
antibiotics. β-Lactamase inhibitors display activity against other multiresistant organisms. Furthermore, studies have shown that addition of lactoferrin to the antibiotics can lower the high minimum inhibitory concentrations (MICs) of resistant microorganisms. Lactoferrin is an iron binding protein found in milk, and has been found to exert anti-microbial activity.

**Purpose:** (1) To investigate the susceptibility of *Aeromonas* isolates to ampicillin/sulbactam (UNASYN), amoxicillin/clavulanic acid (Augmentin), ticarcillin clavulanate (Timentin), and tazobactam-tazobactam (ZOSYN); (2) To study the effect of lactoferrin on the potency of the above antibiotics.

**Methods:** 76 environmental and clinical isolates of motile aeromonads were used in this study (*Aeromonas caviae* = 30; *Aeromonas hydrophila* = 28; *Aeromonas sobia* = 18). Bovine lactoferrin (Sigma) was added to distilled water and filter sterilized. A final concentration of 2000 μg/mL was used. The MICs were determined by the standardized microdilution method in Mueller-Hinton broth as recommended by the National Committee for Clinical Laboratory Standards (NCCLS) (Standards). Customized sensititre panels from TREK Diagnostic Systems, Inc (Westlake, Ohio) were used; 50 μL of lactoferrin solution was added to appropriate wells after inoculation of the organisms.

**Results:** Piperacillin/tazobactam: All three species were susceptible with and without lactoferrin. Ampicillin/sulbactam: 70% of *A caviae*, 61% of *A hydrophila*, and 89% of *A sobia* were resistant without lactoferrin; 23% of *A caviae*, 21% of *A hydrophila*, and 44% of *A sobia* were resistant with lactoferrin. Amoxicillin/clavulanic acid: 10% of *A caviae*, 7% of *A hydrophila*, and 33% of *A sobia* were resistant without lactoferrin; 0% of *A caviae*, 4% of *A hydrophila*, and 0% of *A sobia* were resistant with lactoferrin. Ticarcillin/clavulanic acid: 10% of *A caviae*, 0% of *A hydrophila*, and 33% of *A sobia* were resistant without lactoferrin; 7% of *A caviae*, 0% of *A hydrophila*, and 6% of *A sobia* were resistant with lactoferrin.

**Conclusion:** This study shows that lactoferrin has synergistic antimicrobial activities with β-lactamase inhibitors against motile aeromonads. All three species were highly susceptible to piperacillin/tazobactam with and without lactoferrin.

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**Student Contributions**

A Pilot Study Examining the Effects of Neuromuscular Therapy on Patients With Parkinson’s Disease

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**Introduction:** Parkinson’s disease (PD) is a progressive neurodegenerative disorder. Therapies that address motor dysfunction are pharmacological therapy, surgical procedures, and physical therapy. Neuromuscular therapy (NMT), a form of massage therapy, is a well-established complementary and alternative medicine modality; however, it has not yet been studied in PD. The purpose of this study was to determine whether NMT could improve motor symptoms and quality of life of patients with PD. More specifically, the study was designed to determine if NMT could decrease rigidity, tremor, and bradykinesia and increase locomotion and fine motor skills. It was also designed to determine if NMT could decrease depression and anxiety.

**Methodology:** Thirty-two subjects with PD were randomized to either NMT or music relaxation therapy (active control). Subjects received treatment twice a week for 4 weeks. Testing was conducted at baseline, following the final treatment, and 8 days after the final treatment. Testing measures included the United Parkinson’s Disease Rating Scale (UPDRS), Purdue Pegboard, finger-tapping test, Clinical Global Impression Scale (CGI), Beck Depression Inventory, and Beck Anxiety Inventory. The primary outcome measures were the motor subscale of the UPDRS (Part C) and the CGI.

**Results:** NMT resulted in a significant improvement in the UPDRS Part C (motor subscale) (F1,30 = 6.53, P < .05) in comparison to the music relaxation group, which had little change. The NMT group showed improvement in CGI scores (1.14±0.9 [P<.05]) that was not retained a week after the therapy sessions ended (0.53±0.7, P=NS). Music relaxation had no effect on CGI scores (0.22±0.4, P=NS).

**Conclusions:** The findings indicate that NMT can improve motor symptoms in PD. Future studies should examine whether the beneficial effects can be sustained past 5 weeks or with less frequent NMT, thus making treatment more affordable.

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**Gender Differences in the Cannabinoid Modulation of an A-type K1 Current in Neurons of the Mammalian Hypothalamus**

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Cannabinoid signaling exerts profound influence over the hypothalamic control of homeostasis including but not limited to feeding. The hypothalamic feeding circuitry consists of several components. The stimulatory component of appetite includes the neuropeptide Y (NPY), orexin (hypocretin), ghrelin, and melanin-concentrating hormone (MCH) neurons. Inhibitory inputs are predominately from the proopiomelanocortin (POMC) neurons originating in the hypothalamic arcuate nucleus (ARC). POMC neurons synthesize and release anorexigenic peptides such as cocaine amphetamine-related transcript (CART), α-melanocyte stimulating hormone (α-MSH), and β-endorphin.

This study sought to determine whether cannabinoids...
modulate A-type K+ current ($I_a$) in POMC neurons. To this end, whole-cell patch clamp recordings were performed in hypothalamic slices through the ARC prepared from castrated female and male guinea pigs. A robust $I_a$ that was blocked by high concentrations of K+ channel blocker 4-aminopyridine (3mM AND 10mM) was found in 46% of male and 39% of female cells. We observed that the neuronal responsiveness to cannabinoids was sexually differentiated. In recordings of female neurons, bath application of the brain cannabinoid (CB1) receptor agonists WIN 55,212-2 ($1\mu M$) or arachidonyl-2'-chloroethylaminde (ACEA, $1\mu M$) selectively induced a depolarizing rightward shift in the inactivation curve for the $I_a$ significantly increasing the half-maximal voltage ($V_{1/2}$) for inactivation. This effect was completely blocked by the CB1 receptor antagonist AM251 ($1\mu M$) and occurred without affecting the peak current magnitude ($I_{\text{max}}$). Post-hoc immunofluorescent labeling confirmed that these effects occurred in POMC cells. We observed no effect on the $V_{1/2}$ or the $I_{\text{max}}$ for the activation curve. In contrast, recordings from male neurons indicated no discernible effect of cannabinoids on the $V_{1/2}$ or the $I_{\text{max}}$ for either the inactivation or the activation of the $I_a$. Collectively, these data reveal that POMC neurons express a prominent $I_a$, and that cannabinoids positively modulate this current in a sex-specific way by altering the voltage dependence of its inactivation. The resultant inhibitory effect on this neuronal population may shed some insight into the mechanism(s) by which cannabinoids influence appetite.

**Metabolic Syndrome and Postoperative Complications in Cardiothoracic and Vascular Surgical and Percutaneous Interventions**

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**Purpose:** Metabolic syndrome (MS) constitutes a cluster of risk factors for cardiovascular disease such as hypertriglyceridemia, hypertension, diabetes, central obesity, and low high-density lipoprotein levels. Any three of the above risk factors qualifies to be classified as MS. Many patients undergoing cardiovascular surgical and percutaneous interventions may not have been diagnosed and treated adequately for this syndrome so as to prevent perioperative morbidity. The prevalence of MS and postoperative complications has not been described adequately in the literature.

**Methods:** All patients undergoing cardiovascular and thoracic surgery and percutaneous interventions at a specialty hospital over a 12-month period between June 2001 and June 2002 were included in the study.

Data were collected on demographics, risk factors, laboratory data and diagnostics preoperatively and type of intervention and postoperative complications. Patients were classified as having MS with at least 3 or 4 or 5 risk factors. Analysis was performed using SPSS version 12.0.

**Results:** Of the 312 patients, 68.6% were males and 31.4% females. MS was prevalent in 54.4% of patients with 33.3% having 3 risk factors, 14.4% having 4 risk factors, and 6.7% having all 5 risk factors. Most frequent interventions were percutaneous coronary interventions (27%), endoluminal graft of aorta (16%), peripheral arterial interventions (15%), coronary artery bypass surgery (14%), abdominal aneurysm repair (11%), carotid endarterectomy (10%), and femoral popliteal bypass surgery (5%).

Most common postoperative complications include renal dysfunction (20%), blood transfusion requirement (17%), pleuropericardial effusion (12%), atrial fibrillation (7.4%), congestive heart failure (2%), and stroke (2%). Overall prevalence of complications was 42%.

**Conclusions:** (1) Prevalence of MS is high in patients admitted to hospital for cardiothoracic and vascular surgical or percutaneous interventions. (2) Prevalence of postoperative complications is high in this high-risk interventional procedure.

**Clinical Implications:** An opportunity exists to identify patients with MS preoperatively in cardiothoracic and vascular interventions (both surgical and percutaneously) and implement preventive therapeutic strategies to decrease postoperative morbidity and mortality.

**Prevalence of Somatic Dysfunction Among an Elderly Kenyan Population**

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Although the phrase somatic dysfunction (SD) is one that is well known throughout the osteopathic community, the concept is not often addressed in research. Due to the HIV/AIDS crisis in Africa, many Luo elders are given the responsibility...
of caring for one of the 890,000 orphans now living in Kenya, in addition to performing daily activities. The Luo population is thus ideal for research involving SD. This study assessed the prevalence of SD and its association with caregiving status, sex, type of farming, and socioeconomic status (SES) in a population of 103 Kenyan elders. An osteopathic screen performed by a trained osteopathic medical student determined SD. An interview and a complete physical examination were utilized.

We hypothesized that SD would be more prevalent among caregivers than noncaregivers and more present in women than men. It was also predicted that prevalence of SD would be unique to the type of farming performed in a community and that SES would be inversely proportional to SD. It was expected for SD to be positively associated with the body systems in a clinical history. SD was prevalent among all participants and was associated with caregiving intensity, perceived burden, waist-to-hip ratio and occupation. Women and farmers had more SD at the costal cage than men and nonfarmers, respectively. Significant associations were found among millet and rice farmers and SD in a particular region. The relationship between SES and SD was insignificant. SD was not found to be of clinical significance in this population, but more research is needed to verify the results determined by the clinical history. This study has expanded knowledge of the prevalence of SD in the Kenyan population.

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Blood Glucose Correlations With Depression, Body Habitus and Caregiving Status in the Elderly Kenyan Luo Grandparents

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Background: In this study, participants in an established study that evaluated grandparents caring for their orphaned grandchildren due to the high HIV/AIDS in the Western Kenyan Luo community were examined to identify a possible correlation between depression and non–insulin dependent diabetes mellitus (NIDDM) within this population.

Hypotheses: (1) Fasting glucose will be correlated with depression scores; (2) fasting glucose will be correlated to body habitus; and (3) fasting glucose means will differ between caregiving (CG) and non-caregiving (non-CG) grandparents.

Method: A convenient sample of 103 elderly Luo grandparents older than 60 years from the Nyando District in the Nyanza Province in Western Kenya was used in this study. The revised John Hopkins Symptom Checklist-25 Depression Scale was used to identify depressed participants. Anthropometric measurements and one blood glucose measurement (either postprandial or fasting) was taken for each participant.

Results: No significant correlation with blood glucose level and the revised HSCL-25 scores was observed. There were significant correlations with blood glucose level and body habitus. There was also a 23.17 difference between the basic fasting glucose level means for the CG group and the non-CG group.

Discussion: The revised HSCL-25 scores may have been a poor measure for depression in this elderly population, leading to an insignificant correlation between blood glucose levels and the revised HSCL-25 scores. A positive correlation between the blood glucose level and the body habitus were expected. The difference between the basic fasting glucose level means for the CG group and non-CG group was also expected.

Limitations: A small sample size, random blood glucose measurements, and the traditional Kenyan diet could have limited the results in the study.

Future Research: In the future, this study will evolve to examine the correlation between NIDDM and self-reported depression by diagnosing individuals with NIDDM and increasing the sample size.

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