Table of Contents

1. EEG Fluctuations of Wake and Sleep in Mild Cognitive Impairment .............1
   Johnny O'Keeffe, Barbara Carlson, Lisa DeStefano, Michael Wenger, Melissa Craft,
   Linda Hershey, Jeremy Hughes, Dee Wu, Lei Ding, Han Yuan

2. Highly efficient non-viral VEGF gene delivery to human mesenchymal stem cells by
   phage-like nanoparticles .................................................2
   Mengmeng Zhai, Lin Wang, Penghe Qiu, Chuanbin Mao

3. Identifying EEG Correlates to Infant Limb Movements While Learning to Craw........3
   Monique Shotande, Alejandro Patino, Andrew H. Fagg, Lei Ding, David P. Miller, Thubi
   H.-A. Kolobe

4. Electric Field Induced Acoustic Tomography .............................................4

5. Synthetic Peptide Effects on Chondrogenic Differentiation of Stem Cells .............5
   Salma Mahzoon, Michael S. Detamore

6. Resting State Networks Estimated from EEG: A Simulation and Real Experiment
   Study .................................................................6
   Chuang Li, Han Yuan, Diamond Urbano, Yoon-Hee Cha, Lei Ding

7. Examination of Erythrocyte Microparticle Formation In A Microfluidic High Shear
   Environment ............................................................7
   James P. Buerck, Dimitrios V. Papavassiliou, Tyler Snyder, David W. Schmidtke, Edgar
   A. O'Rear

8. Multimodal Imaging of Human Brain Auditory Responses Using Simultaneous EEG and
   fNIRS ........................................................................8
   Jesse Farrand, Yuxuan Chen, Julia Tang, Yafen Chen, Johnny O'Keeffe, Guofa Shou,
   Lei Ding, Han Yuan

9. Real-time EEG artifact correction during fMRI using ICA ..................................9
   Ahmad Mayeli, Vadim Zotev, Hazem Refai, Jerzy Bodurka

10. OSW-1 Analogs for Anti-body Drug Conjugate .............................................10
    Anh T. Le, Cori A. Malinky, Anthony W. G. Burgett

11. Electroencephalography from Infant Motor Brain at Time of Crawling ............11
    Alejandro Patino, Andrew H. Fagg, Thubi, H.A. Kolobe, David P. Miller, Lei Ding

12. The Role of Bioreactor Oscillatory Mechanostimulation on an Engineered Tendon
    Tissue Construct ..........................................................12
    C. Coffey, M. Hover, Z. Mussett, V. Sikavitsas

13. Single Camera System for Infant Motion Capture ........................................13
    Mustafa A. Ghazi, David P. Miller
14. Identification and Assessment of Bone Morphogenic Protein Receptor Binding Peptides in Osteoblast Differentiation Signaling Pathway ........................................ Kegan S. Sunderland, Ph.D. Candidate¹, Chuanbin Mao

15. Development of a Multi-Scale Mitral Valve Computational Model for Patient-Specific Treatment Planning ................................................................................. Devin Laurence, Chung-Hao Lee

16. Amplitude of Resting-State fNIRS Global Signal is Related to EEG Vigilance Measures ........................................................................................................... Yuxuan Chen, Jesse Farrand, Julia Tang, Yafen Chen, Johnny O’Keeffe, Guofa Shou, Lei Ding, Han Yuan

17. Computation of Surface Laplacian for Tri-polar Ring Electrodes on High-density Realistic Geometry Head Model ........................................................................ Junwei Ma, Han Yuan, Sridhar Sunderam, Walter Besio, Lei Ding

18. Nanoscale Photoacoustic Tomography (nPAT) ................................................ Pratik Samant, Armando Hernandez, Shelby Conklin, Liangzhong Xiang


20. .............................................................................................................................. Muhammad Ghani

21. Novel 3-D in vitro system sets the stage for a neoteric therapeutic approach of Idiopathic pulmonary Fibrosis ........................................................................... Rabab Sharif, Alan Betensley, Dimitrios Karamichos


23. Assessing rTMS Effects in MdDS: Cross-modal Comparison between Resting State EEG and fMRI Connectivity ......................................................... Yafen Chen, Chuang Li, Guofa Shou, Diamond Urbano, Yoon-Hee Cha, Lei Ding, Han Yuan

24. X-Ray Induced Acoustic Computed Tomography ........................................... Elijah Robertson, Shanshan Tang, Rowzat Faiz, Liangzhong Xiang

25. Real-time Multi-spectral Fluorescence Microscopy ......................................... Zheng Li, Yuchen Qiu, Shibo Li, Bin Zheng, Hong Liu


27. Functional-MRI and Physiological Evidence of Emotional Hypo-Reactivity in the Offspring of Alcohol Abusers .........................................................
B. Espinoza-Varas, S. Guo

28. Cartilage Hydrogel Pastes for Osteochondral Regeneration in a Rabbit Model ........28
   Emi A. Kiyotake, Donna M. Pacicca, Joshua T. Bunch, Michael S. Detamore

29. Is Visible/Near-infrared Spectroscopy Superior to Ultrasound for Detection of The Onset of Steatosis in a Rat Liver Model? .................................................. 29
   Daqing Piao, Jerry W. Ritchey, D.V.M., G. Reed Holyoak, D.V.M., Corey R. Wall, D.V.M., Kenneth E. Bartels D.V.M.

30. Novel Shape Memory Polymer Devices for Optimal Endovascular Embolization of Intracranial Aneurysms ................................................................. 31
   Chung-Hao Lee, Yingtao Liu, Bradley N Bohnstedt

31. Systematic Review of Multi-planar Reconstruction and Rendering of 3D Ultrasound Datasets: Terminology and Image Layout ................................................. 32
   Kari E. Boyce

32. Novel laparoscopic optical spectroscopy sensing and topography imaging approaches for identification of below-surface tubular structures ................................. 33
   Daqing Piao, Sanjay Patel

33. MOTION ANALYSIS OF WORK PERFORMANCE IN MALES WITH TRANSTIBIAL LIMB LOSS ................................................................. 34
   C.P. Dionne, J. Day, K. Veirs

34. Methods to Study Neurovascular Coupling in Pre-Clinical Models of Aging and Human Patients: From Laser Speckle Contrast Imaging to NIRS ......................... 35
   Andriy Yabluchanskiy, Stefano Tarantini, Han Yuan, Tamas Csipo, Gabor Fulop, Barbara Carson, R.N., Andrew Dentino, William E. Sonntag, Zoltan Ungvari

35. BIOMIMETIC SURFACE MODIFICATION PLATFORM FOR THE DEVELOPMENT OF IN VITRO TUMOR MODELS .................................................. 36
   Cortes Williams, Patrick McKernan, Roger Harrison, Vassilios Sikavitsas

36. Developed a novel treatment method for cementless implant surgery .................. 37
   M. Khandaker, W. Williams, R. Wolf, H. Jamadagni, Amal Swediwah

37. Tuning Photothermal Properties of Gold Nanodendrites for In Vivo Cancer Therapy within a Wide Near Infrared Range by Simply Controlling Their Degree of Branching ................................................................. 38
   Penghe Qiu, Mingying Yang, Xuewei Qu, Yanyan Huai, Ye Zhu, Chuanbin Mao

38. Highly ordered phage films fabricated by a simple dipping-drawing method .......... 39
   Ningyun Zhou, Chuanbin Mao

39. The Laboratory of Biomolecular Structure and Function at OUHSC ..................... 40
   Simon Terzyan, Blaine Mooers

40. Quantitatively Predict Ovarian Cancer Patients’ Responses to Chemotherapy at Early Stage: A Radiomics Based Method .................................................. 41
Abolfazl Zargari, Yue Du, Gopichandh Danala, Theresa Thai, Camille C, Gunderson, Katherine M. Moxley, Kathleen Moore, Robert S. Mannel, Hong Liu, Bin Zheng, Yuchen Qiu, Simon Terzyan, Blaine Mooers

41. Evaluation of the therapeutic effect of theta burst stimulation in Schizophrenia: a randomized sham controlled resting-state EEG study ........................................... 42
Guofa Shou, Qian Guo, Chunbo Li, Kelvin Lim, Jijun Wang, Bin He, Lei Ding

42. Local release of Combretastatin A-4 from NIR-light activatable prodrugs overcomes areal and temporal limitations of photodynamic therapy ........................................... 43
Pallavi Rajaputra, Moses Bio, Gregory Nkepang, Pritam Thapa, Sukyung Woo, Youngjae You

43. Paclitaxel Prodrug for the Combinational Treatment of Photodynamic Therapy (PDT) and Site-Specific Chemotherapy ................................................................. 44
Pritam Thapa, Mengjie Li, Moses Bio, Pallavi Rajaputra, Yajing Sun, Sukyung Woo, Youngjae You

44. Effects of 3D Printing on Flow-Induced Shear Distributions ................................ 45
Cortes Williams, Vassilios Sikavitsas

45. Neurovascular uncoupling predicts cognitive decline and gait abnormalities in a clinically relevant mouse model of whole brain irradiation ........................................... 46
Andriy Yabluchanskiy, Stefano Tarantini, Gabor Fulop, William E. Sonntag, Anna Csiszar, Zoltan Ungvari

46. OUHSC Laboratory for Molecular Biology and Cytometry Research ................. 47
Virginie Sjoelund, J. Acquaviva, III, Jenny Gipson, Jim Henthorn, Huaiwen Wang, Allison F Gillaspy

47. Optimization of Folate Receptor-Mediated Targeting Delivery of Far-Red Light-Activatable Prodrugs to Ovarian Cancer Cells ....................................................... 48
Mengjie Li, Pritam Thapa, Youngjae You, Sukyung Woo

48. Phage as a Genetically Modifiable Supramacromolecule in Chemistry, Materials and Medicine ................................................................. 49
Binrui Cao, Ph.D., Chuanbin Mao

49. Laser Speckle Contrast Imaging (LSCI) to Study Age-Related Microvascular Dysfunction: Potential Applications for Translational Geroscience ................................. 50
Stefano Tarantini, Andriy Yabluchanskiy, Tamas Csipo, Gabor Fulop, Anna Csiszar, Zoltan Ungvari

50. Corneal Tissue Engineering: A Diabetic Keratopathy In Vitro Model ............... 51
Shrestha Priyadarsini, Jian-Xing Ma, Dimitrios Karamichos

51. Genetically engineered viral nanofiber-assisted stem cell differentiation and vascularized osteogenesis ................................................................. 52
Jianglin Wang, Lin Wang, Chuanbin Mao
52. Experimental Neurovascular Uncoupling Promotes Cognitive Impairment in Mice: Implications for Brain and Cerebromicrovascular Aging
Stefano Tarantini, Marta Noa Valcarcel-Ares, Andriy Yabluchanskiy, Zsuzsanna Tucsek, Ferenc Deak, William E. Sonntag, Anna Csiszar, Peter Toth, Zoltan Ungvari


54. Mass Spectrometry Detection of Drugs in Single Bladder Cancer Cells from Patients
Shawna J. Standke, Ning Pan, Naga Rama Kothapalli, Anh T. Le, C. A. Malinky, Jonathan E. Heinlen, Zhibo Yang, Anthony W. G. Burgett

55. Increasing PSA Density Correlates with Prostate Biopsy Pathology on MRI US Fusion Biopsy
Nathan Rademaker, Alexander Parker, Kelly L. Stratton, Justin North, Brian Cross, Michael S. Cookson

56. Pre-clinical studies using laser speckle contrast imaging to investigate mechanisms contributing to impaired neurovascular coupling in mice: role of mitochondrial oxidative stress
Stefano Tarantini, Marta Noa Valcarcel-Ares, Andriy Yabluchanskiy, Gabor Fulop, Peter Hertelendy, Eszter Farkas, William E. Sonntag, Anna Csiszar, Zoltan Ungvari

57. Inhalable Microparticulate SHetA2 Nanocrystals for Lung Cancer Treatment
Manolya Kukut Hatipoglu, Sanjida Mahjabeen, Doris M. Benbrook, Lucila Garcia-Contreras

58. SHetA2 Vaginal Suppositories to Treat Cervical Dysplasia: Influence of Estrous Cycle on Drug Absorption and Pharmacodynamic Endpoint
Sanjida Mahjabeen, Manolya Kukut Hatipoglu, Stanley D. Kosanke, Doris M. Benbrook, Lucila García-Contreras
EEG Fluctuations of Wake and Sleep in Mild Cognitive Impairment

Johnny O’Keeffe, B.S., Barbara Carlson, Ph.D., Lisa DeStefano, B.S., Michael Wenger, Ph.D., Melissa Craft, Ph.D., Linda Hershey, M.D., Jeremy Hughes, M.D., Dee Wu, Ph.D., Lei Ding, Ph.D., Han Yuan, Ph.D.

1University of Oklahoma, Norman, OK, 2University of Oklahoma Health Sciences Center, Oklahoma City, OK

Amnestic Mild Cognitive Impairment (aMCI), a condition in which the memory functions of cognition are significantly impaired, is an established risk factor for Alzheimer’s disease. Electroencephalography (EEG) is a tool capable of measuring the dynamics of the brain’s neural networks, and is thus an important means in analysis and understanding of aMCI. In this proof-of-concept study, we compared the brain activation patterns of four healthy subjects with those of ten aMCI subjects during sleep by employing a 64-channel EEG data collection system. The power spectrum was analyzed to identify sleep stages, while spectral topography and source imaging techniques were employed to study the fluctuating patterns of the brain. Results of this study show a stage-dependent modulation in EEG power across all sleep stages and frequency bands for aMCI subjects. Additionally, results for source imaging analysis indicate decoupling of the default mode network occurring in stage one of sleep for aMCI subjects, earlier than is expected for healthy subjects. In the proof-of-concept study, our exploratory analysis demonstrated the feasibility of imaging dynamic network organization using EEG in aMCI.
Highly efficient non-viral VEGF gene delivery to human mesenchymal stem cells by phage-like nanoparticles

Mengmeng Zhai, Lin Wang, Penghe Qiu and Chuanbin Mao
Department of Chemistry and Biochemistry, University of Oklahoma, Norman, 73019, United State.

Vascular endothelial growth factor (VEGF) is a protein that can induce the formation of blood vessels. Therefore, it is used for treating bone diseases such as bone defects and cardiovascular diseases such as myocardial ischemia. Non-viral VEGF gene therapy using mesenchymal stem cells (MSCs) is considered as a more effective way than the direct injection of VEGF protein into defect sites. In this project, nanoparticles as vectors, which are a kind of novel dendritic nanoparticles in our lab, carry the VEGF gene into the MSCs using the sleeping beauty system (SB). Firstly, a MSCs’ targeting peptide, -DYHDPSLPTLRK(DYH), was found through biopanning. Secondly, the VEGF gene, targeting peptide(DYH), and nuclear localization signal (NLS) will be packed onto the dendritic nanoparticles to form a VEGF-NLS-DYH nano-complex. Thirdly, the VEGF-NLS-DYH nano-complex will be delivered into the MSCs. Ideally, MSCs will produce fresh VEGF proteins through gene expression and differentiate into functional cells such as bone forming cells or cardiac muscle cells. The transfection efficiency, cytotoxicity assay, and osteogenesis assay will be confirmed later. Therefore, the VEGF gene delivery with the dendritic nanoparticles will be a promising method in stem cell therapy.
Crawling is important to an infant's physical and cognitive development because of its role in facilitating the learning of coordinated movement and spatial skills. The objective of our study is to better understand the brain structures involved in the generation of crawling behavior and how these structures change with learning and maturation.

Typically developing infants, ages 4 to 8 months, were exposed to an assistive crawling robot for 15 minutes, twice a week, for up to 12 weeks. An infant lays in a prone configuration in reach of the floor, with the robot supporting her weight. A kinematic capture suit measures the infant's limb and trunk posture in real time. The robot responds to the limb movements and the forces produced by the infant, by carrying the infant in the indicated direction. Once per week, EEG was measured during exposure to the robot using a 124-channel sensor net.

Our goal is to identify the spatial and temporal characteristics of the EEG signals that correlate with the movement of the left arm, right arm, or feet. The power spectrum is computed using the Fourier Transform for a set of ten electrodes arrayed coronally along the central sulcus. We use a machine learning-based classifier (a Support Vector Machine), to predict specific limb movements as a function of the power spectra. We then use a variable importance method to assess which electrode/frequency pairs are most critical for predicting the limb movement.

Preliminary results indicate that the variable importance method is able to identify specific electrode/frequency pairs that are significantly predictive of movement of a specific limb (e.g., discerning movement of the left arm vs not moving the left arm). In addition, in one infant, the importance analysis shows an age-related shift in the significant frequencies from 12-17 Hz (in session weeks 1-6) to 3-8 Hz (in session weeks 7-12). This latter frequency range is consistent with the expected mu-rhythm frequency band for this age of infant. Furthermore, it is during these latter sessions in which the infant is most skillfully engaging with the robot to navigate within her environment.
Electric Field Induced Acoustic Tomography

Nicklas W. Dang¹, Ali Zarafshani¹,², Shanshan Tang¹, Bin Zheng² and Liangzhong Xiang¹
¹True Lab, School of Electrical and Computer Engineering, University of Oklahoma, Norman, OK 73019. ²CAD Lab, Stephenson Cancer Research Center, School of Electrical and Computer Engineering, University of Oklahoma, Norman, OK 73019

Research Purpose: Techniques such as irreversible electroporation and electrochemotherapy are becoming more common place in clinical settings for targeted ablation and drug delivery. As a result, a need has arisen to develop innovative techniques which can adequately characterize, in situ, the applied electric field. Real time monitoring of the applied electric field allows for efficient targeting of the therapy as well as the ability to provide corrective action during treatment. While some other techniques have been proposed, they are not able to provide real time monitoring of the electric field (i.e. CT or sonography) or require expensive equipment (i.e. in MREIT). The proposed technique will provide in situ, real time monitoring of the electric field distribution based on electric field induced acoustic signals.

Methods: An electric field is produced through the application of a high intensity, ultra-short electric pulse at a repetition rate up to kHz. The size and strength of the electric field can be altered using the system setup by varying the pulse width between 10 usEP and 100 nsEP and varying the voltage intensity from 0 – 1200 V. This pulse produces an electric field of approximately 17.1KV/cm at 1mm spacing of the electrodes, typical of the electric fields used in electrotherapy. Application of the pulse to the target through electrodes induces acoustic wave generation. The ultrasound is then captured by a transducer with center frequency of 500 kHz, amplified through a 60dB amplifier, and then processed to produce a recreation of the electric field distribution.

Results: The proposed technique has the possibility of providing real-time, in-situ, affordable, fast, and sensitive feedback for various clinical electrotherapy techniques. The data collected shows that acoustic wave generation through the application of high intensity short duration pulses is feasible. By varying the pulse width between 10us to 100ns and the voltage intensity between 0 -1.2 kV, different electric field intensities were tested for acoustic signal generation. Specifically, acoustic wave amplitudes of approximately 30 mV can be produced at voltages as low as 600V. Further, signal position can be demonstrated to move as the distance between the transducer and electrodes is changed.

Conclusions: Since the same electric pulse applied for therapy can be used to produce the acoustic waves needed for this monitoring technique there is real potential for, real time, in-situ monitoring of various electrotherapies through this method.
Stem cells are a promising cell source for cartilage regeneration. One of the challenges of using stem cells in cartilage repair is promoting their chondrogenic differentiation in a materials-only manner. Our approach focuses on establishing high throughput techniques of finding cell-binding peptides with the ability to stimulate chondrogenic differentiation in stem cells. Decreasing the binding sequences to a small peptide is advantageous due to the peptide’s properties comparing to the large molecules; peptides are easy to synthesize in large quantities, and their small size decreases the chance of any non-specific binding. Incorporating these peptides in a biocompatible and biodegradable frame that provides the structural integrity suitable for native cartilage can lead to enhancing chondrogenic differentiation of scaffolds and promoting clinical translation. By distinguishing overlapping sequences in target molecules of interest, we have identified proprietary peptide sequences. The chondrogenic potential of the peptides was analyzed for both groups of cells cultured on surfaces coated with the peptides and cells cultured on uncoated surfaces with the presence of soluble peptide in their medium. Different concentrations of the peptides were examined to explicate dose effect versus negative and positive (TGF-β3) controls. The preliminary gene expression results have illustrated that the peptides have the potential to promote chondrogenic differentiation of rBMSCs and are promising options for producing bioactive scaffolds with the capacity of advancing stem cell chondrogenic differentiation.
Resting State Networks Estimated from EEG: A Simulation and Real Experiment Study

Chuang Li¹, Han Yuan, Ph.D.¹, Diamond Urbano, M.D.², Yoon-Hee Cha, Ph.D.², Lei Ding, Ph.D.¹

¹University of Oklahoma, Norman, OK, ²Laureate Institute of Brain Research, Tulsa, OK

Spontaneous and large-scale activity fluctuations that are temporally correlated and spatially organized exist in awake and resting brains. Blood oxygen level-dependent (BOLD) functional magnetic resonance imaging (fMRI) has discovered and classified the networks formed by these fluctuations as resting state networks (RSNs). The spatial patterns of RSNs derived from fMRI appear consistent across healthy subjects and altered RSNs have been related to various neurological and psychiatric disorders, such as Alzheimer’s disease, multiple sclerosis, and major depressive disorder. Recently, novel methods of reconstructing RSNs from high-density magnetoencephalography (MEG) and electroencephalography (EEG) have also been developed to investigate electrophysiological aspects of RSNs, and to identify the links between electrophysiological and hemodynamic RSNs. In these studies, independent component analysis (ICA) plays an important role in decomposing mixed data into independent components (ICs), and RSNs were found within these ICs. With increasing researches have derived RSNs from EEG data independently, its capacity to derive RSNs needs to be evaluated. One concern is whether ICA is an accurate method to reconstruct RSNs. In the present study, the performance of two modalities of ICAs was evaluated using both simulated and real EEG data. For simulated experiment, the results were compared with the ground truth. Their spatial, temporal and spectral reconstructions were evaluated through statistical analysis. For real resting-state data obtained from seven healthy participants, the ground truth of RSNs is unknown. Therefore, cortical RSN templates constructed from resting-state fMRI data were used as reference because fMRI RSNs were well studied and showed great consistency in different studies. The results generated from real EEG data were compared to the templates and the spatial similarity with the templates were evaluated. Results from simulated data indicated that ICA has great performance in reconstructing spatial, temporal, and spectral feature of RSNs. Results from resting-sate EEG data in seven healthy participants also showed the spatial similarity to the RSN templates from fMRI to certain degree. Moreover, the comparison between two modalities indicated that different ICA method may generate different results and their performance also varies.
Examination of Erythrocyte Microparticle Formation In A Microfluidic High Shear Environment

James P. Buerck¹, Dimitrios V. Papavassiliou¹ PhD, Tyler Snyder² PhD, David W. Schmidtke³ PhD, Edgar A. O'Rear¹ PhD
¹University of Oklahoma, Norman, OK, 73019 USA; ²Integris Advanced Cardiac Care, Oklahoma City, OK, 73112 USA; ³The University of Texas at Dallas, Richardson, TX, 75080 USA

Introduction: Blood flow in ventricular assist devices (VAD) exposes red blood cells (RBC) to uncharacteristic high shear that increases the likelihood of RBC damage. Trauma to RBCs as the result of increased shear flow can result in membrane damage or hemolysis. Other groups have shown an increase in microparticles found in circulating blood of patients with implanted VADs. One suggested reason for this phenomenon is that the erythrocytes use the microvesicle formation as a means to prevent premature clearance of a still otherwise healthy cell. In working towards the improvement of VADs, developing a quantifiable response to trauma on RBCs is critical.

Materials and Methods: Venous blood was collected from a pool of healthy, human donors then separated and re-suspended with a final hematocrit of ~37% for studies. Microfluidic channels were created using poly-dimethyl siloxane (PDMS) at a 1:10 ratio of curing agent to base poured over a mold made through use of negative photolithography. The microfluidic channels used a constriction to control shear rate, with exposure times of 0 ms, 1 ms, 5 ms, 10 ms and 15 ms in a high shear region. For each channel, while the exposure time to high shear was varied, the shear rate was kept constant. The 0ms exposure time was used as a control where the micro channel had no constriction. After washed RBCs were flowed through the channel they were analyzed using a BD C6 flow cytometer and fluorescent markers.

Results and Discussion: Using a gating scheme to ensure positive marking for the two fluorescent markers, Annexin V-Alexa Fluor 647 and CD235A-FITC, we witnessed a positive trend in microparticle formation. The Annexin V is a positive marker for exposure of external phosphatidylserine that suggests apoptotic activity in other cells. CD235A serves to absolutely mark RBCs and RBC microparticles. The data collected from the flow cytometer indicated a trend in RBC microparticle formation in the size range from 0.5 to 1.0 μm. The increase in microparticle formation from the control to the longest exposure time showed over a ten-fold increase.

Conclusions: Microparticle formation with exposed phosphatidylserine occurs after high shear of 0-15 millisecond duration. Future sights are set on determining a molecular marker for positive shear damage to erythrocyte membrane damage and cell clearance.
Measuring and quantifying the neurovascular relationship in cerebral tissue is important in understanding brain function at normal and diseased conditions. The study of neurovascular coupling requires simultaneous measurement of neuronal activity and vascular momentum. In human studies, the images are typically achieved by electroencephalography (EEG), which directly records electrical neural activity, and functional Magnetic Resonance Imaging (fMRI), which acquires hemodynamics when using the blood-oxygenation-level-dependent contrast. fMRI, however, is currently prohibitively expensive, and the equipment is too large to be used in a standard clinical setting. Additionally, fMRI has poor temporal resolution that leads to aliasing in hemodynamic measurement. Like fMRI, functional near-infrared spectroscopy (fNIRS) can be used to measure hemodynamic activity in the human brain, including oxygenated and deoxygenated status. EEG and fNIRS are reasonably affordable and adaptable, and both have adequate temporal resolution. A multimodal neuroimaging device utilizing EEG and fNIRS can potentially be used in various clinical settings, including outpatient clinics. In our current study, we investigated the auditory response in simultaneously acquired EEG and fNIRS data.

Three healthy subjects were recruited and completed data collection in two sessions, an eyes-open and eyes-closed session. Sessions consisted of three trials with different body positions: standing, sitting, and supine. A mixed block- and event-related design was used to accommodate the dramatically different temporal scale for electrical and hemodynamic responses. EEG and fNIRS data of the brain response to the monaural auditory stimulus was simultaneously recorded. Our results showed auditory evoked potentials (AEP) to individual stimuli from EEG recordings. Meanwhile, concurrent hemodynamic response to a train of auditory stimuli was observed. Topographies of AEP and fNIRS showed a common pattern with peaks originated from the auditory regions.

To our knowledge, this is the first demonstration of simultaneous, whole-brain and high-density recording of EEG and fNIRS responses to auditory stimuli. Our study demonstrated the feasibility of the multimodal neuroimaging using simultaneous EEG and fNIRS. Such noninvasive, broadly accessible imaging capability holds potential for studying neurovascular coupling in the human brain.
Real-time EEG artifact correction during fMRI using ICA

Ahmad Mayeli, MS1,2, Vadim Zotev, Ph.D.1, Hazem Refai, Ph.D.2, Jerzy Bodurka, Ph.D.1,3
1Laureate Institute for Brain Research, Tulsa, OK, 2Department of Electrical and Computer Engineering, University of Oklahoma, Tulsa, OK, 3Biomedical Engineering Center, University of Oklahoma College of Engineering, Norman, OK.

Concurrent EEG-fMRI provides an important tool for investigating human brain function. However, simultaneous recording EEG and fMRI results in EEG signal contamination from imaging and ballistocardiogram (BCG) artifacts [1]. Automatic artifact correction of EEG data in real time for different real-time applications, such as neurofeedback studies, is the subject of ongoing research. To date, average artifact subtraction (AAS) is the most widespread real-time method used to partially remove BCG and imaging artifacts without requiring extra hardware equipment; no alternative real-time methods for removing other artifacts are available.

We introduce a novel and improved approach for real-time EEG artifact correction during fMRI (rtICA). The rtICA is based on real-time independent component analysis (ICA) and it is employed following AAS method. The proposed method was implemented and validated during EEG and fMRI experiments on healthy subjects. In real time, we detect and suppress eye blinks, motion artifacts, and residual BCG, imaging and muscle artifacts. We compared our real-time artifact reduction results with various offline methods using multiple evaluation metrics, including power analysis. Our results demonstrate that the rtICA employed after the rtAAS can obtain 98.4% success in detection of eye blinks, 4.4 times larger INPS reductions compared to RecView-corrected data, and effectively reduce motion artifacts, as well as imaging and muscle artifacts, in real time on six healthy subjects [2]. Specifically, the EEG frequency bands power comparison shows a significant reduction in delta and theta frequency bands, and removes muscle and residual imaging artifacts in high frequency bands (30 Hz and higher) when compared to real-time AAS (rtAAS) correction alone. Importantly our correction does not affect brain neuronal activities as reflected in other EEG bands, including alpha band.

References:
OSW-1 Analogs for Anti-body Drug Conjugate

Anh T. Le¹, Cori A. Malinky¹, Anthony W. G. Burgett, Ph.D.¹
University of Oklahoma, Norman, OK

OSW-1 is a potent anti-cancer natural product isolated from the bulbs of *Ornithogalum saundersiae* flower. OSW-1 exerts its biological effects through binding to oxysterol-binding protein (OSBP) and oxysterol-binding protein related protein 4 (ORP4), without selectivity. While OSBP is ubiquitously expressed in all tissues, ORP4 only shows high expression level in leukemia cells and plays an essential role in cancer cell proliferation. This suggests that ORP4 can be a promising target for precision medicine in cancer treatment. Hence, there is a need to understand how OSW-1 interacts with the cellular components to result in its potent anti-cancer activity, and to increase the specificity of OSW-1 to cancer cells.

One of our strategy to increase the specificity of OSW-1 compound to cancer cells is through antibody-drug conjugation (ADC) approach. For ADC, OSW-1 analogs with a linker were synthesized. The linker was installed on the 3” or 4” position in the disaccharide moiety of OSW-1 through a carbamate functional group with free amine handle. With the free amine handle installed, the potent anti-cancer molecule OSW-1 can be conjugated to antibody that specifically target cancer cells, allowing the selective delivery of OSW-1. Besides the possibility to be conjugated to antibodies, the amine functional group will allow us to synthesize different OSW-1 analogs for various biological studies. We can attach a fluorescent label on OSW-1, or use this amine handle to enhance the solubility of the compound in water.
Electroencephalography from Infant Motor Brain at Time of Crawling

Alejandro Patino\textsuperscript{1}, Andrew H. Fagg, Ph.D.\textsuperscript{1}, Thubi H.A. Kolobe Ph.D.\textsuperscript{2}, David P. Miller, Ph.D.\textsuperscript{1} and Lei Ding, Ph.D.\textsuperscript{1}

\textsuperscript{1}University of Oklahoma, Norman, OK, \textsuperscript{2}University of Oklahoma Health Sciences Center, Oklahoma City, OK

Crawling is an important first step in locomotion capability development of infants as their primary method of interaction with the environment. Infants that lack necessary skills for crawling can be severely hindered in the development of cognitive functions. Electroencephalography (EEG) can be used to study the development of various regions of the human brain. Extensive studies have been performed on EEG signals from adults regarding the function of motor cortex, but few have been conducted on infants, especially with the focus on development. The purpose of this longitudinal study is to monitor the development of infants between 4-9 months old immediately before they develop crawling skills. High-density EEG (i.e., 124 channels) was recorded in multiple infants and EEG power densities at various frequency bands were analyzed and compared longitudinally. Spatial topographic visualization was performed alongside power spectrum densities to identify locations of scalp activity in each frequency band. Our preliminary results indicated that the mu rhythm in infants was characterized at lower frequencies than that of adults. As an infant learns to crawl, a shift in this frequency band of the mu rhythm toward higher frequencies is observed. It is suggested that the maturation and frequency band shifts of the mu rhythm can be used as biomarkers for monitoring infant motor skills development. EEG signatures of individual infants can be compared to those of a group average of healthy infants to identify their differences. Our ongoing study is aimed to collect more EEG data from both typical developing infants and those at risk of cerebral palsy to evaluate the clinical value of infant mu rhythm in neuromotor disorders.
Introduction: Tendon injuries are a common occurrence among both athletes and the general population. In this present research, we utilize the vein from the human umbilical cord (HUV) as a scaffold for an engineered tendon construct as it promotes cell adherence and has a mechanical strength one order of magnitude less than certain tendons. Longitudinal stretching of these constructs in a bioreactor has shown to promote Mesenchymal stem cell (MSC) differentiation into tenocytes. We have investigated the effects of stretching frequency and duration on cell proliferation and differentiation.

Materials and Methods: We are investigating the time dependent effects of mechanostimulation. For this study, human umbilical veins (HUVs) are seeded with MSCs and cultured within custom bioreactors. During culture, the HUVs are subjected to cyclical 2% strain for differing durations and frequencies before being removed and examined for cellularity, maximum tensile strength and tendon gene expression. Stretching times will vary from 5 minutes to 2 hours at a time with frequencies varying from 2 cycles/min to 0.5 cycles/min. The shorter durations will be done in groups that add up to 1 total hour (5min 12/day, 15min 4/day, etc).

Results and Discussion: The HUV constructs exhibited significant differences in cellularity, ECM alignment and tendon gene expression compared to traditional stretching regimens of 1 hour every 24 hours. Shortening the stretch time while increasing the frequency of stretching beneficially reduces the stress the cells are under, increasing proliferative capabilities without reducing the differentiation potential. Longer stretching times of 2 hours a day greatly reduce proliferative capabilities but tendon gene expression (scleraxis, tenomodulin) is significantly increased under those conditions. Unfortunately, after 7 days there are no significant differences in tensile strength of construct. Literature implies that large differences will be evident at 14 days after the cells have had ample time to remodel the surrounding ECM.

Conclusions: The duration of mechanical stimulation has a profound effect on the tendon properties of our constructs. Currently, the highest maximum tensile strength we’ve achieved in lab is roughly 6 MPa, a bit less than the 10 MPa needed to be an acceptable rotator cuff tendon replacement, but further alterations could lead to a large enough increase for clinical applications.

References:
1 Bureau of Labor Statistics, Number of nonfatal…affected by injury or illness, private industry, 2009.
This work presents a new motion capture system designed for crawling infants. The motivation for the development of this system is an experimental robotic device called SIPPC. SIPPC provides physical therapy to crawling-age infants with motor disorders. As part of the therapy process, it monitors crawling motion. Currently, this is done by using a motion capture system with wearable inertial sensors. In certain environments, the sensors are prone to electromagnetic interference. This new motion capture system was developed to augment the existing system in such environments.

The new system captures motion with the help of planar square-shaped markers. These markers are placed on the body and tracked by cameras. The markers have unique black and white patterns similar to QR codes. A single camera can track these markers in 3D. The use of a single camera is made possible by solving a Perspective-n-Point (PnP) problem. The PnP problem is set up as follows. By comparing the apparent geometry of the 4-corner marker in the camera image to its known geometry, the 3D location and orientation of the marker can be computed.

This new motion capture system has several advantages. It is very suitable for use with infants since they cannot be engaged for extended periods. The markers can be put on very quickly as compared to commercial marker-based systems. And unlike commercial marker-based systems, calibration is not required every time the system is used. The new system is very portable. All the hardware can fit inside a backpack for easy transport. It is small enough to be used onboard a mobile robot like the SIPPC. The advantages of this system come at a cost. The wearable markers must be larger than those in typical marker-based systems. So these need more space on the body. In addition, the accuracy is less than that of the best commercial marker-based systems.

Preliminary testing of the system has shown that under various conditions, the tracking accuracy for this method varies between 10 mm and 30 mm up to a distance of 1 m. A cursory literature review suggests that this is sufficient to capture crawling motions of infants. For future work, the system will be tested more thoroughly and under more realistic testing conditions. With a more detailed literature review, this will help determine optimum marker sizes for infant motion capture.
Identification and Assessment of Bone Morphogenic Protein Receptor Binding Peptides in Osteoblast Differentiation Signaling Pathway

Kegan S. Sunderland, Ph.D. Candidate¹, Chuanbin Mao, Ph.D.¹,²
¹University of Oklahoma, Norman, OK, United States, ²Zhejiang University, Hangzhou, Zhejiang, 310027, China

Bone morphogenetic proteins (BMPs) are part of the transforming growth factor-β superfamily and function as key regulators of cellular growth, differentiation, and tissue formation. While recombinant BMPs can be used to induce osteoblast differentiation, they are expensive to produce. In contrast, peptides can be efficiently produced in massive quantities through phage display. It would therefore be highly valuable to develop short peptides capable of mimicking BMPs which could be displayed on human-safe M13 phage. I have discovered 4 short, 12-mer peptide sequences (full sequences are not given for patent purposes) through a selective process called biopanning. These 12-mer peptides have been shown to bind the BMP receptors on live cells by immunofluorescence and bind the BMP receptors in an ELISA when genetically displayed on the p3 coat protein of M13 bacteriophage. In addition, the peptides were genetically displayed on the p8 (~2700 copies) coat protein of M13 bacteriophage to greatly increase the number of copies available for binding relative to the p3 (5 copies) coat protein. These newly engineered phages were incorporated into phage films having aligned ridge groove surface topographies. Human mesenchymal stem cells cultured on these films demonstrated varying levels of differentiation according to the peptide displayed as well as the concentration of the peptide. Differentiation has so far been confirmed by immunofluorescence of the marker protein osteopontin, but future studies will also study osteocalcin in addition to real-time PCR results for osteoblast marker genes.
Development of a Multi-Scale Mitral Valve Computational Model for Patient-Specific Treatment Planning

Devin Laurence¹ and Chung-Hao Lee, Ph.D.¹
¹School of Aerospace and Mechanical Engineering, University of Oklahoma, Norman, OK 73019

The mitral valve (MV) is an essential component to the regular function of the heart as it regulates unidirectional blood flow and prevents the heart function from regurgitation. There exists an approximate 5% annual mortality rate for severe mitral regurgitation without surgical intervention[1]. Approximately 40,000 mitral valve surgical repairs are performed annually in the United States; however, recent long-term studies showed an unsatisfactory recurrence rate of severe mitral regurgitation 3–5 years after initial surgery[1-2].

Varying valve structure between patients results in a wide array of modes of regurgitation such that one method of treatment may not be commutable across all patients. To address this variation, it is necessary that patient-specific treatment be emphasized to allow for better realization of the patient’s pathologies and valvular geometry in a computational modeling framework. The present study aims to develop a multi-scale model, which includes both the tissue-level micromechanics and the cellular metabolism & biosynthesis to assess the mechanical and biological conditions of the MV. A computational framework for tissue-cellular microenvironment will be constructed by taking account of various aspects of the valve, such as valve interstitial cell (VIC) mechanical responses and VIC-regulated collagen biosynthesis & tissue remolding. Such innovative development will enable investigations of the effects of alterations to the valve tissue’s structure & micromechanics in response to surgery, effects of disease progression on the overall heart valve function, and of the underlying stresses experienced in the tissue. The developed computational framework will help to facilitate predictive computer simulation tools, integrated with clinical patient's image data and experimental measurements, for timely diagnosis and patient-specific surgery planning for optimally treating valvular heart diseases, including ischemic and functional mitral regurgitation, with enhanced long-term durability and reduced mortality.

References

Amplitude of Resting-State fNIRS Global Signal is Related to EEG Vigilance Measures

Yuxuan Chen, M.S.¹, Jesse Farrand², Julia Tang², Yafen Chen, B.S.², Johnny O’Keeffe, B.S.¹, Guofa Shou, Ph.D.¹, Lei Ding, Ph.D.², and Han Yuan, Ph.D.²
¹School of Electrical and Computer Engineering, University of Oklahoma, Norman OK, USA, ²Stephenson School of Biomedical Engineering, University of Oklahoma, Norman OK, USA

Global signal regression (GSR) has been widely utilized as a pre-processing approach in human brain studies using functional magnetic resonance imaging (fMRI). However, removal of the global signal in GSR and thereby introduced spurious anti-correlation become increasingly concerned. Recent studies using simultaneous electroencephalogram (EEG) and fMRI measures in human subjects suggest that global signal amplitudes of cerebral hemodynamics, as measured by fMRI, is related to physiological condition of vigilance. The nature of the global signal in cerebral hemodynamics is still not clear. In our current study, we investigated the physiological factor of hemodynamic global signal, as measured by near-infrared spectroscopy (NIRS), across eyes-open and eyes-closed resting states and in different body positions. Three healthy subjects participated in eyes-closed and eyes-open sessions. In each subject, six sessions of 10-minute resting state data were obtained, including eyes-closed and eyes-open conditions and three different body positions (standing, sitting, and supine), yielding a total of 18 datasets in the current study. Whole-brain and high-density NIRS and EEG signals were recorded simultaneously in the study participants. Our results found a negative correlation between the eye-opened and eyes-closed (EO-EC) changes in the global signal amplitudes and the respective changes in EEG vigilance. The correlation values between them was -0.53.

In this paper, we report that spontaneous fluctuation exists in resting-state global signal of functional brain images and that the fluctuation varies across eyes-closed and eyes-open conditions as well as different body positions. Moreover, we observed an inverse relationship between the changes across EO and EC in NIRS global signal amplitude and the changes in relative EEG vigilance. Our findings have important implications in the understanding of hemodynamic global signal, as measured by either fMRI or NIRS.
Neural activity inside the human brain generate electrical signals that can be detected on the scalp. Electroencephalograph (EEG) is one of the most widely utilized techniques helping physicians and researchers to diagnose and understand various brain diseases. Due to its nature, EEG signals have very high temporal resolution but poor spatial resolution. To achieve higher spatial resolution, a novel tri-polar concentric ring electrode (TCRE) has been developed to directly measure Surface Laplacian (SL). TCRE has a center disk, a middle ring and an outer ring that can measure the potentials on skin, then SL can be derived. TCRE has shown excellent performance on higher spatial resolution, higher signal-to-noise ratio (SNR) and other aspects in our previous works. The objective of the present study is to accurately calculate SL for TCRE based on a realistic geometry head model. A locally dense mesh was proposed to represent the head surface, where the local dense parts were to match the small structural components in TCRE. Other areas without dense mesh were used for the purpose of reducing computational load. We conducted computer simulations to evaluate the performance of the proposed mesh and evaluated possible numerical errors as compared with a low-density model. Finally, with achieved accuracy, we presented the computed forward lead field of SL for TCRE for the first time in a realistic geometry head model and demonstrated that it has better spatial resolution than computed SL from classic EEG recordings. The simulation of forward calculation in this work could allow us to solve high density inverse problem with SL and conventional EEG. These high density inverse mapping are going to play an important role in many clinical applications that could help doctors to diagnose brain diseases.
Super-resolution microscopy has made strides recently, providing novel avenues with which optical imaging entered the nanoscale. There does not yet exist a 3D label-free imaging modality that can image single cells at nanoscale resolutions beyond the diffraction limit. This study introduces nanoscale photoacoustic tomography, a novel imaging modality that obtains optical absorption with high frequency ultrasound resolution in order to enable label-free 3D axial super-resolution imaging.

Theoretical analysis was performed to ensure that a strong photoacoustic signal could be generated without inducing a high temperature rise in a biological sample (red blood cell). Next, a photoacoustic imaging system was built from an ultrashort (7ps) pulsed laser (532nm), two photodiode detectors, an optical delay line, and beam geometry/polarization components. The laser set-up featured a pump-probe imaging motif with a chopped pump beam and confocal pump and probe beams. Experimental results have demonstrated the system's first signal of a thin steel sheet.

The theoretical analysis demonstrated the thermal safety of the system. The resolution of the system, was calculated to be ~10nm in the axial direction. In experiment, the system was able to obtain a detectable and repeatable signal on the order of GHz.
A Novel Bioreactor System for Rotator Cuff Tendon Tissue Engineering

Jin Liu¹, Julien Arrizabalaga¹, Matthias Nollert¹,²

¹ Stephenson School of Biomedical Engineering, ² School of Chemical, Biological and Materials Engineering
University of Oklahoma, Norman, OK 73019

Rotator cuff tears are a common cause of pain and disability caused by injury and degradation of rotator cuff tendons. More than 250,000 patients require surgery each year, a majority of whom are over the age of 40. With failure rates estimated between 20% and 54%, rotator cuff tendon repair remains a challenge in orthopedic surgery. We propose a novel solution to engineer rotator cuff tendon by using the human amniotic membrane as a scaffold, adipose-derived stem cells as a cell source, and mechanical stimulation as a signal to promote tenocyte differentiation. Four layers tissue constructs are obtained by adhering together membranes with fibrin glue in between each layer. Tissue constructs are then placed in the bioreactor for culturing and stretching over time, promoting the tenocyte differentiation of the adipose-derived stem cells. The object of this study is to design a four-channel bioreactor providing adjustable cyclic mechanical stretching and appropriate tissue culture environment for four tissue samples at the same time.
The objective of this study was to compare the detectability of simulated objects within a dense breast phantom using high energy x-rays for phase sensitive breast imaging in comparison with conventional imaging systems. Two phantoms of 5 cm thickness were used which represented a compressed breast consisting of 70% glandular and 30% adipose tissue ratio by weight in homogenous and heterogeneous backgrounds. The phantom had a 6 × 6 matrix of holes with milled depths ranging from 1 to 0.1 mm and diameters ranging from 4.25 to 0.25 mm representing simulated tumors. The phase contrast images were acquired using a prototype with a micro-focus x-ray source operated at 120 kVp, 4.5 mAs and 1.3 mGy with a magnification factor (M) of 2.5 and a detector with a 50 µm pixel pitch. Conventional images were acquired with the Hologic Selenia system at 29 kV, 142mAs, 1.57mGy and with the prototype at 40 kV, 12.5 mAs. The observer study and CNR analyses indicated that the phase contrast image in a homogenous background had higher disk perceptibility and detection as compared to conventional images. The Hologic acquired image had a CNR values and higher disc detection as compared to the conventional image acquired with the prototype. The CNR values of the disks with the phase contrast image in the heterogeneous background dropped at a smaller rate which allowed the perceptibility and detection of more disks as compared to the conventional image. The potential demonstrated by this study for imaging a dense breast with a high energy phase sensitive x-ray imaging to improve tumor detection in warrants further investigation of this technique.
Novel 3-D in vitro system sets the stage for a neoteric therapeutic approach of Idiopathic pulmonary Fibrosis

1Rabab Sharif, 2Alan Betensley, 1,3Dimitrios Karamichos
1Department of Cell Biology, University of Oklahoma Health Sciences Center, Oklahoma City, OK 73104, USA.
2Nazih Zuhdi Transplant Institute, Integris Baptist Medical Center, Oklahoma City, OK 73112, USA. 3 Department of Ophthalmology/Dean McGee Eye Institute, University of Oklahoma Health Sciences Center, Oklahoma City, OK 73104, USA.

Idiopathic pulmonary fibrosis (IPF) is a progressive interstitial lung disease with a median survival of approximately 3 years. IPF is a disease associated with aging and an estimated prevalence as high as 42.7 per 100,000 and 22.72 per 100,000 in the United States. It is characterized by fibroblast proliferation and extracellular matrix (ECM) remodeling. This promotes irreversible damage of the lung’s architecture, leading to respiratory failure in IPF patients. Current studies suggest that IPF is a result of abnormal wound healing in response to various sites of ongoing alveolar epithelial injury that ultimately evolves to fibrosis. Only two antifibrotic agents are approved for human use in the United States, and they do not arrest or reverse IPF progression but only slow it down; nintedanib, a tyrosine kinase inhibitor and pirfenidone, a known TGF-β inhibitor. In this study we developed a novel 3D in vitro tissue engineering model consisting of normal human lung fibroblasts (NHLF) or idiopathic lung fibroblasts (IPF). Both cell types were stimulated by TGF-β1, and TGF-β3, and a stable derivative of vitamin C to stimulate ECM assembly. Western blot, qRT-PCR, Indirect-immunofluorescence (IF), and Transmission electron microscopy (TEM) were performed. Untreated NHLFs and IPFs served as controls. Data revealed that IPF constructs express significantly higher levels of collagen III when compared to NHLF that showed no expression of the fibrotic marker. In addition, IPF cells were able to secrete and assemble a thicker ECM when compared to NHLF, mirroring what is seen in vivo. Our preliminary data shows that when testing the role of TGF-β3, significant downregulation of specific fibrotic markers such as collagen III and smooth muscle actin (SMA) is observed. TGF-β3 also led to significant increase in ECM assembly for NHLF, while preserving the same amount of ECM for IPF. Future studies will utilize this model to determine whether TGF-β3 is a valid therapeutic candidate. We will also use this model as a drug screening tool to determine the efficacy and safety of current and future IPF drugs. Overall, this is a very promising tissue engineering approach as we attempt to clinically improve lung tissue turnover and delay or prevent IPF.
Bone Regeneration in Critical Size Calvarial Defects Utilizing Natural Materials and Growth Factors In Vivo

J. M. Townsend¹, S. C. Dennis², J. Whitlow², M. S. Detamore¹, C. J. Berkland³

¹Stephenson School of Biomedical Engineering, University of Oklahoma, Norman, OK, ²Bioengineering Program, University of Kansas, Lawrence, KS, ³Pharmaceutical Chemistry, University of Kansas, Lawrence, KS.

Novel paste-like scaffolds incorporating natural materials such as demineralized bone (DBM), decellularized cartilage (DCC), hydroxyapatite (HAp), and hyaluronic acid (HA), were used to bridge the gap in fracture healing mechanics. This technology can have an important impact in treating traumatic brain injuries (TBI). Current TBI treatments include removal of a portion of the calvarial bone to allow brain swelling. Current methods require a secondary surgery to close the cranial vault once brain swelling has subsided, increasing the cost and risk to the patient. Our paste-like material can be implanted with the initial surgery, providing a single-stage surgical intervention to treat TBI and eliminate the need for a secondary surgical procedure. The paste material is able to regenerate bone in critical size defects and remain pliable during the brain swelling process. 7.5 mm calvarial defects were created in rat models and paste-like scaffolds were implanted containing a combination of ECM materials and BMP-2 and/or VEGF. Results were analyzed after 8 weeks using micro-computed tomography (microCT) to assess and quantify bone formation. Contrasting and reconstruction analysis are used to track newly formed bone within the defect site and bone that has formed and migrated out of the desired area. Histology and immunohistochemistry were used to assess deposition of desired ECM components in the regenerated tissue. MicroCT results indicate a statistically significant increase for the group containing BMP-2 without ECM materials, and for the group containing decellularized cartilage without growth factors, compared to the sham (Fig. 1). Comparable bone growth between the ECM only group and growth factor containing groups highlight the potential of naturally derived materials in regenerative medicine. The bone regeneration rate shows promise for applications requiring tailored bone growth.

Figure 1. A) In vivo regenerated bone volume. Bone regeneration was evaluated after 8 weeks using micro-computed tomography (µCT) analysis. Asterisks (**) represent statistically significant results (p<0.01) compared to the sham (n = 4-5). Error bars represent standard deviations. B) µCT analysis using Avizo Fire software. Orange coloring indicating the regenerated bone, and blue coloring defining original colloidal particles. Scale bar = 5 mm.
Assessing rTMS Effects in MdDS: Cross-modal Comparison between Resting State EEG and fMRI Connectivity

Yafen Chen, B.S.¹, Chuang Li, B.S. ¹, Guofa Shou, Ph.D.¹, Diamond Urbano, B.S.², Yoon-Hee Cha, M.D.², Lei Ding, Ph.D.¹,², Han Yuan, Ph.D.¹,²
¹University of Oklahoma, Norman, OK, ²Laureate Institute of Brain Research, Tulsa, OK

Background: Repetitive transcranial magnetic stimulation (rTMS) has been explored in treating several neurological and neuropsychiatric conditions, including a chronic neurological disorder of imbalance, called Mal de Debarquement Syndrome (MdDS). However, optimizing the therapeutic effect of rTMS depends on reliably assessing brain state conditions. Neuroimaging has been suggested for identifying biomarkers related to the rTMS treatment.

Materials and Methods: Ten subjects were recruited in the clinical trial (NCT02470377) and completed the recording of simultaneous EEG and fMRI both before and after rTMS protocol. Raw EEG data were de-noised from MR-related and cardiac artifacts. Resting state fMRI connectivity was computed as Pearson’s correlation with respect to a seed region in the entorhinal cortex. Based on scalp EEG measurements, the source dipoles were constructed by using individual physical forward models. Sources of EEG traces were down-sampled to time points at microstates and then concatenated microstates were subject to group-level independent component analysis. Spectral dynamics of the independent component were reconstructed from temporally concatenated EEG data and were reorganized into new pre- and post-TMS sessions for each subject.

Results: Based on the post-rTMS assessments using the visual analog scales, three subjects were classified as positive responders (symptom reduction ≥ 10), three as negative responders (symptom exacerbation ≥ 10), and 4 as neutral responders (change of symptoms < 10). The EEG network with highest spatial correlation coefficient to the fMRI default mode network (DMN) template was chosen as the EEG DMN of interest. Comparing EEG data to fMRI data, the results yielded significant negative correlation between frequency shift of EEG network due to rTMS treatment and connectivity changes in fMRI network (r = -0.75, p = 0.01). Both EEG and fMRI modulations were marginally related to the symptoms changes in the cohort of 10 subjects (r = 0.49 for EEG and r = -0.60 for fMRI).

Conclusion: Our study demonstrated the feasibility of imaging dynamic network organization using multimodal neuroimaging in MdDS. With the goal of optimizing an rTMS protocol, both EEG and fMRI hold promise as therapeutic markers in overseeing rTMS treatment effects, while EEG is more broadly accessible than fMRI.
The detection of breast cancer early in its development remains to be a pervasive challenge of modern medicine. Current breast cancer screening methods such as mammography have been extremely useful in the detection of cancer in its early stages, but these methods have unfortunate limitations such as the sheer amount of false-positive results, particularly in the case of ductal carcinoma in situ (DCIS), where the cancer is detected by the presence of micro-calcifications, small accumulations of calcium. According to the National Cancer Institute, more than 50% of women screened annually in the United States will at some point in their lives receive a false-positive result for breast cancer. Improved screening methods are therefore essential to preventing these unfortunate calls. Taking up this challenge, we present a new imaging technique, X-ray induced Acoustic Computed Tomography (XACT), which, unlike mammography, creates three-dimensional images of tissue, yielding vastly more information than conventional mammography. XACT runs off a groundbreaking new physics discovery, that X-rays can generate ultrasound waves within tissue. In conventional mammograms, X-rays are used to generate mere two-dimensional images of breast tissue. XACT, in contrast, uses a much smaller X-ray dose to generate three-dimensional images via acoustic waves. Using XACT, we have demonstrated the successful reconstruction of gold fiducial markers and the OU logo with using an effective new system. Simulation results have also illustrated that the XACT system has the ability to detect micro-calcifications as small as 100µm in size. The dose required by the proposed XACT configuration was calculated to be 0.4mGy for a 4.5cm-thick compressed breast. This is one-tenth dose level of a typical two-view mammography for the breast with same compression thickness. Our study results indicate that this imaging device and methodology provide a rapid and high resolution approach for imaging dynamic information, and may have potential for becoming a new promising noninvasive imaging modality used in future applications.
Real-time Multi-spectral Fluorescence Microscopy

Zheng Li\(^1\), Yuchen Qiu\(^1\), Shibo Li\(^2\), Bin Zheng\(^1\), and Hong Liu\(^1\)

\(^1\)College of Engineering, University of Oklahoma, Norman, OK. Email: liu@ou.edu, \(^2\)University of Oklahoma Health Sciences Center, Oklahoma City, OK.

Fluorescence microscope is an essential tool for studying biological specimen in state-of-the-art biological and biomedical research. Fluorescence imaging relies on capturing the re-radiation of fluorescence light, which is of a particular spectrum, to localize and visualize the substances of interests. In order to achieve spectral imaging, a typical fluorescence microscope requires switching among multiple filter sets while a more sophisticated approach, such as a confocal microscope, may rely on lasers and electro-tunable filters for such task. The imaging technologies are designated for imaging fixed specimen because the imaging processes are, temporally and/or spatially, inconsistent.

We developed a custom fluorescence microscope that can simultaneously acquire multiple fluorescence spectrum for situations where temporal resolution and acquisition efficiency are critical. Instead of alternating filter sets and illuminating spots with multiple lasers, our approach utilizes configuring advanced optical components to simultaneously acquire multiple spectra. As a result, functional fluorescence microscopic imaging under x60 and x100 can be achieved at 45 frames per second rate, which should be applicable for applications where real-time performance is required.
Extracellular matrix stiffness regulates cancer cell behavior by altering membrane structures

Yuchao. Yang, M.S.¹ ², Jun. Ouyang, M.D, Ph.D.² and Xin.A. Zhang, M.D, Ph.D.¹*
¹Department of Physiology, Stephenson Cancer Center, The University of Oklahoma Health Science Center, Oklahoma City, OK.²Department of Anatomy, Southern Medical University, Guangzhou, China.

Introduction
Extracellular matrix (ECM) stiffness potently regulates cellular behaviors in various biological events. However, because of difficulties in modulating the stiffness, composition, and architecture of ECM environment independently, the role of stiffness of mechanical inputs in the cell behavior remains unclear. Here we take advantage of interpenetrating networks (IPNs), made by collagen gel or matrigel and alginate, to tune ECM stiffness independently of composition and architecture. In preliminary experiments, we found that changes in ECM stiffness affect the cell morphology by altering membrane structures such as blebs, filopodia, and lamellipodia. Membrane blebs and filopodia in three dimensional (3D) microenvironment may affect cell migration and invasion in vivo. It is known that, in human breast tumor, increased ECM stiffness may induce malignant phenotypes of normal mammary epithelial cells. So in this study, we hypothesize that changing ECM stiffness in tumor microenvironment may have a significant effect on cancer cell behaviors by altering membrane structures such blebs, filopodia, and lamellipodia.

Method
All IPNs in this study consisted of 1.5 mg/ml rat-tail collagen-I (BD Biosciences) and 10 mg/ml alginate. A solution, containing calcium sulfate dihydrate (CaSO₄·2H₂O) (Sigma), was used to crosslink the alginate network. These two solutions were mixed and immediately deposited into a well of a 48-well plate. The plate was then transferred to the incubator (at 37°C and 5% CO₂) for 1-2 hours to allow gelation, following by addition of culture media to each gel. Then MDA-MB-231 human breast cancer cells were seeded on the gel, and culture media were replenished every two days during each experiment.

Objective
1) Determine how cancer cells utilize membrane structures such as blebs, filopodia, and lamellipodia to modulate for their motility.
2) Determine how cancer cells sense the 3D microenvironment stiffness by generating various membrane structures.

Future plan
We plan to use scanning electron microscopy(SEM) and energy dispersive spectroscopy(EDS) to identify the composition and architecture of IPNs. We will also use immunofluoresensence, Western blot, and signaling approach to explore the mechanism by which altered membrane structures affect cancer cell morphology and behaviors.

References

Acknowledgement
Dr. Brian P. Grady, School of Chemical, Biological, and Materials Engineering, University of Oklahoma.
A Research Grant from Oklahoma Center for Adult Stem Cell Research (OCASCR).
Functional-MRI and Physiological Evidence of Emotional Hypo-Reactivity in the Offspring of Alcohol Abusers

B. Espinoza-Varas,1 and S. Guo2
1Communication Sciences and Disorders, 2College of Medicine, OU Health Sciences Ctr.

Introduction: Family history of alcohol use disorder (AUD) is a genetic liability expressed as a neurobehavioral-disinhibition phenotype exhibiting cognitive, behavioral, and emotional dysregulation. The hypothesis assumes lower emotional reactivity in participants with positive (FHP) than with negative (FHN) family history of AUD. Testing entails comparing FHP to FHN samples in terms of task-related or resting-state fMRI brain-activation patterns, emotion-laden probe-task performance and emotion dysregulation indices (EDI) by reviewing existing studies.

Methods: Twenty relevant studies were reviewed for differences between FHN and FHP participants in four criteria: probe-task performance, demographics and matching variables, task-related fMRI brain-activation patterns/regions, and EDI scores. Against this evidence, the following predictions were tested: a) lower probe-task performance in FHP samples; b) significant fMRI activation-pattern differences between FHP and FHN samples, in emotion-processing or behavioral-control brain regions; c) significant EDI differences; and d) significant correlation between the three predicted differences.

Results: Nine different probe tasks and six different EDIs were used with demographically-matched FHP and FHN teenagers/young adults. The existing evidence showed: a) five probe tasks produced significant between-sample performance differences; b) three studies reported between-sample activation differences in emotion-processing regions; five studies showed sample differences in behavioral-control regions; and c) fMRI activation patterns correlated closely with EDIs and physiological probe-task responses but not with behavioral probe-task performance.

Conclusion: Currently, there is conflicting and insufficient evidence demonstrating that FHP individuals exhibit emotional hypo-reactivity. Studies employing physiological emotion indices provide stronger support to the hypothesis than those employing behavioral responses.

This project was supported by the National Institute of General Medical Sciences of the National Institutes of Health through Grant Number 8P20GM103447.
Natural hydrogels, such as those made from cartilage extracellular matrix, are advantageous for cartilage regeneration because of their intrinsic bioactivity, which eliminates the need for incorporating additional biological factors. However, liquid hydrogel precursor solutions tend to leak out of defects before they are crosslinked and are difficult to deliver. To counteract this, cartilage hydrogels with a paste-like precursor were previously developed and synthesized from bovine devitalized (DVC) and decellularized cartilage (DCC). DVC hydrogels consisted of methacyrlated DVC with DVC particles, and DCC hydrogels consisted of methacyrlated DCC with DCC particles. Both materials demonstrated chondroinductivity in vitro and were further hypothesized to promote chondrogenesis in vivo; therefore, a 5-week pilot study in rabbits was conducted for an initial proof-of-concept to confirm the chondrogenic properties previously seen and to demonstrate the efficacy of a paste-like hydrogel precursor in delivery and placement within osteochondral defects. During the surgical procedures, surgeon feedback demonstrated the ease of delivery of the paste-like precursor to the defect site through a needle without leakage prior to UV crosslinking. Histological analyses by H&E and Safranin O staining showed a non-inflammatory response and the potential chondroinductivity of cartilage hydrogels after 5 weeks.
Is Visible/Near-infrared Spectroscopy Superior to Ultrasound for Detection of The Onset of Steatosis in a Rat Liver Model?

Daqing Piao, Ph.D.¹, Jerry W. Ritchey, D.V.M., Ph.D.², G. Reed Holyoak, D.V.M., Ph.D.², Corey R. Wall, D.V.M., M.S.², Kenneth E. Bartels D.V.M., M.S.²

¹ School of Electrical and Computer Engineering, Oklahoma State University, ² Department of Veterinary Pathobiology, Center for Veterinary Health Sciences, Oklahoma State University.

Background: With the increased discussion for using fatty livers to address the severe donor liver shortage, there is also an arising challenge for robust and rapid assessment of fatty liver in donors that may not be suitable for grafts. Currently, ultrasound is the de facto modality for bedside imaging evaluation of steatosis, which is highly reliable for identifying steatosis of moderate or severe grades. However, ultrasound poorly differentiates livers with mild steatosis from lean livers. Mild macrovesicular steatosis may be related to adverse outcomes in living donors who undergo right hepatectomy, which poses a potential concern in donor safety. This study used a rat fatty liver model to assess the potential of optical reflectance spectroscopy in detection of the onset of hepatic steatosis.

Methods and Materials: To facilitate longitudinal and direct sampling of the liver tissue, reflectance spectroscopy (540-940nm) was performed percutaneously by using a single-fiber probe with ultrasound guidance. Sixteen rats were fed a methionine-choline-deficient (MCD) diet and eight control rats were fed a normal diet. Longitudinal in vivo percutaneous single-fiber spectroscopy (SfS) and transabdominal ultrasonography of the rat livers were performed on day-12, 13, 27, 28, 41, 48, 55, and 77 after diet initiation. On each of these examination-days two MCD-diet treated rats and one control rat were euthanized for histological sampling.

Results: In the MCD-diet treated group (n=16), mild lipid infiltration in the liver was confirmed in 7 rats, moderate in 3 rats, and severe in 6 rats. None of the control rats (n=8) developed steatosis. Images of haemotoxylin & eosin stained specimens were analyzed morphometrically to extract the area fraction, total count, and mean size of the lipid droplets. The mean droplet size increased proportionally with the increase of the lipid area fraction (R=0.93), but the total count followed a bi-phasic pattern with the increase of the lipid area fraction. The SfS resolved scattering-slope for the MCD-diet treated livers combined (0.33±0.21, n=16) were significantly greater (p<0.0088) than that for the control livers (0.036±0.25, n=8). When measured at day-12&13 with none of the MCD-treated livers revealed steatosis-diagnostic patterns on ultrasonography but 4 MCD-treated livers had histologically-determined mild steatosis, the SfS-resolved scattering-slope of the MCD-diet treated livers (0.32±0.17, n=16) were significantly greater (p<0.03) than those of the control livers (0.10±0.11, n=8).

Conclusions: The elevation of scattering-slope, which is detectable by interstitial or surface reflectance spectroscopy, may serve as an early diagnostic indicator of the onset of hepatic steatosis.
Novel Shape Memory Polymer Devices for Optimal Endovascular Embolization of Intracranial Aneurysms

Chung-Hao Lee, Ph.D.¹, Yingtao Liu, Ph.D.¹, Bradley N Bohnstedt, M.D.²

¹School of Aerospace and Mechanical Engineering, University of Oklahoma, Norman, OK 73019
²Department of Neurosurgery, University of Oklahoma Health Sciences Center, Oklahoma City, OK 73104

Stroke is a time-sensitive, medical emergency, a leading cause of serious, long-term disability, and was the fifth leading cause of death (6% of all deaths) in Oklahoma in 2012[1]. Incidental rupture of an intracranial aneurysm results in subarachnoid hemorrhage (SAH), which accounts for ~5% of all strokes and causes about 10% of individual’s death before reaching medical attention. Endovascular embolization based on Guglielmi detachable coils[2] is a surgical procedure for treating the abnormal blood vessel by excluding the aneurysmal space and neck from intracranial circulation by means of complete and lasting occlusion. Although it has become an established therapy in the past decade, current clinical challenges associated with disappointing long-term therapeutic outcomes still remain[3], including high risk of aneurysm’s rupture and recurrence (about 41%), and incomplete occlusion due to the complex geometry of the treated aneurysms.

Synthesized materials, such as shape memory polymers (SMPs), possess excellent mechanical properties and shape memory features, can be applied to therapeutic management of cerebral aneurysms. Despite the tremendous evolution in materials synthetization, limited strategies exist for manufacturing biomaterials that can be tailored to patient-specific pathological conditions and aneurysm’s geometry. Therefore, the objective of this project is to develop an innovative computer simulation-guided manufacturing pipeline for systematic fabrications of SMPs with applications to patient-specific endovascular embolization of intracranial aneurysms. The developed SMP-based embolic devices are expected to achieve optimal complete occlusion of the treated aneurysms with desired properties and geometry, short preparation time before surgical operation, and a lower aneurysm recurrence rate. Such development will eventually facilitate individual-optimized endovascular embolization therapies with improved long-term durability, which will be beneficial to the health care of Oklahomans with intracranial aneurysms and reduce the corresponding in-hospital expenditure for managing SAH-induced stroke.

References


Introduction: Sonography students and other healthcare trainees learning to perform 3D and 4D ultrasound acquisition and post-processing are typically exposed to equipment from multiple vendors during clinical practicums at a variety of healthcare facilities. Terminology and presentation of volumetric ultrasound datasets are currently not standardized across vendors. This variation adds complexity for learners.

Methods: This investigation compared terminology and image presentation of basic non-cardiac ultrasound volume navigation controls and functions obtained during a systematic review of contemporary equipment produced by five sonographic equipment companies (GE, Philips, Siemens, Toshiba and Zonare).

Results: Controls and functions explored included 3D volume acquisition, multi-planar reconstruction and rendering. This poster presentation will present examples related to multi-planar reconstruction and rendering.

Discussion: While several controls and functions were identified with common terms across vendors, there were numerous proprietary trademarked terms restricted to a single vendor’s equipment. In addition, default image presentation formats varied across vendors, which require the sonographer or trainee to “reorient” to the image layout prior to evaluating the content of the actual images.

Conclusion: The author proposes familiarity with volumetric terms and image presentation for only a single vendor’s equipment is a disadvantage to students. Familiarity with the variety of common and trademarked terms used to identify volume navigation controls and functions is more useful and an important step in accelerating the learning curve of sonography students and other healthcare trainees.
Novel laparoscopic optical spectroscopy sensing and topography imaging approaches for identification of below-surface tubular structures

Daqing Piao, Ph.D.¹, Sanjay Patel, M.D.²

¹ School of Electrical and Computer Engineering, Oklahoma State University ² Department of Urology, University of Oklahoma Health Sciences Center

Background: Pure or robotic-assisted laparoscopic procedures have increasingly become the preferred surgical approach for management of solid-organ urologic malignancy including prostate, bladder and kidney cancers. When compared to open procedures, laparoscopic procedures face more challenges in the proper identification of anatomical “danger zones” which include large arteries/veins, ureter, bowel, etc, due to absence of or poor tactile feedback from robotic assisted and pure laparoscopic techniques respectively. Techniques that help identify these structures below tissue surfaces that the surgeon cannot see would have the potential to reduce iatrogenic injury and reduce operative times. This work demonstrates two new laparoscopic sensing and imaging approaches in laboratory settings with the potential of identifying below-surface tubular structures.

Methods and Materials: The first approach is laparoscopic diffuse optical spectroscopy (DOS) using two optical fibers attached to the jaws of a laparoscopic bipolar device. One 3-meter long 320 µm fiber was fixed to each of the grasping jaw of a robotic precise bipolar forceps device (Da Vinci® 420110), allowing passage through a 12-mm trocar port. An ultra-bright broadband light source was used for DOS at a fiber-probe (jaw) separation up to 10mm. The second approach is laparoscopic en-face diffuse optical topography (DOT) using densely packed optical fibers. A standard blunt-tip trocar fitting a 12mm port was modified to house 128 copper-coated 750 µm fibers that form radially alternating illumination (70 fibers) and detection (58 fibers) channels. Simultaneously illuminating the source channels and concurrently measuring the light diffusely propagated to the detector channels render near-surface tissue heterogeneity to be imaged at a sub-millimeter resolution. These two laparoscopic devices were tested in laboratory settings on tissue phantoms and avian tissues.

Results: When tested on a 4mm-tubing embedded at an edge depth of 2mm within avian muscle tissue, the laparoscopic DOS discriminated among the following contents of the tubing: water simulating a ureter, air simulating bowel, 33% red-dye simulating a blood vessel, 33% yellow-dye simulating a bile duct, and 33% green-dye. When tested on a thermally coagulated avian egg tissue with a strip of absorption contrast formed by a needle-track of ink injection at an approximate depth of 1mm, the laparoscopic en-face DOT resolved the sub-surface spatial extent of the attenuation track.

Conclusions: Two novel independent laparoscopic approaches for sensing or imaging of below-surface tubular structures are demonstrated in laboratory settings. When combined, the two approaches may bring new potential for identifying below-surface tubular structures at depths informing safe dissection.
MOTION ANALYSIS OF WORK PERFORMANCE IN MALES WITH TRANSTIBIAL LIMB LOSS

C.P. Dionne,¹ J. Day,² K. Veirs¹
¹Department of Rehabilitation Sciences, ²Department of Orthopedic Surgery & Rehabilitation, University of Oklahoma Health Sciences Center, Oklahoma City OK

BACKGROUND: Despite advances in technology and rehabilitation, two-thirds of employable people with transtibial limb loss (TTLL) are not in the workforce. Residuum injury suffered during work-related activity (WRA) performance is one factor precluding work participation. Health care and engineering must develop predictive mechanisms underlying biomechanical residuum injury risk which can translate into effective interventions. The next step is to determine how working-age people with TTLL perform specific WRAs.

PURPOSE: To examine relationship of GRFs and hip/knee kinematics between residual /prosthetic limb and intact limb (and controls) during 2 walking conditions, lifting, and carrying.

METHODS: TTLL inclusion criteria (goal n=38; controls=8): English-speaking; “working-age” adults; unilateral TTLL; ≥12mo post-amputation; actively use prosthesis. Exclusion criteria: significant medical conditions that prevent brisk walking, lifting or carrying tasks; lower limb open wounds; use of walking assistive devices; inability to give formal consent. Participants’ personal, employment, medical, surgical, prosthetic, and rehabilitation information, heart rate, and VAS-pain data are documented at baseline and after testing. Investigators attach reflective markers to participants for 3-D motion analysis and record biomechanical data (ground reaction forces, hip/knee kinematics) during WRA performance. Data are analyzed with descriptive statistics, Qualisys and Visual3D packages.

RESULTS: At time of abstract submission, 2 TTLLs lifted 64lbs and carried 54lbs; 8 controls lifted 59.1lb and carried 49.7lb. Updated results will be reported at conference.

DISCUSSION: This ongoing study will elucidate relationships of GRFs, joint kinematics between residual and intact limbs during WRA performance never before documented in this population.
Energy demand of the brain varies both spatially and temporally with changes in neuronal activity, which requires prompt cerebral blood flow adjustments in a highly regulated fashion to maintain cellular homeostasis and function. This is accomplished through a process termed neurovascular coupling (or “functional hyperemia”), which is orchestrated by an inter-cellular signaling network comprised of neurons and astrocytes, as well as smooth muscle cells and endothelial cells of cerebral microvessels. There is strong clinical and experimental evidence that neurovascular coupling is impaired in aging and in age-related diseases (Alzheimer’s disease, diabetes mellitus, hypertension), which contribute significantly to cognitive decline. Despite its clinical importance, the mechanisms underlying impairment of neurovascular coupling responses remain elusive and no treatments are available for prevention. The Translational Geroscience Laboratory at Department of Geriatric Medicine fosters innovative collaborative relationships between basic scientists, bioengineers and clinical investigators to develop bi-directional translational research projects to elucidate critical mechanisms of neurovascular dysfunction and develop and test novel therapeutic interventions to protect cerebromicrovascular function and to promote cognitive health. To study neurovascular coupling in pre-clinical mouse models of aging and age-related diseases we established a protocol using Laser Speckle Contrast Imaging (LSCI). This type of imaging technique allows rapid and minimally-invasive visualization of cerebromicrovascular blood perfusion while combining high resolution and speed to provide great spatiotemporal accuracy to measure moment-to-moment changes in cerebral blood flow in response to sensorimotor stimulation. To study neurovascular coupling in human patients methods using near-infrared spectroscopy (NIRS) are developed. fNIRS is a powerful non-invasive technique for optical estimation of task-evoked changes in regional blood flow and cortical activity. It relies on the low optical absorption of the biological tissue of the infrared radiation in the 650–1000 nm wavelength window. fNIRS measures the relative changes in concentration of oxygenated and deoxygenated hemoglobin, taking advantage of their absorption coefficients in the optical window. We discuss potential use for LSCI- and NIRS-based methods for bi-directional translational studies focusing on a critical mechanism contributing to the pathogenesis of vascular cognitive impairment and Alzheimer’s disease.
BIOMIMETIC SURFACE MODIFICATION PLATFORM FOR THE DEVELOPMENT OF IN VITRO TUMOR MODELS

Cortes Williams¹, Patrick McKernan¹, Roger Harrison², and Vassilios Sikavitsas¹.
¹Stephenson School of Biomedical Engineering, ²Department of Chemical, Biological & Materials Engineering
The University of Oklahoma

Traditional chemotherapy regimens put a high degree of emphasis on the use of historical data to predict a cancer patient’s response to a proposed therapy. Unfortunately for the patients, this often leads to continuous rounds of trial-and-error in the search for a compatible treatment, decreasing their chances for survival. Tumor engineering seeks to alleviate this issue by growing patient tumors outside of the body, providing a high throughput avenue for treatment discovery. Utilizing a variety of techniques for 3D culture, researchers have created models that more closely resemble and predict in vivo tumor drug responses; however, there is still more room for improvement. In particular, these in vitro models consistently exhibit poor cell proliferation and distribution, which severely limits their predictive capabilities.

The ability to seed and culture in vitro patient tumor cells on 3D scaffolds presents unique challenges due to the inert nature of commonly used polymeric or ceramic biomaterials. Mimicking the natural microenvironment of the target tissue can be of great benefit to not only improve the seeding efficiency, but also the drug response of the engineered tumor. The choice of scaffold used to support cells in culture plays a significant role in cell viability and ex vivo tumor development. Scaffold properties, such as rate of degradation, hardness, and biocompatibility must be manipulated to match desirable tissue properties and the rate of tissue growth to scaffold degradation. Surface modification primarily addresses the interface that the cells directly interact with, coating underlying material with undesirable properties.

To combat this major issue, we have leveraged our patented biomimetic surface modification platform for tumor engineering applications. In particular, we have identified various moieties specific to certain tumors that are integral to cellular adhesion, and have used these to modify our scaffolds and trick the cancer cells into exhibiting higher rates of adhesion. For instance, in terms of prostate cancer, poly(L-lactic acid) (PLLA) scaffolds were modified to express n-cadherin, which is a highly upregulated protein used for cellular adhesion. After cell seeding, we were able to significantly increase PC3 seeding efficiency and potentially improve cell physiology without compromising the mechanical and degradation properties of the underlying PLLA. For the study as a whole, we will be seeding various cancer cell lines (PC3, MDA, MB49, and B16) on both 2D and 3D PLLA scaffolds, and culturing them under increasing shear levels in perfusion bioreactors with samples taken intermittently.
Developed a novel treatment method for cementless implant surgery

M. Khandaker, Ph.D.¹, W. Williams, Ph.D.², R. Wolf, H. Jamadagni, Amal Swediwah, M.D., Ph.D.¹

¹University of Central Oklahoma, Edmond, OH, ²University of Kansas, Lawrence, KS, ³University of Pennsylvania, Philadelphia, PA

This study invented a method to coat a metal implant with nano scale coating material (NSCM) made with collagen polycaprolactone nanofiber matrix (CG-PCL NFM). The goal of the study was to evaluate the biomechanical performances of titanium (Ti) implant by CG-PCL NFM treatments on Ti. Specific Aims were to find the CG-PCL NFM treatments effect on the biomechanical performances of Ti implant for cementless surgeries from in vitro biocompatibility and in vivo tests using rabbits. This invention implements a set of grooves that are created on Ti at the circumferential direction to increase the surface area of the implant in contact with bone [1]. CG-PCL NFM was coated along the sub-micrometer grooves on the Ti implant using the electrospin setup, which was awarded with a US patent [2]. A 2.2 mm diameter and 12 mm length medical grade Ti was used for in vivo implant. A total eighteen parallel circumferential microgrooves (71.31 ± 13.62 µm, n=4) at a distance 0.5 mm from one end of Ti wire were cut by a saw machine. Three groups of Ti samples were prepared: without grooves (control), with grooves (groove) and grooves coated with CG-PCL NFM (groove-NFM). PCL NFM was collected between two parallel wires using electrospin unit [3]. Six layers of PCL NFM were deposited on CG coated Ti by rotating the Ti rod. Each group of samples was implanted in the femoral condyle of rabbits. The animal was euthanized after 8 weeks of surgery. Soft tissues were cleaned and trimmed using saw to prepare a mechanical and histological test samples. The Ti/bone sample was embedded in an acrylic cup using a low-viscosity acrylic bone cement. After curing the cement, pull out tension tests were conducted on Ti/bone samples using a mechanical tester. The shear strength was calculated by dividing the maximum pull-out force at the point of failure of implant by the surface area of Ti in contact with bone. Sectioning, staining, and imaging for histomorphometric analysis of bone samples with implant was done using standard histological techniques. We found that the in vitro rat osteoblast cell adhesion, proliferation, mineralization, and protein adsorption as well as the in vivo mechanical stability and promoting osseointegration on Ti were significantly improved by the CG-PCL NFM coating (p <0.05). The outcomes of this study will enable clinicians to use improved implants for orthopedic and orthodontic surgeries, thereby lowering implant loosening and preventing expensive revisions.

Reference:

Tuning Photothermal Properties of Gold Nanodendrites for In Vivo Cancer Therapy within a Wide Near Infrared Range by Simply Controlling Their Degree of Branching

Penghe Qiu, Ph.D.¹, Mingying Yang, Ph.D. ², Xuewei Qu¹, Yanyan Huai¹, Ye Zhu¹, Chuanbin Mao, Ph.D.¹

¹University of Oklahoma, Norman, OK, ²Zhejiang University, Hangzhou, Zhejiang, China

Although dendritic nanoparticles have been prepared by many different methods, control over their degree of branching (DB) is still impossible, preventing us from understanding the effect of the DB on the properties of the nanodendrites. Herein, we developed a novel seed-mediated method to prepare gold nanodendrites (AuNDs) in an organic solvent using long chain amines as a structural directing agent. We discovered that the DB could be tuned facilely by simply adjusting synthetic parameters, such as solvent type, the type and concentration of the long chain amines. We found that DB tuning resulted in dramatic tunability in the optical properties in the near infrared (NIR) range, which resulted in significantly different performance in the photothermal cancer therapy. Our in vitro and in vivo studies revealed that AuNDs with a higher DB were more efficient under lower wavelength NIR irradiation. In contrast, those with a lower DB performed better under higher wavelength NIR irradiation, indicating that AuNDs of even lower DB should have even better photothermal efficiency within the second NIR window. Thus, the tunable optical properties of AuNDs in the NIR range allow us to selectively determine a suitable laser wavelength for the best cancer therapeutic performance.
Highly ordered phage films fabricated by a simple dipping-drawing method

Ningyun Zhou¹, Chuanbin Mao¹
¹University of Oklahoma, Norman, OK

M13 phage is a type of semi-flexible linear-shaped bionanofiber with negatively charged protein shell. It is about 900 nm long and 7 nm wide. Previous studies have reported that the M13 phage can assemble in a specific pattern and function as a substrate to regulate cell growth. However, the reported phage film pattern is not well organized. In this project, we simply use the dipping-drawing method to draw the poly-lysine coated glass slides from the M13 phage solution to achieve the phage assembly, through the interaction between positively charged poly-lysine substrate and negatively charge phage. By doing this, we discovered a highly-organized phage assembled pattern which has never been reported before. We hypothesize that the formation of this unique structure is due to liquid crystal sematic phase formation of phage. During the dipping-drawing process, the phages also experienced an external strain helping them forming this new structure via flow-induced crystallization (FIC). We will use this unique highly organized phage film pattern to manipulate cell growth in the future.
The Laboratory of Biomolecular Structure and Function at OUHSC

Simon Terzyan, Ph.D, Blaine Mooers, Ph.D
Oklahoma University Health Science Center

We would like you to know about the services in protein purification and structural studies available to OUHSC in the Protein Expression Production (PEP) Facility and the Laboratory of Biomolecular Structure and Function (LBSF). Dr Simon Terzyan (BRC 406) manages both facilities. He has 25 years of experience in protein science and crystallography (co-author on 40+ papers, 40+ structures in the Protein Data Bank). The PEP has shakers for bacterial growth and chromatography equipment for non-structural biology labs to make pure protein on the milligram scale. The pure protein can be used for biological, biophysical, or crystallographic studies. Training in the use of this equipment is available from Dr. Terzyan, or he can do this work on a fee-for-service basis. Dr. Terzyan can do standard molecular modeling tasks (e.g., design mutants, compare structures). He can make images of protein structures for your grant applications and manuscripts. For advanced molecular modeling projects (e.g., homology modeling, structure-based drug design), the LBSF has a Molecular Modeling Unit run by Dr. Timothy Mather. This unit has a workstation with four GPUs optimized to run four independent molecular dynamics simulations. Dr. Terzyan can also provide training in dynamic light scattering, protein crystallization, X-ray data collection (the LBSF has a new X-ray generator with two detectors), and structure determination, refinement, and analysis. He can also provide these services on a fee-for-service basis. Dr. Terzyan serves as an interface with the OU Crystallization facility in Norman and the Physical Biochemistry Laboratory in the Department of Biochemistry and Molecular Biology (BMB). The latter facility has CD spectroscopy, microcalorimetry, and microscale thermophoresis instruments. The LBSF is a Vice President of Research Core Facility and a core facility of the Oklahoma COBRE in Structural Biology. The PEP is supported by the BMB.
Quantitatively Predict Ovarian Cancer Patients’ Responses to Chemotherapy at Early Stage: A Radiomics Based Method

Abolfazl Zargari\(^1\), Yue Du\(^1\), Gopichandh Danala\(^1\), Theresa Thai M.D.\(^2\), Camille C, Gunderson M.D.\(^3\), Katherine M. Moxley M.D.\(^3\), Kathleen Moore M.D.\(^3\), Robert S. Mannel M.D.\(^3\), Hong Liu M.D.\(^1\), Bin Zheng M.D.\(^1\), Yuchen Qiu M.D.\(^1\)
\(^1\)School of Electrical and Computer Engineering, University of Oklahoma, Norman, OK 73019, \(^2\)Department of Radiology, Health Sciences Center of University of Oklahoma, Oklahoma City, OK 73104, \(^3\)Department of Obstetrics and Gynecology, Health Sciences Center of University of Oklahoma, Oklahoma City, OK 73104

In gynecologic oncology, ovarian cancer is the second most prevalent malignancy with leading mortality rate. For clinical practice, early stage prognostic evaluation is critically important to improve the overall survival rate of ovarian cancer patients. However, the current method, response evaluation criteria in solid tumors (RECIST), cannot achieve satisfied predicting accuracy in the cancer treatment. In order to overcome this limitation, a radiomics based quantitative image analysis scheme was developed to accurately predict the tumor response to chemotherapy. During the experiment, we retrospectively assembled a dataset containing pre- and post- treatment CT images acquired from 91 advanced stage, recurrent ovarian cancer patients. A total of 149 features were firstly estimated on the metastatic tumors depicted on the CT images. Next, principal component analysis (PCA) algorithm was applied on the estimated features to reduce the redundancy. The best performing components was used as radiomics feature signature for the following classification. Finally, based on the feature signature, we use support vector machine (SVM) to predict the tumor response to chemotherapy. The results indicate that our new scheme is able to achieve a predicting accuracy of 82%. This initial study demonstrates that quantitative radiomics method may be able to improve the performance of predicting tumor responses to chemotherapy at early stage for ovarian cancer patients.

**Keywords:** Computer aided diagnosis (CAD), ovarian cancer, quantitative radiomics, chemotherapy response evaluation at early stage, Response Evaluation Criteria in Solid Tumors (RECIST), principal component analysis (PCA), support vector machine (SVM)
Evaluation of the therapeutic effect of theta burst stimulation in Schizophrenia: a randomized sham controlled resting-state EEG study

Guofa Shou, Ph.D.¹, Qian Guo, Ph.D.², Chunbo Li, Ph.D.², Kelvin Lim, M.D.³, Jijun Wang, M.D.², Bin He, Ph.D.³, Lei Ding, Ph.D.¹

¹University of Oklahoma, Norman, OK, ²Shanghai Jiao Tong University, Shanghai, China, ³University of Minnesota, Minneapolis, MN

Repetitive transcranial magnetic stimulation (rTMS) is a promising neuromodulation technology to treat multiple neuropsychiatric disorders. The objective of the present study is to evaluate the therapeutic effect of theta burst stimulation (TBS) in schizophrenia by probing the neural activations and frontal-parietal network using resting-state EEG (rsEEG), along with clinical measurements of cognitive functions and severity of symptoms.

Thirty-five schizophrenia patients (all males) were enrolled and randomly assigned into two groups, i.e., TBS (18 patients) and sham (17 patients), with a five-day rTMS treatment (1200 stimulations per day) at 80% of motor threshold on left dorsolateral prefrontal cortex. Before and after rTMS, 32-channel rsEEG (3 minutes) with closed eyes were recorded. To evaluate the cognitive functions and the severity of symptoms, visual spatial working memory (VSWM), verbal fluency test (VFT), and the Positive and Negative Syndrome Scale (PANSS) were performed.

After preprocessing, rsEEG from all subjects were temporally concatenated for a group-level analysis using the complex independent component analysis (ICA). Thereafter, spectral powers of individual ICs from pre- and post-rTMS were statistically compared. In addition, frontal-parietal network was investigated. Four frequency bands, i.e., theta (4-7 Hz), low alpha (8-10 Hz), high alpha (11-13 Hz) and beta (14-30 Hz) band, were separately studied. Meanwhile, the scores in VSWM, VFT and PANSS from pre- and post-rTMS were compared.

Between TBS and sham groups, there were no significant differences in demographic characteristics and the baseline scores. Between pre- and post-rTMS measures, there was significant increase in VFT score (22.2±7.7 vs. 26.6±9.4; p<0.0005) and significant decrease in negative symptoms (23.6±5.4 vs.21.0±4.5; p<0.0001) following TBS but not following sham.

In rsEEG, 7 of 15 ICs of interests were identified with significant power changes in TBS but none in sham or in baseline powers between TBS and sham. For examples, two prefrontal ICs have significant increased theta power, suggesting increased executive functions as also reflected in the significant score changes in VFT and negative symptoms. The parietal IC and bilateral temporal-parietal IC have significant increased alpha powers, indicating the therapeutic effect of TBS since reduced alpha powers are observed in schizophrenia than healthy controls. Moreover, significantly enhanced frontal-parietal connectivity in high alpha (p<0.05) was detected in TBS but not in sham, indicating the increased executive control after TBS.

In conclusion, the findings clinical measurements and EEG signals demonstrate potential therapeutic effect of the present TBS paradigm in schizophrenia, especially for negative symptoms.
Local release of Combretastatin A-4 from NIR-light activatable prodrugs overcomes areal and temporal limitations of photodynamic therapy

Pallavi Rajaputra,1 Moses Bio,1 Gregory Nkepang,1 Pritam Thapa,1 Sukyung Woo,1 Youngjae You

1Department of Pharmaceutical Sciences, College of Pharmacy, University of Oklahoma Health Sciences Center, Oklahoma City, OK 73117

A unique prodrug strategy for treating localized cancers, in which NIR light-illuminated prodrug effectively ablates tumors through the combined effects of photodynamic therapy (i.e., singlet oxygen [SO]) and locally released anticancer drugs has been proposed. Due to short distance of action (< 0.04 μs) and short lifetime (< 0.02 μm) of SO, direct damage of PDT is both areally and temporally limited. We hypothesized that the locally released anticancer drugs would overcome the areal and temporal limits of SO. Near IR-activatable prodrug of combretastatin A-4 (CA4), Pc-(L-CA4)₂, and its pseudo-prodrug, Pc-(NCL-CA4)₂, were evaluated in vitro and in vivo. After partial illumination of a 24 well, all the cells in the prodrug-treated well were killed by the released CA4. Limited areal damage was observed in the pseudo-prodrug-treated wells. A time-dependent cell survival study revealed more extensive cell death in the prodrug-treated cells, due to the sustained damage from the released CA4. Cell cycle analysis and microscopic imaging data demonstrated the typical damage patterns of CA4 in the prodrug-treated cells. A time-dependent histological study showed that prodrug-treated tumors lacked mitotic bodies. The prodrug caused broader and more long-lasting tumor size reduction than did the pseudo-prodrug. These data consistently support that the released CA4 overcomes the areal and temporal limits of SO, providing far superior antitumor effects.
Paclitaxel Prodrug for the Combinational Treatment of Photodynamic Therapy (PDT) and Site-Specific Chemotherapy

Pritam Thapa, Mengjie Li, Moses Bio, Pallavi Rajaputra, Yajing Sun, Sukyung Woo, Youngjae You
Department of Pharmaceutical Sciences, College of Pharmacy, University of Oklahoma Health Sciences Center, Oklahoma City, OK, USA

Paclitaxel (PTX), a well-known and widely used anti-cancer drug, has several systemic side effects similar to other anticancer drugs. On the other hand, PDT suffers from incomplete ablation and subsequent recurrence of tumors in part due to the short half-life and poor diffusion rate of singlet oxygen. To overcome these drawbacks, we prepared a photo-activatable prodrug of PTX where PTX was conjugated with phthalocyanine photosensitizer via a singlet oxygen cleavable amino acrylate linker. The PTX prodrug was tested for tubulin polymerization, the release rate of PTX from prodrug upon illumination at 690 nm, stability in complete media, and the combination effect in killing ovarian cancer cells in vitro (SKOV-3). As anticipated, the PTX prodrug did not enhance the tubulin polymerization unlike PTX. From the stability test, the prodrug was stable in the complete media under dark. Upon illumination with 690 nm lasers at 5.6 mW/cm², > 90% of PTX was released in 30 min. The prodrug showed very potent phototoxicity (IC₅₀ = 3.9 nM), through the combinational effect of PDT and PTX whereas less dark toxicity (without illumination) compared to PTX. We can conclude that the prepared PTX prodrug has essential properties for further study to develop as a light-activatable prodrug possessing combinational effect of PDT and local PTX chemotherapy.
Effects of 3D Printing on Flow-Induced Shear Distributions
Cortes Williams and Vassilios Sikavitsas
Stephenson School of Biomedical Engineering, The University of Oklahoma

As 3D printing gains more exposure as the future of scaffold fabrication for tissue and tumor engineering, the ability to preemptively model the constructs microenvironment becomes of vital importance. Particularly when using perfusion based bioreactor systems, the most important properties to obtain are the fluid flow and wall shear fields that potential cells will experience during and after seeding. Traditionally, 3D printing has given users the impression that the scaffold obtained after printing will exhibit the same, or closely similar, architecture as the designed model. While this may be the case in terms of the macroscopic view, we hypothesized that the local fluid dynamic environment would differ greatly. The intention of this study was to 3D print scaffolds of various pore size gradients, image these constructs utilizing micro-computed tomography, and perform computational fluid dynamic simulations on the resulting reconstructions and the initial designs in order to compare the average shear on the fibers, where the cells would be adhered. As a determining factor, the probability density function (p.d.f.) of each was compared to not only highlight the major differences, but also to show the reproducibility of the printing process.

PPF scaffolds were fabricated with varying gradient designs. The scaffolds were scanned non-destructively with micro-computed tomography (CT), filtered, thresholded, and stacked to form the 3D reconstructions. Simulations were then conducted with flat velocity profiles based on a flow rate of 0.1 mL/min, no-slip boundary conditions imposed at the walls, and also an outlet with constant static pressure. Probability density functions were tested using previous published methods, and a previously identified matched was tested for accuracy.

Average shear stress values, both per layer and per construct, were compared, and confirmed major differences between the idealized model and reconstructions. In a few instances, localized regions of higher shear stress were observed near the edges (eg. SML top layer) and near defects within the 3D printed scaffolds (eg. MMM middle layer). To accompany these findings, p.d.f.s of the wall shear stress for the resulting reconstructions and the initial designs were compared, highlighting confirming the disparities between the two. Finally, we proved that a previously identified standard gamma distribution describes the p.d.f. of flow induced stresses in the reconstructions. Statistically, however, this distribution does not describe those of the CAD designs within acceptable limits.
Neurovascular uncoupling predicts cognitive decline and gait abnormalities in a clinically relevant mouse model of whole brain irradiation

Andriy Yabluchanskiy, Ph.D.¹, Stefano Tarantini, Ph.D.¹,
Gabor Fulop, M.D.¹, William E. Sonntag, Ph.D., Anna Csiszar, M.D., Ph.D.¹,
Zoltan Ungvari, M.D., Ph.D.¹

¹Translational Geroscience Laboratory, Reynolds Oklahoma Center on Aging/Department of Geriatric Medicine,
University of Oklahoma Health Sciences Center, Oklahoma City, OK

Whole brain irradiation (WBI) is known to promote accelerated brain senescence and leads to progressive cognitive dysfunction in up to 50% of tumor patients surviving long-term after treatment, due to, at least in part, gamma-irradiation induced cerebromicrovascular injury. Moment-to-moment adjustment of cerebral blood flow (CBF) via neurovascular coupling (NVC) has a critical role in maintenance of healthy cognitive function. To determine whether cognitive decline induced by WBI associate with impaired NVC, C56BL/6 mice (3 mo) subjected to a clinically relevant protocol of fractionated WBI (5 Gy twice weekly for 4 weeks) and control mice were compared. Mice were tested for spatial memory performance (radial arm water maze), sensorimotor coordination (computerized gait analysis, CatWalk) and NVC (laser Doppler flowmetry, laser speckle contrast imaging) at 3 and 6 months post-irradiation. We found that mice with WBI exhibited impaired NVC at 3 months post-irradiation, which was associated with impaired performance in the radial arm water maze, while gait parameters remained unaltered. At 6 months post-irradiation persisting neurovascular dysfunction and cognitive impairment were evident, which were associated with significant impairment in gait coordination (including changes in the regularity index and phase dispersion). Collectively, our findings provide evidence for early and persisting neurovascular un-coupling after a clinically relevant protocol of fractionated WBI, which predict early manifestations of cognitive impairment.
The goal of the Laboratory for Molecular Biology and Cytometry Research (LNBCR) is to facilitate high-quality, state-of-the-art research by providing expertise and access to technology for a variety of specialized biomedical research applications. It is a full-service, research-oriented, institutionally-managed core dedicated to enhancing research and providing education and training to all users. The LNBCR assists investigators in all aspects of research, including initial project consultation, assistance with experimental design, training in protocols and instrumentation usage, sample preparation, and assistance with analysis and interpretation of results. The core also assists manuscript preparation and grant submission for applications that require heavier usage of specialized technology or techniques.
Optimization of Folate Receptor-Mediated Targeting Delivery of Far-Red Light-Activatable Prodrugs to Ovarian Cancer Cells

Mengjie Li, Ph.D.1, Pritam Thapa, Ph.D.1, Youngjae You, Ph.D.1, Sukyung Woo, Ph.D.1
1University of Oklahoma Health Science Center, Oklahoma City, OK

Background and Purpose: We developed a far-red light-activatable prodrug which combines photosensitizer of phthalocyanine (Pc) and Paclitaxel (PTX) with a singlet oxygen-cleavable linker (L). Our prodrug Pc-(L-PTX)2 demonstrated characteristics of fluorescence imaging, light-controlled local PTX release and combined effect of photodynamic damage and chemotherapy. However, the uptake of Pc-(L-PTX)2 on ovarian cancer cell line was low. To further enhance its intracellular uptake, a more effective targeting delivery system is needed. Folic acid (FA) has been widely used as a targeting moiety for various anti-cancer drugs where folate receptors has been known to be overexpressed in several human tumors including ovarian cancer. Therefore, we designed and prepared five FA-conjugated prodrugs (PTX-L-Pc-PEGn-FA, n=0,1,2,3 and 5K). The differences in the polyethylene glycol (PEG) length can influence the accessibility of the folate ligand binding to the folate receptors on cells. In this study, we evaluated the prodrug efficacy in terms of PEG spacer length and find out the optimal PEG chain length. Furthermore, we provided a mechanism based kinetic model to guide the optimal design and targeting delivery of FA-conjugates.

Methods: Human ovarian cancer cells SKOV-3 was used to evaluate the in vitro uptake and cytotoxicity of the FA-conjugates. The time-dependent intracellular accumulation of FA-conjugates with or without FA were measured via fluorescence plate reader. The phototoxicity of FA-conjugates after illumination were evaluated by performing methylthiazol-tetrazolium (MTT) assay. An in vitro kinetic model was developed based on mechanism of folate-receptor mediated endocytosis to characterize the intracellular time-concentration profiles of FA-conjugates.

Results: The uptake of FA-conjugates was improved by increasing PEG chain length (1K, 2K and 3K). The optimal chain length is between 1K and 3K. The uptake was decreased when the PEG length reached to 5K. For 1K, 2K and 3K FA-conjugates, over 50% of the prodrugs were internalized through folate receptor mediated endocytosis while only 22% of 5K conjugate uptake was receptor mediated.1K, 2K and 3K FA-conjugates showed a higher cytotoxicity (IC50~125 nM) than 0K and 5K (IC50~400 nM) which was consistent with the in vitro uptake result. Our model suggests the higher intracellular accumulation could be achieved from a combination of both folate-mediated and non-specific endocytosis.

Conclusions: Our result indicated that the optimal PEG chain length is around 1K to 3K. Over prolonged PEG chains could comprise the targeting moiety on folate receptor-mediated delivery of the conjugates. Moderate percentage of folate-mediated endocytosis is a key factor for higher intracellular accumulation of FA conjugates.
Filamentous bacteriophage (phage) is a genetically modifiable supramacromolecule. It can be pictured as a semi-flexible bionanofiber (~900 nm long and 7 nm wide) made of a DNA core and a protein shell with the former genetically encoding the latter. Powered by our well-established expertise in displaying a foreign peptide on its surface through genetically engineering its DNA, we have employed phage to identify target-specific peptides, construct novel organic-inorganic nano-hybrids, develop nano- and bio-materials for disease treatment, and generate bioanalytical methods for disease diagnosis. Compared with conventional biomimetic chemistry, phage-based supramolecular chemistry represents a new frontier in chemistry, materials science and medicine. In this poster, we introduce our recent successful efforts in phage-based supramolecular chemistry, by integrating the unique nanofiber-like phage structure and powerful peptide display technique to the fields of chemistry, materials science, and medicine. Specifically, we describe our following seminal contributions: (1) successfully synthesized silica, nucleated hydroxyapatite, and assembled gold nanoparticles on/along phage nanofiber templates to form novel functional materials; (2) introduced azo units to phage nanofibers to form functional azo-phage nanowires via a diazotization reaction between aromatic amino and the tyrosine residues genetically displayed on phage surfaces; (3) assembled bioengineered phages into 2D films for studying the effects of both biochemical (the peptide sequences displayed on phage) and biophysical (the topographies of phage film) cues on the proliferation and differentiation of both mesenchymal stem cells (MSCs) and induced pluripotent stem cells (iPSCs) and identified several functional peptides that can induce the osteogenic differentiation of stem cells; (4) seeded MSCs into a 3D hydroxyapatite scaffold decorated with phage displaying cell-signaling peptides and found the phage could induce both angiogenesis and osteogenesis of MSCs for bone regeneration; (5) identified breast cancer cell-targeting and MSC-targeting peptides by phage display technique and used the identified peptides to significantly improve the efficiency of targeted cancer therapy and MSC-based gene delivery, respectively; (6) synthesized biomarker-capturing magnetic nanoparticle-decorated phage and applied such phage to magnetically enrich biomarkers for ultra-sensitive disease diagnosis; and (7) magnetically construct centimeter-scale 3D multi-layered phage assemblies for bone regeneration and device fabrication. Our findings demonstrated that phage is indeed a very powerful bionanomaterial for developing novel nanostructures and advancing the important fields in medical research, including molecular targeting, cancer diagnosis and treatment, drug and gene delivery, tissue regeneration, and stem cell niche study.
Laser Speckle Contrast Imaging (LSCI) to Study Age-Related Microvascular Dysfunction: Potential Applications for Translational Geroscience

Stefano Tarantini, Ph.D.¹, Andriy Yabluchanskiy, Ph.D.¹, Tamas Csipo, M.D.¹, Gabor Fulop, M.D.¹, Anna Csiszar, M.D., Ph.D.¹, Zoltan Ungvari, M.D., Ph.D.¹

¹Translational Geroscience Laboratory, Reynolds Oklahoma Center on Aging/Department of Geriatric Medicine, University of Oklahoma Health Sciences Center, Oklahoma City, OK

The microcirculation is of vital importance to deliver oxygen and nutrients to all parts of receiving end-organs. The endothelium, the dynamic cell layer that covers the inside of all blood vessels, is the largest endocrine organ in the human body. Among the many functions that it performs, the endothelial cell layer is responsible for controlling vasomotor tone, blood pressure, tissue perfusion, trafficking of cells and nutrients in- and out- of the blood vessels, maintenance of blood fluidity, and angiogenesis (the growth of new blood vessels). There is strong experimental evidence that age-related microvascular dysfunction contributes to the pathogenesis of various diseases of aging. To translate these findings there is a growing need for quick, non-invasive, reproducible tests of microvascular function that can be used in geriatric patients. The Translational Geroscience Laboratory at the Department of Geriatric Medicine has launched a bi-directional translational initiative to bridge the knowledge gap in microvascular aging between animal and human studies. We are currently establishing methods to characterize microvascular functional impairment in human patients that could be used to test mechanistic hypotheses derived from pre-clinical studies. Here we present a method using Laser Speckle Contrast Imaging (LSCI) to assess age-related changes in microvascular reactivity and endothelial function in patients. Non-invasive tests to interrogate microvascular responses include post occlusive reactive hyperemia, cold-induced hyperemia, Lewis triple response and reactive hyperemia following ball squeezing exercises. Microvascular perfusion is measured by LSCI in the forearm and/or under the corpus unguis. Using this method will provide a quick, non-invasive, reproducible test battery to investigate a clinically highly relevant outcome measure involved in the pathogenesis of numerous age-related diseases.
Corneal Tissue Engineering: A Diabetic Keratopathy In Vitro Model

Shrestha Priyadarsini¹, Jian-Xing Ma², Dimitrios Karamichos¹,³.

¹ Department of Ophthalmology/Dean McGee Eye Institute, University of Oklahoma Health Sciences Center, Oklahoma City, OK, USA, ²Department of Physiology Harold Hamm Diabetes Center, University of Oklahoma Health Sciences Center, Oklahoma City, OK, USA, ³Department of Cell Biology, University of Oklahoma Health Sciences Center, Oklahoma City, OK, USA,

Purpose: Tissue engineering applications has garnered great interests in recent years across various fields of medical science. Treatment for various ocular dystrophies demands effective tissue engineering approaches. Diabetes mellitus is one of the common systemic diseases, which is known to cause a number of ocular dysfunctions, often leading partial or complete vision loss. Today, 284 million people are severely visually impaired worldwide, of which 39 million are completely blind. Diabetes severely affects the human cornea leading to what is known as diabetic keratopathy. Studies on diabetic keratopathy are limited and mainly focused around the epithelium and nerves. Several animal models have also been developed however, in many occasions, the strategies and treatments that were successful in rodents failed in humans. The development of a model more relevant to the patient population is absolutely necessary. In this study we developed a 3D self-assembled corneal tissue substitute that mirrors the basic anatomical and physiological of the corneal tissue in vivo and can be used as a tool for investigating the defects in human diabetic keratopathy as well as screening the efficacy of various agents before animal testing.

Methods: Human corneal stromal cells from healthy (HCF), Type I (T1DM) and Type 2 (T2DM) diabetic donors were isolated and seeded on polycarbonate membrane inserts at 1million cells/well with Vitamin C to promote collagen assembly. Human corneal nerve cells (HCNs) were seeded on top of the constructs at the 3 weeks’ time point and differentiated. At week 4, all constructs were processed for real time PCR and WB.

Results: Our results show upregulated nerve markers (βIII tubulin and Nestin) in T2DM co-cultures (3 fold; p≤0.001) whereas T1DM co-cultures showed significant downregulation (1 fold; p≤0.05) for both the nerve markers, compared to healthy controls. We also observed increased Col I (4 fold; p≤0.0001) and Col V (2.5 fold; p≤0.001) expression in T2DM which was correlated with significant increase in ECM assembly and deposition.

Conclusion: We have successfully developed a novel, first of its kind, innervated 3D in vitro corneal substitute that helps better understanding of the crucial parameters that often gets altered during hyperglycemic conditions and are essential for the normal corneal environment maintenance. Targeting these parameters would pave the way for developing therapeutic measures in treating diabetic keratopathy.
M13 filamentous phage, a virus that specifically infects bacteria and is harmless to human beings, is a bionanofiber (880 nm long and 6.6 nm wide). It is made of DNA as a core and protein coat as a sheath that wraps the core. The coat protein constituting the side wall of phage is termed pVIII and encoded by gene VIII of the phage DNA which can be genetically engineered to display foreign peptides all over the surface of phages. Based on these features, we report the design of a unique matrix, assembled from engineered M13 phage bionanofibers with specific cues of nanotopographies and versatile signal peptides to simulate native niche for directing the osteoblastic differentiation of mesenchymal stem cells (MSCs) and induced pluripotent stem cells (iPSCs). In addition, by filling a 3D printed bioceramic scaffold with phage nanofibers displaying high-density RGD peptide, we also developed a virus-activated 3D matrix. After it is seeded with MSCs and implanted into a bone defect, the phage nanofibers successfully induce osteogenesis and angiogenesis by activating endothelialization and osteogenic differentiation of MSCs.
There is increasing evidence that vascular risk factors, including aging, hypertension, diabetes mellitus and obesity, promote cognitive impairment, however, the underlying mechanisms remain obscure. Cerebral blood flow (CBF) is adjusted to neuronal activity via neurovascular coupling (NVC) and this mechanism is known to be impaired in the aforementioned pathophysiological conditions. To establish a direct, causal relation between impaired NVC and cognitive decline, we induced neurovascular uncoupling pharmacologically in mice by inhibiting the synthesis of vasodilator mediators involved in NVC. Treatment of mice with the epoxygenase inhibitor MS-PPOH, the NOS inhibitor L-NAME and the COX inhibitor indomethacin decreased NVC by 75% mimicking the aging phenotype, which was associated with significantly impaired spatial working memory (Y-maze), recognition memory (Novel object recognition) and impairment in motor coordination (Rotarod). Blood pressure (tail cuff), basal cerebral perfusion (arterial spin labeling perfusion MRI) and synaptic activity (evoked field potential) were unaffected. Thus, targeted experimental disruption of NVC per se leads to significant impairment of cortical function, including cognitive decline, recapitulating neurological symptoms and signs observed in brain aging and pathophysiological conditions associated with accelerated cerebromicrovascular aging.
A flexible applicator-probe with nine channels of side-by-side photo-diode and fiber-optics sensors for deployment in the spinal canal of cadaverous dogs after hemilaminectomy is being developed. This flexible applicator-probe will be used for simultaneously measuring transcutaneous light irradiance at nine positions within the spinal canal for evaluating the potential of photobiomodulation (PBM) of the spinal cord at a clinically acceptable surface irradiance. The demonstration of PBM treatment of spinal cord injury has been limited to rodent models. In contrast, the transcutaneous use of PBM to treat spinal cord conditions in children or adults is expected to be challenging due to the significant attenuation of light energy as it travels through the thick and heterogeneous layers of tissue and bone to reach the level of the spinal cord. A pilot study performed on one cadaverous dog using a single photo-diode sensor has suggested a potential narrow treatment window for transcutaneous PBM of spinal cord at the shallowest T12/13 and T13/L1 intervertebral disc positions. This present study, by developing a flexible applicator-probe with multiple channels for intra-spinal dosimetry in multiple positions, will assess if the recommended photo-bio-stimulatory treatment dose reaches the spinal canal under a surface dose that is clinically acceptable. This flexible applicator-probe is built upon a ¼” outer-diameter plastic tube consisting of 9 sets of photodiodes and fiber-optical diffusers epoxied over an 8-centimeter length at a 1cm interval. Each photodiode of peak responsivity at 900nm, which has a surface mounting profile of 4.4mm×2.1mm, is connected to a signal-conditioning module by 30AWG wires of 4-meters length for computer-based acquisition of the photodiode readout of the local irradiance from all photodiode channels. The 9 photodiode sensors are conditioned to three subsets with one of three levels of responsivities scaled by 10 times (i.e., 1V per 1mW/cm2, 1V per 100µW/cm2, and 1V per 10µW/cm2). Each fiber-optical diffuser sensor, which is a 1.75mm cylinder of 200 µm in diameter, is connected to a 9×1 fiber-switch through 4-meter long 200µm fiber for spectrophotometry measurement of the irradiance. The photodiode sensor allows for absolute dosimetry, but it has limited dynamic range of approximately 3-decades. The limited dynamic range of the photo-diode measurement is supplemented by the relative fiber-optical spectrophotometry that has a much greater (2 decades greater) dynamic range. The performance of this flexible applicator probe for simultaneous multi-site dosimetry will be calibrated using tissue phantoms in a setting simulating intra-spinal deployment, prior to dog studies.
The ability to personalize therapeutic administration of anti-cancer compounds would be extremely beneficial to both patients and doctors. In order to develop personalized care and determine the effectiveness of the drug, the anti-cancer compound’s interactions with cancer cells must be explored at the single-cell level. We have developed a method utilizing the Single-probe, which is a miniaturized, multifunctional device that can be coupled to a mass spectrometer for the analysis of live, single cells under ambient conditions. We have used single cell mass spectrometry (SCMS) to detect a variety of anti-cancer compounds in a various cell lines, including T24 and SW780. One major goal of this project includes single-cell quantification of anti-cancer compounds by utilizing an internal standard in the sampling solvent. Another major goal is the adaptation of a suspension cell analysis system for the SCMS determination of anti-cancer compounds in the urine of bladder cancer patients.
Increasing PSA Density Correlates with Prostate Biopsy Pathology on MRI US Fusion Biopsy

Nathan Rademaker, M.D.¹, Alexander Parker, B.S.², Kelly L. Stratton, M.D.¹, Justin North, M.D.¹, Brian Cross, M.D.¹, Michael S. Cookson, M.D., M.M.H.C.¹

¹University of Oklahoma, Department of Urologic Surgery, Oklahoma City, OK, ²University of Oklahoma College of Medicine, Oklahoma City, OK

Purpose
MRI Fusion biopsy has been studied in patients with prior negative biopsies; however, its noted improvement in cancer detection may benefit other subsets of patients with clinical concern for prostate cancer. PSA Density has been linked to grade reclassification upon repeat biopsy and in post-prostatectomy pathology. This study evaluated PSA density and cancer detection in men undergoing MRI fusion biopsy across a spectrum of indications.

Materials and methods
We retrospectively reviewed a prostate cancer database of 157 patients undergoing MRI US fusion biopsy. Results were compared across indications including: initial biopsy, active surveillance, and prior negative biopsy. PSA density was calculated using the most recent PSA and the prostate volume at fusion biopsy.

Results
Median PSA density in clinically-significant prostate cancer was 0.23 and 0.12 in those with negative pathology or Gleason 6 disease. Increasing PSA density correlated with increasing gleason score. Clinically significant prostate cancer (GS >/= 7) was discovered in a third of patients with statistical significance via a chi-squared analysis (P: < 0.001). Systematic biopsy outperformed targeted biopsy in prostate cancer and clinically-significant prostate cancer detection in the biopsy naïve and active surveillance subgroups, while targeted biopsy was superior in patients with prior negative biopsy.

Conclusion
Fusion biopsy has clinical usefulness in a variety of settings. Increased PSA density is associated with worse pathologic grade on MRI US fusion prostate biopsy. Incorporating PSA density into decision-making regarding fusion biopsy may improve the detection of clinically significant prostate cancer and decrease risk of repeat prostate biopsy.
Pre-clinical studies using laser speckle contrast imaging to investigate mechanisms contributing to impaired neurovascular coupling in mice: role of mitochondrial oxidative stress

Stefano Tarantini, Ph.D.¹, Marta Noa Valcarcel-Ares, Ph.D.¹, Andriy Yabluchanskiy, Ph.D.¹, Gabor Fulop, M.D.¹, Peter Hertelendy, M.D.¹, Eszter Farkas, Ph.D.¹, William E. Sonntag, Ph.D., Anna Csiszar, M.D., Ph.D.¹, Zoltan Ungvari, M.D., Ph.D.¹

¹Translational Geroscience Laboratory, Reynolds Oklahoma Center on Aging/Department of Geriatric Medicine, University of Oklahoma Health Sciences Center, Oklahoma City, OK

Moment-to-moment adjustment of cerebral blood flow (CBF) via neurovascular coupling has an essential role in maintenance of healthy cognitive function. In advanced age increased oxidative stress and cerebromicrovascular endothelial dysfunction impair neurovascular coupling, likely contributing to age-related decline of higher cortical functions. There is increasing evidence showing that mitochondrial oxidative stress plays a critical role in a range of age-related cellular impairments, but its role in neurovascular uncoupling remains unexplored. The present study was designed to test the hypothesis that attenuation of mitochondrial oxidative stress may exert beneficial effects on neurovascular coupling responses in aging. To test this hypothesis 24 month old C57BL/6 mice were treated with a cell permeable, mitochondria-targeted antioxidant peptide (SS-31; 10 mg/kg/day, i.p.) or vehicle for 2 weeks. Neurovascular coupling was assessed by measuring CBF responses (laser speckle contrast imaging) evoked by contralateral whisker stimulation. We found that neurovascular coupling responses were significantly impaired in aged mice. Treatment with SS-31 significantly improved neurovascular coupling responses by increasing NO-mediated cerebromicrovascular dilation, which was associated with significantly improved spatial working memory, motor skill learning and gait coordination. These findings are paralleled by the protective effects of SS-31 on mitochondrial production of reactive oxygen species and mitochondrial respiration in cultured cerebromicrovascular endothelial cells derived from aged animals. Thus, mitochondrial oxidative stress contributes to age-related cerebromicrovascular dysfunction, exacerbating cognitive decline. We propose that the antioxidant peptide SS-31 should be considered for pharmacological microvascular protection for the prevention/treatment of age-related vascular cognitive impairment (VCI).
**Inhalable Microparticulate SHetA2 Nanocrystals for Lung Cancer Treatment**

Manolya Kukut Hatipoglu¹, PhD, Sanjida Mahjabeen¹, MSc, Doris M. Benbrook², PhD and Lucila Garcia-Contreras¹, PhD

¹Department of Pharmaceutical Sciences and ²Department of Obstetrics and Gynecology, University of Oklahoma Health Sciences Center, Oklahoma city, OK, USA

**Introduction:** Treatment of lung cancer by chemotherapy is associated with severe side effects. Thus, alternative approaches to improve the treatment of lung cancer are needed. SHetA2 is capable to kill cancer cells without harming normal cells. We developed a microparticulate SHetA2 nanocrystal (NCs) formulation for pulmonary administration to maximize concentrations at the site of drug action and enhance the therapeutic efficacy of SHetA2.

**Methods:** SHetA2 NCs were prepared by the bottom-up approach. The process was optimized to yield NCs with the smallest possible geometric diameter (dg) using Design of Experiments (DoE) software. The parameters optimized by the DoE were drug concentration in the organic phase, sonication power, sonication time and ultrasonication time. Morphology of the NCs was evaluated by Scanning Electron Microscopy (SEM). The batch of NCs with the smallest dg was used to manufacture NCs into inhalable microparticles by spray drying. The parameters optimized by the DoE were feed concentration, temperature and mannitol ratio. Three different formulations were obtained: SHetA2 NCs alone, SHetA2-NCs-mannitol 50% and SHetA2-NCs-mannitol 10%. The morphology of the microparticles was evaluated by SEM.

**Results:** Optimized SHetA2 NCs were rectangular prism shaped crystals with dg between 157nm and 710nm and size distribution (GSD) between 1.54 and 2.00. SHetA2 NCs with dg=157 and GSD= 2.0 were produced with 60 min of sonication time, SHetA2 concentration of 75mg/mL, 20 min of ultrasonication time and 10W sonication power. This SHetA2 NC formulation was selected to be spray dried to produce microparticles. The best microparticle formulation for pulmonary delivery was SHetA2-NCs-mannitol 50%, having an average volume diameter, dv=2.53µm±0.08, 95% respirable fraction and manufacturing yield of 35%.

**Conclusion:** The resulting inhalable SHetA2 microparticulate NCs are suitable for pulmonary delivery to mice to evaluate pharmacokinetics and preliminary efficacy in a lung cancer mouse model.
SHetA2 Vaginal Suppositories to Treat Cervical Dysplasia: Influence of Estrous Cycle on Drug Absorption and Pharmacodynamic Endpoint

Sanjida Mahjabeen, MSc¹, Manolya Kukut Hatipoglu, PhD¹, Stanley D. Kosanke, PhD², Doris M. Benbrook, PhD³, Lucila Garcia-Contreras, PhD¹
¹College of Pharmacy, ²Department of Pathology, ³Department of Obstetrics and Gynecology
The University of Oklahoma Health Sciences Center, Oklahoma City, OK

**Purpose:** We demonstrated that vaginal administration of SHetA2 as a suppository achieved cervix concentrations that were >100-fold the therapeutic concentration. However, large variability was observed among mice receiving the same dose, at the same time point. In this study, we evaluated the influence of the stage of estrus cycle on the extent of drug absorption and reduction in cyclin D.

**Method:** FVB female mice received SHetA2 (15mg/kg) in a suppository by the vaginal route. Untreated mice and those receiving suppository without drug (placebo) were used as controls. Mice were euthanized at 0.5, 1, 4, 8h after administration and gynecological tissues collected. A piece of the uterus was used to prepare hematoxylin-eosin slides to determine the stage of estrus cycle in each animal. Drug content in tissues was determined by HPLC. Cyclin D1 expression was determined by double-antibody sandwich ELISA.

**Results:** Overall, drug concentrations in all gynecological tissues were above the therapeutic concentration at all time points, with the highest concentrations observed in the cervix at 0.5h (C_{max,cervix}= 54.84±17.64 µg/g). As in our previous study, significantly large variability (standard-deviation, SD) was observed among mice from same time point, with the smallest SD=30% at 0.5h and the largest SD=100% at 1 and 4 h. Histological analysis revealed that the highest absorption was in the diestrus stage and the lowest absorption at the estrus stage. Both, SHetA2 and placebo suppository induced approximately 70% reduction in cyclin D1 in cervix compared to untreated controls.

**Conclusion** The extent of drug absorption in gynecological tissues after suppository administration appears to be significantly influenced by the stage of the estrus cycle. However, regardless of the stage, drug concentrations at the cervix were above therapeutic level (≥4µM) at all times. Cyclin D1 expression was not affected by these changes.