**Courtney Smith**  
Iowa State University, Undergraduate Student, Dr. Jeff Trimarchi

**Title**  
CRISPR/Cas9 genome editing in the zebrafish Danio rerio as a tool to examine ALS-associated gene function in motor neurons

**Abstract**  
Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disease characterized by the deterioration of the upper and lower motor neurons in the central nervous system. Although the pathogenesis of ALS is largely unknown, exome-sequencing studies have revealed new genes with mutations that are linked to familial or sporadic cases. The current challenge is to understand the in vivo function of these genes, with the hope that this insight will shed light on their connection to ALS pathology. The zebrafish is a great model organism to study this disease because its genome has been sequenced and is very similar to that of humans. To begin our investigation, we used in situ hybridization to visualize the expression of 20 genes throughout zebrafish development. Specifically, we focus on expression in the motor neurons as these are the cells that die in ALS. Next, we have used clustered regulatory interspaced short palindromic repeats (CRISPRs) technology to engineer targeted mutations in a few of these ALS-linked genes in zebrafish. These mutations were induced in an Mnx1:GFP line of transgenic fish to visualize the motor neurons both in development and in possible death and dysfunction. Ultimately, these experiments in zebrafish will provide insight into the role of these genes in motor neuron development and, we hope, ultimately point us toward a definitive link with ALS.

**Matt Jefferson**  
Iowa State University, Graduate Student, Dr. Marian Kohut

**Title**  
PKR as an Early Neuroinflammatory Mediator in Diet-Induced Obesity: Implications for Parkinson’s Disease?

**Abstract**  
Neuroinflammation resulting from host factor detriments, such as obesity, has emerged as a well-recognized component of Parkinsonian pathogenesis. While it’s understood that low-grade, systemic inflammation from obesity is capable of perturbing CNS homeostasis, the characterization of these inflammatory pathways in the brain is enigmatic and highly context-dependent. Our lab has previously identified the expression of double-stranded RNA-dependent protein kinase (PKR) in murine brains fed a long-term high fat diet. PKR may potentially serve as a neuroinflammatory target for therapeutic intervention in obese populations at risk for Parkinson’s disease (PD). PKR activation has been suggested to
serve as an intracellular danger-sensing mechanism and may precede NLRP3 inflammasome activation. To determine its clinical relevancy to Parkinsonian pathogenesis, we established a chronic model of diet-induced obesity in C57BL/6 mice over 14 weeks, followed by an acute MPTP challenge (5 mg/kg, i.p.). Brains were micro-dissected at 1 and 7 days post-MPTP challenge and surveyed for gene (qPCR) and protein (western blot) expression, revealing PKR up-regulation in the hippocampus at 1 day post-MPTP. Additionally, this expression appears regionally selective to the hippocampus, is met by concurrent microglial activation in combination of high fat feeding and MPTP, and does not correspond to NLRP3 expression. This suggests PKR as an early, innate response in the obese brain following Parkinsonian insult. Given the current absence of disease-modifying therapies for PD, focusing on modifiable lifestyle factors such as obesity and their neuroinflammatory signature may yield meaningful preclinical insights into the progressive nature of this disease.

| Name (University & Position) | Dr. Auriel Willette  
Iowa State University, Assistant Professor |
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<tr>
<td>Title</td>
<td>Insulin resistance and longitudinal brain pathology in Alzheimer's disease</td>
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<td>Abstract</td>
<td>BACKGROUND: Half of Americans by age 65 develop some degree of insulin resistance (IR) related to pre-type 2 diabetes, increasing Alzheimer’s disease (AD) risk. We have shown in aged adults across the AD spectrum that IR cross-sectionally predicts brain atrophy in AD-sensitive brain regions, which is a disease hallmark. Yet, it is not known how IR longitudinally tracks brain atrophy in these areas, as well as amyloid and tau proteins in the brain that are thought to cause AD. OBJECTIVES: We examined longitudinal associations between IR, biomarkers of amyloid and tau using cerebrospinal fluid, and brain volume using Magnetic Resonance Imaging. We predicted that higher IR would be related to more regional brain atrophy and higher protein biomarker levels over 3 years. METHODS: Data was obtained from 205 aged adults (55-89 years) from the Alzheimer’s Disease Neuroimaging Initiative (ADNI). Structural brain networks were derived from FreeSurfer 5.1 data. Growth curve modeling in R regressed IR main effects against outcomes, as well as interactions with clinical diagnosis. Fasting glucose and insulin, which compose IR, were also examined. RESULTS: Higher IR was related to progressive atrophy along the AD spectrum in AD-sensitive areas like medial temporal (R²=0.406),</td>
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inferior temporal (R^2=0.40), parietal (R^2=0.235), and frontal (R^2=0.302) structural networks. Effects were strongest in patients with mild AD. Fasting glucose but not IR was strongly associated with amyloid (R^2=0.55) and tau species like ptau-181 (R^2=0.32).

CONCLUSION: Higher IR may be a relevant predictor to track longitudinal brain changes in AD and a potential therapeutic target.

| Name (University & Position) | Mark Hartman  
Iowa State University, Graduate Student, Dr. Panteleimon Ekkekakis |
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<td>Title</td>
<td>Using transcranial direct current stimulation (tDCS) during exercise to modulate affect and performance: A pilot study</td>
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| Abstract                    | Research on psychological and neurological models of exertional fatigue are extremely limited. Exercise evokes changes in affect (i.e. pleasure-displeasure) which are directly related to the exercise intensity. Significant declines in affect during exercise coincide with exhaustion. Based on multiple lines of evidence, during exhaustive exercise, the rate of decrease in dorsolateral pre-frontal cortex (dIPFC) oxygenation or hypo-metabolism, correlates with decreases in affect. Transcranial Direct Current Stimulation (tDCS) increases regional brain activation and improves regional blood oxygenation. Therefore, tDCS applied on the dIPFC during exercise may influence affective responses and performance. METHODS: In a randomized counterbalanced-order, 7 participants performed an incremental cycling exercise to volitional exhaustion while receiving either anodal tDCS applied over the dIPFC (AF3) or sham stimulation. Time to exhaustion (tLim), pulmonary gas-exchange, and self-ratings of affective valence were recorded. Ventilatory threshold (VT), respiratory compensation point (RCP), and maximal oxygen uptake (VO2max) were identified from the gas exchange data to demarcate exercise domains (i.e, moderate, heavy, and severe). RESULTS: tDCS resulted in 14.3% (0% - 29.1%) improvement in affect during heavy exercise, 4.5% (-5.7% - 48.5%) improvement during severe exercise, and 4.7% (-3.9% - 29.4%) improvement during moderate exercise. tDCS improved performance (tLim) by 1.8% (-11.2% - 6.3%). No changes were observed in VO2max, VT, or RCP. CONCLUSION: Based on preliminary pilot data, increased activation of the dIPFC attenuates feelings of displeasure during high intensity exercise. Larger studies with greater statistical power are needed to validate the affect modulation effects induced by tDCS during exercise and associated performance gains.
| Name (University & Position) | Steve Anderson  
Iowa State University, Graduate Student, Dr. Eric Cooper & Dr. Elizabeth Stegemoller |
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<td>Title</td>
<td>Testing the hypothesis: Disruptions in complex object recognition which occur in PD are the result of abnormal dopaminergic signaling in the ventral visual pathway</td>
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<td>Abstract</td>
<td>While Parkinson’s disease (PD) is most commonly associated with deficits in motor functioning, in many cases it is also characterized by a variety of deficits in cognition and perception. Specifically, a broad spectrum of visual symptoms has been observed in individuals diagnosed with PD, ranging from impairments in low-level vision (e.g., deficits in visual acuity and contrast sensitivity) to difficulty with relatively high-level visual processing (e.g., object recognition and mental object manipulation). Many of the signature effects of PD result from the death of dopaminergic neurons in the substantia nigra pars compacta (SNC), a structure in the basal ganglia (BG). The pathology in these neurons produces a profound dopamine (DA) deficiency in the areas to which they project. The fact that DA receptors have been discovered in a variety of regions throughout the human visual system thus lends credence to the view that abnormal dopaminergic transmission could be responsible for the impairments that individuals with PD display on visual tasks. Additionally, several pathways have been identified which strongly connect areas of the ventral visual pathway with structures in the BG which are known to exhibit DA deficiency in individuals with PD. This talk will briefly describe a study designed to investigate the hypothesis that disruptions in complex object recognition which occur in PD are the result of abnormal dopaminergic signaling in the ventral visual pathway.</td>
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| Name (University & Position) | Emir Malovic  
Iowa State University, Graduate Student, Dr. Anumantha Kanthasamy |
| Title                      | Manganese exposure induces neuroinflammation by impairing mitochondrial dynamics in astrocytes |
| Abstract                   | Chronic manganese (Mn) exposure induces neurotoxicity, which is characterized by Parkinsonian symptoms resulting from impairment of the basal ganglia. Mitochondrial dysfunction and oxidative stress are considered key pathophysiological features of Mn neurotoxicity. Recent evidence suggests astrocytes are a major target of Mn neurotoxicity since Mn accumulates predominantly in astrocytes; however, these mechanisms of metal neurotoxicity are not
completely understood. In this study, we examined the interrelationship between mitochondrial dysfunction and astrocytic inflammation in Mn neurotoxicity. We first evaluated whether Mn exposure alters mitochondrial bioenergetics in cultured astrocytes. Metabolic activity results by MTS assay prompted us to use 100 μM Mn for both primary mouse astrocytes and U373 human astrocytes. Furthermore, Mn treatment reduced mitochondrial mass, indicative of impaired mitochondrial function and biogenesis, which is substantiated by the significant reduction in mRNA of mitofusin-2, a protein that serves as a ubiquitination target for mitophagy. Seahorse analysis of bioenergetics status in Mn-treated astrocytes revealed that Mn significantly decreased the basal mitochondrial oxygen consumption rate as well as ATP production. Since astrocytes regulate immune functions in CNS, we also evaluated whether Mn modulates astrocytic inflammation. Mn exposure in astrocytes not only stimulated the release of proinflammatory cytokines, but also exacerbated the inflammatory response induced by aggregated ß-synuclein. Lastly, the novel mitochondria-targeted antioxidant, mito-apocynin, significantly attenuated Mn-induced inflammatory gene expression in U373s, further supporting the role of mitochondria dysfunction and oxidative stress in mediating astrogliosis. Collectively, we demonstrate for the first time that Mn drives proinflammatory events in astrocytes by impairing mitochondrial bioenergetics.