Peter J Bergeaux
Medical, L2
LSUHSC School of Medicine, New Orleans, LA

Dr. Liz Simon:
LSUHSC, Associate Professor of Physiology
Comprehensive Alcohol-HIV/AIDS Research Center

Chronic Binge Alcohol Impairs Myoblast Differentiation: Role of microRNA-206

Background: With recent advances in antiretroviral therapy, people living with HIV (PLWH) now have a near-normal life expectancy. As such, PLWH experience aging-related comorbidities, such as metabolic disorders and frailty earlier in life than the general population. At-risk alcohol use is twice as likely in PLWH compared to the general population and alcoholic myopathy occurs in 40-60% of people with an alcohol use disorder. Previous studies demonstrate that chronic binge alcohol (CBA) in simian immunodeficiency virus (SIV)-infected rhesus macaques causes decreased myoblast differentiation and downregulation of microRNA-206 (miR-206) in skeletal muscle. Additionally, lower miR-206 expression is associated with increased expression of myocyte enhancing factor 2C (MEF2C), a transcription factor important for differentiation.

Hypothesis: We hypothesize that increasing miR-206 expression in CBA-derived primary macaque myoblasts will increase differentiation.

Methods: Eight SIV-infected female rhesus macaques received either water (VEH) or CBA (13-14 g/kg/week) for 14.5 months. Three months into either VEH or CBA treatment, animals were vaginally infected with $SIV_{mac251,}$ and antiretroviral therapy was started 2.5 months later. Animals were sacrificed after 9 months, and primary cells were derived from vastus lateralis skeletal muscle tissue 24 hours after the last dose of alcohol (blood alcohol=0 mM). Myoblasts at passage 4 (VEH & CBA) were proliferated for three days. Transfection with either miR-206 or miR scramble as a control occurred on day 0 of differentiation creating four groups: VEH+scramble, CBA+scramble, VEH+miR-206, and CBA+miR-206. Cells were allowed to differentiate for five days before analysis. Expression levels of miR-206 were determined using qRT-PCR. Fusion index was calculated by determining the percentage of myonuclei that have fused into myotubes after HEMA 3 staining.

Results: Two-way ANOVA revealed increased miR-206 levels (about 150 fold, $p<0.01$) and increased fusion index ($p<0.01$) with miR-206 transfection. No significant effects of miR-206 expression or fusion index were found between VEH and CBA groups likely due to the effect of transfection.

Conclusions: Our results suggest that miR-206 is important for myoblast differentiation. Future studies will determine the effect of miR-206 transfection on histone deacetylase 4 (HDAC4), a target of miR-206, and MEF2C expression.