Name: Falk W. Lohoff Email: falk.lohoff@nih.gov

Additive Effects of Stress and Alcohol on Epigenetic Aging

Jeesun Jung, Ph.D.[1], Daniel L. McCartney, Ph.D.[2], Josephin Wagner, M.D.[1], Joyce Yoo, B.A.[1], Andrew S. Bell, B.A.[1], Lucas A. Mavromatis, B.S.[1], Daniel B. Rosoff, B.S.[1], Colin A. Hodgkinson, Ph.D.[3], Hui Sun, M.D.[3], Melanie Schwandt, Ph.D.[3], Nancy Diazgranados, M.D.[3], Alicia K. Smith, Ph.D.[4,5], Vasiliki Michopoulos, Ph.D.[5], Abigail Powers, Ph.D. [5], Jennifer Stevens, Ph.D.[5], Bekh Bradley, Ph.D.[5], Negar Fani, Ph.D.[5], Rosie M. Walker, Ph.D. [2], Archie Campbell, Ph.D.[2], David J. Porteous, Ph.D. [2], Andrew M. McIntosh, M.D.[2], Steve Horvath, Ph.D.[6,7], Riccardo E. Marioni, Ph.D.[2], Kathryn Evans, Ph.D.[2], David Goldman, M.D.[3*], Falk W. Lohoff, M.D.[1*#]

[1]Section on Clinical Genomics and Experimental Therapeutics, National Institute on Alcohol Abuse and Alcoholism, National Institutes of Health, Bethesda, MD, USA; [2]Centre for Genomic and Experimental Medicine, Institute of Genetics and Cancer, University of Edinburgh, Edinburgh, United Kingdom; [3] Laboratory of Neurogenetics, National Institute on Alcohol Abuse and Alcoholism, National Institutes of Health, Bethesda, MD, USA; [4]Emory University School of Medicine, Department of Gynecology & Obstetrics, Atlanta, GA, USA; [5]Emory University School of Medicine, Department of Psychiatry and Behavioral Sciences, Atlanta, GA, USA; [6]Department of Human Genetics, David Geffen School of Medicine, University of California Los Angeles, Los Angeles, CA, USA; [7]Department of Biostatistics, Fielding School of Public Health, University of California Los Angeles, Los Angeles, CA, USA

Stress contributes to premature aging and susceptibility to alcohol use disorder (AUD) and AUD itself is a factor in premature aging; however, the interrelationships of stress, AUD and premature aging are poorly understood.

We constructed a composite score of stress (CSS) from thirteen stress-related outcomes in a discovery cohort of 317 individuals with AUD and controls. We then developed a novel methylation score of stress (MS Stress) as a proxy of CSS comprising 211 CpGs selected by a penalized regression model. The effects of MS Stress on health outcomes and epigenetic aging were assessed in a sample of 615 AUD patients and controls using epigenetic clocks and DNAm telomere length (DNAmTL). Statistical analysis with an additive model using MS Stress and a methylation score for alcohol consumption (MS Alcohol) were conducted. Results were replicated in two independent cohorts (Generation Scotland GS n=7028 and the Grady Trauma Project GTP n=795).

CSS and MS Stress were strongly associated with heavy alcohol consumption, trauma experience, epigenetic age acceleration (EAA) and shortened DNAmTL in AUD. Together, MS Stress and MS Alcohol additively showed strong stepwise increases in EAA. Replication analyses showed robust association between MS Stress and EAA in the GS and GTP cohort.

A methylation-derived score tracking stress exposure is associated with various stress-related phenotypes and EAA. Stress and alcohol have additive effects on aging, offering new insights into the pathophysiology of premature aging in AUD, and potentially, other aspects of gene dysregulation in this disorder.