Newborn blood DNA methylation correlates likelihood of autism spectrum disorder with grandparental cigarette smoking

George E. Kuodza[1-4], Julia S. Mouat[1-4], Nickilou Y. Krigbaum[6], Viki Haghani[1-4], Emily G. Thrall[1-4], Ray Kawai[1-4], Sophia Hakam[1-4], Yunin J.L. Rodriguez[5], Timothy N. Sullivan[4], Aron Judd P. Mendiola[1-4],Dag H. Yasui[1-4], Taylor A. Tran[1-4], Osman Sharifi[1-4], Christina G. Torres[1-4], Deborah H. Bennett[3-5], Rebecca J. Schmidt[3-5], Michele A. La Merrill[7], Piera M. Cirilo[6], Irva Hertz-Picciotto[5], Barbara A. Cohn[6],Janine M. LaSalle[1-4]*

[1]Department of Medical Microbiology and Immunology, School of Medicine, University of California, Davis, CA, USA; [2]Genome Center, University of California, Davis, CA, USA; [3]Perinatal Origins of Disparities Center, University of California, Davis, CA, USA; [4]MIND Institute, University of California, Davis, CA, USA; [5]Department of Public Health Sciences, University of California, Davis, CA, USA; [6]Child Health and Development Studies, Public Health Institute, Berkeley; [7]Department of Environmental Toxicology, University of California, Davis, CA, USA

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***Abstract*:** Autism spectrum disorder (ASD) cases have been on the rise, and the underlying causes are not fully understood. Grandparental non-genetic factors, such as smoking and advanced age at conception, have been associated with likelihood of ASD in grandchildren, but potential molecular mechanisms warrant further investigation. We examine ASD versus typically developing (TD) newborn dried blood spots (NDBS) DNA methylation (DNAm) patterns to make biological connections between grandparental exposures and grandchildren's ASD diagnosis. We used two unique three-generation cohorts from California. We obtained NDBS from 456 grandchildren (245 ASD, 211 TD) and assayed DNAm using whole-genome bisulfite sequencing. To analyze the DNAm, we identified correlation methylation (co-methylation) networks using Comethyl, which were further correlated with grandchildren's neurodevelopment and grandparental exposures. We identified five co-methylation networks significantly associated with ASD. The light-yellow module was negatively correlated with both ASD (Bicor=-0.1, p=0.04) and paternal grandparents' smoking (Bicor=-0.23, p=0.04). Some of the genes in this network, such as *CD164*, have previously been associated with schizophrenia and smoking. The results from this study provide biological insight into grandparental factors associated with the likelihood of ASD and show that altered DNAm may be a mechanism by which grandparental factors, such as smoking transfers across generations.

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***Contact Information for Presenting Author**: gekuodza@health.ucdavis.edu*