

Name: Fang Fang

Email: ffang@rti.org

Reversibility of DNA Methylation Changes Associated with Cannabis Use

Stephanie N. Giamberardino¹, Aino E. Heikkinen², Mikaela Hukkanen², Miina Ollikainen²,
Jaakko Kaprio², Dana B. Hancock¹, Eric O. Johnson^{1,3}, Fang Fang¹

¹GenOmics and Translational Research Center, RTI International

²Institute for Molecular Medicine Finland, University of Helsinki

³Fellow Program, RTI International

Cannabis use may induce changes across the DNA methylome, suggesting that DNA methylation (DNAm) could serve as a sensitive biomarker for cannabis use. Previously, we identified DNAm associations with lifetime cannabis use (ever vs. never), but these changes may be reversible upon cessation. Using DNAm data from 2,216 human blood samples across two cohorts, we conducted two meta-analyses of epigenome-wide association study (EWAS) for (1) current cannabis users (n=385) vs. never users (n=1,475); and (2) former users (quit >1 year, n=356) vs never users. Our comparison revealed that most DNAm changes associated with current cannabis use attenuated after a year of quitting, with a correlation of 0.26 between the effect sizes from the two EWAS meta-analyses. Using the EWAS Toolkit, we examined the overlap between our top 20 CpGs associated with current cannabis use and epigenome-wide results from the EWAS Atlas. Our analysis identified significant enrichment in various health conditions and traits, including cardiovascular and metabolic health, immune diseases, substance use, pain syndrome, carcinoma, aging, and air pollution. Pathway analysis indicated that the most enriched pathway for genes implicated in current cannabis use is cholesterol metabolism.

This study is the first to investigate the reversion of DNAm changes associated with cannabis use upon quitting. We found that DNAm levels of most CpGs revert to those observed in never users after a year of cessation, providing preliminary evidence for the reversibility of DNAm associated with cannabis use.