Candidate Genes for Cocaine and Methamphetamine Preference in *Drosophila melanogaster*

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In the United States, drug use accounts for monetary losses exceeding 193 billion dollars annually. Stimulant use disorder is characterized by drug-seeking behavior and consumption of psychostimulants in the face of deleterious effects. Our understanding of the mechanistic and physiological causes of stimulant use disorder is imperfect due to the difficulty in performing population genetic analyses in humans and vertebrate models. The *Drosophila melanogaster* Genetic Reference Panel (DGRP) consists of 205 sequenced inbred lines derived from a natural population. These lines are largely unrelated, highly polymorphic, and exhibit a rapid decay in local linkage disequilibrium – all favorable for genome wide association (GWA) mapping. We modified the Capillary Feeding (CAFÉ) assay to quantify consumption, preference, and tolerance for cocaine and methamphetamine relative to sucrose, and assessed variation in consumption, preference, and tolerance for 47 of the most genetically diverse DGRP lines. Briefly, each line was given a choice between sucrose and sucrose plus cocaine or methamphetamine for three consecutive days. Preference was scored as the proportion of drug consumed and tolerance as the difference in preference on days one and three. We observed significant genetic variation, including genetic variation in sexual dimorphism, for both traits, enabling us to perform GWA analyses for consumption, preference, and tolerance. We identified 830 variants in 411 genes at a nominal $P < 10^{-5}$. Several candidate genes were involved in dopamine signaling; others were novel. We used RNAi to knock down of gene expression for a sample of candidate genes with ubiquitous and brain-specific drivers and observed that a large proportion of RNAi targets affect preference and tolerance. Thus, *Drosophila melanogaster* can be developed as a powerful model system to identify evolutionarily conserved genes that affect drug-seeking behavior.