Investigation of Nicotine's Effect on *Drosophila Melanogaster* Motor Function and Aggression

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Introduction

- *Drosophila melanogaster* (*D. melanogaster*) is a popular model organism in neurobiology.
- The CDC (2019) claims that nicotine in e-cigarettes can cause adverse health effects in children and teenagers.
- Acetylcholine, a neurotransmitter increased by the use of nicotine, impacts the regulation of motor movement.
- Dopamine and serotonin are two neurotransmitters that are tied to aggressive behavior, and both of these transmitter levels are affected by the use of nicotine as well.
- **Hypothesis:** We hypothesized that nicotine from e-cigarette vapor will reduce D. melanogaster motor function and increase aggression.

Methods

- **Culturing:** Weekly, 5 female and 5 male *D. Melanogaster* were placed into a vial containing yeast and a growth medium, with the purpose of continuing the population for experimentation.
- **RING Assay:** *D. Melanogaster* were treated with the assigned banana nicotine treatment as seen in Fig 2. Then we began to video record the RING assay. The vial (with a marking every cm) was hit on the table 3 times, and 6 seconds after the last hit, the measurement of where the flies were located was recorded.
- **Aggression Latency Assay:** Aggression was measured through placing two adult male *D. Melanogaster* into a well plate as seen in Fig 4, and treated with the assigned banana nicotine treatment. Then a video recording was set up, and the time it took for each well to display charging or boxing behavior was recorded.

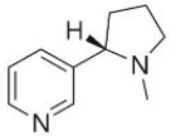

Figure 1. Chemical structure of the compound nicotine.

Figure 2. The diagram above shows our method for vaping *D. melanogaster*.

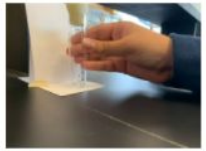

Figure 3. The diagram above shows our method for the RING assay.

Figure 4. The diagram above shows our method for observing the *D. melanogaster* aggression assay.

Results

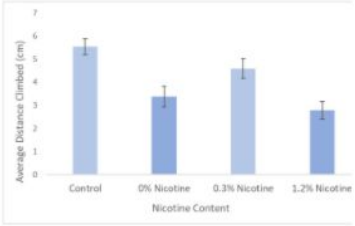

Figure 5. Results of RING assay after exposure to various nicotine concentrations with average distance climbed (cm). Error bars display standard error. t-tests assuming equal variances were performed between the control and the 0% nicotine group (p-value < 0.001), the control and the 0.3% nicotine group (p-value = 0.084), the control and the 1.2% nicotine group (p-value < 0.001), the 0% nicotine group and the 0.3% nicotine group (p-value = 0.052), the 0% nicotine group and the 1.2% nicotine group (p-value = 0.31), and the 0.3% nicotine group and the 1.2% nicotine group (p-value = 0.0021).

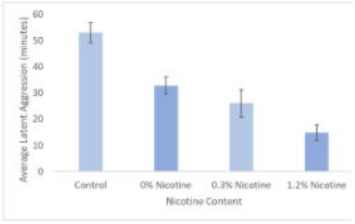

Figure 6. Results of aggression latency assay after exposure to various nicotine concentrations with average aggression.latency. Error bars display standard error.t-tests assuming equal variances were performed between the control and the 0% nicotine group (p-value = 0.0022), the control and the 0.3% nicotine group (p-value = 0.0019), the control and the 1.2% nicotine group (p-value < 0.001), the 0% nicotine group and the 0.3% nicotine group (p-value = 0.29), the 0% nicotine group and the 1.2% nicotine group (p-value = 0.0021), and the 0.3% nicotine group and the 1.2% nicotine group (p-value = 0.094).

Analysis/Conclusion

- There was no statistical difference between the motor ability of the 0% nicotine group and the 0.3% and 1.2% nicotine group.
- We did find a statistical difference between the motor ability of the control group and the 0% and 1.2% nicotine group.
- It was noticed that the flies who were under the influence of nicotine did demonstrate erratic behavior.
- There was a statistical difference in the latent aggression assay between the 0% nicotine group and the 1.2% nicotine group.
- We also found a statistical difference in the latent aggression assay between control group and the three nicotine groups.
- While our results cannot definitively say that nicotine concentration is the sole factor in affecting *D. melanogaster* behavior, we do have strong evidence that vaping does.

References

Dierick, Herman. "A method for quantifying aggression in male Drosophila melanogaster." Nature protocols 2, no. 11 (2007): 2712-2718.

Gargano, Julia Warner, Ian Martin, Poonam Bhandari, and Michael S. Grotewiel. "Rapid iterative negative geotaxis (RING): a new method for assessing age-related locomotor decline in Drosophila." Experimental gerontology 40, no. 5 (2005): 386-395.

National Center for Biotechnology Information (2021). PubChem Compound Summary for CID 89594, Nicotine.

"Quick Facts on the Risks of E-Cigarettes for Young People" Centers for Disease Control and Prevention (CDC), (2019),

Roberts, David. "Drosophila melanogaster: the model organism." Entomologia experimentalis et applicata 121, no. 2 (2006): 93-103.

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