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Effect of Nicotine on Spitz Gene Expression in *Drosophila melanogaster*

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Introduction

- The multicellular eukaryotic organism, *Drosophila melanogaster*, commonly known as the fruit fly, is a popular model organism in biology research.
- The *Spitz* gene, found in the brain, codes for a RAS signaling protein and is involved in a variety of differentiation processes and ventralization of the embryo.
- Nicotine, a known carcinogen, affects Nicotinic Acetylcholine Receptors, and can cause a variety of health issues. As nicotine is being found in more products, including electronic cigarettes and vape pens, it is important to continue studying the effect it has on the developing brain.
- **Hypothesis:** If *Drosophila melanogaster* are exposed to nicotine, then there will be an increase in expression of the *Spitz* gene, as well as increased aggression in adult flies.

Methods

- **Primer synthesis:** Primers for *Spitz* were designed using IDT Oligoanalyzer software.
- **Culturing:** *Drosophila melanogaster* cultures were created using 10 mL of potato flakes, yeast granules, and a 0.01% nicotine/water mixture. Flies were exposed to nicotine for 24 hours at 22.5 °C.
- **RNA extraction:** RNA was extracted using Trizol and purified using Qiagen's RNeasy Mini Kit.
- **Reverse transcription:** cDNA was synthesized using RevertAid.
- **qPCR** was performed using PowerUp SyBr Green Master Mix along with primers targeting *Spitz* and *GAPDH*.
- **Aggression Assays** were performed to determine if nicotine exposure caused an increase in aggressive behaviors in the flies. The aggressive behaviors quantified were lunging, head butting, and wings up.

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Results

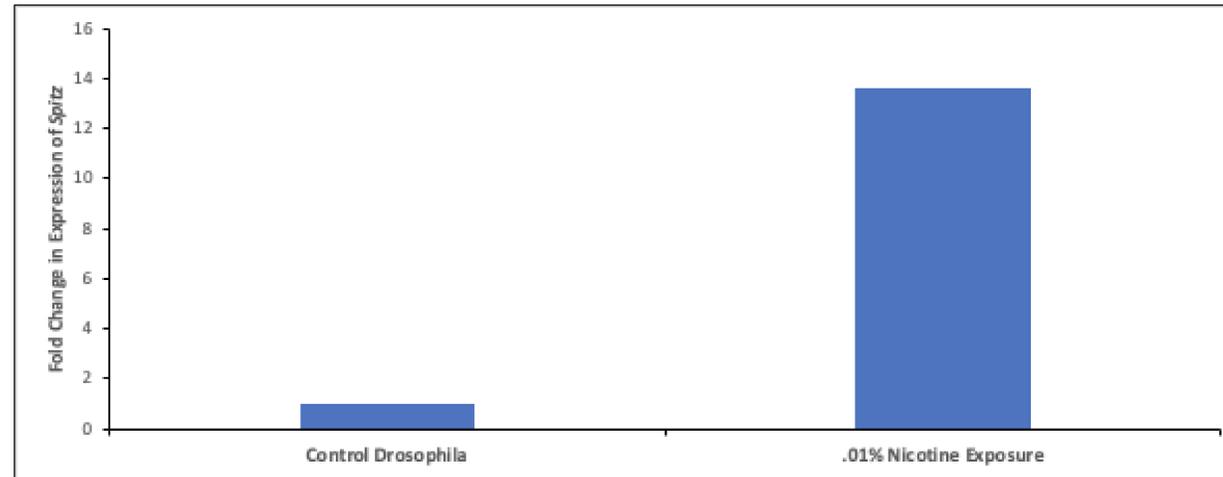


Figure 1: Fold change in *Spitz* expression in both control and nicotine exposed groups of *Drosophila melanogaster*. ($p=0.416$). ($n=8$ per group)

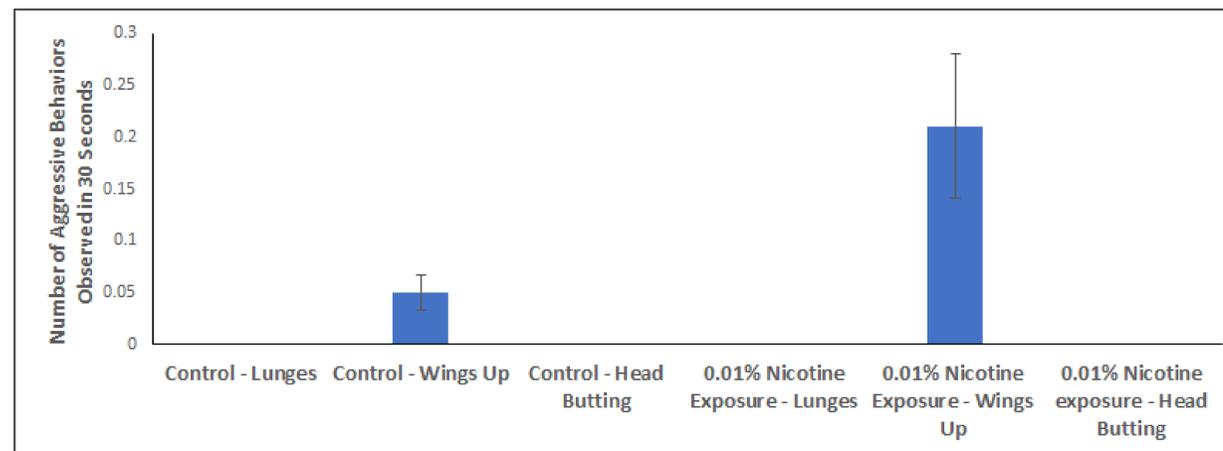


Figure 2: Results of aggression assay performed on control and nicotine exposed groups of *Drosophila melanogaster*. ($p=.535$) ($n=20$ per group)



Figure 3. Image of control *Drosophila melanogaster*.

Conclusion

- The aggression assay results were insignificant and showed no increased aggression in flies exposed to nicotine compared to control flies, as seen in Figure 2, but nicotine exposed flies did seem to have more erratic movements.
- There was a 13 fold increase in the quantitative relative expression of *Spitz*, as shown in Figure 1, but it was not statistically significant ($p=0.416$).
- The results disagreed with our hypothesis on gene expression and this is likely due to the small sample size used.
- For future research, larger sample sizes would be recommended to yield significant results.

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