Introduction

Hypothesis

Exposure to GenX will increase expression of Torsin in male fruit fly brains due to their much stronger reaction to dopamine compared to female brains.

Drosophila melanogaster (fruit fly)

- Shares 75% of human disease-related genes
- Easy to take care of and exposure to various treatments
- Sexually dimorphic
- Commonly-used model organism

Torsin

- Conserved in both humans and fruit flies
- Involved in ATP binding and ATPase activity
- Regulates dopamine metabolic process, neuron cellular homeostasis, and positive regulation of cellular growth

Gen-X

- Used to make non-stick coatings, firefighting foam, food packaging
- Mammals chemical and created to be a replacement for PFAS/PFOA chemicals
- Has been found in both rain and groundwater

Materials and Methodology

Primers used:

Forward: 3’ TCCGGGCGAATAGATGAATTAG 5’
Reverse: 3’ TTCAATGTGGGGACGATATG 5’

qRT-PCR

Results

- Does exposing Drosophila melanogaster (fruit flies) to GenX have any effects on the expression of Torsin?
- How does the expression of Torsin differ between male and female fruit flies?
- Gene expression of Torsin in male and female fruit flies with and without exposure to GenX was measured using qRT-PCR.
- Torsin was downregulated 1048.03 fold from female to male controls.
- Fold difference in gene expression was calculated using 2^ΔΔCt

RNA was extracted using the RNeasy kit (QiaGen).

The flies were separated by sex ≤ 4h post-eclosion

Control: Fed cornmeal-based food for 7 days
Experimental: Fed cornmeal-based food dosed with 1000mg/kg of GenX for 7 days

Flies were sacrificed via freezing at -80°C. Brains were dissected and pooled with 100 brains in each sample

Data Analysis

- Torsin expression was normalized to Actin
- Fold difference in gene expression was calculated using 2^ΔΔCt

- ΔΔCt was calculated for the pairs of conditions that were compared by taking the ΔCt difference between female and control males, as well as between experimental condition and control condition for both males and females.
- Student’s t-test was performed using ΔCt values. If the p-value is <0.05, the data is considered significant.

Study Limitations and Future Directions

- GenX exposures were only conducted on adult life stages.
- Future experiments can begin exposing the fruit flies at earlier developmental stages.
- The effects of GenX exposures was only studied on the fruit fly brain.
- RNA can be extracted from other organs to study changes in gene expression
- Flies were only exposed to GenX for 7 days and other time points were not measured.
- Flies can be exposed to GenX for varying lengths of time

Discussion and Conclusion

- Female fruit fly brains have a statistically significant lower Torsin expression level than male flies at baseline (p<0.05).
- There is no significant effect on the gene expression of Torsin due to the GenX exposure through fly food.
- For female exposure vs. control groups, p = 0.0549
- For males exposure vs. control groups, p = 0.8405
- Torsin has a part in ATP binding and ATPase activity which lets us conclude that
- Female fruit flies tend to utilize ATP less compared to male fruit flies.
- GenX exposure did not affect ATP utilization in fruit flies.
- Torsin relates to proteins like strol and CN3 (Fig. 3) which are related to many cellular metabolic and developmental processes.
- Because Torsin is related to ATP utilization, difference in Torsin expression level could have implications on cellular energy management and affect the other cellular functions as mentioned above.

Summary

- Sexually dimorphic fold difference in gene expression was calculated using 2- ΔΔCt related to ATP utilization, difference in gene expression was calculated using 2^ΔΔCt.
- Torsin was downregulated 1048.03 fold from female to male controls.
- Fold difference in gene expression was calculated using 2^ΔΔCt.
- There is no significant effect on the gene expression of Torsin due to the GenX exposure through fly food.
- For female exposure vs. control groups, p = 0.0549
- For males exposure vs. control groups, p = 0.8405
- Torsin has a part in ATP binding and ATPase activity which lets us conclude that
- Female fruit flies tend to utilize ATP less compared to male fruit flies.
- GenX exposure did not affect ATP utilization in fruit flies.
- Torsin relates to proteins like strol and CN3 (Fig. 3) which are related to many cellular metabolic and developmental processes.
- Because Torsin is related to ATP utilization, difference in Torsin expression level could have implications on cellular energy management and affect the other cellular functions as mentioned above.

Torsin is significantly upregulated in Male fruit fly brain compared to Female fruit fly brains

Kotamarti A.1, Contreras J.2, Zida S.3, Perez Orozco D.4, Hirata K.5

1 U.S. Grant High School, 2 Castle Park High School, 3 Oguaa Senior High School, 4 Sweetwater High School, 5 Boz Life Science Research and Teaching Institute.

1 U.S. Grant High School, 2 Castle Park High School, 3 Oguaa Senior High School, 4 Sweetwater High School, 5 Boz Life Science Research and Teaching Institute.