

GenX treatment resulted in unaltered expression of protein-modification gene *sir-2.1* in *Caenorhabditis elegans*

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Summary

- Does feeding *C. elegans* with GenX-exposed bacteria affect the expression of *sir-2.1*?
- Quantified gene expression of larval *C. elegans* via qRT-PCR.
- No significant change in *sir-2.1* expression was observed.

Abstract

GenX, a potential toxic chemical used to manufacture many products, such as cookware, laptops and cell phones, has raised many questions surrounding its safety. We investigated its effects on gene expression. *C. elegans* were exposed to GenX during their early lives, from L1 to the beginning of adulthood. Our results show that there was no significant change in *sir-2.1* expression level, but expression of other genes were affected.

Introduction

Hypothesis: GenX exposure through *Escherichia coli* (*E. coli*) OP50 changes the expression of *sir-2.1* in *C. elegans*.

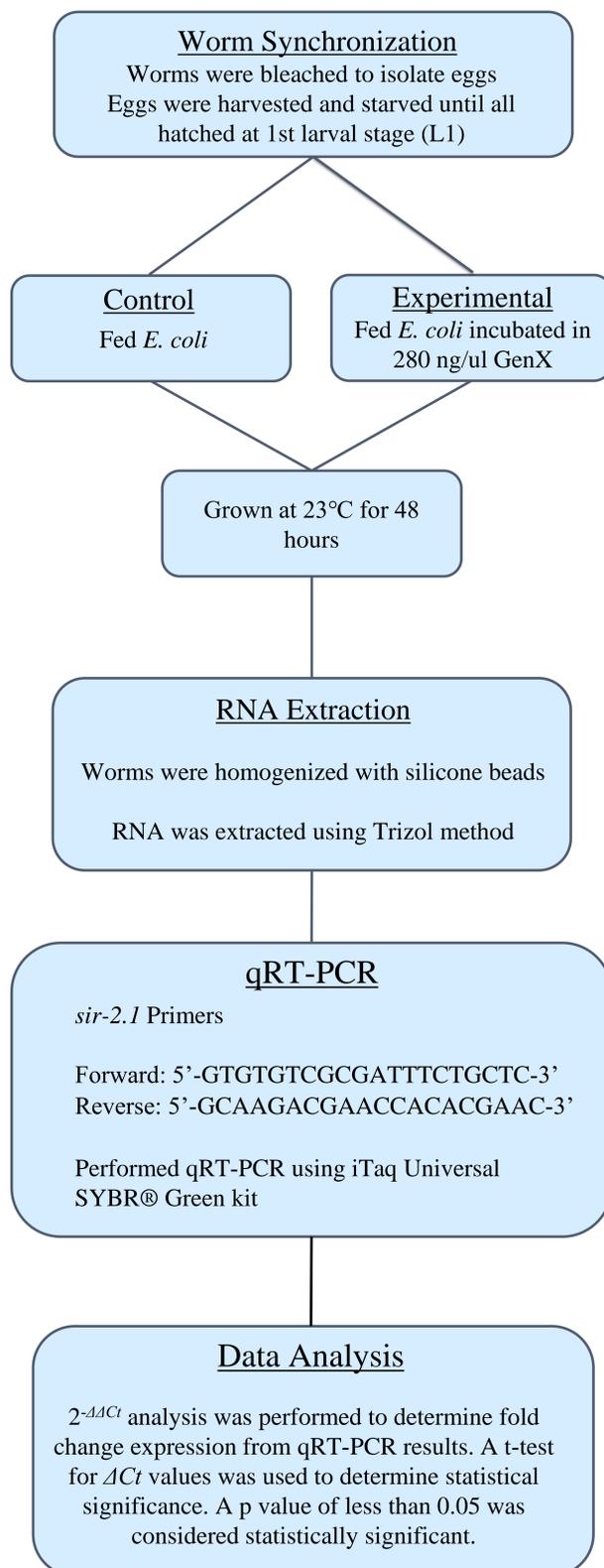
- **GenX** - a potentially toxic chemical that is used to make food packaging, paint, and non-stick coatings.¹
- ***C. elegans*** - our model organism. *C. elegans* have neurons and muscles that are similar in function to those of humans.²



Figure 1. Photo of a *C. elegans* at its third larval stage (L3). This photo was taken with an inverted microscope at 10X magnification.

- **Target gene** - Our target gene is *sir-2.1*. It is associated with cellular protein modification, and with the signaling in response to DNA damage. The *sir-2.1* gene is involved in chromatin silencing and aging.^{3,4,5}
- **Treatment** - *C. elegans* were treated by feeding them *E. coli* incubated with 280 ng/L GenX (EPA draft health advisory level).^{6,7}

Materials and Methodology



Results

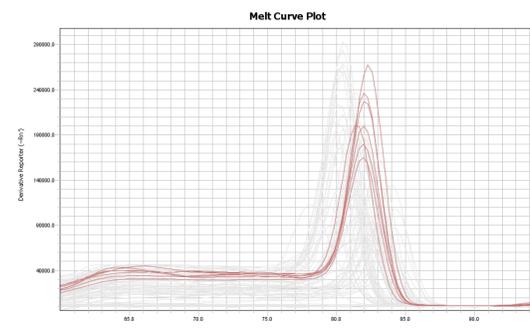


Figure 2. Melt curve plot showing melting temperatures of amplified products of qRT-PCR. Overlapping peaks at the same temperature indicate same or highly similar amplification. This figure was generated by QuantStudio™ Design & Analysis Software v1.5.1.

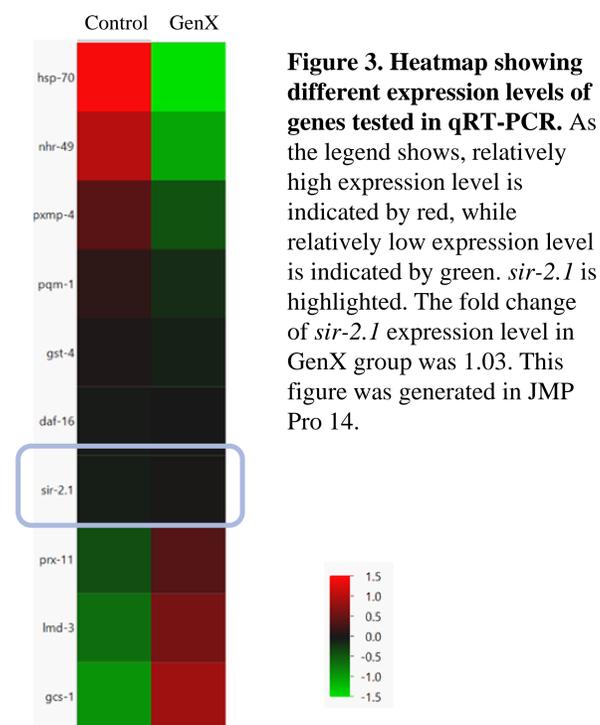


Figure 3. Heatmap showing different expression levels of genes tested in qRT-PCR. As the legend shows, relatively high expression level is indicated by red, while relatively low expression level is indicated by green. *sir-2.1* is highlighted. The fold change of *sir-2.1* expression level in GenX group was 1.03. This figure was generated in JMP Pro 14.

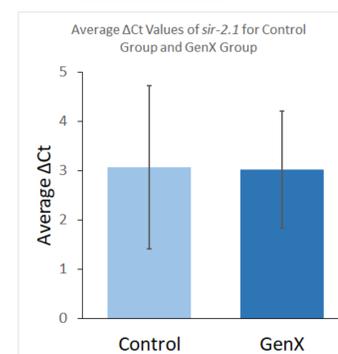


Figure 4. Bar graph comparing average ΔC_t values of the control group and the experimental group. Error bars indicate standard deviation. There is no statistically significant differences between the two groups ($p=0.972$).

Discussion & Conclusion

- As upregulation of the *sir-2.1* gene is shown to slow down the process of aging in mice and yeast, downregulation of the gene expression due to GenX exposure may affect the process of aging in *C. elegans*.
- The homolog in mammals for this gene is known as *SIRT1*.
- Since the exposure concentration, 280 ng/L, was a theoretical concentration in drinking water calculated from the EPA draft reference dose for human exposure to GenX, *sir-2.1* expression changes in the model organism *C. elegans* will indicate potential harmful effects from GenX exposures in humans via the mammalian homolog of *sir-2.1*, *SIRT1*.
- Gene expression of *sir-2.1* was not significantly altered in GenX exposed *C. elegans*.
- We further infer that a short-term exposure to GenX at low concentrations will not affect *SIRT1* expression in humans.

Study Limitations & Future Directions

Limitations:

- *C. elegans* were pooled for analysis; individual variation was not accounted for.
- *C. elegans* were treated with only one dosage of GenX.
- The exposure was via *E. coli* and indirect.
- *C. elegans* were only exposed to GenX for 48 hours, while the EPA draft reference dose is a draft standard for chronic exposure.

Future Directions:

- Conduct similar experiments with
 - Multiple GenX concentrations to develop a dose-response relationship.
 - Direct GenX exposure via liquid medium for *C. elegans*.
 - Long-term exposure throughout the adulthood of *C. elegans* to check if there is any effects of chronic exposure to low-concentration GenX.

References

