Yellow Food Dye Damages Cardiac Cells

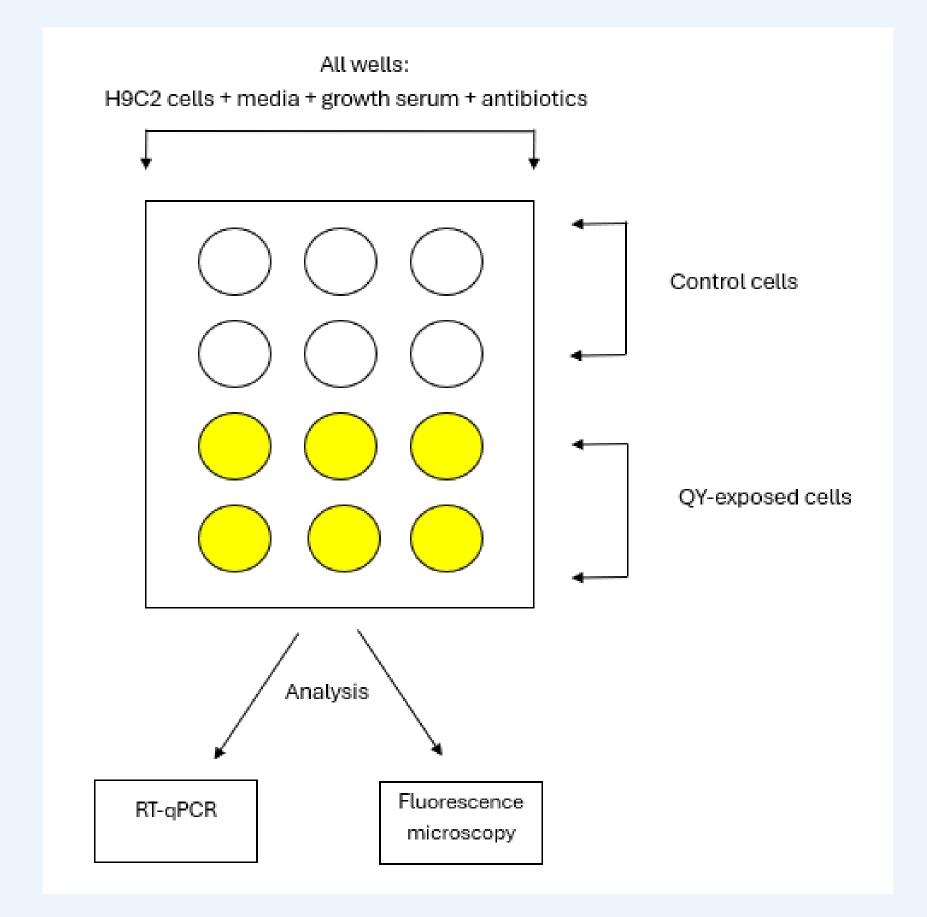
Morgan Walz and Colton Martens, PhD



Background

Poor diet is increasingly common in America and is linked to many chronic diseases. A growing concern is the widespread consumption of synthetic dyes, artificial sweeteners, and other additives (Dunford et al., 2023). Several food dyes, including FD&C Red No. 40, Yellow No. 5, and Yellow No. 6, have been associated with cancer risk, ADHD, and other health issues (Kobylewski & Jacobson, 2012). Mitochondrial dysfunction is a common factor in these conditions and plays a role in diseases like Parkinson's, Alzheimer's, and cardiomyopathy (Popov, 2020). Quinoline yellow (QY), a less-studied synthetic dye, has been shown to cause DNA damage and alter signaling pathways indirectly related to mitochondrial function, which suggests that QY could contribute to mitochondrial dysfunction and disease (Chequer et al., 2015). This project aimed to test this potential link between QY and mitochondrial dysfunction in cultured mammalian cells. Through a combination of qPCR and fluorescent cell staining, our results suggest that QY may alter mitochondrial dynamics and negatively influence cellular health.

General Approach



CR Methods

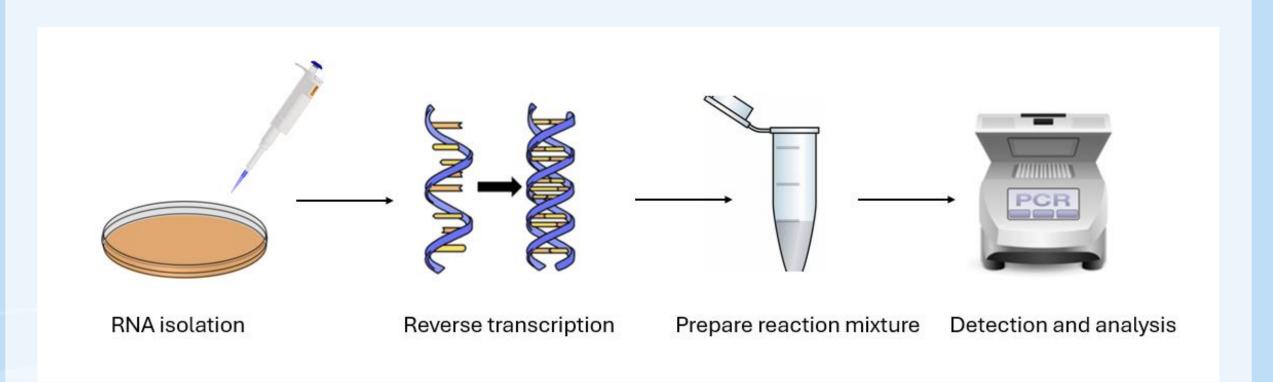


Figure 1. Quantitative PCR analysis of gene expression. Total RNA was extracted using TRIzol reagent according to the manufacturer's protocol. The isolated RNA was reverse transcribed into complementary DNA, and qPCR was performed using SYBR Green dye. Gene expression levels were quantified based on fluorescence intensity.

qPCR Results

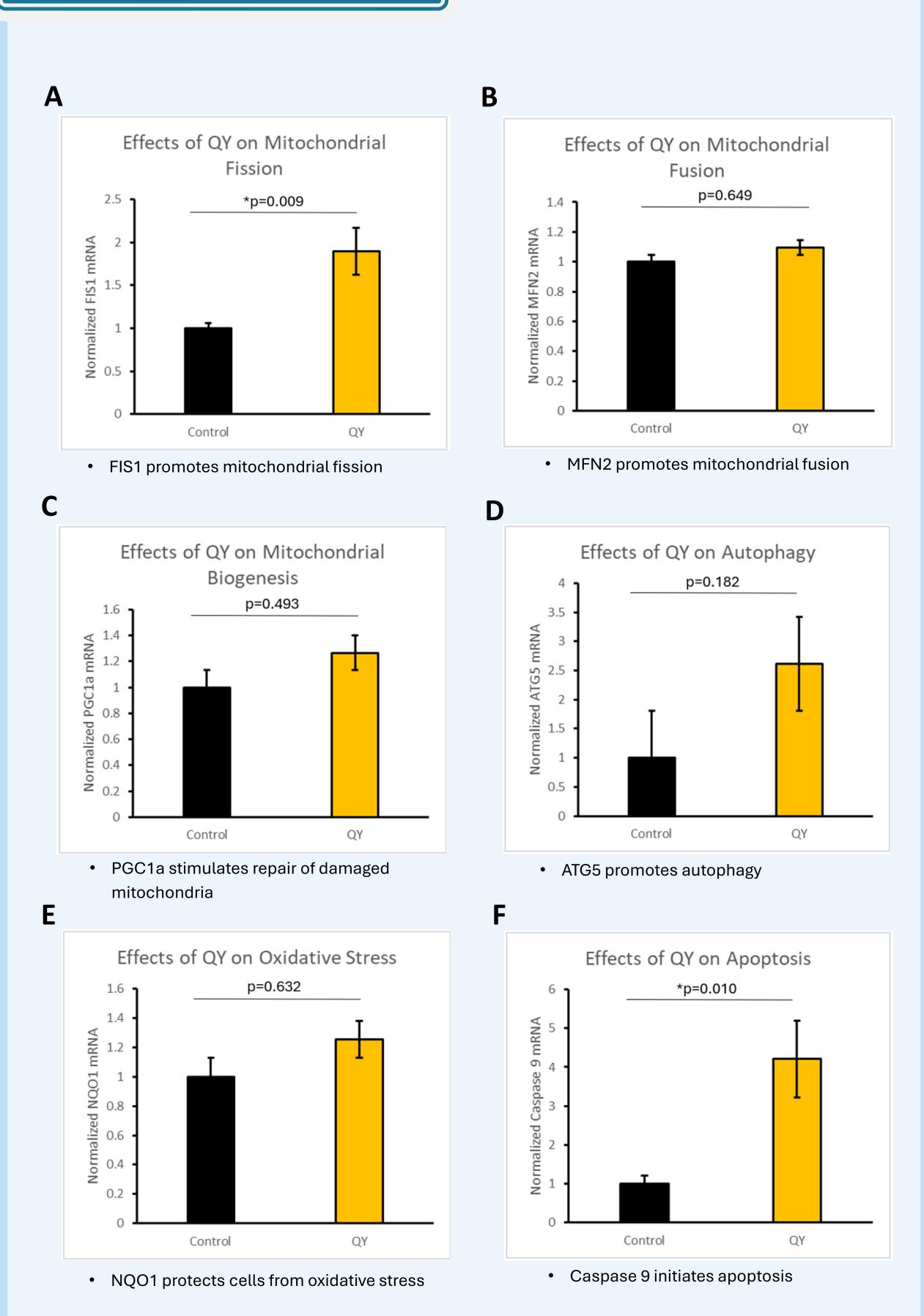
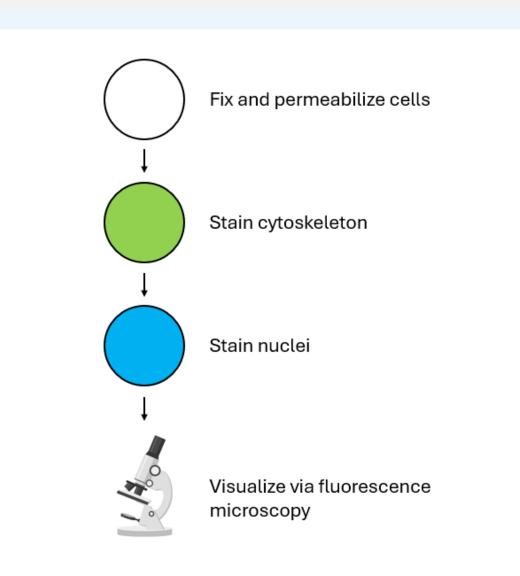


Figure 2. qPCR results for control cells and 24-hour QY-exposed cells. A) Results for normalized FIS1 mRNA. B) Results for normalized MFN2 mRNA. C) Results for normalized PGC1a mRNA. D) Results for normalized ATG5 mRNA. E) Results for normalized NQO1 mRNA. F) Results for normalized Caspase 9 mRNA. n=6. Error bars indicate SEM.

Microscopy Methods



3. Fluorescent staining protocol for H9C2 cells. Cells were fixed with 3.7% paraformaldehyde and permeabilized using Triton X-100. They were then treated with phalloidin green actin stain and DAPI stain. Imaging was performed with a ZOE Fluorescent Cell Imager.

Microscopy Results

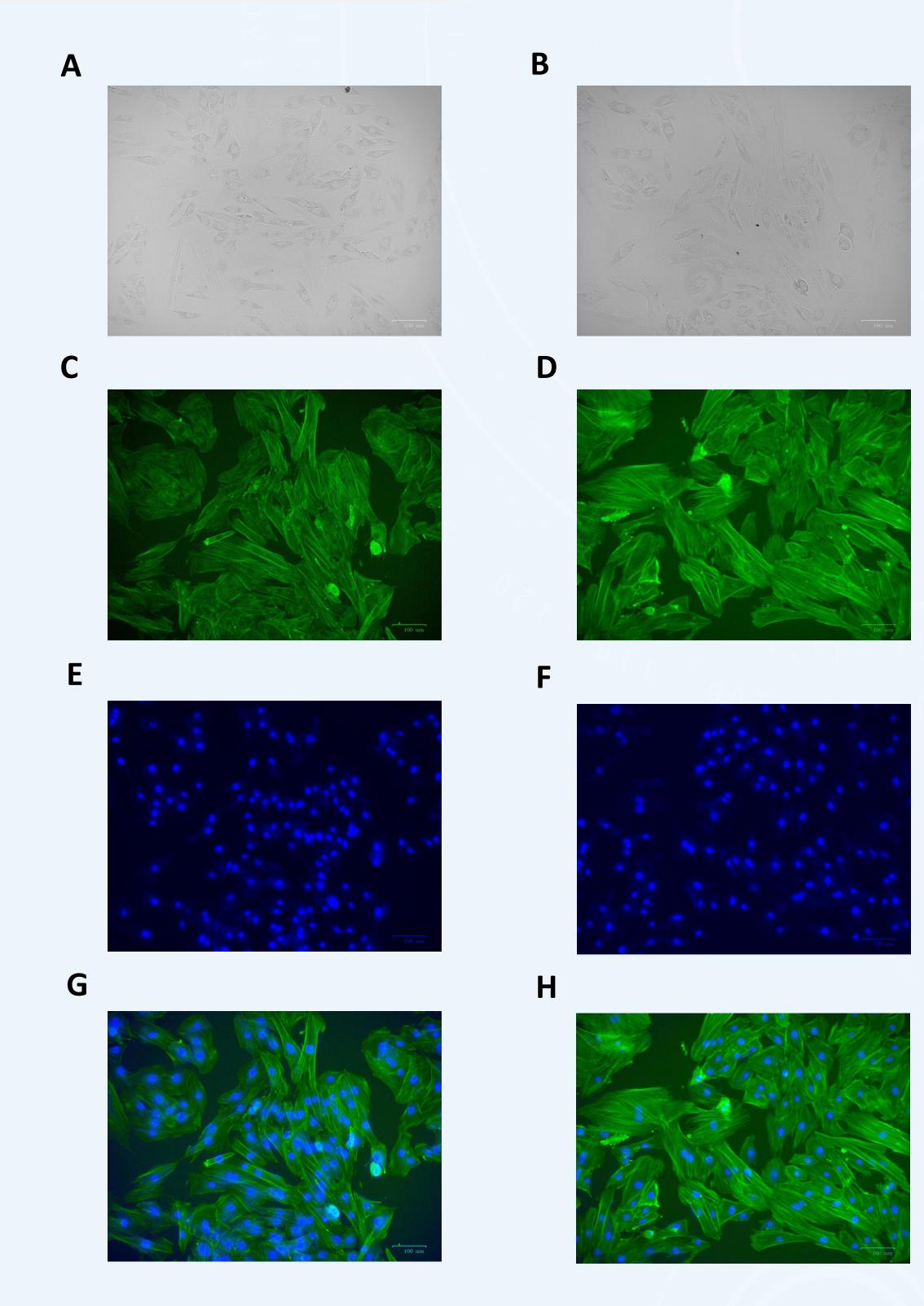


Figure 4. Fluorescent images of control and 24-hour QY-exposed cells. A) Microscope image of QY-exposed cells. B) Microscope image of control cells. C) QY-exposed cells with actin stain. D) Control cells with actin stain. E) QY-exposed cells with nuclear stain. F) Control cells with nuclear stain. G) QY-exposed cells with both actin and nuclear stain. H) Control cells with both actin stain and nuclear stain.

Discussion/Future Directions

Discussion

- QY is harmful to cardiac cells
- QY induces apoptosis
- QY causes mitochondrial fission

Future Directions

- Confirm qPCR results via Western blot
- Perform fluorescence microscopy with mitochondrial and apoptotic stains
- Evaluate the effects on additional metabolic and apoptotic genes
- Perform experiment on other cell types
- Perform experiment on rodents
- Evaluate the effects of additional synthetic dyes

References

Chequer, F. M. D., Venâncio, V. de P., de Souza Prado, M. R., Campos da Silva e Cunha Junior, L. R., Lizier, T. M., Zanoni, M. V. B., Rodríguez Burbano, R., Bianchi, M. L. P., & Antunes, L. M. G. (2015). The cosmetic dye quinoline yellow causes DNA damage in vitro. Mutation Research/Genetic Toxicology and Environmental Mutagenesis, 777, 54–61. https://doi.org/10.1016/j.mrgentox.2014.11.003 Dunford, E. K., Miles, D. R., & Popkin, B. (2023). Food Additives in Ultra-Processed Packaged Foods: An Examination of US Household Grocery Store Purchases. Journal of the Academy of Nutrition and

Kobylewski, S., & Jacobson, M. F. (2012). Toxicology of food dyes. International Journal of Occupational and Environmental Health, 18(3), 220–246. https://doi.org/10.1179/1077352512Z.000000000034

Popov, L.-D. (2020). Mitochondrial biogenesis: An update. Journal of Cellular and Molecular Medicine, 24(9), 4892–4899. https://doi.org/10.1111/jcmm.15194