

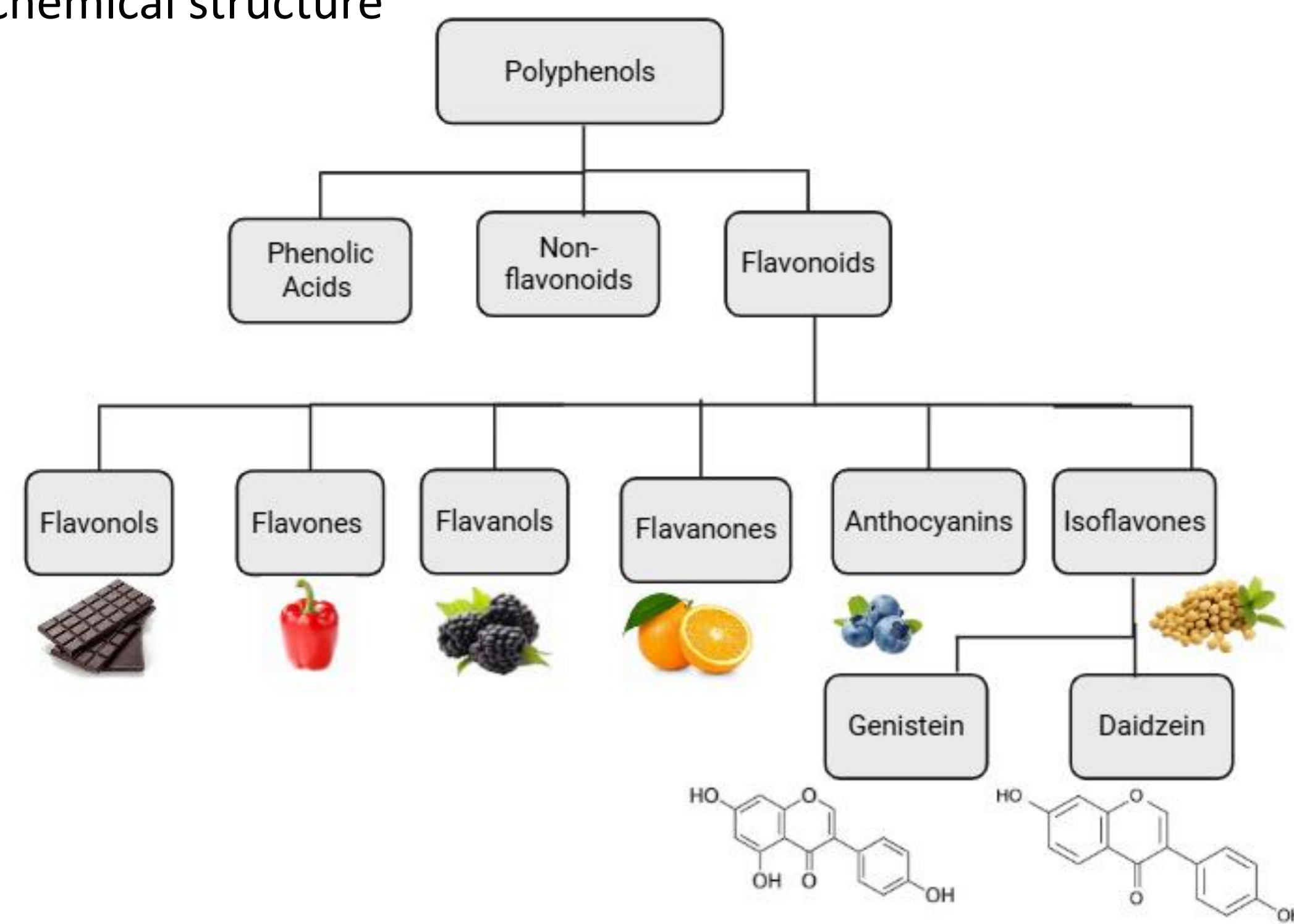
# Dietary Isoflavones Alter Stress Resistance in *C. elegans*

Coco Malnar and Dr. Strand

## Abstract

Oxidative stress occurs when reactive oxygen species (ROS) accumulate and damage cells, contributing to diseases such as obesity and type 2 diabetes. In this study, we investigated how exposure to two polyphenols, genistein and daidzein, influenced stress resistance in *C. elegans*. Oxidative stress was assessed using hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) which generates ROS in cells. When ROS concentrations in the cell exceed its antioxidant capacity, it can be very damaging to the cell and lead to oxidative stress. A heat stress assay was used to assess survival under elevated temperature conditions. We studied the differences in survival between treatments of genistein, daidzein, genistein and daidzein combined, and the control of dimethyl sulfoxide (DMSO). A choice assay was conducted to analyze if the nematodes showed a preference in polyphenol. These experiments aimed to understand how exposure to polyphenols impacted *C. elegans* under oxidative and heat stress. The results of the oxidative stress assay showed trends that when *C. elegans* were exposed to daidzein, there were some protective benefits, and the genistein treated nematodes exhibited the greatest protective effects. The results of the heat stress assay showed trends that combined treatment of genistein and daidzein provided the greatest protection, whereas with genistein alone resulted in the least protection. The results between the treatment groups were not found to be statistically significant for either assay, but differences in survival were shown. The implication of this study is that genistein and daidzein may show protective effects when under heat and oxidative stress in *C. elegans*.

- Polyphenols are small molecules produced by plants during metabolism that function as defense and protection
- Polyphenols are the most abundant antioxidants in the human diet
- Polyphenols are classified into classes based on differences in chemical structure



**Figure 1.** Classes of polyphenols and common food sources.

## Acknowledgements

Thank you to my advisor, Dr. Strand, for your constant support and guidance, making this project possible. I am also very grateful to have received Copeland Funding.

## Overall Question

Does polyphenol treatments of genistein, daidzein, and genistein with daidzein provide protective effects for *C. elegans* when under oxidative and heat stress?

## Methods

Add 200µM of polyphenols with *E. coli* OP50 to NGM plates & incubate for 24 hours  
- Genistein + OP50, Daidzein + OP50, Genistein + daidzein + OP50, DMSO + OP50

Expose *C. elegans* to polyphenols for 24 hours

### Oxidative Stress

Expose *C. elegans* to 30% H<sub>2</sub>O<sub>2</sub> for 30 minutes

Remove H<sub>2</sub>O<sub>2</sub> & transfer *C. elegans* to new seeded plates with polyphenols

Analyze *C. elegans* survival across plates for 8 days

### Heat Stress

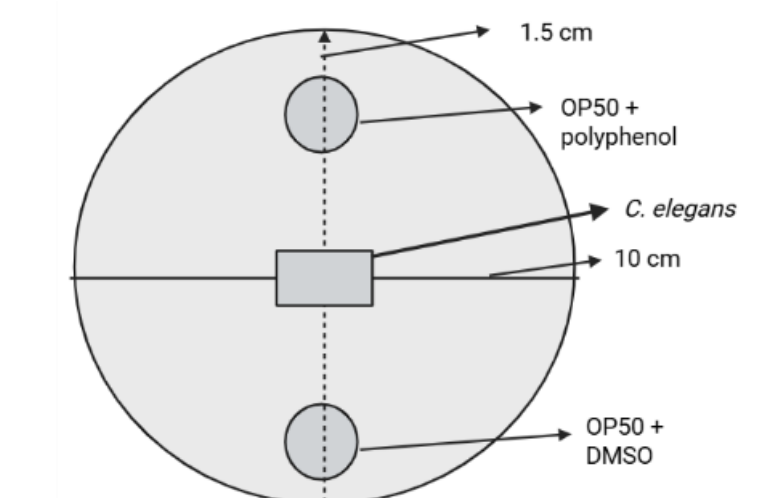
Incubate *C. elegans* at 34°C for 10 hours

Analyzing survival of *C. elegans* across experimental plates every 2 hours for 10 hours

### Choice Assay

Add 200µM polyphenol + OP50 on one side of plate, other side consisted of DMSO + OP50 & incubated for 24 hours

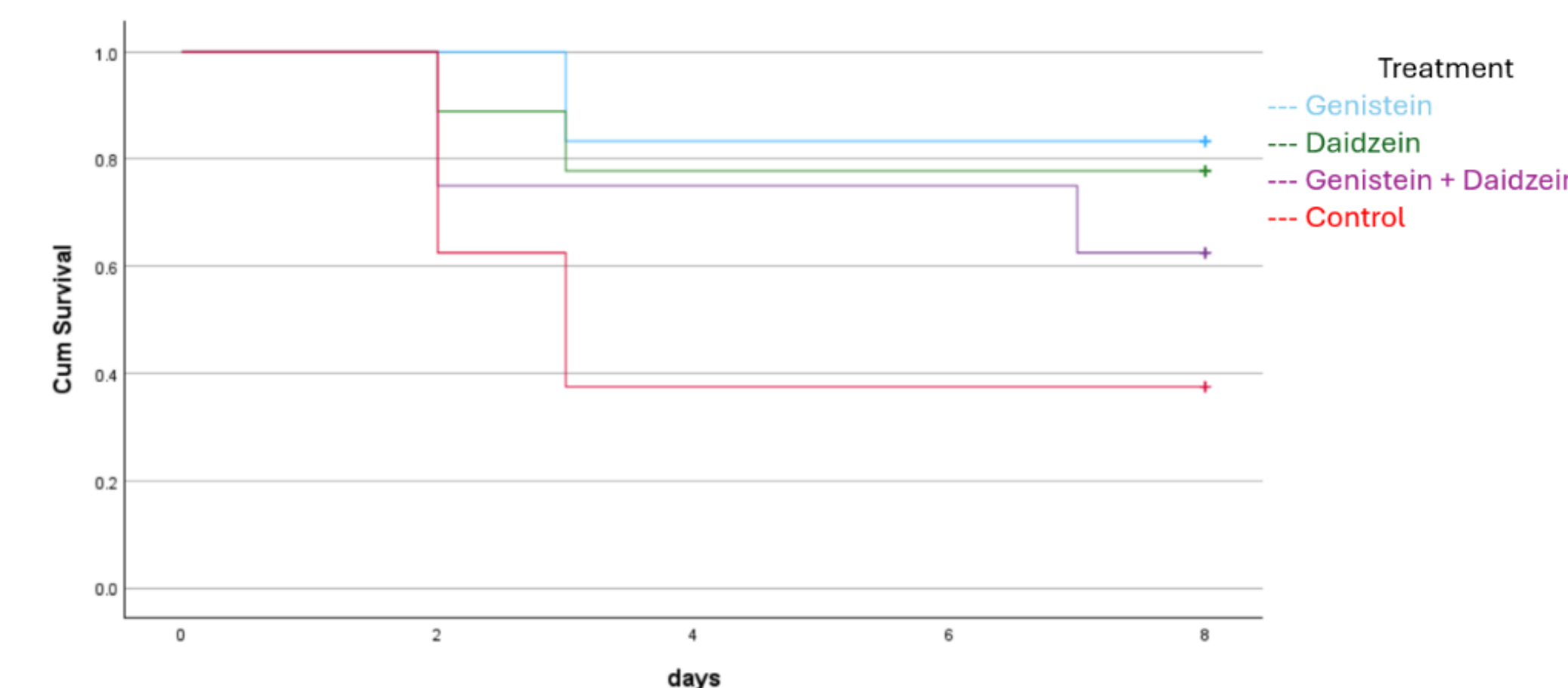
Add chunk of previous *C. elegans* seeded plate to middle of plate for 30 minutes



Analyze migration of *C. elegans* to treatment after 24 hours

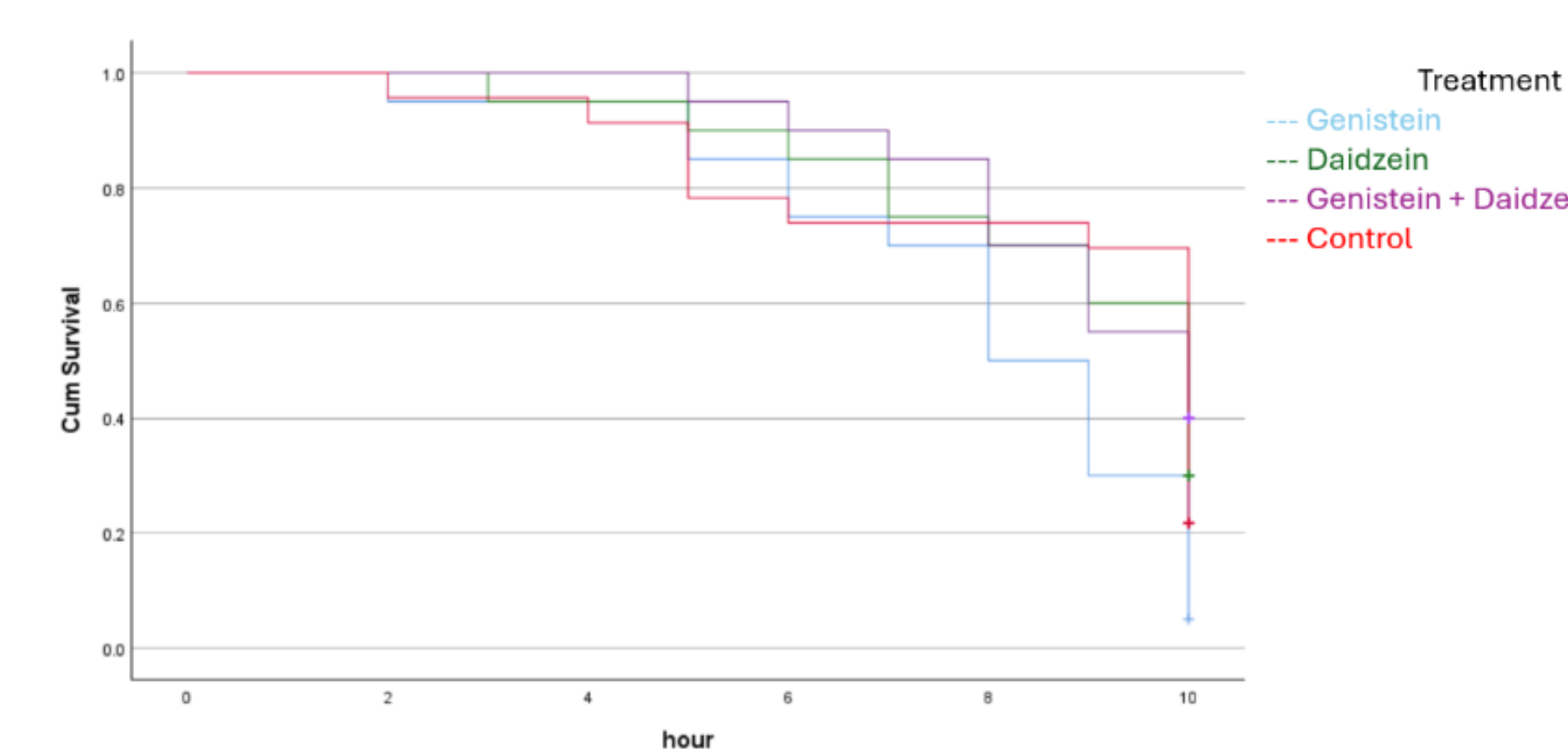
$$CI = \frac{(\# \text{ of } C. \text{ elegans in bacteria} - \# \text{ of } C. \text{ elegans in treatment})}{\text{total } \# \text{ of } C. \text{ elegans in experiment}}$$

## Oxidative Stress



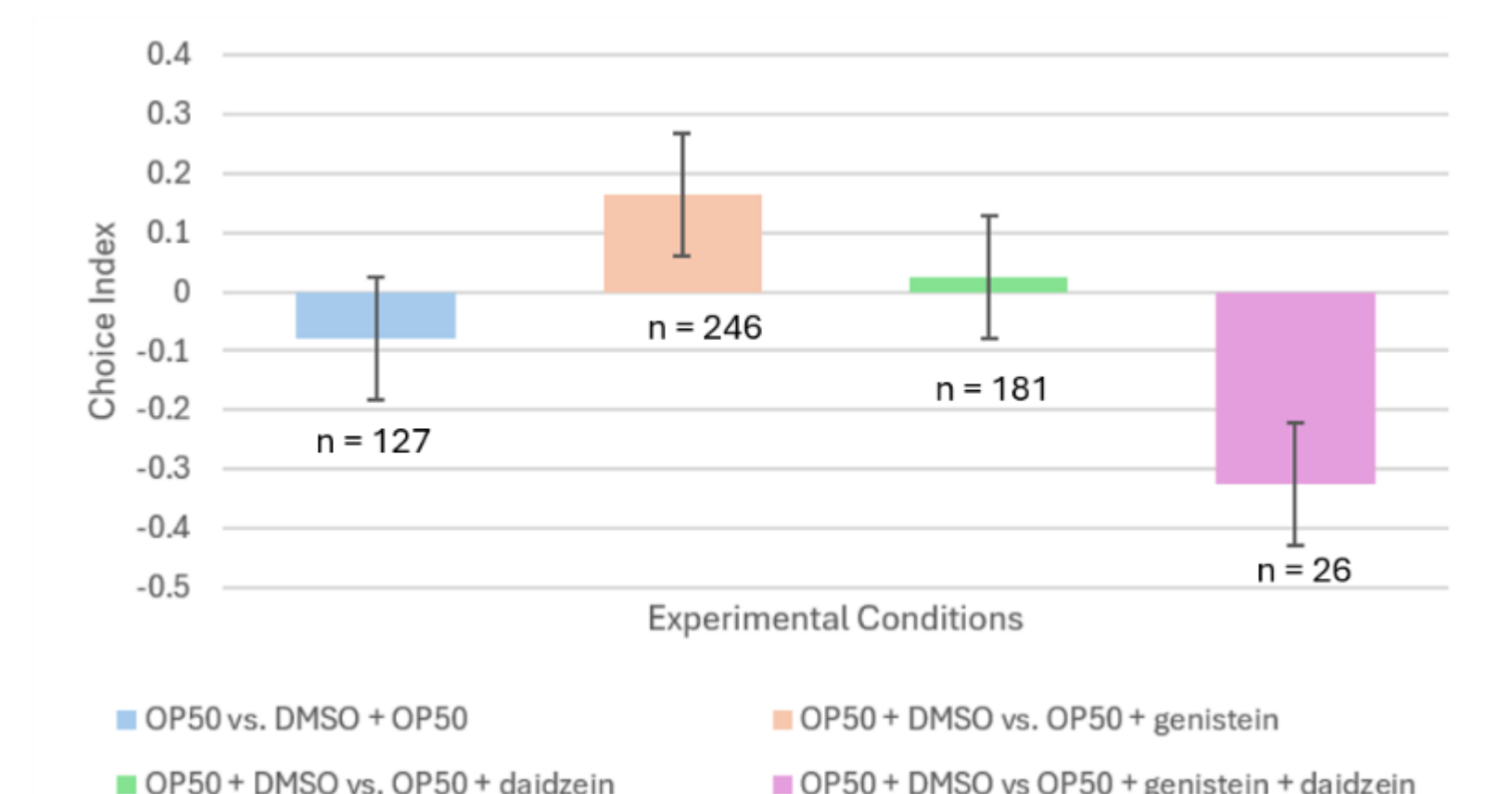
**Figure 2.** Kaplan-Meier survival curves represent the number of worms alive over time for each experimental group post oxidative stress analysis. (p = 0.208).

## Heat Stress



**Figure 3.** Kaplan-Meier survival curves represent the number of worms alive over time for each experimental group during heat stress analysis (p = 0.049).

## Choice Assay



**Figure 4.** Choice index of *C. elegans* towards polyphenols.

Our results suggest polyphenols have some protective effects against heat and oxidative stress.

- In the oxidative stress assay, the trend suggests genistein may be more protective than the other treatment groups based on more nematodes surviving following the assay.
- In the heat stress assay, the trend suggests that genistein may be less protective than the other treatment groups based on fewer nematodes surviving following the assay.
- In the choice assay, the trend suggests that *C. elegans* showed preference towards genistein & avoidance behavior towards genistein with daidzein.
- This study can be used as a framework for future studies as it provides information on the survival of *C. elegans* when exposed to two polyphenols at once, something that has not been previously assessed.

**References:** Rodriguez-Mateos et al., (2024). Dietary (poly)phenols and cardiometabolic health: from antioxidants to modulators of the gut microbiota. Emad Abd-elmoniem Abada et al., (2009). *C. elegans* Behavior of Preference Choice on Bacterial Food.