

# Examining the Impact of Antiproliferative Agents on the Replication of HeLa Cells

Carson Krefft, Drake Teal, Raida Sugule & Suhaylah Marty

Faculty: Dr. Bergerson

Department Of Science, Concordia Saint Paul

## Abstract

Common supplements, including resveratrol, gentisic acid, and flaxseed oil, have proven to demonstrate anti-cancerous properties in relevant doses. The antiproliferative properties of these supplements were tested on HeLa cells growing in glucose media to verify similar claims.

## Hypothesis

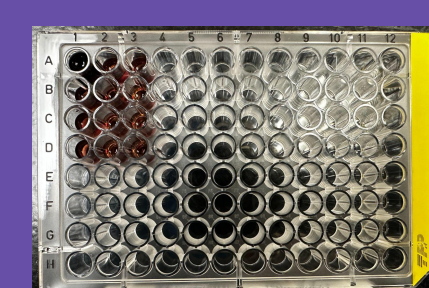
Of the supplement variables—resveratrol, gentisic acid, and flaxseed oil—added to HeLa cells growing in media, it was hypothesized that resveratrol would impede HeLa cell proliferation at a level higher than the other reagents tested.

## Background

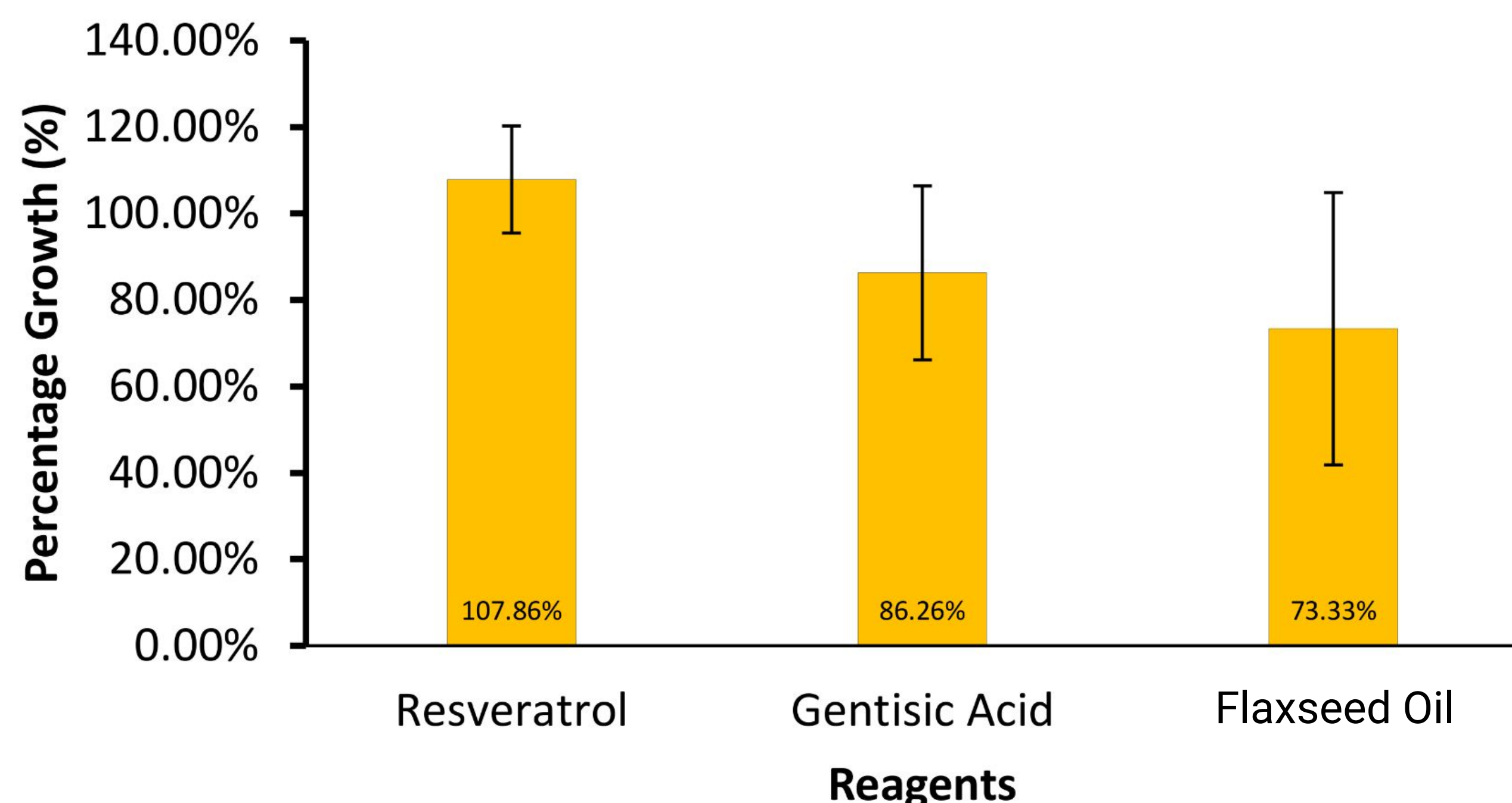
- **HeLa cells:** These cells are the earliest and most widely used human cell line. They were obtained from cervical cancer cells of the renowned Henrietta Lacks.
- **Resveratrol:** A natural phenol produced by plants, resveratrol is most commonly found in grapes, berries and select nuts and legumes. It is known to damage pathogens. Resveratrol revealed astounding anticancer properties when tested on cancerous cell viability (2).
- **Gentisic acid:** As a chemical compound caused by the breakdown of aspirin, gentisic acid is excreted by the kidneys. A recent study, *Gentisic Acid Increases Proliferation of REK Cells* by the Science Dept. of CSP, found a significant shift in proliferation of rat epidermal keratinocytes (REK).
- **Flaxseed oil:** Most commonly used to decrease inflammatory diseases, flaxseed oil demonstrated possible anti-carcinogenic properties and malignant MCF-7 breast cancer cells administered *in vitro* (1).

## Methods

- HeLa cells were grown in T-25 flasks.
- Stock solutions of 0.01g/mL resveratrol, 0.01g/mL gentisic acid, and  $5.38 \times 10^{-4}$  g/mL flax oil were added to the wells in triplicate along with a control.
- 100uL of cells with media was added to the 12 wells.
- Following 24 hour incubation at 37 degrees Celsius, cytotoxicity assay was ran and data was collected using a microplate reader.



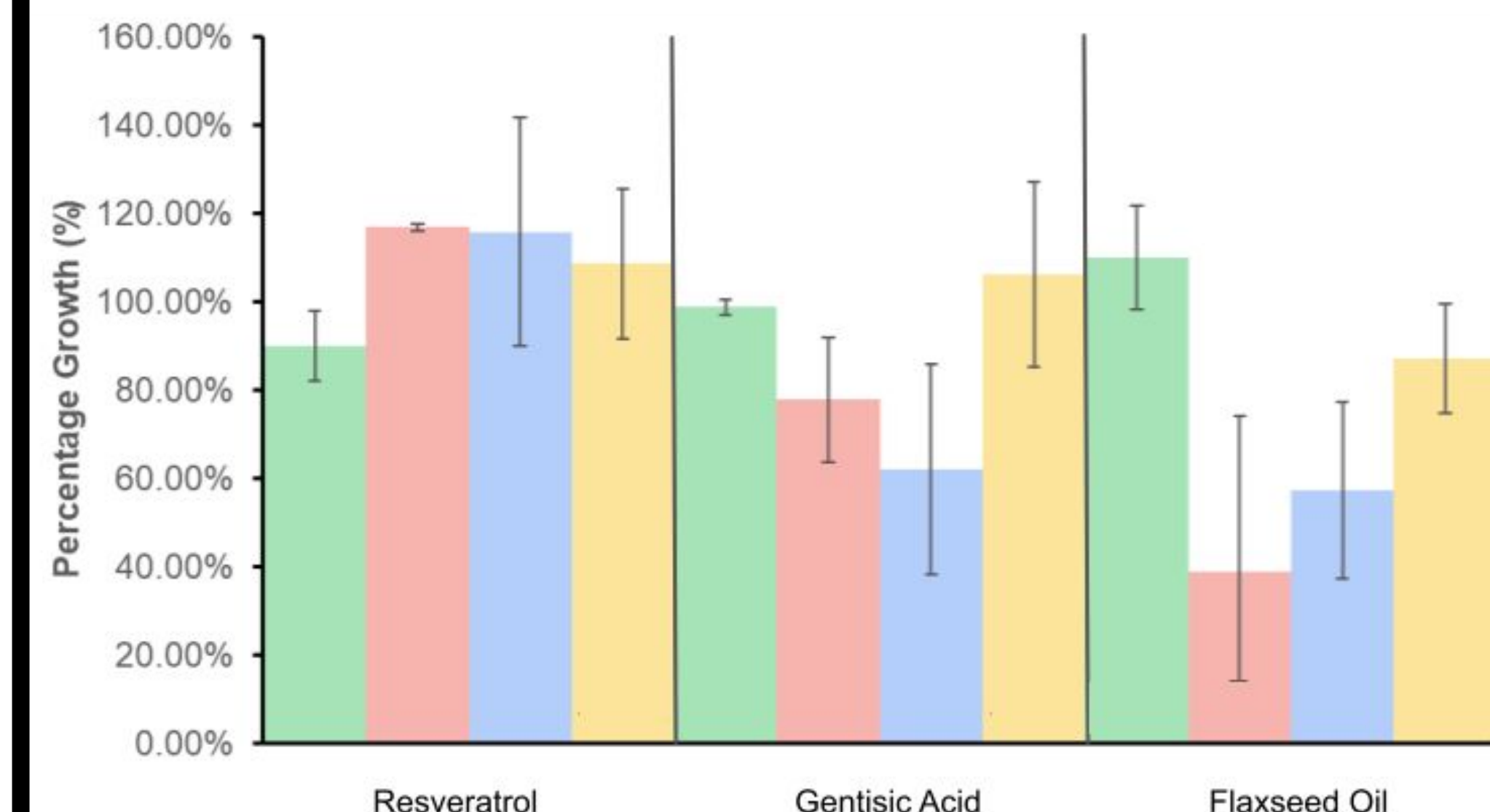
# Resveratrol, Gentisic Acid, And Flaxseed Oil: Antioxidative Agents Against HeLa Cell Proliferation



**Figure 1. Effects of reagents on HeLa cell proliferation.** The bar graph displays the average growth of HeLa cells when treated with resveratrol, gentisic acid, or flaxseed oil compared to a control group. The results are based on four independent trials, and raw data was found using a microplate reader to analyze the cells and media at 490 micrometers wavelength. These growth values were then compared to the growth of a control group. Averaging the four trials led to the full intermediate of growth in percentages. Resveratrol provided HeLa cell growth of 107.86% (error bars: 116.9% and 90.12%). Gentisic acid HeLa cell growth was 86.26% (error bars: 106.32% and 62.12%). Flaxseed oil also provided a proliferation decrease at 73.33% (error bars: 110.0% and 38.96%). The data presented highlights the potential use of these reagents in modulating cell proliferation, which could have significant implications in developing novel therapies for diseases such as cancer.

## Results

Results showed that treatment with resveratrol increased cell proliferation with an average growth of 107.86%. Only one trial performed with resveratrol saw a decrease in percentage growth in comparison to the control. On the other hand, HeLa cells treated with gentisic acid as well as flaxseed oil saw a significant decrease in cell viability. HeLa cells treated with flaxseed oil saw a consistent decrease in percentage growth throughout the trials with an average growth of 73.33%, whereas cells treated with gentisic acid saw average growth of 86.26% but had a slight increase in percentage growth in trials one and four. Further studies are needed to explore the mechanisms underlying these effects and to validate their potential clinical applications.



**Figure 2. Analysis of individual trial results.** Figure 2 presents an analysis of the individual trial results of HeLa cell proliferation using a cell titer cytotoxicity assay. The experiment was carried out by adding stock concentrations of resveratrol, gentisic acid, and flaxseed oil to HeLa cells with media in a well plate in concentrations of 2.86%, 3.00%, and 1.80%, respectively. A control group was also included in the experiment, in which no additional reagent was added to the cells and media, that was used as a reference to calculate cell proliferation. Each trial was performed in triplicate, and four trials were conducted. The resulting bar graph displays the average growth of HeLa cells in each trial, with green bars representing the average growth of trial one, red bars representing the average growth of trial two, blue bars representing the average growth of trial three, and orange bars representing the average growth of trial four. The error bars were calculated using standard deviation; their values are indicated on the graph.

## Conclusion

Contrary to the proposed hypothesis, results suggest that resveratrol may have minimal effects in decreasing the proliferation of HeLa cells. Instead, resveratrol may promote the growth of these cells significantly, as observed by the increase in cell proliferation compared to the control group. In contrast, gentisic acid demonstrated a significant decrease in the proliferation of HeLa cells, supporting its potential anti-cancer properties. Notably, flaxseed oil showed the most promising results in impeding HeLa cell growth, indicating its potential as an effective anticancer agent. However, it is essential to note that the mechanisms underlying the observed effects of these reagents on HeLa cell proliferation require further investigation. Additional studies are needed to elucidate these effects' underlying pathways and molecular targets.

## References

- (1) Buckner, A. L., Buckner, C. A., Montaut, S., & Lafrenie, R. M. (2019). Treatment with flaxseed oil induces apoptosis in cultured malignant cells. *Heliyon*, 5(8), e02251. <https://doi.org/10.1016/j.heliyon.2019.e02251>
- (2) Carter, L. G., D'Orazio, J. A., & Pearson, K. J. (2014). Resveratrol and cancer: focus on in vivo evidence. *Endocrine-related cancer*, 21(3), R209–R225. <https://doi.org/10.1530/ERC-13-0171>