

Isolation of Health-benefiting Bacteria from Probiotic Foods

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ABSTRACT

Probiotic Foods are known to have many health benefits such as bolstering intestinal health, reducing risk to many diseases, and reducing antibiotic resistance, to name a few. Because these foods have so many benefits, I hypothesized that by isolating bacteria from them, these health benefits could be extended to all kinds of different foods. The bacteria of interest were isolated from Great Value Greek Plain Nonfat Yogurt by using MRS medium to encourage the growth of lactic acid bacteria. After several strains had been isolated, they were used to ferment orange juice and carrot juice for about three days. By using HPLC (High-Performance Liquid Chromatography), several differences between the original juices and the fermented juices were found. E. Coli Bacteria were tested against antibiotics as well as salt concentrations after being exposed to one of two treatments: fermented juice and non-fermented juice. Through this experiment, I have been able to observe positive results from three bacterial strains. I was able to observe a decrease in antibiotic resistance to ampicillin, hygromycin, carbenicillin, and kanamycin when I exposed E. Coli bacteria to juices fermented by my isolated bacteria.

Introduction

Probiotic foods are known for their beneficial impacts on human health. From that fact arose my research question: Can bacteria isolated from probiotic foods be used to produce positive health effects in other foods as well? My hypothesis in this experiment was that bacteria isolated from yogurt, the probiotic food of choice, could be used to ferment other food to produce beneficial health effects. The “health effect” I wanted to test in this experiment was antibiotic resistance. According to the CDC in 2019, more than 2.8 million antibiotic-resistant infections occur in the United States alone each year, and more than 35,000 people die as a result. It is because of this, that finding an easy and efficient way to resolve the issue of antibiotic resistance has very big implications in our world today. My decision to ferment other juices with the bacteria I isolated stemmed from the fact that transforming the food we eat into our medicine makes for the most convenient method of providing beneficial health impacts to people all around the world. The goal of this study is to provide a convenient and easy way for people to reduce their risk to different kinds of health issues.

Materials and Methods

Materials:

- Probiotic food (Great Value Greek Plain Nonfat Yogurt)
- Food of interest to be fermented (Orange juice and Carrot juice)
- MRS Medium
- HPLC

Methods:

- Isolate lactic acid bacteria from probiotic food by using MRS agar to facilitate the growth of lactic acid bacteria.
- Transfer colonies to a liquid MRS and incubate at 37°C for a day or two.
- Centrifuge to sink cells to the bottom and replace the liquid MRS medium with the food of interest (Orange juice and Carrot juice). Store at 37°C for a couple of days to allow the juice to ferment (In this experiment, the flasks were stored for about 4 days.)
 - The orange juice and carrot juice were first autoclaved for 15 minutes at 121°C before being fermented by the isolates. Samples of the autoclaved juices were run through the HPLC and results were compared to those after fermentation.
- Run samples of the fermented juices through the HPLC and compare the results to those of the non-fermented juices.
- Pour 5mL of fermented and non-fermented juices into respective LB agar plates to expose E. Coli bacteria to them and observe the difference in their impacts on antibiotic resistance.

Results

First off, I isolated two strains of bacteria from the yogurt that were able to produce beneficial effects in my experiment. The HPLC results of the original autoclaved juices in comparison to the results of the fermented autoclaved juices are shown below (All graphs represent HPLC results of wavelengths of 210, 230, and 275 nm, respectively):

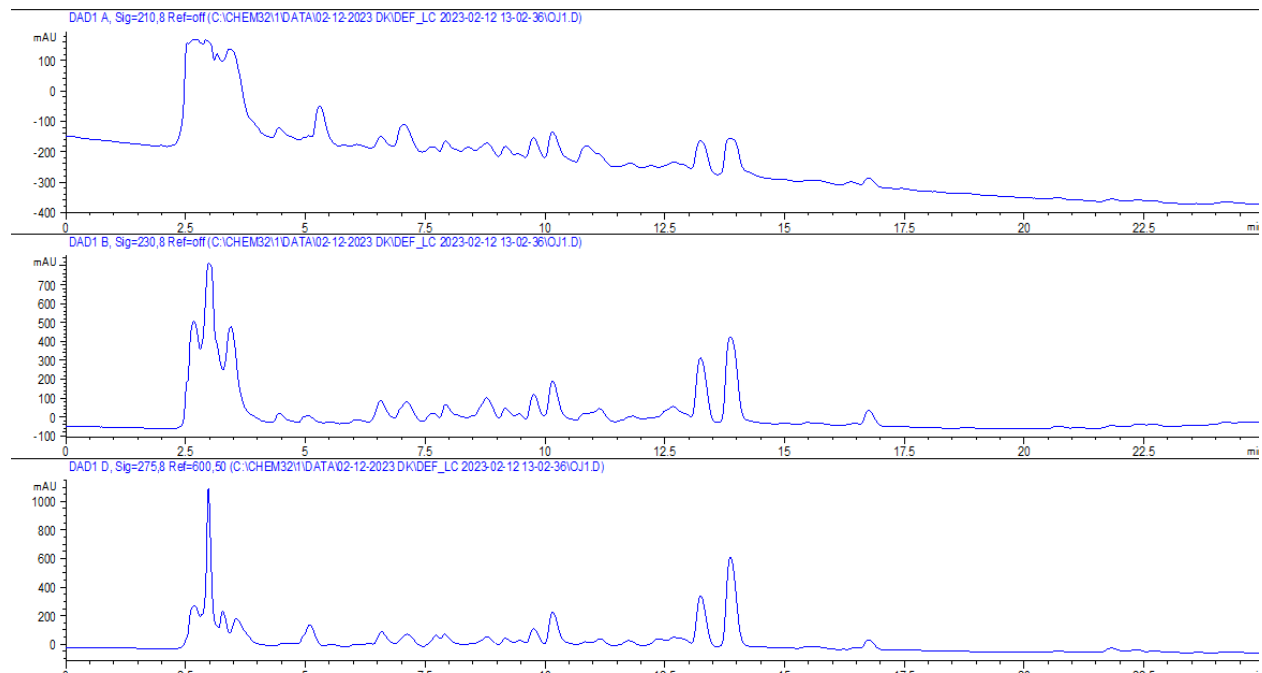


Figure 1 - Autoclaved orange juice that was not fermented.

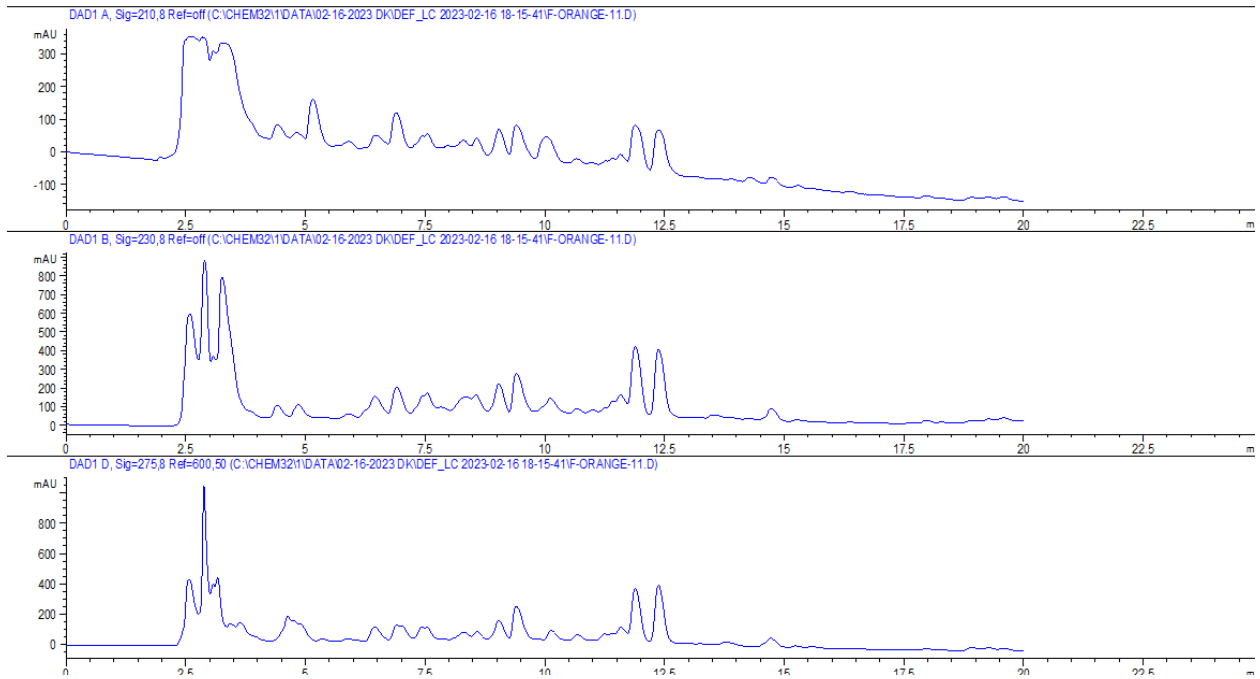


Figure 2 - Autoclaved orange juice that was fermented by isolate #1

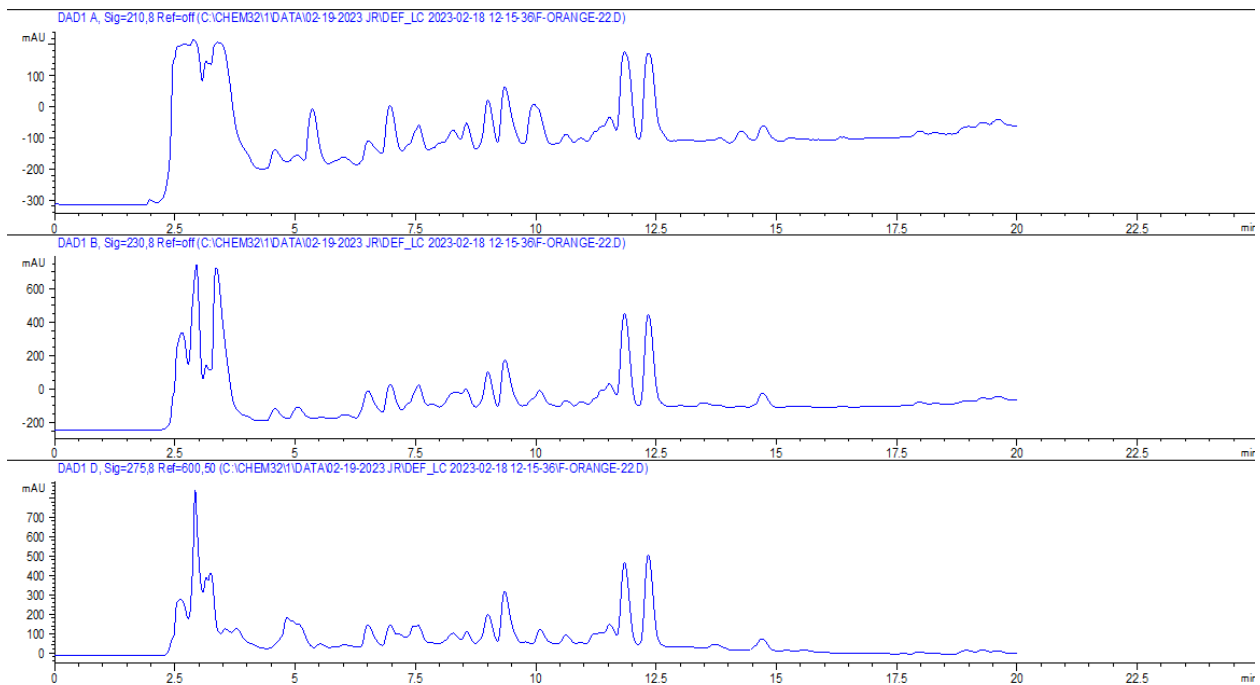


Figure 3 - Autoclaved orange juice that was fermented by isolate #2

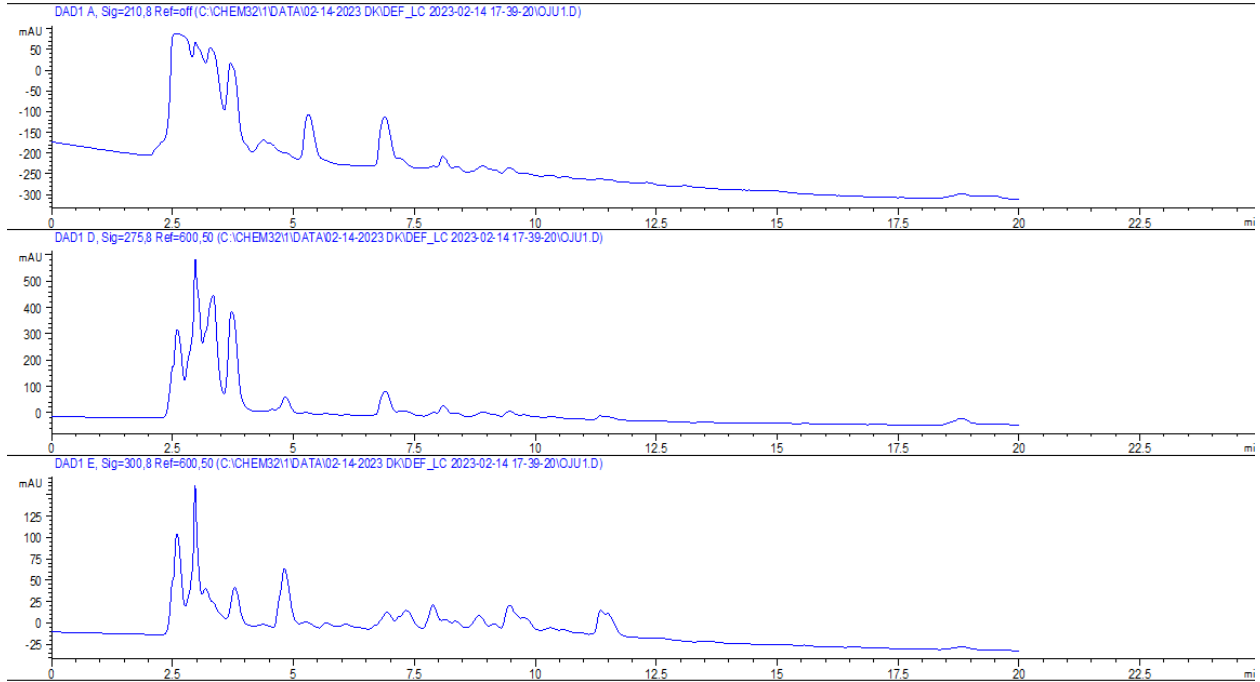


Figure 4 - Autoclaved carrot juice that was not fermented.

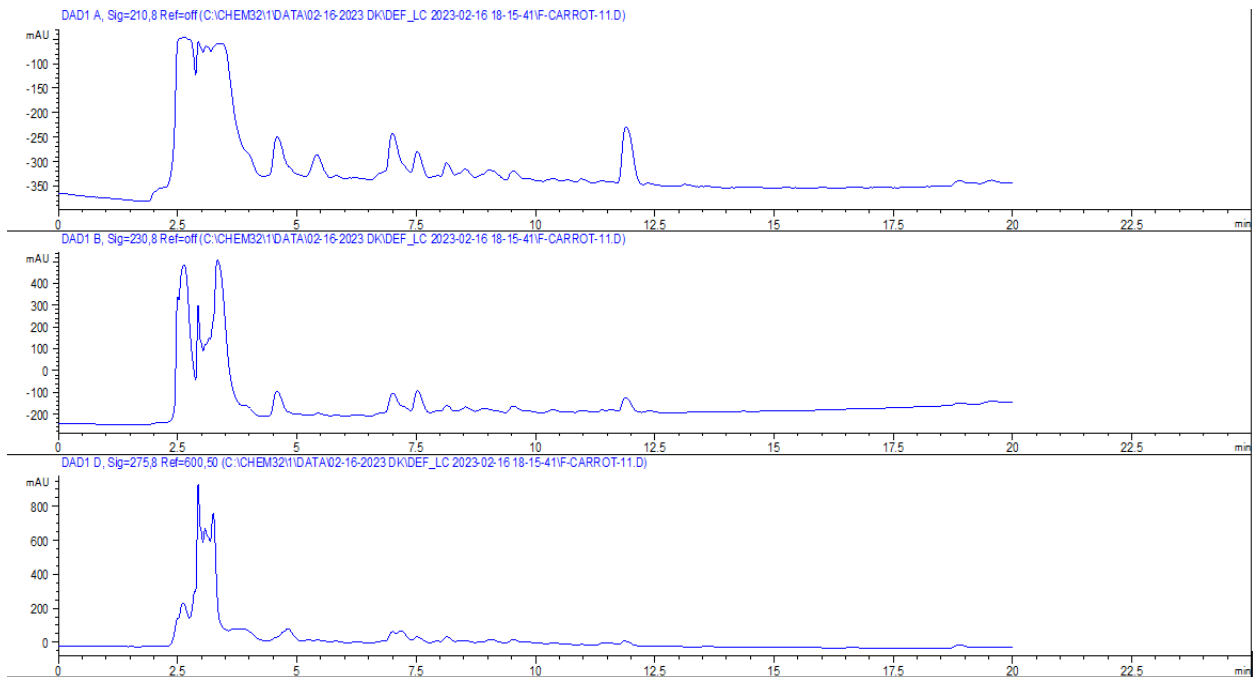


Figure 5 - Autoclaved carrot juice that was fermented by isolate #1

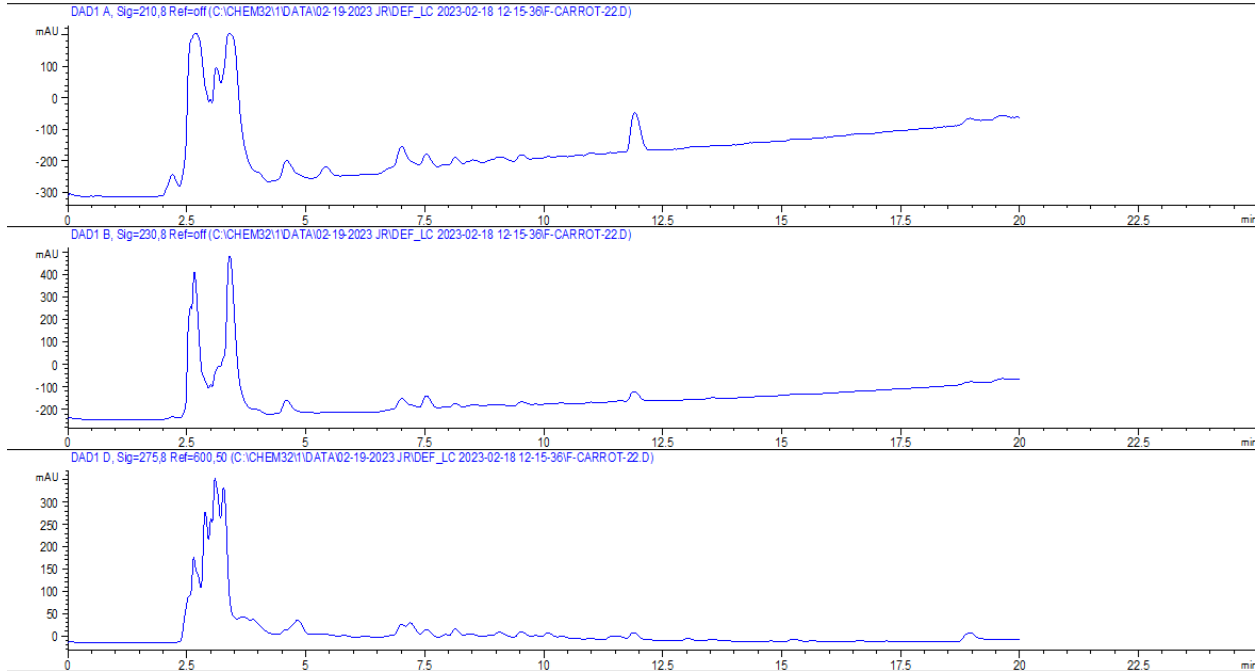
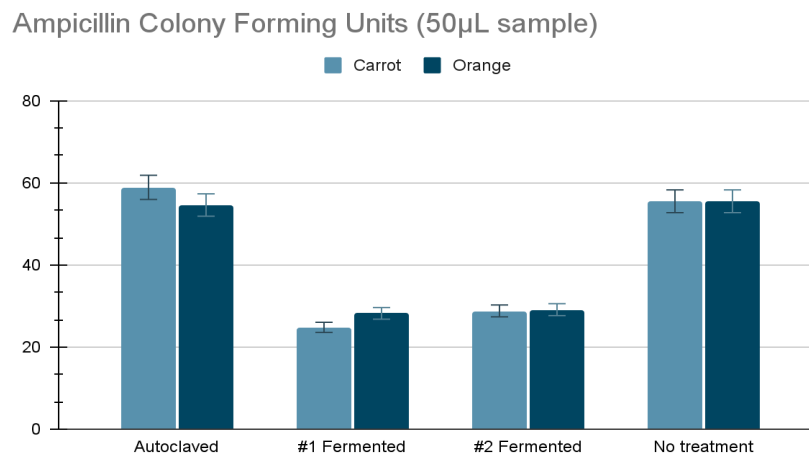


Figure 6 - Autoclaved carrot juice that was fermented by isolate #2

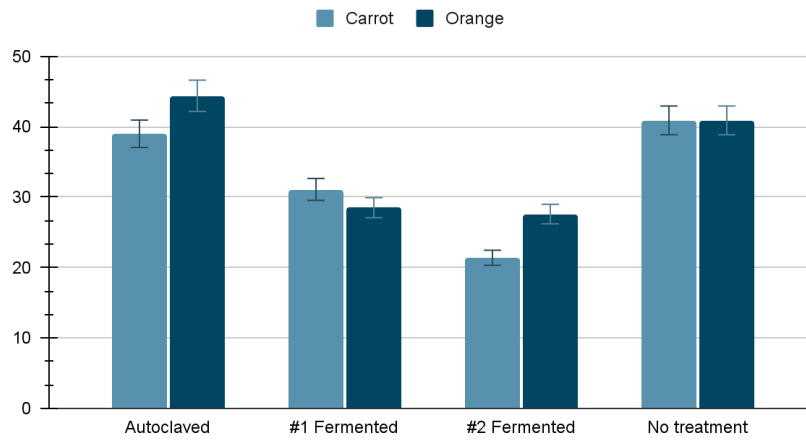
Both isolate #1 and #2 made noticeable changes to the makeup of these juices, and through my experiment I’ve been able to see that these changes can lead to positive health results.

Antibiotic Resistance

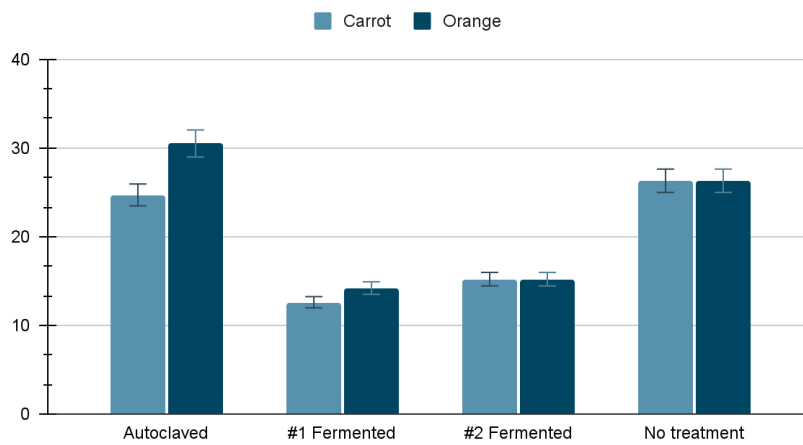
To test the effects of my isolates on antibiotic resistance, I first exposed E. Coli bacteria to four antibiotics: ampicillin, hygromycin, carbenicillin, and kanamycin. Once some colonies grew, I repeated this process four more times to select for the most resistant bacteria. Afterward, I picked up colonies from that plate and randomly exposed them to one of two treatments: the autoclaved juice that wasn’t fermented, or the autoclaved juice that was. Finally, I performed serial dilutions for both of the treatments as well as the control group (resistant E. Coli that was not exposed to a treatment) and plated them on LB agar plates with the antibiotic of choice in them. I repeated this process 50 times for each antibiotic and treatment. The results are as follows:



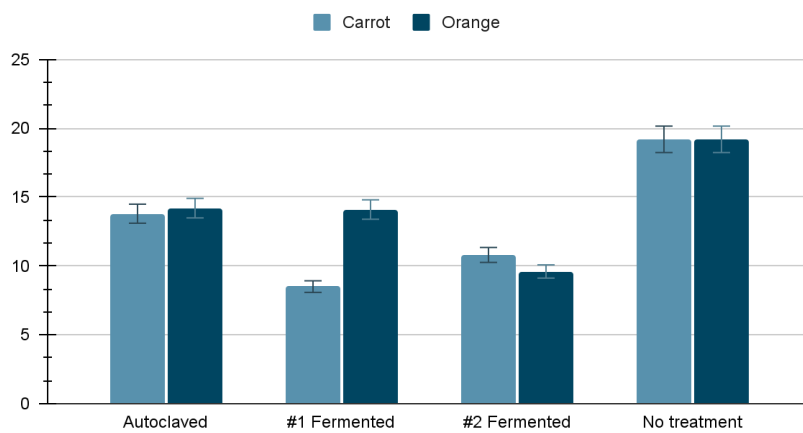
Hygromycin Colony Forming Units



Carbenicillin Colony Forming Units



Kanamycin Colony Forming Units



Discussion and Conclusions

From my experiment, I was able to observe a decrease in resistance to four antibiotics across the board. The conclusion I can draw from these results is that my hypothesis was correct: by isolating certain bacteria from health-benefiting probiotic food, we can potentially use those bacteria to provide health-benefits to all kinds of other foods. I am extremely happy with the results I have gotten, but my experiment does not end here.

In terms of how my research can be applied in the real world, my research can be used to do three things:

1) Reduce resistance to a variety of medications

While my experiment solely focused on the issue of antibiotic resistance, the principles, and methods that I used can be applied to other medications as well.

2) Reduce the need for a variety of medications

The goal of my research was to implement health benefits into the food people eat on a daily basis. If people are able to reap health benefits from every meal of every day, we can reduce the need for a variety of medications in the first place. And in turn, we can reduce resistance to these medications as well.

3) Providing health benefits to those with allergies to certain foods or medications

Using lactose intolerance as an example, many people with lactose intolerance have trouble consuming dairy products which are known to have various health benefits. My research can potentially be used to project the health benefits of these dairy products to foods that people with lactose intolerance are more comfortable consuming. In the future, I would like to sequence the DNA of these two isolates so that I can identify them. In addition to this, I would like to test the effects of these isolates against other health issues, such as halotolerance which has big implications in the world today regarding heart disease.

References

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