Spice, Spice, Baby: A Study on the Effects of Spices on Bacterial Growth

OBJECTIVES

The purpose of the experiment is to identify spices that promote the growth of *Escherichia coli*.

HYPOTHESIS:

Garlic powder, black pepper, and cinnamon will promote the growth of *E. coli*, while cumin and ginger will inhibit *E. coli*'s growth.

BACKGROUND INFORMATION

Spices have been used to enhance flavor for centuries, and some have become central to the cuisine of many distinct cultures. The usage of spices for purposes aside from food preparation is commonplace; however, the effect of specific spices on bacterial propagation is not often considered in cuisine. Spices may be used to effectively kill harmful, or even deadly, microbes. Studies have concluded that these spices would be of value, even in modern medicine, due the the ability of bacteria to develop resistance to conventional antibiotics. (Agabalogin, 2018; Liu, 2017; Sonal, 2018) Consequently, effective methods of utilizing the antimicrobial activities in spices would be advantageous for advancing medical treatment of bacterial infections. Due to the natural presence of *E. coli* in the human gut that lives in a mutual relationship to aid with metabolism and digestion, the link between bacterial growth and food consumption is especially relevant to human health.

It is true for most human beings that the number of *Escherichia coli* cells far outnumber the somatic cells. (E.coli, 2018) *E. coli* bacteria are normally found in human intestines, protecting the host from other pathogenic microorganisms. This symbiotic relationship can transform into a parasitic relationship if the *E. coli*. Population exceeds the toxicity limits for its human hosts, causing vomiting, diarrhea, and other illnesses. There are also virulent strains of *E. coli* that are known to cause a multitude of diseases and foodborne illnesses. It is important to find ways to manage the *E. coli* population in human intestines without the use of antibiotics and prevent bacterial resistance. (Sonal, 2018)

Several studies have been performed on the link between bacterial growth and spice in the environment. Specifically, garlic extract has been shown to kill about 75% of the *E. coli* and non-pathogenic Salmonella on chicken meat as well as a majority of the contaminating bacteria on the meat after 10 minutes of exposure. This study also detailed the possibility that garlic could limit the growth of all the bacteria tested. Garlic was also shown to be effective in combating *Helicobacter pylori*, which has been linked to stomach cancer. (Agabalogun, 2012) Other research was done on spices and their antimicrobial abilities preventing food spoilage. It was shown that a dominant chemical in the spice inhibited some unknown function of bacterial reproduction. (Liu, 2017) These studies have shown that garlic possesses some antimicrobial properties.

Along with the study of food, a study from Cornell University claimed that those who ate food seasoned with spices with antibacterial properties were more likely to have greater health, live longer, and have more children. This correlation was shown to be especially prevalent in warmer climates. The study concluded that this trend likely exists due to the chemical compounds found in spice plants that protect them from their natural enemies being at work in food today, and in part kill food-borne bacteria and fungi. Phytochemicals, or essential oils, are the main antimicrobial agents in spices; most of which appear to derive antimicrobial properties from benzene functional groups. Foodborne illnesses are reduced by the incorporation spices to recipes, while at once altering the taste of the food, and making the people who learn to like the new taste are healthier for it. (Food, 1998)

The chi squared values for cumin, cinnamon, black pepper, and ginger, and garlic powder were 11.9, 80.6, 21.1, 63.6, and 80.6, respectively. By analyzing the critical value, each group was shown to be statistically significant. The null hypothesis - the results from each petri dish with a different spice is statistically significant - is rejected.

<u>Procedure</u>

In order to test the hypothesis stated above, five plates of agar, divided into four equally sized sections, were prepared with spices (cumin, cinnamon, garlic powder, black pepper, and ginger). Each quadrant contained a distinct amount of the petri dish's designated spice (1.0 g, 0.5 g, 0.25 g, 0.1 g). One petri dish was prepared without any spices in any of the quadrants. Then, agar was poured in and mixed thoroughly with the spices after being microwaved in intervals of 30 seconds.

Next, each section of the petri dish was inoculated with an equal amount of *E. Coli* bacteria in a specified circular region to measure how the bacteria grew. The dishes were then placed in an incubator upside-down to prevent condensation for 48 hours. After the incubation period, the dishes were removed, observed, and photographed. The growth of the bacteria was then measured in each quadrant with a ruler over through the top lid. There was a repetition of the smallest concentration to create an average and a more accurate range of the bacterial growth.

Safety and cleanliness was an important issue in this experiment. To prevent contamination, an inoculating loop was placed in an incinerator for ten seconds and then used to stir the agar and spice in each quadrant. Between each petri dish, the loop was wiped with an antibacterial wipe to clean off remaining spice and kill any bacteria from the surroundings. While handling inoculation, we wore gloves to prevent any direct contact with the bacteria.

RESULTS

There was a distinct variance between the different spices and bacterial growth. The control had the most growth out compared to all of the set ups with spices mixed into the agar. It had an average of 37.1 mm. Conversely, cinnamon and ginger had no bacterial growth at all.

The next greatest amount of growth was in the petri dish with cumin. It's smallest concentration (0.1 g in a 15 mL quadrant) yielded a growth diameter of 25.5 ± 6.4 mm. There was a strong positive correlation between the concentration of cumin and the growth diameter with the slope of the regression line being 6.9 mm/g.

Less than cumin, the black pepper petri dish also yielded distinct bacterial growth. It's smallest concentration had a bacterial growth diameter of 21.5 ± 4.9 mm. There was also a strong positive correlation between the concentration of pepper and growth diameter with the slope of the regression line being 6.4 mm/g.

Uniquely, the petri dish with ginger was the only petri dish that yielded quantifiable bacterial growth in its smallest concentration and had no growth at higher concentrations. The smallest ginger concentration had 4.5 mm growth diameter, and the concentration had a negative correlation with the growth diameter with the slope of the regression line being -3.5.

DISCUSSION

The results of the study suggest that the best environment for the bacteria to propagate involves no spices. The results suggest that to promote *E. coli* growth in human intestines, one should have no spices, or stick to spices like cumin and black pepper.

These results are crucial to human health because it has implications on the foods that should be integrated into a person's diet based on their intestinal bacterial needs. Especially in cultures where multiple spices are eaten at every meal and areas that are switching from antibiotics to natural remedies, these results have indications that certain spices should be consumed over others.

In India, research was conducted on some of the most common spices in everyday food. They tested the inhibition of *E. coli* and the spices, including the spices that were tested in out experiment. The results were similar to ours, and they mentioned the importance of spreading the knowledge of bacterial inhibition from certain spices across India where people are already using foods and spices as cures for bacterial infections. (Sonal, 2018)

In the future, this study could inspire some more research. This was only a correlational study on spices and *E. coli* growth. Future research could include the cellular processes involved with taking in the chemicals in the spices and how the cell reacts to them, which would be causational.

Also, other scientists could study the effects of spices on different species of bacteria. This experiment was only on one type of bacteria that has a relationship with humans. There are foodborne illnesses and other bacterial diseases that are caused by other bacteria, and research should be conducted on the amount of growth or inhibition of these different species with different spices.

CONCLUSIONS

This experiment was designed in order to determine the effects of different spices on *E. Coli* growth. Petri dishes were prepared with various spices and spice concentrations in order to test the hypothesis of spices and their concentrations affecting *E. Coli.*The results of this experiment are inconclusive, as when compared to the control all spices used decreased the growth rate of bacteria. Contradictory to this, bacteria growth was increased in direct correlation with increased spice concentrations of black pepper and cumin (the only spices which showed growth in each concentration). As the difference between the control and the other spices was shown to be statistically significant, it can be concluded that spices affect bacterial growth. This has a major implication for human health worldwide and natural treatments for diseases from *E. coli*.

WORKS CITED

Agabalogun, L. (2018). *THE SPICES OF LIFE: TESTING THE ANTIMICROBIAL EFFECTS OF GARLIC (ALLIUM SATIVUM), CINNAMON (CINNAMOMUM ZEYLANICUM), AND CLOVE (SYZYGIUM AROMATICUM) AGAINST STREPTOCOCCUS MUTANS*. Retrieved from DREW website: http://www.drew.edu/wp-content/uploads/sites/99/Team3Sandler12.pdf

This source details the effects of spice concentration on bacterial growth and explains the scientific makeup of spices. This research was used in the background information to help explain the effect of spice on bacterial growth.

E.coli (Escherichia coli). (2018, February 26). Retrieved April 8, 2018, from Centers for Disease Control and Prevention website: https://www.cdc.gov/ecoli/general/index.html

This source was used to collect basic information about the bacterium *Escherichia coli*. The facts given by this website were used to open up the background information to provide readers with basic knowledge about the species.

Food bacteria-spice survey shows why some cultures like it hot. (1998, March 4). Retrieved April 9, 2018, from Cornell Chronicle website: http://news.cornell.edu/stories/1998/03/food-bacteria-spice-survey-shows-why-some-cultures-it-hot

This source details the results of a scientific study performed on cultural preferences on spice. It explains the purpose of spices being used in cooking, as well as the effects of consuming spices.

Liu, Q. (2017, June). *Antibacterial and Antifungal Activities of Spices*. Retrieved from U.S. National Library of Medicine database. (Accession No. 10.3390/ijms18061283)

The experiment performed in this article tested different spices and their effect on food spoilage as a result of bacterial and fungal growth. The information from this article was used in the background research to write about the effects of spices on bacterial inhibition.

Sonal, D. (2018, April). *Antibacterial Effect of Herbs and Spices Extract on Escherichia coli*. Retrieved from iMedPub website: http://ejbio.imedpub.com/antibacterial-effect-of-herbs-and-spices-extract-on-escherichia-coli.php?aid=5943

This is a research report on an experiment of *E. coli* inhibition when introduced to spices and herbs. Different paper discs were soaked in solutions with herbs and spices and then placed on inoculated plates. The inhibition ring was measured after incubating the discs for 24 hours, and certain spices showed a strong positive correlation between concentration and inhibition.