Coffee VS. Bacteria

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Question

What is the effect of different concentrations of coffee on bacteria?
Background Information

Independent Variable-

- The concentrations of coffee, also known as the coffee to water ratio. (10ml, 8ml, 6ml, 4ml, and 2ml.)

Dependent Variable-

- The bacteria made using saliva and agar on the petri dish.

Controlled Variable-

- The cleanliness of the lab, the amount of time each colony of bacteria has to grow, and the amount of bacteria being produced.
Background Information

Vocabulary

**Bacteria** - a member of a large group of unicellular microorganisms which have cell walls but lack organelles and an organized nucleus, including some which can cause disease.

**Acrylamide** - a colorless crystalline solid which readily forms water-soluble polymers.

**Nitrogen** - a colorless, odorless unreactive gas that forms about 78 percent of the earth's atmosphere.

**Chlorogenic Acid** - the major polyphenolic compound in coffee, isolated from the leaves and fruits of dicotyledonous plants. This compound, long known as an antioxidant, also slows the release of glucose into the bloodstream after a meal.
Vocabulary

Antioxidant - a substance that inhibits oxidation, especially one used to counteract the deterioration of stored food products.

Dicaffeoylquinic Acid - naturally occurring polyphenolic compound found in plants like fennel and coffee. It is characterized by two caffeic acid molecules linked by ester bonds to one quinic acid molecule.

Acid - a chemical substance that neutralizes and dissolves some metals, and turns litmus red; typically, a corrosive or sour-tasting liquid of this kind.
How does coffee kill bacteria? Coffee has many chemical components that cause bacteria to react and die. Coffee contains Acrylamide. This is a large component in bacteria cell death. Acrylamide is in many foods including coffee and can have a potential risk on you if you consume too much of it. It is a chemical used in water purification which helps state that it kills bacteria in water. Coffee also contains nitrogen. Nitrogen gas is required 24/7 for bacteria growth prevention. But is it required to kill oral bacteria? Nitrogen in this case would help kill the bacteria because the researcher grew the bacteria in a warm oven the nitrogen would then take action. The last large component in the coffee that kills the bacteria is Chlorogenic Acid.
Background Information

How it works

This acid, once decomposed is called dicaffeoylquinic acid. Coffee is considered an antioxidant and this acid is a huge component to coffee that helps it keep its name. This acid kills bacteria in your blood and reduces blood sugar levels.
Background Information

Scientific concepts

What is the process of killing bacteria called? This process is called sterilization. Bacteria are single celled prokaryotic microorganisms. This concept involves many physical and chemical changes in a cell which finally lead to the destruction of that cell. These changes include destruction to the cells protein, alteration of the distribution of a cell’s metabolic processes, and damage of the cell's nucleoid. By adding coffee to the bacteria it will damage the cell in chemical and physical ways.
Background Information

**Why this project was chosen**

The researcher chose this project because she was very interested in seeing how the coffee would react on bacteria. The researcher is also very much into skin care and coffee is used a lot in facial cleansers and scrubs and she always wanted to know if it actually worked.

**How this project will benefit society**

This project will benefit society by showing society that Coffee has the capacity to kill oral bacteria and research has shown that it also kills gut bacteria. This will show society that having coffee in large amounts can be detrimental to your life span. The reason why restaurants offer you coffee after you eat food is because it kills oral bacteria that was built up in your mouth.
Hypothesis

If coffee with the most concentration of caffeine (tested using the Optical power meter) interacts with bacteria then most of the bacteria will be killed because there will be more nitrogen, dicafeoylquinic acid, and Acrylamide (harmful chemicals in coffee that can kill bacteria) in the coffee. These chemicals can harm the bacteria in many ways.
Hypothesis

According to https://www.thestar.com.my/lifestyle/health/2019/05/02/killing-bacteria/

Coffee contains many of the chemical components needed to harm the bacteria’s proteins a nucleus. The nucleoid contain most of the cells genetic materials. Without these genetic materials the cell cannot stay alive. It will also kill the proteins in the cytoplasm of the cell. The cell of bacteria would not have the energy to stay alive.
Materials

- Coffee
- 6 Cotton swabs
- 6 Petri dishes with agar in them
- Graduated cylinder
- Pipette
- 1 black sharpie marker
- Optical power meter
- Water
- Incubator
- French press
- Digital scale
- Boiling pot

- 6 Clear glass vials
- Beaker
- Tape
- Stove
- Heat protection gloves
- Flashlight
- Ruler
- Camera
Procedure (Making the coffee and testing the opacity)

1. Boil water with the stove.
2. Measure 12 grams of coffee grounds using the digital scale.
3. Put the grounds in a french press.
4. Measure 100 ml of boiling water using a graduated cylinder and pour the water into the french press.
5. Allow the coffee to steep in the french press for 10 minutes.
6. Push the plunger down on the french press.
7. Pour out the coffee into the graduated cylinder.
8. Label five clear glass vials from 1-6.
9. Use a pipette to measure 10 ml of the original coffee and deposit it into vial number 1.
10. Use a pipette to measure 8 ml of the original coffee and deposit it into vial number 2.
11. Use a pipette to measure 6 ml of original coffee and deposit it into vial number 3.
12. Use a pipette to measure 4 ml of original coffee and deposit it into vial number 4.
13. Use a pipette to measure 2 ml of original coffee and deposit it into vial number 5.
14. Use a pipette to measure 2 ml of boiled water and deposit it into vial number 2.
15. Use a pipette to measure 4 ml of boiled water and deposit it into vial number 3.
Procedure (Making the coffee and testing the opacity)

1. Use a pipette to measure 6 ml of boiled water and deposit it into vial number 4.
2. Use a pipette to measure 8 ml of boiled water and deposit it into vial number 5.
3. Use a pipette to measure 10 ml of boiled water and deposit it into vial number 6 as a control to measure the opacity of the concentrations of coffee.
4. Hold the control vial (vial number 6) right in front of the photodiode which is connected to the optical power meter.
5. Place the flashlight 7.5 cm away from the glass vial so the light shines directly on the vial.
6. Turn the optical power meter on and set the optical power to 0.
7. Turn the flashlight on and read the optical power meter and note the amount of power.
8. Repeat step 20, five times each for each of the glass vials.
9. The higher concentration coffee should have a lower optical power because it is less opaque. This will be used to define the concentration of the coffee.
The different concentrations of coffee
Using the pipette to pick up different amounts of coffee.
The optical power meter that is used to test the opacity of the coffee concentrations.
The results of the opacity of each coffee concentration were shown here.
Procedure (Making the bacteria)

1. Label each Petri dish with the numbers 1-6.
2. Take a cotton swab and dip the tip of the swab in distilled room temperature water.
3. Using that same cotton swab, take a swab of the inside of your cheek.
4. Then take that swab and put it in the center of the dish, touch the swab to the agar and turn it 180 degrees.
5. Using the same swab make a 4 line star using the center dot as a guideline to where the lines you are drawing with the swab should cross.
6. Close the petri dish, flip it upside down, and tape it closed.
7. Repeat steps 26-29 for all 6 Petri dishes.
8. Allow the bacteria colonies to accumulate over the span of 30 hours in the incubator with a heat source set to 34°C.
9. Take a picture of each one of the 6 Petri dishes.
10. Use a pipette to add 2 ml of coffee to each one of the correspondingly labeled Petri dishes.
11. Take a picture of the dishes after letting them sit in the incubator for 30 hours.
Making the four line star with the cotton swab on the agar in the petri dish.
The petri dishes numbered from 1-6.
The incubator
And the dishes stacked in the incubator.
Procedure  (Collecting data)

1. Edit the *before* and *After* pictures changing the opacity, tint and skin tone so you can better see the colonies.
2. Locate the six longest lines of culture and number them on the *before* image.
3. Find the corresponding image of the dish after adding coffee and edit the image the same way.
4. Repeat steps 35-38 for all of the dishes.
5. Count each of the six longest lines separately on your *before* image of your bacteria and record the data.
6. Count each of the six longest lines separately on your *after* image of your bacteria and record the data.
7. Repeat steps 40 and 41 for all of the data.
This is an example of dish number 5.

An example of what the dishes looked like and how the researcher distinguished between each line of bacteria colonies she was counting.
All of the petri dishes.
Average Percentages of Colonies killed with Different Concentrations of Coffee

![Graph showing the relationship between concentrations of coffee and percentage of colonies killed. The percentage of colonies killed increases as the concentration of coffee increases. The graph is a line chart with concentrations of coffee (ml) on the x-axis and percentage change on the y-axis. The line starts near 0% at 0 ml, increases gradually to around 10% at 4 ml, plateaus at around 15% from 4 to 6 ml, then sharply increases to around 30% at 8 ml, and continues to rise steeply to about 40% at 10 ml.](image-url)
## Average Table

<table>
<thead>
<tr>
<th>Coffee Concentrations</th>
<th>Without Coffee</th>
<th>With Coffee</th>
<th>Percentages</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 ml (Water/control)</td>
<td>164</td>
<td>162</td>
<td>1.22%</td>
</tr>
<tr>
<td>2 ml</td>
<td>191</td>
<td>184</td>
<td>3.66%</td>
</tr>
<tr>
<td>4 ml</td>
<td>133</td>
<td>121</td>
<td>9.02%</td>
</tr>
<tr>
<td>6 ml</td>
<td>144</td>
<td>122</td>
<td>15.28%</td>
</tr>
<tr>
<td>8 ml</td>
<td>221</td>
<td>190</td>
<td>14.03%</td>
</tr>
<tr>
<td>10 ml</td>
<td>124</td>
<td>78</td>
<td>37.10%</td>
</tr>
</tbody>
</table>

## Data Analysis

The data shows that 10 ml of coffee to no water killed an average of 37.10% of the colonies of bacteria compared to 2 ml of coffee to 8 ml of water, which killed 3.66% of the colonies of bacteria. 10 ml of coffee killed the most bacteria thus far.
Data Analysis

- 10 ml of coffee to no water killed 37.10% of the colonies of bacteria.
- 8 ml of coffee to 2 ml of water killed 14.03% of the colonies of bacteria.
- 6 ml of coffee to 4 ml of water killed 15.28% of the colonies of bacteria.
- 4 ml of coffee to 6 ml of water killed 9.02% of the colonies of bacteria.
- 2 ml of coffee to 8 ml of water killed 3.66% colonies of bacteria.
Conclusion

- The research question was, “What is the effect of different concentrations of coffee on bacteria?”
- The hypothesis states that 10 ml of coffee to 0 ml of water would kill the most bacteria compared to 2 ml of coffee to 8 ml of water.
- The data was supported because of the difference in the amount of nitrogen, Acrylamide, and dicaffeoylquinic acid. They all have coffee killing components that help the coffee break down barriers of proteins.
Conclusion

- As stated in the previous slide, Acrylamide, nitrogen, and Dicaffeoylquinic acid are some main components in killing bacteria in the coffee.
- These components affect the researchers' project in many ways, mainly helping kill the bacteria but also helping understand why coffee has the capacity to kill the bacteria. This is how the scientific principles discussed in the background information connects to the researchers' project.
Conclusion

- During the process of conducting this experiment there were many errors including the difference in length of lines of colonies made by the researcher.
- Another error was the comparison of the lines made by the researcher.
- Some more errors include accuracy of counting the colonies and the exclusion of fully understanding the effect of the optical power meter.
- If the researcher were to conduct this experiment again they would measure out the lengths on the lines drawn onto the agar in the petri dish. They would also use a more efficient way of counting the colonies on the dish.
Conclusion

- Many things were learned in the process of conducting this experiment not only about bacteria and coffee, but also that the smell of bacteria growing in agar smells extremely bad. Some comparisons to the smell include moldy bread and socks or unbrushed teeth. (bad breath)

- This project benefits society in many ways, like helping explain why restaurants give out coffee after dinner. The answer is to help kill oral bacteria. This project benefits society also by explaining that coffee isn’t necessarily bad for you, but not to over use it, rather consume it in small amounts.

- Ideas for future research include testing different brands of coffee on bacteria, or really experimenting with the optical power meter, and testing opacities of daily used objects.
Thank you for your attention

Are there any questions?