The Effects of Mentha and Mentha Piperita on Heart Rate and Hemoglobin Levels in *Daphnia magna*

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Abstract

Heart diseases have a large impact on modern society and have continued to affect millions of lives. The purpose of this experiment was to test the effects of Mentha and Mentha piperita on the heart rate and hemoglobin levels in *Daphnia magna*. It was hypothesized that Mentha and Mentha piperita solutions would be able to increase heart rate and hemoglobin levels in *D. magna,* as mint is known for having anti inflammatory properties, which are useful in fighting heart diseases. Three solutions were made for both types of mint, a 0.05% solution, a 0.5% solution, and a 5% solution. Three solutions were placed onto the *D. magna* and were then observed under a microscope for one minute for their heart rate, followed by being placed into a spectrophotometer for their hemoglobin levels to be collected. It was found that Mentha and Mentha piperita were able to increase the heart rate and hemoglobin levels of the *D. magna*. As the concentration of the mint solutions increased, the heart rate and hemoglobin levels of the *D. magna* also increased. The 5% mint solution caused the *D. magna* to have a much greater heart rate than the control. This can perhaps lead to more humans adding Mentha or Mentha piperita to their diet and potentially improving their health along with decreasing the chances of getting a heart disease.

Keywords: *Daphnia magna*, heart rate, heart disease, hemoglobin levels, *Mentha, Mentha piperita*

Introduction

Heart diseases are an ongoing issue in our society that continues to affect millions of lives, even today. Heart diseases have a large impact on the modern day world.Approximately 2.6 million people in the United Kingdom struggle with the buildup of fatty wastes in their

coronary arteries, leading them to get heart diseases (Goodall, 2018). In a group of 75 million Americans, 1 in every 3 Americans suffer from a heart disease (Goodall, 2018). There are many different types of heart diseases and these usually form due to the buildup of fatty wastes in the heart's arteries, causing the arteries' walls to thicken and the lumen to narrow. Sometimes they form when the arteries begin to narrow, causing the heart rate to slow down. Among the most typical illness of the circulatory system is arteriosclerosis, where the fatty deposits in the arteries triggers the walls to stiffen and thicken (Goodall, 2018).

Mentha, otherwise known as mint, is known for having many healing properties and has been used for thousands of years. Mint has been effective in helping with indigestion in the stomach and is able to increase bile flow, decreasing the pain caused by indigestion. Indigestion may occur when food sits in the stomach for too long before passing through the rest of the digestive tract (Pearson, 2017). Due to heart diseases having a large effect on the modern day world, in my experiment, I will be aiming to find a cheaper alternative to help prevent heart diseases using mint.

Mint is the most well known herb for medicine. It is used in folk and traditional medicines.(Mokhtar M. Bishr and others, 2018). Along with being able to help with indigestion, mint is also said to treat cough, inflammation of the throat, bronchitis, and other internal issues. Mint vapor is used as an inhalant for respiratory congestion, while its tea is used to treat bronchitis, and the inflammation of the oral mucosa and throat (Mokhtar M. Bishr and others, 2018). In addition to their medical uses, they are widely used in the pharmaceutical industry as flavoring agents (Mokhtar M. Bishr and others, 2018). Mint and *Mentha piperita* (peppermint) have flavonoid agents, flavonoids are needed in order to prevent heart diseases. In my experiment, I used mint and peppermint.

The goal of my research is to find a new way to prevent heart diseases that can be a cheaper alternative compared to expensive medicines and diets. Pearson and others (2017) have done multiple studies testing the effects of mint on Irritable Bowel Syndrome. Irritable bowel syndrome (IBS) is a common digestive tract disorder. It is characterized by digestive symptoms like stomach pain, gas, bloating and changes in bowel habits (Pearson, 2017). Many studies have been conducted due to mint's well known healing properties. One study found that 75% of patients who took peppermint oil for four weeks showed improvements in IBS symptoms, compared to 38% of the patients in the placebo group (Pearson, 2017). In my experiment, I tested to see if mint and peppermint are able to help with heart disease and increased heart rate, allowing one to have a lower chance of getting a heart disease.

Studies have also been done testing the effects of mint on indigestion. A clinical study in people with indigestion showed that a combination of peppermint oil and caraway oil taken in capsules had effects similar to medications used to treat indigestion. This helped improve stomach pain and other digestive symptoms (Pearson, 2017). Mint has similar effects to medicines and is able to help reduce the amount of pain caused by indigestion. Since heart disease has such a large impact on the people in today's society, my research can provide a

cheaper alternative to help one prevent getting heart disease without spending a large amount of money.

Heart diseases usually occur due to the buildup of fatty wastes in the heart's arteries and the narrowing of the heart's arteries. This does not allow the exchange of blood and oxygen to occur and results in a lower heart rate. Hemoglobin levels are also negatively affected by heart diseases. Therefore, In my experiment I used mint and peppermint to test to see if they can increase heart rate and can allow the exchange of blood and oxygen to occur.

Daphnia magna, otherwise known as Daphnia, are tiny water fleas that live in freshwater. *D. magna* has a clear, external structure, allowing one to see its internal organs. The body is enclosed by a transparent shell-like structure, called a carapace, which is mostly made of chitin (Elenbass, 2013). One of the internal organs that is visible is the *D. magna*'s heart and one is able to count its heart beat. These water fleas are very small, usually 2-5 mm long, with an overall shape similar to a kidney bean (Elenbass, 2013). Since *D. magna* is small and their heart is visible, I used *Daphnia magna* for my experiment to test the effects of mint and peppermint on their heart rate and hemoglobin levels.

Hypothesis:

Mentha is known for having many healing properties. In a study done by Bishr and Salama (2018), different types of mints were analyzed for their properties and it was found that *Mentha* and *Mentha piperita* have many inflammatory properties along with it being able to help with cough and bronchitis. If I place *Mentha* and *Mentha piperita* solutions on *Daphnia magna*, then it will cause Daphnia's heart rate to increase. This is because inflammation of the heart is usually a result of heart diseases. Since *Mentha* and *Mentha piperita* have anti-inflammatory properties, this will result in the *Daphnia magna* having a lower chance of getting heart diseases.

Materials and Method

Preparation of the *Mentha* and *Mentha piperita* solutions:

In this experiment, the independent variable is the type of mint. *Mentha*-mint, and *Mentha piperita*- peppermint, (Amazon) solutions were made. Three solutions were made, a 0.05% solution, 0.5% solution, and 5% solution (refer to Table 1). The 5% solution was made by having 180 mL of water filled into a beaker. 10 mL of the mint extract was then placed into the beaker with the water. For the 0.5% solution, 180 mL of water was filled into a second beaker, then 20 mL of the liquid from the 5% solution was added. For the 0.05% solution, 180 mL of water was filled into a third beaker, 20 mL of the liquid from the 0.5% solution was added. These steps were repeated to make the 0.05% , 0.5% , and 5% peppermint solutions, using different

beakers (refer to Table 2). A control group was also made. The control group was 180 mL of water.

Table 1: Solutions for *Mentha*

Control group	5% *Mentha* solution	0.5% *Mentha* solution	0.05% *Mentha* solution
180 mL water	180 mL of water with 10 mL of *mentha* extract.	180 mL of water with 20 mL of liquid from the 5% *mentha* solution.	180 mL of water with 20 mL of the liquid from the 0.5% *mentha* solution.

Table 2: Solutions for *Mentha piperita*

Control group	5% *Mentha piperita* solution	0.5% *Mentha piperita* solution	0.05% *Mentha piperita* solution
180 mL water	180 mL of water with 10 mL of *mentha piperita* extract.	180 mL of water with 20 mL of the liquid from the 5% *mentha piperita* solution.	180 mL of water with 20 mL of the liquid from the 0.5% *mentha piperita* solution.

Experimental Setup:

In this experiment, the dependent variable is the heart rate and hemoglobin levels of *Daphnia magna*. *Daphnia magna* (Connecticut Valley Biological Supplies), otherwise known as Daphnia, will be used. The solutions that were prepared were tested on the *D. magna*. Four groups of 5-6 *D. magna* each were transferred to four small cups. The first group was a control group, only water was added to the *D. magna* while their heart rate and hemoglobin levels are being collected (refer to table 3 & 6).

After this, a single *D. magna* from another group was transferred to an empty slide using a pipette. Before placing them into the solutions, I ensured they were alive under a hand-held microscope. Two drops of the 0.05% mint solution were placed onto the *D. magna.* The *D. magna* was then observed under the microscope for one minute. During this minute, their heart rate was collected, for it gave me their heart rate in beats per minute (bpm) (refer to table 4).

Once the heart rate has been collected, the hemoglobin levels of each *D. magna* were collected using a spectrophotometer. After that, the *D. magna* was placed into a test tube to then be placed into the spectrophotometer to have the *D. magna* hemoglobin levels taken (refer to table 7). The hemoglobin levels were in grams per deciliter (g/dL). After that, the *D. magna* was gently cleaned and returned to the small cup that it was placed in earlier. The slide was then cleaned and wiped dry. Followed by another *D. magna* from the same cup was placed onto the slide. For the heart rate, these steps were repeated for a total of five trials. For the hemoglobin levels, these steps were repeated for a total of two trials. Once the 0.05% solution trials are finished, these steps were repeated with the 0.5% solution and the 5% solution using the *D. magna* from the unused groups. Once finished, new groups of *D. magna* were made and the used *D. magna* had their hemoglobin levels taken, then cleaned and returned to their culture. All these steps were then repeated using new groups of *D. magna* with the peppermint solutions (refer to tables 5 & 8).

Daphnia magna will be used in my experiment, stating that no sterilization will be needed and there is no sort of open flame or culturing. Low concentrations of *Mentha* and *Mentha piperita* will be used, they pose no threat to the experimenter or the environment. Solutions will be prepared under the supervision of the designated supervisor. All safety procedures will be followed and all solutions and chemicals will be returned properly to the research lab.

Results

The hypothesis of this experiment stated that if *D. magna* were placed in *Mentha* and *Mentha piperita* solutions, then their heart rate and hemoglobin levels would increase. The heart rate of the *D. magna* is presented in table 4 and 5, the heart rate gradually increased as the concentration of *Mentha* solutions and *Mentha* piperita solutions also increased. The different *Mentha* solutions increased heart rate, resulting in the *D. magna* having a lower chance of the buildup of fatty wastes in their heart. Fatty wastes in the heart and its arteries usually result in a decreased heart rate, since *Mentha* caused the heart rate in *D. magna* to increase, then the *D. magna* are less likely to have a heart problem.

The hemoglobin levels of the *D. magna* is presented in tables 7 and 8, similar to the heart rate, the hemoglobin levels also increased as the concentration of *Mentha* and *Mentha piperita* solutions also increased. This shows that *Mentha* solutions were able to increase heart rate and they also increased hemoglobin levels. For if *Mentha* and *Mentha piperita* is able to increase hemoglobin levels, then my hypothesis was supported and results in *D. magna* having a lower chance of getting a heart disease.

Table 3: Control Group for heart rate

Trials	Number of heartbeats (bpm)
1	156 bpm
2	120 bpm
3	148 bpm
4	134 bpm
5	188 bpm

Table 4: Heart rate in *Daphnia magna* using *Mentha* solutions

Trials	0.05% mentha solution	0.5% mentha solution	5% mentha solution
1	124 bpm	150 bpm	175 bpm
2	135 bpm	203 bpm	193 bpm
3	149 bpm	177 bpm	210 bpm
4	168 bpm	156 bpm	192 bpm
5	165 bpm	184 bpm	219 bpm

Table 5: Heart rate in *Daphnia magna* using *Mentha piperita* solutions

Trials	0.05% Mentha piperita solution	0.5% Mentha piperita solution	5% Mentha piperita solution
1	61 bpm	126 bpm	172 bpm
2	75 bpm	170 bpm	217 bpm
3	83 bpm	151 bpm	183 bpm
4	167 bpm	203 bpm	206 bpm
5	189 bpm	119 bpm	194 bpm

Table 6: Control group for Hemoglobin levels

Trials	Hemoglobin levels (g/dL)
1	3.4 g/dL
2	3.2 g/dL

Table 7: Hemoglobin levels in *Daphnia magna* using *Mentha* solutions

Trials	0.05% mentha solution	0.5% mentha solution	5% mentha solution
1	4.6 g/dL	5.4 g/dL	7.8 g/dL
2	3.7 g/dL	6.1 g/dL	8.4 g/dL

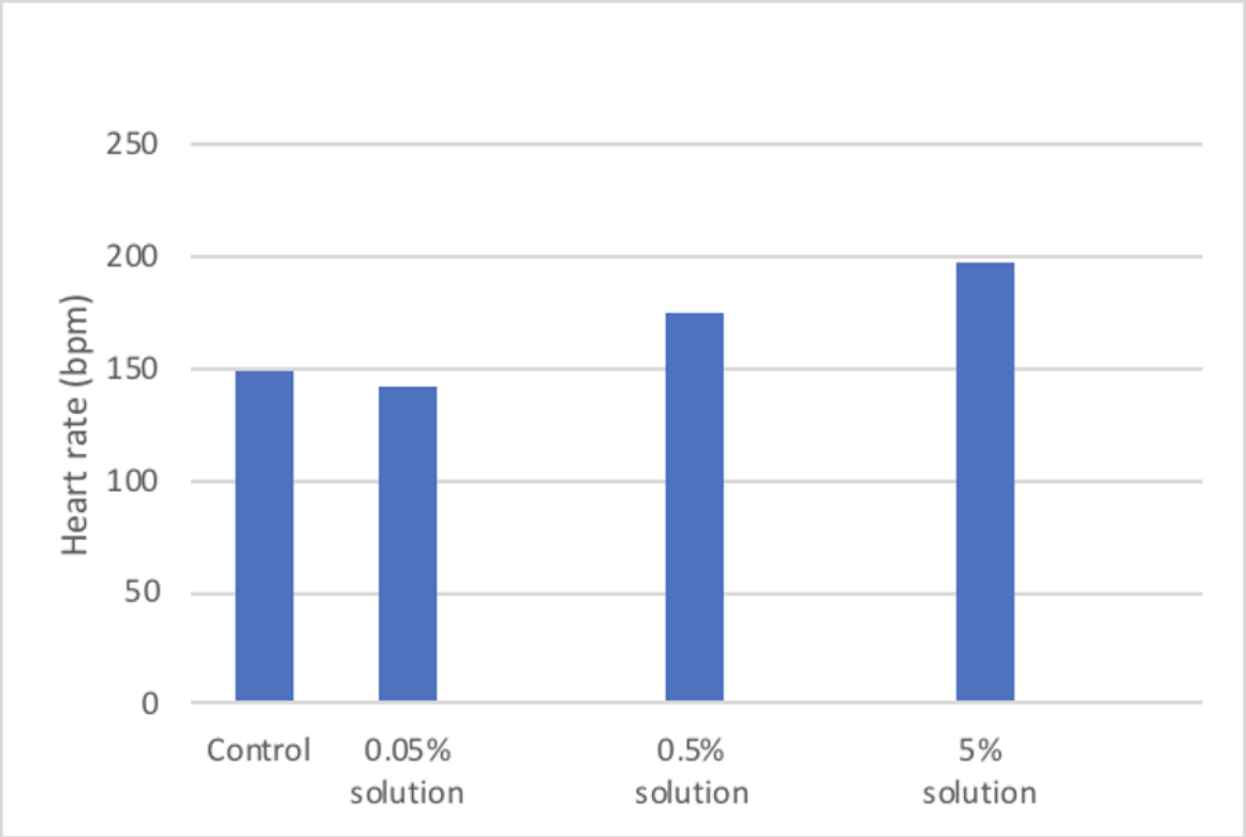
Table 8: Hemoglobin levels in *Daphnia magna* using *Mentha piperita* solutions

Trials	0.05% mentha piperita solutions	0.5% mentha piperita solution	5% mentha piperita solution
1	5.0 g/dL	6.2 g/dL	8.2 g/dL
2	5.1 g/dL	7.4 g/dL	8.6 g/dL

Table 9: Averages and Standard Deviation

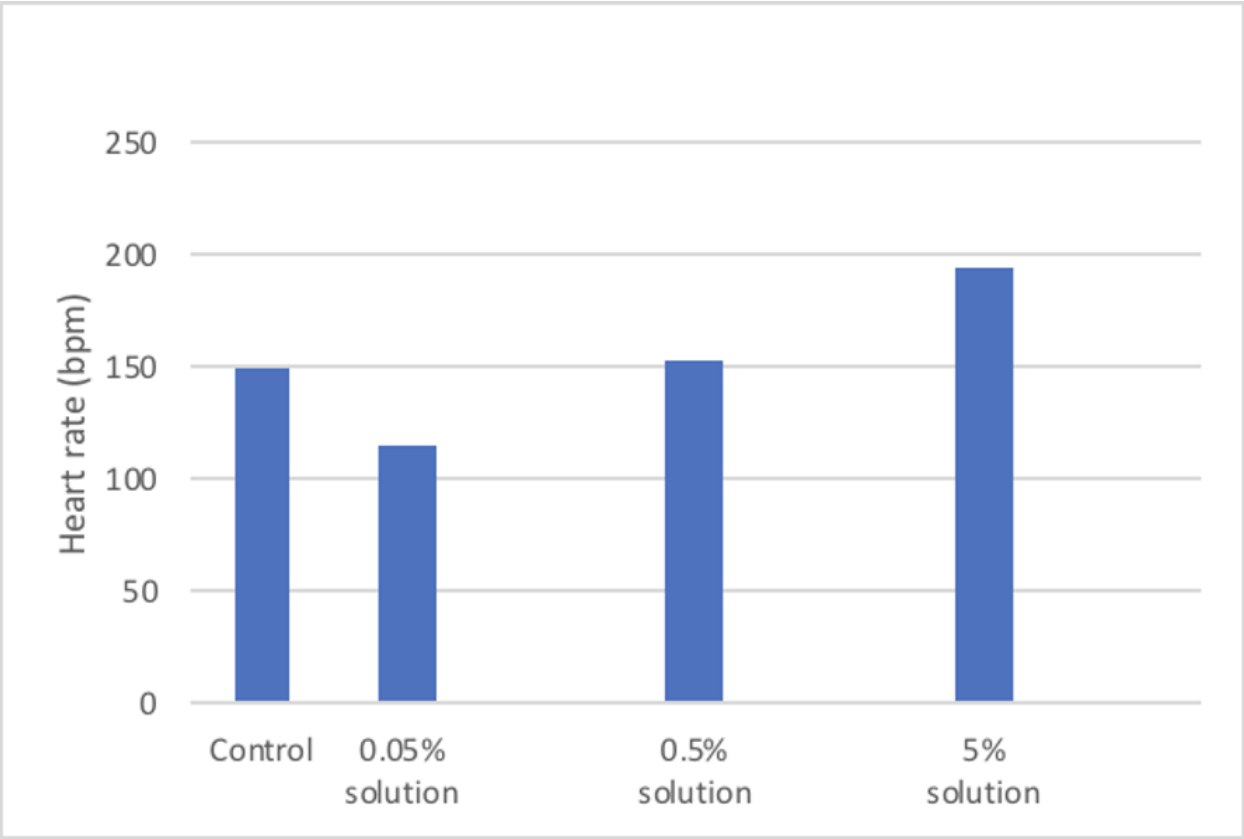
Group	Type of mint	Average ± Standard Deviation
1	Mentha	199 ± 20
2	Mentha piperita	200 ± 15

Figure 1: Average number of heart beats for mentha in *D. magna* in beats per minute (bpm)



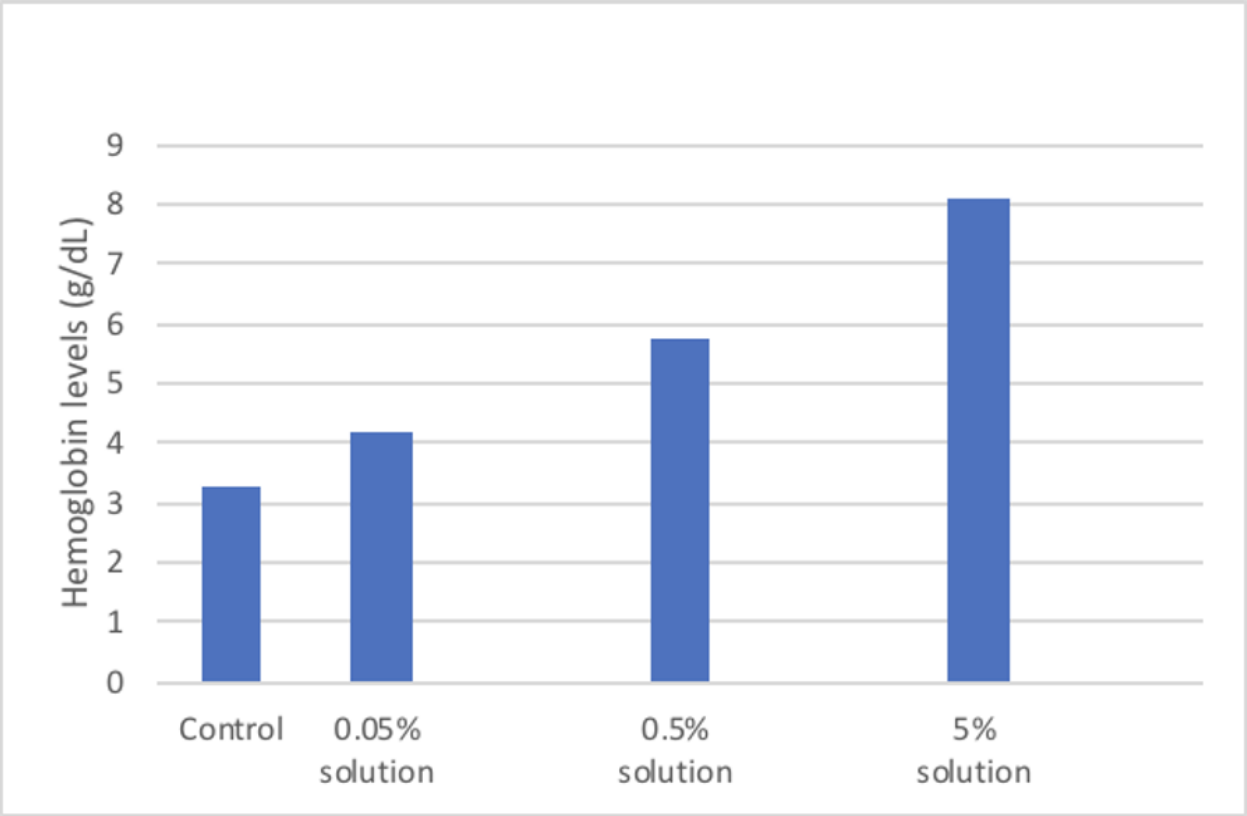
This graph shows the average heart rate of the *D. magna* that was in the *Mentha* solutions. The 0.05% solution didn't greatly affect the average heart rate. As the different mint solutions were added to the *D. magna*, their heart did gradually increase. The standard deviation for the control was 157 ± 23, for the 0.05% *Mentha* solution it was 160 ± 9, for the 0.5% solution it was 167 ± 16, and for the 5% it was 199 ± 20.

Figure 2: Average number of heart beats for mentha piperita in *D. magna* in bpm



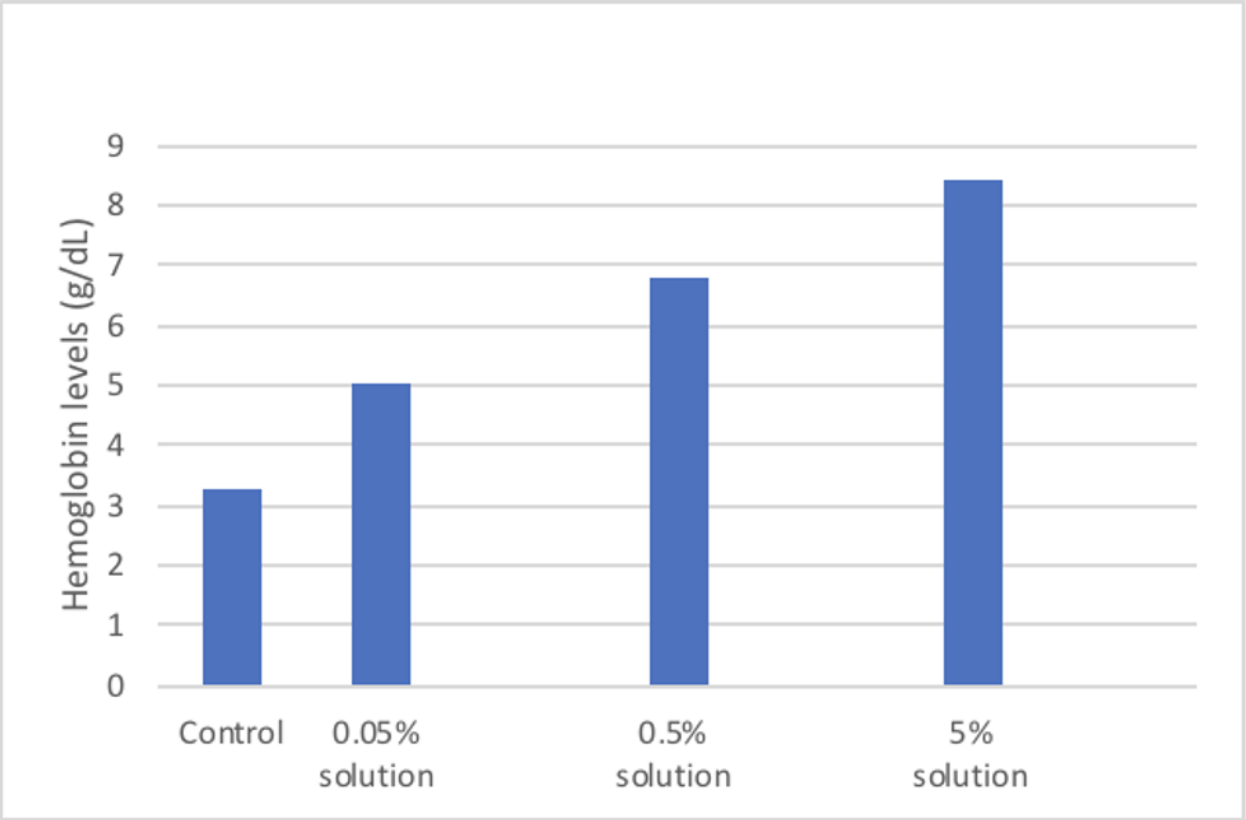
This graph shows the average heart rate of the *D. magna* that was in the *Mentha piperita* solutions. When the 0.05% peppermint solution was given to the *D. magna*, their heart rate did not increase. Yet, when the 0.5% and 5% peppermint solutions were added to the *D. magna*, their heart rate increased. The standard deviation of the 0.05% *Mentha piperita* solution was 128 ± 58, for the 0.5% solution was 162 ± 32, and for the 5% it was 200 ± 15.

Figure 3: Average hemoglobin levels for mentha in *D. magna* in g/dl



This graph shows the average hemoglobin levels of the *D. magna* in the *Mentha* solutions. When the Daphnia were placed in the 0.05%, 0.5%, and 5% mint solutions, their hemoglobin levels increased. The higher concentration of the solution, the higher hemoglobin levels of the *D. magna* were.

Figure 4: Average hemoglobin levels for mentha piperita in *D. magna* in g/dl



This graph shows the average hemoglobin levels of the *D. magna* in the *Mentha piperita* solutions. As the *D. magna* were placed in different peppermint solutions, their hemoglobin levels gradually increased.

Discussion

The outcome of this experiment shows that *Mentha* and *Mentha piperita* were able to increase heart rate and hemoglobin levels in *D. magna*. This shows that the hypothesis was supported. As the different mint and peppermint solutions were added to the *D. magna*, their average number of heart beats and their hemoglobin levels increased. The standard deviation also showed that my hypothesis was supported. For example, the standard deviation of the control was 157 ± 23, while the standard deviation for the different *Mentha* solutions were 160 ± 9, 167 ± 16, and 199 ± 20; also, the standard deviation for the different *Mentha piperita* solutions were 128 ± 58, 162 ± 32, and 200 ± 15. Comparing the standard deviation of the control to the different solutions, the standard deviation has increased, stating that *Mentha* and *Mentha piperita* were able to increase heart rate in the *D. magna*.

It was predicted that *Mentha* and *Mentha piperita* would be able to increase heart rate and hemoglobin levels in Daphnia magna because mint is said to have had many healing properties. Since heart diseases usually result in a decreased heart rate and lower hemoglobin levels, mint might've been able to increase heart rate for it is known for having healing abilities. In this experiment, the healing properties of mint were tested by having different mint solutions placed on *D. magna* to see if their heart rate and hemoglobin levels would increase. As a result, the mint did cause the average heart number of heart beats and hemoglobin levels of the *D. magna* to increase. Since the mint and peppermint both increased heart rate in *D. magna*, then it could be said that mint can help with heart disease and help one with a low heart rate.

Although it was found that *Mentha* and *Mentha piperita* are able to help with heart problems, in a study done by Pearson and others (2017), they tested the effects of peppermint oil on Irritable Bowel Syndrome (IBS) and if it could improve the symptoms of IBS. IBS is when one suffers from issues in the digestive track and experiences stomach pain, bloating, and gas. Two groups were made; one group was given capsules with peppermint oil and the other group was given a regular capsule. One study found that 75% of patients who took peppermint oil for four weeks showed improvements in IBS symptoms, compared to 38% of the patients in the placebo group (Pearson, 2017). The results stated that overall, individuals who suffered from IBS in the experimental group did see improvements in their digestive system. More members in the experimental group saw improvements than the members in the control group. My results agree with the results of a Pearson's study because both Pearson and I were testing the healing properties of mint on different issues. Both results concluded that mint is able to help with internal issues, whether it is in the heart or the stomach. My results are similar to Pearson's in which we both found that mint is able to help with internal body problems. An exception between my experiment and theirs is that they tested their hypothesis on humans while I tested my hypothesis on *D. magna*. Another exception is that Pearson did a test on a group of 700 people with IBS while I only tested five *D. magna* for each mentha solution.

Conclusions & Future Studies

This study established that *Mentha* and *Mentha piperita* were able to increase the average number of heart beats and the hemoglobin levels in *Daphnia magna*. As different mint and peppermint solutions were added to the *D. magna*, their heart rate gradually increased. The average number of heart beats and hemoglobin levels for the mint solutions were greater than the average number of heart beats and hemoglobin levels for the control group.

A possible error made in this experiment could be that during the timespan of collecting data, the *Mentha* and *Mentha piperita* solutions began to evaporate, resulting in changed concentrations of the solutions.. These errors could have resulted in my data not being completely accurate. A possible solution to minimize this error is to place the mint solutions in a cooler place. The solutions were kept in a small cabinet during time periods where experimentation did not occur, yet it was a bit warm in the cabinet which might have been the reason for the evaporation. One way to improve this experiment would be to create more trials for testing the hemoglobin levels of the *D. magna*. In this experiment only two trials were done for the hemoglobin levels, yet my data could have been more accurate if I had done more trials.. As mentioned before, the solutions evaporated over time, to prevent this from occurring again the solutions should be kept in a different area. Perhaps in further study, *Mentha* and *Mentha piperita* should be tested on humans to see if it'll help their health. Since mint is known for having many healing abilities and is now possible for helping with the heart of *D. magna*, then in future study, mint could be able to help humans decrease their chances of getting a heart disease and overall improve their health.

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