Journal of Undergraduate Biology Laboratory Investigations

2018

Different Concentrations of Caffeine Affecting the Heart Rate of Daphnia

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Caffeine occurs naturally in many plants, therefore it is also found in many food products, and many humans consume some form of caffeine everyday. For this reason many studies have been conducted on caffeine and how it affects the humans body. Some studies use Daphnia to test the effects of caffeine because the Daphnia's heart is similar to a humans and are easy to use for lab use. We predicted that by adding different concentrations of caffeine to the Daphnias environment we would speed up the heart rate in beats per minute (BPM) of the Daphnia. We conducted four trials per each concentration of caffeine finding the percent change in the Daphnia heart rate. Based on our results we found that there was not a significant difference between the two concentrations; however, we found that caffeine did have an overall increase in heart rate, this may be helpful to know for those who have a higher BPM that might want to avoid increasing it further.

Introduction

Caffeine is widely consumed substance and a known stimulant. It has physiological and psychological effects such as causing sympathetic nervous system activity. (Sondermeijer, et al., 2002). Humans usually consume caffeine in the form of a beverage or pill (Powell et al., 2001). It can be found in many common household items such as coffee, soda, tea, and energy drinks. When caffeine enters the body, it has drug like effects interacting with the nervous system by triggering dopamine receptors.

Caffeine greatly interacts with catecholamines that then influence the nervous system. Catecholamines are hormones that are produced from our adrenal glands. The three types of catecholamines are dopamine, epinephrine, and norepinephrine. Adenosine, a known neurotransmitter, is the common cause for why most people feel tired (Green and Suls, 1996). Neurotransmitters are used during the process of action potential propagation. An action potential is an electrical potential that helps the passage of an impulse along the membrane of a muscle, like in the heart. Caffeine binds with adenosine receptors and stops it from reaching the brain. When the adenosine is blocked, excitatory neurotransmitters move freely and cause the pituitary glands to release hormones that aid in adrenaline production. The production of adrenaline can increase action

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potentials firing throughout the body. Production of adrenaline and an increase in action potentials can increase blood pressure which can cause an increase in heart rate. (Scott, 2015).

The human heart is myogenic, meaning it it controlled by the muscle. Most arthropods have a neurogenic heart which are controlled by the nervous system (Stein, et al., 1966). Daphnia, which are planktonic crustacean has a myogenic heart like humans (Ebert, 2005). The Daphnia's heart is located posteriorly to the intestine. The Daphnia are much easier to test the effects on rather than humans since their hearts are similar. Since the Daphnia is transparent we were able to observe how the caffeine affected the heart rate. We decided to test the effects that caffeine would have on the Daphnia's heart rate. We believed that testing different strengths of caffeine would best display how chemicals can influence heart rate because testing several types of stimuli could lead to complicated results. The question we attempted to answer was how will adding different stimuli affect the heart rate of a Daphnia? We hypothesized by increasing the amount of caffeine the heart rate will also increase because caffeine activates neurotransmitters that excite the body which then increases heart rate.

Methods

Our study system included each Daphnia used in each specific trial, the two concentrations of caffeine, and the plain water solution. The experimental variable was the concentration of caffeine, .005M and .01M. The dependent variable that was measured was the change in heart rate. We measured the heart rate before the chemical was added and also measured the heart rate after it was added. We believed this was the best method to see how the rate changed when the caffeine was present. We used a different Daphnia for each chemical trial. There is high variability between each Daphnia so we chose to change the Daphnia for each solution to ensure accuracy. The negative control group was the Daphnia measured in the water solution only. This was used to compare the effect of the caffeine compared to no caffeine present.

We referred to the Daphnia Heart Rate Procedure (Shaw and French, 2018) to see how to properly record the Daphnia's heart rate. Once the Daphnia was acquired, we placed them on the slide and added our chemicals. First, we tested water and followed by testing two concentrations of caffeine. We let the solution sit for 7 minutes before any data was collected. We tested the heart rate of the Daphnia with just water. After, we collected our heart rate before the .005M caffeine solution was added. We recorded the Daphnia on a phone from the microscope. We recorded the Daphnia heart beat for 10 seconds, slowed down the video, counted the recorded heart beats, and then multiplied our number of heart beats by six. By slowing down the video, we received a more accurate view for heart rate calculation. We then added the solution and repeated the heart rate counting procedure again. We followed by repeating the same procedure steps for the .01M concentration of caffeine. We ran 4 trials for each solution to ensure we had enough data for comparison. The same lab member counted the heart rates for every trial with the same hand counter.

To calculate the heart rate, we calculated the percent change from the pre heart rate to the post heart rate of each trial. We made a Box and Whisker graph and also calculated averages for each concentration.

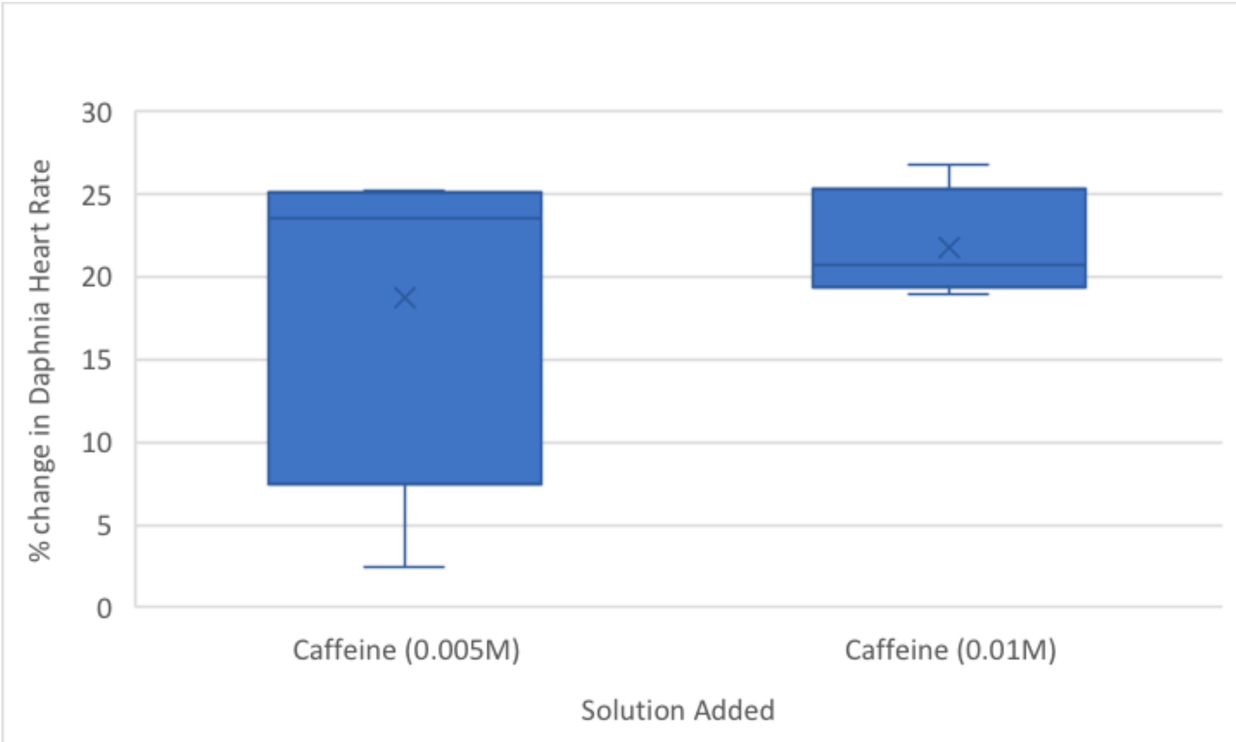

Fig. 1a. A Box and Whisker graph showing the percent change in Daphnia heart rate, after different concentrations of caffeine were added. The two concentrations produced similar percent changes and specific data trends cannot be concluded.

We ran two paired t-tests to compare the pre and post heart rates for both the low and high concentrations. This was a paired t-test because the pre and post heart rates came from the same Daphnia (before and after). We ran a unpaired T-test to analyze our data's statistical significance by comparing the percent change values for our high and low concentrations.. We used an Unpaired t-Test because we had two different nominal groups. These tests were conducted using PAST3.

Results

There was not a major difference in the percent change in heart rate between the two concentrations of caffeine. As shown in figure 1a. the average percent change for the 0.005M was 18.7 percent and the average percent change for the 0.01M was 21.7 percent.

We ran a paired t-Test for each of the concentrations to compare the before and after heart rates in order to test the overall effect of caffeine being added. We ran one for each of the two concentrations of caffeine. For the 0.005 M caffeine, there was significant difference between the pre and post exposure heart rates of Daphnia; $t(3) = -3.45$, $p = 0.041$. A paired t-Test was conducted to compare the effect of 0.01 M caffeine on Daphnia heart rate in treated (after caffeine) and untreated (before caffeine). There was a significant difference between the two condition; $t(3) = -11.048$, $p = 0.0016$. We then ran an unpaired t-Test to compare the two different concentrations (0.005M and 0.01M) of caffeine on the heart rate percent change values of Daphnia. There was not a significant difference between the two concentrations when comparing the percent changes; $t(3) = 2.45$, $p = 0.61$.

Discussion

Based on our statistical analysis, our hypothesis was not supported. The difference found between the two concentrations of caffeine was not great enough to be consider significant. However we did find a significant difference in heart rate between before and after caffeine was added. Referring to our initial question, the heart rate of Daphnia was increased when caffeine was present but altering the concentration did not have a great enough influence. Our data was not statistically significant but could be used to explore other experiments or studies testing caffeine and heart muscle interactions.

On trial 4 of the .005M caffeine, the Daphnia gave birth during the 7 minute waiting period which may have influenced its heart rate. During the recording the Daphnia still had an offspring inside trying to get out, which could have also influenced heart rate. If this experiment was to be performed in the future, we would recommend making sure the Daphnia was not in a state of labor or activity that could have altered its heart rate or state of being. Aside from this experiment, the results from this trial could be used in other experiments testing the effects of chemicals on labor.

Further exploration of this subject is recommended because caffeine is a prominent factor in today's society and requires further examination. Society as a whole digests and consumes caffeine in many forms with little education on the chemical. Caffeine can have other medical influences other than simply increasing heart rate. Many people could be unaware that they possess a caffeine sensitivity and digest too much. In a case study which examined the influence of caffeine intake on the heart, it was discovered by his doctor that a 33 year old man with a heart arrhythmia was digesting several various forms of caffeine. When the caffeine intake was reduced, his arrhythmia became much less severe (Kinugawa, et al., 2011). Another cause for further exploration of this topic is the wide variety of experiment results. A similar study that tested the effects of both stimulants and depressants came to the results that caffeine had no influence on heart rate. (Corotto, et al., 2010). The wide array of results suggests that the topic has inconsistent results and requires further testing.

Literature Cited

Corotto, F., Ceballos, D., Lee, A., & Vinson, L. (2010). Making the Most of the Daphnia Heart Rate Lab: Optimizing the Use of Ethanol, Nicotine & Caffeine. *American Biology Teacher (National Association of Biology Teachers)*, *72*(3), 176–179.

Ebert, D. (2005). Chapter 2, Introduction to Daphnia Biology. In Ecology, Epidemiology, and Evolution of Parasitism in Daphnia [Internet]. National Center for Biotechnology Information (US). Bethesda, MD.

Green, P., Suls, J. (1996). The Effect of Caffeine and Ambulatory Blood Pressure, Heart Rate, and Mood in Coffee Drinkers. Journal of Behavioral Discussion. 19(2): 111-128

Hammer & Harper. (2013). PAST (3.2) [Computer software]. Oslo, Norway: https://folk.uio.no/ohammer/past

Kinugawa,T., Kurita, T., Nohara, R., Smith, M. (2011). A Case of Atrial Tachycardia Sensitive to Increased Caffeine Intake. National Center for Biotechnology Information. 52(6): 398-400

Powell, K., Iuvone, P., Holtzman, S. (2001). The role of dopamine in the locomotor stimulant effects and tolerance to these effects of caffeine. Pharmacology Biochemistry and Behavior. 69 (1-2): 59-70

Scott. (2015). Three Ways That Caffeine Affects Your Brain. https://driftaway.coffee/brain/ (accessed 10/29/18).

Sondermeijer, H., J van Marle, A., Kamen, P., Krum, H. (2002). Acute effects of caffeine on heart rate variability. The American Journal of Cardiology. 90(8): 906-907