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Chemical control: observing the excitatory effects of MSG compared to the inhibitory effects of KCl in the heart rate of *Daphnia magna*

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Abstract

From tiny *Daphnia magna* to humans and other mammals, heart rate is a quantitative characteristic for several species. In this study, we measured the effect of two different chemicals on the heart rate of *Daphnia* by comparing the heart rate of the Daphnia before and after the addition of each chemical. Our experimental design was unique to our goal by comparing the effects of a KCl solution and an MSG solution to *Daphnia* in a normal environment without either chemical to see the average percent change in heart rate that was caused by the chemical. *Daphnia* heart rate was measured under a light microscope and recorded with an iPhone in slow motion to accurately determine the number of beats per minute. Once averaged, the results indicated that there was a 16.76% increase in heart rate due to the addition of MSG and a 34.51% decrease when put in a KCl solution. Our results may be beneficial to those wishing to study the effects of other excitatory or inhibitory chemicals on subjects such as humans, as a strong correlation has been made between the change in heart rate for both humans and *Daphnia*.

Introduction

The human heart is a central organ that is interconnected to other parts of the body to relay messages and receive action potentials from the nervous system. The heartbeats are controlled by specialized cardiac cells that carry electrical signals to or from the nervous system, according to the frequency of the action potential ("Electrical System of the Heart - Topic Overview," n.d.). The cardiac ganglion is stimulated in order to contract the heart, followed by a chain reaction of passing electrical signals through the body until they reach the heart (Baptista & Kirby, 1997). These action potentials are regulated by sodium-potassium pumps that line the axons carrying the signals, which allow sodium ions to rush into the cell and potassium ions to flow out as an action potential passes through (Hoefnagels, 2017). In humans, MSG is a known neuroexcitatory amino acid, found in many foods, that degrades the taurine found naturally in the heart (Schaffer et al., 2010). Taurine is an essential amino acid that has a heavy influence on cardiac function. When taurine is degraded by excessive MSG, the heart rate increases (Schaffer et al., 2010). Additionally, increased levels of K+ cause an imbalance in the sodium-potassium pump, which impacts the rate of action potential occurrence (Yamasaki & Narahashi, 1959). As a result, the addition of KCl decreases heart rate.

Daphnia Magna, on the other hand, possess a globular myogenic heart, which operates independently of neural, metabolic, or hormonal stimulation (Kaas, 2009). Rhythms in this type of heart are initiated directly in the cardiac tissue. Instead of a heartbeat regulated in relationship to other organ systems, the Daphnia heart acts almost completely alone to regulate heartbeat. The daphnia heart beats about two-hundred times per minute (Ebert, 2005), which is excessive when compared to a human heart, which beats 60-100 times per minute ("Electrical System of the Heart - Topic Overview," n.d.)

The distinction between the human and *Daphnia* cardiac systems lead us to question if we can observe the same trends observed in humans exposed to potassium chloride (KCl) and MSG in *Daphnia*. The vagus nerve will play a crucial role in heartbeat regulation as it is a main component of the parasympathetic nervous system in both humans and *Daphnia* (Breit et al., 2018). This is why *Daphnia* serve as a good comparison to the human heart because

despite their differing anatomy, they both share a main nerve in regulating action potential driven heart rhythms. We hypothesize that KCl will decrease the heart rate of Daphnia while MSG will increase it, as it does in humans, because both Daphnia and humans have a vagus nerve to regulate their heartbeat. This hypothesis will be supported if the Daphnia exposed to KCl have a lower average heart rate than those in water and if Daphnia exposed to MSG have a higher average heart rate than those in water. Inverse results in which KCl leads to a higher heart rate and MSG leads to a lower heart rate or the lack of a correlation between the chemicals and heart rate would not support the hypothesis.

Method

In this experiment, potassium chloride (KCl) and monosodium glutamate (MSG) solutions were tested on *Daphnia* to observe the effects they had on heart rate. Each daphnia was observed in pure aquarium water before exposing them to the KCl and MSG, so the water-only control group would be directly compared to the change induced by the specific chemical used. The amount of KCl and MSG used each time did not change, nor did the concentration.

For each trial, one *Daphnia* was randomly selected from the aquarium to be observed under a light microscope. The *Daphnia* was placed on a microscope slide and recorded (by an iPhone) for 10 seconds in aquarium water only. A very small piece of cotton ball was added to the microscope slides in order to keep the *Daphnia* in place and allowed us to easily see the heart moving. After this, the same *Daphnia* was exposed to either KCl or MSG and left for 7 minutes (Shaw & French, 2018). A 0.5% potassium chloride solution and a 1% monosodium glutamate solution were used. Another video was taken, and the heart rate was recorded after the specimen was exposed to the chemical. The videos were taken in slow motion and played back to accurately calculate the heart rate. The number of beats counted during the 10-second interval (before and after the experimental chemical was added) was multiplied by six to get a heart rate in beats per minute. This process was repeated 3 times for each chemical.

Data was recorded in beats per minute and averaged between all the trials. This data was then displayed in a box and whisker plot for easy visual comparison between heart rates for each chemical. Finally, because the control and experimental groups were tested on the same subject, a paired t-test for each chemical was used to determine the statistical significance of the experiment.

Results

The average percent change of Daphnia heart rate shows the experimental group of Daphnia that was introduced to MSG had an increase in heart rate, which was 16.76% (Fig 1). The group of Daphnia introduced to KCl showed a decrease in heart rate, which was reported as 34.51%. A Paired t-Test was conducted to compare the effect of MSG on Daphnia heart rate, tested before and after MSG introduction. There was a significant difference between the two conditions; t(df)=2, p=0.01105109. An additional Paired t-Test was conducted to compare the effect of KCl on Daphnia heart rate, tested before and after KCl introduction. There was a significant difference between the two conditions; t(df)=2, p=0.045522349.



Fig 1. The results of heart rate change from KCl and MSG. On average, heart rate increased by 16.76% when the *Daphnia* were in the MSG, while KCl caused an average of a 34.51% decrease in heart rate. Both numbers were compared to the average heart rate of Daphnia in pure aquarium water.

Discussion

As hypothesized, adding KCl to the water decreased the heart rate of the Daphnia, while adding the MSG solution slowly increased the heart rate. According to our data results, our hypothesis was supported, and it was seen that both KCl and MSG affect *Daphnia* the same way that they affect humans. This trend can be attributed to the fact that both humans and Daphnia have a vagus nerve, which is plays a very important role in regulating heartbeat in both species. Although human hearts work together with other systems in our body, Daphnia heart rate increased and decreased similarly despite having a heart that is connected to their central nervous system. The 16.76% increase in heart rate and 34.51% decrease in heart rate shows us that even without a Daphnia heart rate being regulated by sodium-potassium pumps, substances like MSG and KCl still drastically change heart rate through affecting action potential.

However, like any species there is variation in the population and there were some outliers seen in our data. The population of Daphnia that were used for this experiment were all female and some were pregnant, some younger than others, some more active than others. Because of this variation, some of the Daphnia reacted to the chemicals differently. The younger Daphnia were already more active than the older Daphnia, and they were also affected by KCl and MSG more than the older Daphnia. Daphnia that were smaller and younger had base heart rates of 516 bpm and 576 bpm, compared to 474 bpm and 396 bpm seen in the older, larger Daphnia. Just

like *Daphnia*, there is extreme variation in the human population and not every human's heart rate and action potential frequency will be affected the same way by different chemicals such as KCl, MSG, nicotine, ethanol, etc.

There are plenty more studies to be done after seeing the results of the newfound Daphnia heart rate data. Because there is a correlation between KCl and MSG affecting heart rate, other compounds known for their excitatory or inhibitory effects could be studied in humans to see how they affect human heart rate. Different diets may change sodium intake and replace it with potassium, and an overconsumption of food with MSG may cause health affects which are currently undiscovered (van Buren et al., 2016). Further studies could look into the long-term heart rates of individuals that intake such chemicals; studying heart rate can act as appropriate measurement for biological changes from the chemicals due to their ions and molecules interacting with the nervous system of the body, whether it be Daphnia or humans. Further research could also investigate the effects of other chemicals such as ethanol (in drinking alcohol) or nicotine from a cigarette addiction. These compounds also have excitatory/inhibitory properties but do not directly affect the body with potassium and sodium ions, as the ones tested in this experiment did.

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