Effects of Caffeine Consumption on Physiological Markers of Stress

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Abstract

Caffeine is known to be used by college students to help stay awake, focus and increase academic performance. Caffeine's biological mechanisms are fairly well-understood, but its effect on a stress response should be better explored. The purpose of this study is to examine the effect of caffeine on key physiological indicators of stress- blood pressure (BP), heart rate (HR), and electrodermal activity (EDA)- in healthy college students. Forty-five students participated in this experiment, which began with baseline measurements of the three variables mentioned, followed by a stressor in the form of a brief math test, and a retesting of those three variables. The participants also completed a brief questionnaire to determine their caffeine usage, and were sorted into different groups based on their caffeine consumption. Statistically significant interactions could not be shown for most of the variables; however, there was a significant difference in the change of systolic blood pressure in participants when divided based on their intake of caffeine on the day of the experiment. Participants who consumed very high levels of caffeine (286mg and above) on the day of the experiment had a greater increase in systolic blood pressure than those who had less caffeine. High caffeine consumption is therefore correlated with a larger increase in systolic blood pressure in response to stress.

Introduction

Caffeine is a psychoactive substance many people use on a regular basis. In the United States, estimates show that more than 85% of adults consume caffeine regularly with a daily intake of 180mg per day, or roughly two cups of coffee (Temple, 2017). Caffeine is found in many beverages and foods, with varying levels of caffeine content. Coffee is the most commonly consumed caffeinated beverage and in many studies was the primary source of caffeine (Mahoney, 2018, Mitchell, 2013). Other sources of caffeine include: teas, soft drinks, energy drinks, supplements, and chocolate. The abundant sources of caffeine marketed in America allow for readily available consumption. Many new caffeine products are specifically targeted toward adolescents and young adults (Temple, 2017). With this in mind, college students may use caffeine to help cope with stress and increase academic performance.

Considering the physiological effects caffeine exerts on the body, a point of interest we considered was why college students use caffeine during stressful periods, such as in preparation for a difficult exam or project. According to one study, 74.9% of college students drink caffeine (Bucher, 2019). A likely reason why is for the stimulating effects that offer help with cognitively demanding tasks that college students are constantly exposed to. One study found that college students reported taking caffeine to stimulate mood and performance,

specifically to feel more awake and improve concentration (Mahoney, 2018). However, the perceived positive benefits of caffeine are achieved only at low to moderate intake doses, up to 400mg per day (Bucher, 2019). If college students consume excess amounts of caffeine (>400mg/day), negative consequences start to outweigh the benefits. Excess caffeine consumption has been associated with anxiety, headaches, nausea, and restlessness as well as unpleasant withdrawal symptoms after habitual use is discontinued (Mitchell, 2013). In Bucher's study, a reinforcement pattern was observed, that when students experience inadequate sleep, they tend to use caffeine despite knowing the associated risks, such as impaired daytime function.

Despite the known risks of caffeine consumption, we were interested in understanding why students continue to use caffeine in academic settings. A probable reason is due to the perceived benefits exerted by peers during exam periods. Evidence for this behavior is shown by students' willingness to take caffeine pills for neuroenhancement (NE), non-medical use of a psychoactive substance to enhance psychological function (Brand & Koch, 2016). Although this study is limited to caffeine pills, the reinforced benefits of caffeine by peers cannot be dismissed because of potential negative consequences. Research is limited on the association between caffeine and stress in college students. Therefore, a greater understanding of how caffeine and stress are associated is needed in order to assess positive and negative physiological effects in college students. Upon exploration of potential negative effects of excess caffeine consumption, education could then be provided to students for better study tools.

Biologically, caffeine works by antagonizing adenosine receptors (Cappelletti, 2015). This mechanism occurs through the central and peripheral nervous systems and exerts effects on cardiovascular organs, such as the heart and blood vessels (Temple, 2017). Adenosine is a neuromodulator that promotes sleep and suppresses arousal. When caffeine is ingested, adenosine can no longer bind to its receptor, therefore eliciting wakefulness. Caffeine regulates the cardiovascular system by increasing levels of intracellular calcium concentrations, releasing norepinephrine, and sensitization of dopamine receptors (Temple, 2017). An observed effect of this mechanism is vasoconstriction of blood vessels. Increase in systolic blood pressure (BP) resulting from

vasoconstriction was observed to be associated with self-reported stress in both men and women after caffeine ingestion (Bennett, 2013). Therefore, we were interested to observe how these results compare in a college student population.

In addition to caffeine's effect on BP, we wanted to observe the effect on heart rate (HR). However, findings in the literature are inconclusive regarding caffeine's effect on HR due to the dependent nature of experimental conditions. Although there is ambiguity in the literature, we hope to see a pattern emerge that caffeine consumers will have a greater HR response to an induced stress situation than non-caffeine consumers.

Another physiological parameter we were interested to observe was electrodermal (EDA) response. Davidson and Smith mention consistent findings of increased EDA after caffeine consumption. Their research findings were consistent with a prediction that caffeine increases arousal and enhances the arousing effects of situational or psychological agents (Davidson & Smith, 1991). The psychosituational agent relevant to our study is stress. Davidson and Smith reference a previously observed synergistic effect between caffeine and stress to increase both BP and forearm blood flow. Therefore, we predict caffeine increases EDA and coupled with stress will result in increased BP. We additionally have to consider our participants are college students and Davidson and Smith did not specify participant demographics regarding the synergistic effect of caffeine and stress.

The purpose of this study was to examine the effects of caffeine on the change in BP, HR, and EDA in healthy college students given induced stressors. A further distinction we aim to address is the difference in these measurements between habitual caffeine users, non-habitual caffeine users and non-caffeine users. We predicted for all individuals that the measurements (BP, HR, EDA) would have a greater response than baseline measurements and remain elevated during the post-stress period, and caffeine users would have significantly increased biomarkers compared to non-caffeine users both at baseline and post-stress. We hope this experiment will provide an accurate representation of how college students' caffeine consumption relates to their physiological response during a stressful situation.

Materials

In order to measure physiological differences in stress responses, heart rate (HR), blood pressure (BP), and electrodermal activity (EDA) were examined using two different measurement devices. A Series 10 Blood Pressure Monitor (Model Number: BP785N, Serial Number: 20150310128LG, Omron Healthcare, Inc., Lake Forest, IL, USA) was used to measure systolic blood pressure and diastolic blood pressure in millimeters of mercury (mmHg), and heart rate in beats per minute (bpm). BSL EDA finger electrodes with Xdcr leads (Model Number: SS3LA, Serial Number: 1709004276, Biopac Systems, Inc., Goleta, CA, USA) in conjunction with Isotonic Recording Electrode Gel (Model Number: GEL101, Biopac Systems, Inc., Goleta, CA, USA) was used to measure electrodermal activity in microsiemens (μS). The Biopac Systems, Inc. Student Manual (Biopac Systems Inc. ISO 9001: 2008) was used to learn correct equipment usage. Electrodermal activity was measured and analyzed using Biopac Student Lab System (BSL 4 Software, MP 36). A singular Dell computer from the laboratory room was used for the collection and analysis of electrodermal activity data within the BSL 4 application. A three page math test was developed by the experimenters, using Microsoft Word (Microsoft, 2017), as a stressful stimulus. Each page of this math test contained three lines of multiplication problems containing 10 problems each, for a total of 90 multiplication problems. These problems were selected at random, with each term kept between zero and twelve. To increase the difficulty of the math test, a math-based logic question was inserted between each line of multiplication problems, for a total of 9 logic questions. This stimuli was presented to the participants on three separate pieces of 8.5- by 11-inch paper. Microsoft Word (Microsoft, 2017) was also used for the development of the Post-Experiment Questionnaire. The Post-Experiment Questionnaire was one page in length, and was used to collect each participant's age, sex, weight, and caffeine consumption patterns. Questions regarding caffeine consumption were used to sort the participants into groups corresponding to the amount of caffeine they consumed.

Methods

Participants

Forty-five participants were recruited voluntarily from a pool of students enrolled in Physiology 435 during the spring semester of 2019 at the University of Wisconsin-Madison. There were 17 male and 28 female participants, with ages ranging from 20 to 28 years of age (Mean = 21.68, SD = 1.26). All experimental procedures and collection of data took place in the UW-Madison Medical Sciences Center. Each participant was required to read and sign a consent form stating the confidentiality precautions taken and the possible risks associated with participating. The consent form was completed prior to the beginning of the experiment.

Procedure

All voluntarily recruited participants from Physiology 435 that agreed to sign the consent form were accepted into the study. Before beginning the experiment the BSL 4 software was opened on the computer, and the BSL EDA finger electrodes with an Xdcr lead was connected to channel 1 of the Biopac Student Lab System. The computer was faced away from the participant at all times. To measure electrodermal activity, "Electrodermal activity" (EDA, SS3L, SS3LA, SS57L, 0-35 Hz) was selected. EDA was consistently measured in microsiemens (μS). Isotonic Recording Electrode Gel was applied to the BSL EDA Finger Electrodes, which were subsequently secured to the participant's index and middle fingers on their non-dominant hand. The BSL EDA Finger Electrodes were calibrated for each participant, following the Biopac Systems Inc. Student Manual. The Series 10 Blood Pressure Monitor was plugged into an outlet and strapped to the participant's non-dominant arm, three inches above the elbow. The Series 10 Blood Pressure Monitor was then turned on, and required no calibration. The Series 10 Blood Pressure Monitor was also faced away from the participant at all times. Two experimenters conducted each experiment and remained in the testing room through the duration of the experiment to record the data, and provide instructions to the participant. The experiments were all conducted in a quiet, private room to eliminate distractions and confounding variables. For this same reason, the participant sat facing away from the experimenters for the duration of the experiment.

The participant's baseline heart rate, blood pressure, and electrodermal activity were recorded over a one minute period. The experimenters recorded the blood pressure and heart rate readings by hand, and saved the BSL EDA data to a private drive for later analysis. The participant was given a pencil and the three-page multiplication task upside down, and were told not to flip it over until instructed to do so by the experimenters. The experimenters explained to the participant that they were to take a brief and simple math test, and that it was critical that they finish each problem on the test for their data to be viable. These instructions, in conjunction with an unreasonable time limit of five minutes, were utilized to elicit a stress response from the participant. The participant was instructed to begin the test, and the experimenters started a timer as the participant flipped the page. A warning every thirty seconds was given verbally to the participant during the task. When the five minutes had passed the experimenters instructed the participant to place their pencil down, and place their test to the side. The experimenters waited thirty seconds after the completion of the multiplication task and recorded the participant's blood pressure, heart rate, and electrodermal activity over a one minute period. Again, blood pressure and heart rate were recorded manually from the Blood Pressure Monitor's display, and the Biopac BSL EDA data was saved to a private drive. Experimenters took the Blood Pressure Monitor and the BSL EDA Finger Electrodes off of the participant, and allowed the participant to clean the Isotonic Recording Electrode Gel off of their own hands using a hand sanitizing wipe. The participant were given the Post-Experiment Questionnaire and were instructed to answer each question to the best of their ability. After completing the Post-Experiment Questionnaire the participant was thanked for their time, and was instructed to leave. For a graphical depiction of this experimental timeline, see Figure 1.

Data Analysis

Each participant's blood pressure (mmHg), heart rate (bpm), and average EDA (μS) were collected both before and after the multiplication task. Self reported caffeine consumption (mg) was collected during the Post-Experiment Questionnaire. The measurements recorded prior to the multiplication task served as the baseline BP, HR, and EDA. Each participant's change from baseline to experimental readings was calculated

and recorded. This data was compiled into a single Microsoft Excel spreadsheet (Microsoft, 2017) for data analysis. The Blood Pressure Monitor was used to record both heart rate, systolic blood pressure, and diastolic blood pressure directly following the one minute collection of EDA data. Average EDA was determined by use of a one minute sample and the "mean" tool within the Biopac software. In order to determine any differences between groups, and using the answers to the exit survey, four separate statistical analyses were done, based on: average daily caffeine intake, caffeine intake on the day of participation, whether participants overall did or did not consume caffeine, and if the participant consumed caffeine within four to six hours prior to their participation in the study. The former two tests grouped participants based upon which "level" of caffeine consumption they indicated, from 0-5, each level indicating approximately a cup of coffee worth of caffeine. The latter two grouped participants into two categories, a simple yes or no. In each analysis category, an analysis of variance (ANOVA) test was ran on the participants' change from baseline to post-test readings for heart rate, systolic blood pressure, diastolic blood pressure, and electrodermal activity. The final result of this was sixteen separate ANOVA tests, which provide both an F statistic and a P-value. ANOVA results are found to be statistically significant if the F value is greater than the F-critical value determined by the test. Additionally, a P-value of less than 0.05 is generally regarded as statistically significant.

Positive Controls

Positive control tests were run on each of the four experimenters in order to demonstrate that the Biopac equipment was functioning properly. Baseline measurements for BP, HR, and EDA were taken over a one minute period. These baseline averages were 102.67 ± 22.19 mmHg for systolic blood pressure, 69.00 ± 12.00 mmHg for diastolic blood pressure, 70.00 ± 10.44 bpm for heart rate, and 7.15 ± 2.26 μS for electrodermal activity (n= 4). The experimenters recorded the same measurements following a stressful five minute multiplication task. These post-stress averages were 105.67 ± 20.74 mmHg for systolic blood pressure, 71.00 ± 12.77 mmHg for diastolic blood pressure, 72.67 ± 10.07 bpm for heart rate, and 7.24 ± 1.65 μS for electrodermal activity (n= 4). Each variable measurement was elevated after exposure to a stressful stimulus.

This demonstrated the functionality of all equipment and the experimenter's ability to accurately collect the desired measurements.

Negative Controls

One experimenter volunteered to wear the BSL EDA Finger Electrodes and the Blood Pressure Monitor for five minutes (the length of the experiment) to collect a negative control. The experimenter did not complete the multiplication task. The data for electrodermal activity was recorded in the Biopac system, and the blood pressure and heart rate were collected from the Blood Pressure Monitor. This demonstrated that while no experimental manipulation was occuring, the Biopac system and the Blood Pressure Monitor were accurately measuring and recording the physiological parameters required for the experiment.

Results

For this experiment, F values were used in order to determine statistical significance for all parameters. F values greater than F-critical as determined by ANOVA are accepted as statistically significant. As there are thirty-two total F values, insignificant F values are recorded only in Table 1 in an effort to remain concise. There were a few data points that were not used for data analysis to be noted. First, when participants were grouped by caffeine intake on the day of the study, only one participant responded to consume caffeine in level three, and one in level five. ANOVA requires a mean for each level, and as such one data point was insufficient and went unused. Also to note, extraneous data points were removed from two participants that exhibited an extreme drop of 42 mmHG from baseline to post-stress systolic and 40 mmHG diastolic, which were likely due to instrumental error. For analysis by average daily and day-of caffeine consumption, participants were placed in levels, each level corresponding to approximately one cup of coffee worth of caffeine, or ninety-five mg.

There was found to be no statistical significance in changes in heart rate, electrodermal activity, or diastolic blood pressure for participants from baseline to post-stress when grouped by average daily intake, intake on the day of the trial, if they consumed caffeine in general, or if they consumed caffeine within four to six hours before the trial.

No statistical significance was found in the change in systolic blood pressure under groupings by if the participants consumed caffeine overall, if they had consumed caffeine in the four to six hours prior, or by average daily intake. However, there was a significant difference in the change in systolic blood pressure of participants when grouped by their intake on the day of the trial. Average changes when grouped by consumption on the trial date (from levels 0, 1, 2, and 4 respectively) are: -2.720 (variance 49.62), -3.000 (variance 7.00), -8.250 (variance 157.07) and 5.2857 (variance 11.90). The F-value was 3.7792, with an F-critical value of 2.8450.

After running analysis on all of these data points from pre to post stress, it was thought that using percent change rather than raw change may give a different result. ANOVA was re-ran using percent changes, but the results were found to be the same.

Discussion

As discussed earlier in this paper, there is evidence in scientific literature that caffeine consumption can cause increased electrodermal activity and systolic blood pressure. Our study aimed to determine whether it would also cause greater changes in these variables, as well as diastolic blood pressure and heart rate, under stressed conditions. This information could then be used to advise on potential issues college students may encounter when consuming caffeine before stressful events, such as exams.

It was hypothesized that caffeine consumers would exhibit more significant changes in the physiological markers of heart rate, systolic and diastolic blood pressure, and electrodermal activity than non-consumers.

Heart Rate

In all grouping schemes, change in heart rate after a stressful stimulus was not found to be significantly different between varying levels of caffeine consumption. This result is contrary to our hypothesis. As seen in Figure 2, changes in heart rate between day-of caffeine consumption categories are generally similar, and the means follow no trend. Additionally, when grouped by caffeine consumers versus non-consumers, distribution and means are very similar (Figure 3).

Systolic Blood Pressure

When grouped by average daily intake, whether they consume caffeine overall, and if they had consumed caffeine in the last four to six hours, participants showed no difference in change in systolic blood pressure. Figure 5 shows an example of this, with similar sample distributions and means. These results are in opposition to our original hypothesis.

However, one statistically significant difference across groups was found between systolic blood pressures. It was found that when grouped by caffeine intake on the day of the trial, participants who consumed very high levels of caffeine (four cups of coffee worth) had a greater increase in systolic blood pressure than other levels of consumption. This result was found to be statistically significant by ANOVA test (F>F-critical). This elevated change in systolic blood pressure can be seen in Figure 4, and is in agreeance with our hypothesis.

Diastolic Blood Pressure

Diastolic blood pressure was also found to change equally between groupings across all analysis schemes, contrary to our original hypothesis. Figure 6 shows there is no general trend between means or data ranges when grouped by day-of caffeine consumption, and similarly in Figure 7 there is no trend between overall caffeine consumers and non-consumers.

Electrodermal Activity

As with heart rate and diastolic blood pressure, EDA change in response to a stressful stimuli was not found to vary between groups by any analysis variable. This is contrary to our hypothesis. Figure 8 and Figure 9 show that there is no discernable trend between either day-of caffeine consumption or consumption versus non-consumption and change in EDA respectively.

Limitations

The largest limitation associated with this study is the reliability of the caffeine dosage information. Caffeine consumption was recorded on a self-reporting basis from the administered survey, which could be unreliable due to a number of factors: inaccurate memory or estimation, poor addition, imprecise caffeine

content (such as brewed coffee), et cetera. It is also possible that participants did not find the test they were administered to be stressful enough to elicit a stress response. This however can be refuted by the EDA skin conductance graph in Figure 10, which shows a dramatic increase in EDA at the moment the subject is informed they will be taking a test. This is strong evidence the subject is mounting a physiological response to the stressor. Another limitation inherent in this study is the small sample size made up only of UW-Madison students.

Future Research

Further study in this area of research could be done using caffeine dosing rather than a self-reporting survey. This would allow for precise measurements regarding caffeine intake as well as timescales. A larger sample size of participants from different age categories and demographics would also be beneficial. Due to our one significant result coming from high dosage caffeine on the day of the trial coupled with change in systolic blood pressure, more research should be done in that avenue. A similar study could be done with two participant groups - one with no caffeine dosage, on with a high caffeine dosage (4+ cups of coffee worth). These groups could be exposed to a stressful stimulus, and biomarkers measured to see if the caffeinated group exhibits a more dramatic physiological response than non-caffeinated. This would provide further insight into whether students who consume large amounts of caffeine show heightened physiological responses to stressors such as tests than those who do not.

Conclusions

Based on the results of this study, it can be concluded that the change in heart rate, blood pressure, and electrodermal activity in response to a stressor is no different between those who do and do not consume caffeine. The same can be said for those who do and do not consume caffeine four to six hours before being exposed to a stressor, and between those who consume caffeine at varying levels on an average daily basis. People who consume caffeine in varying levels on the same day as receiving a stressor have the same response in regards to heart rate, diastolic blood pressure, and EDA. However, it can be said that there is a greater

increase in systolic blood pressure in response to a stressor for persons who consume large amounts of caffeine on that same day than those who do not. This shows that students who consume caffeine in large quantities before a stressor such as an exam will have a greater rise in their systolic blood pressure than those who do not. More specific research should be conducted into this physiological response as outlined above.

Relevance

This research is relevant to Physiology 435 curriculum in that it deals with physiological responses to stressors. Specifically, it looks at cardiovascular regulation and temperature regulation in the form of sweat, increasing EDA. This research applies information on the sympathetic and parasympathetic nervous systems, particularly in how it is utilized in response to stress. Caffeine, which affects adenosine receptors, acts as a parasympathetic nervous system antagonist, an extension of topics covered in unit one of Physiology 435. We have learned about these topics already, so this research helps us to understand how exogenous substances affect the regulation of homeostasis. This research is directly applicable to our peers that drink caffeine during stressful events in school.

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Tables

Table 1.

Grouping	**F-Values by Physiological Response Tested (F/F-critical)**			
	Heart Rate	Systolic BP	Diastolic BP	Skin Conductance (EDA)
By Average Daily Intake	0.9966/2.4558	1.9870/2.4625	0.0823/2.4558	1.0730/2.4558
By Today's Intake	0.7993/2.8450	3.7792/2.8450	2.3485/2.8450	0.9124/2.8450
By Overall Consumption (Y/N)	0.6263/4.0670	0.1755/4.0670	0.3035/4.0670	2.8420/4.0670
By Consumption in Last Six Hours (Y/N)	0.0457/4.0670	0.1198/4.0670	3.4100/4.0726	0.2545/4.0670

Table 1: ANOVA test F-value results. Statistically significant results are highlighted.

Figures and Legends:

Figure 1. Chronological outline of experimental design

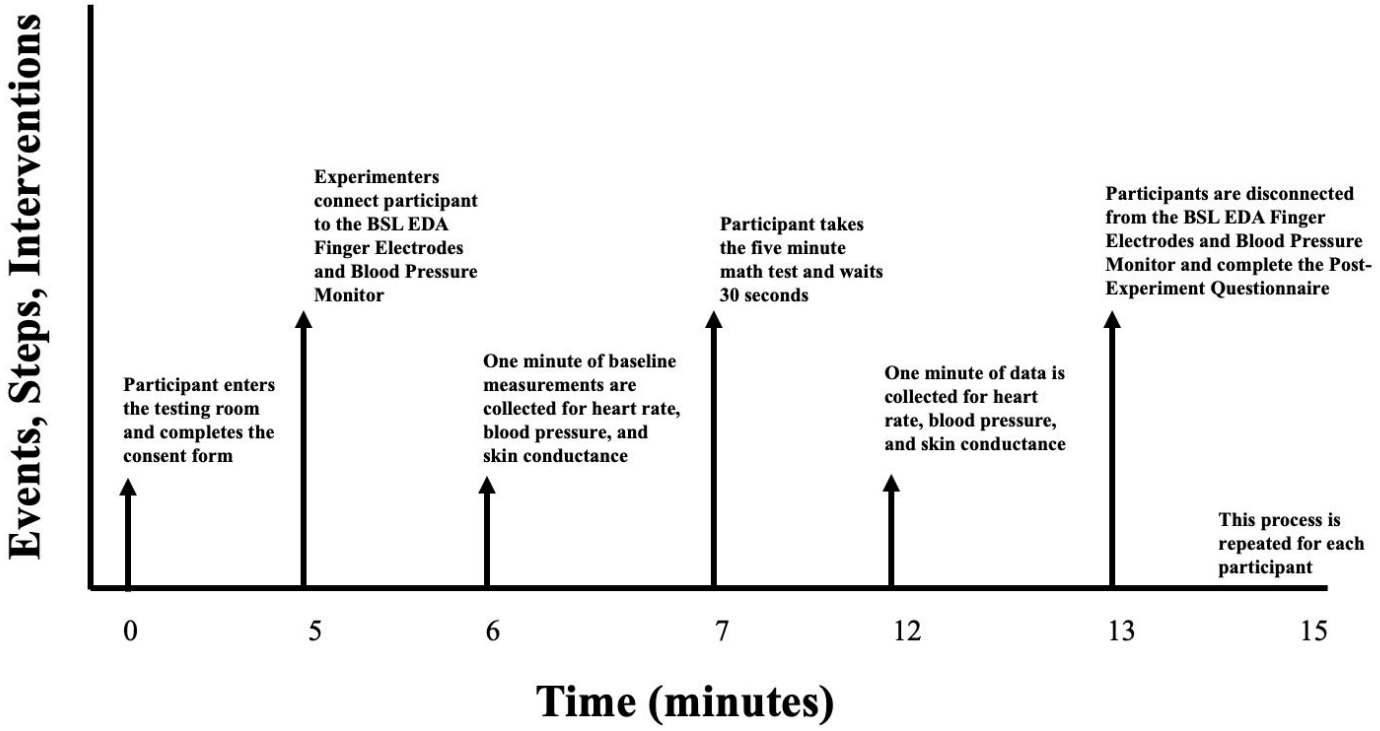

Figure 1: Describes procedure for the study. The total time for each participant was around 15 minutes. During the positive control trials, those participants only did the baseline measurements before being disconnected from equipment.

Figure 2.

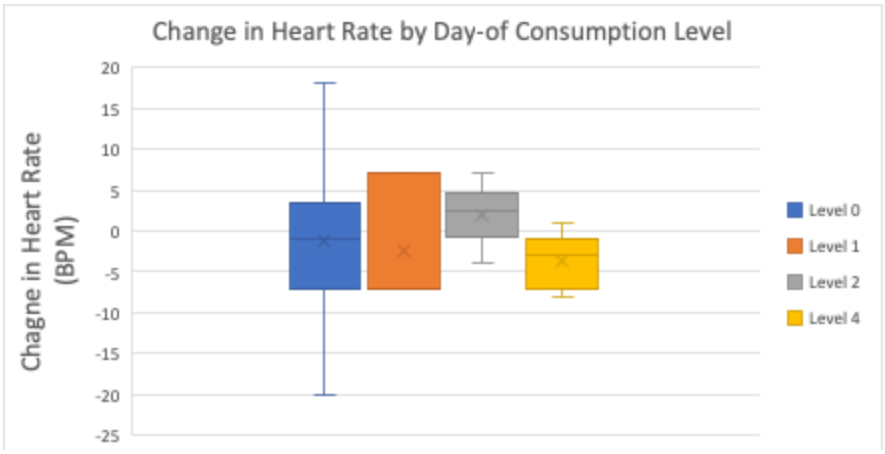

Figure 2: A box and whisker plot showing the mean and distribution from each of the significant treatment groups' heart rate data. No statistically significant difference was found between any of these groups. (Groups 3 and 5 did not have enough participants to do valid statistical analysis.)

Figure 3.

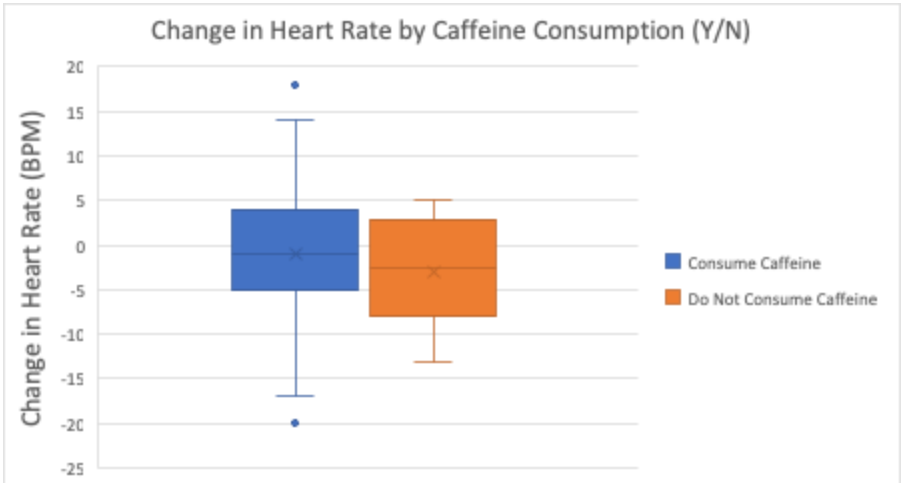

Figure 3: A box and whisker plot showing the mean and distribution from each of the treatment groups' change in heart rate data. No statistically significant difference was found between these groups.

Figure 4.

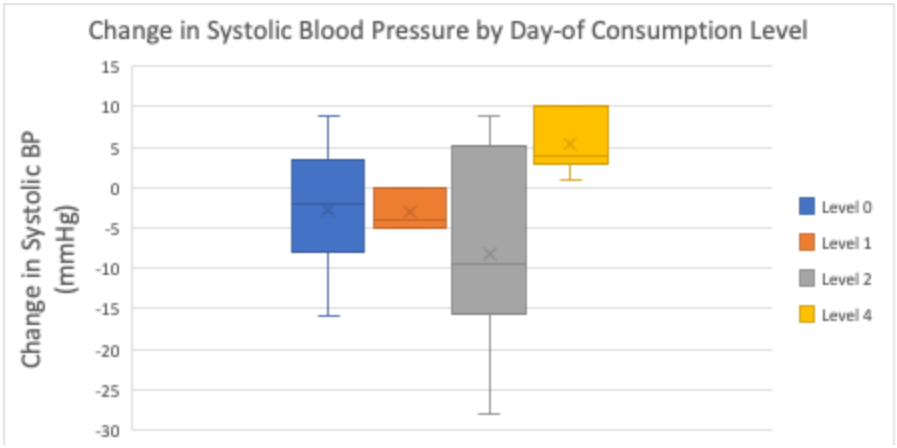

Figure 4: A box and whisker plot showing the mean and distribution from each of the treatment groups' change in systolic blood pressure data. Group 4 shows statistical significance. (Groups 3 and 5 did not have enough participants to do valid statistical analysis.)

Figure 5.

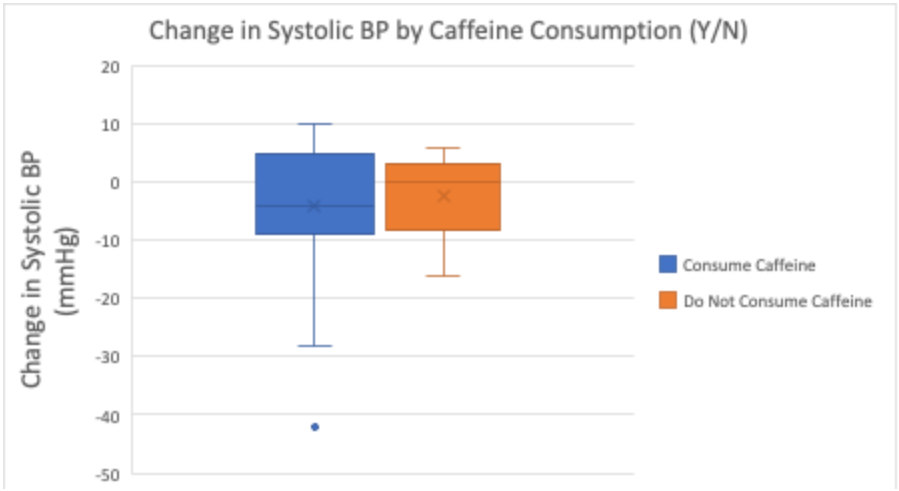

Figure 5: A box and whisker plot showing the mean and distribution from each of the treatment groups' change in systolic blood pressure data. No statistically significant difference was found between these groups.

Figure 6.

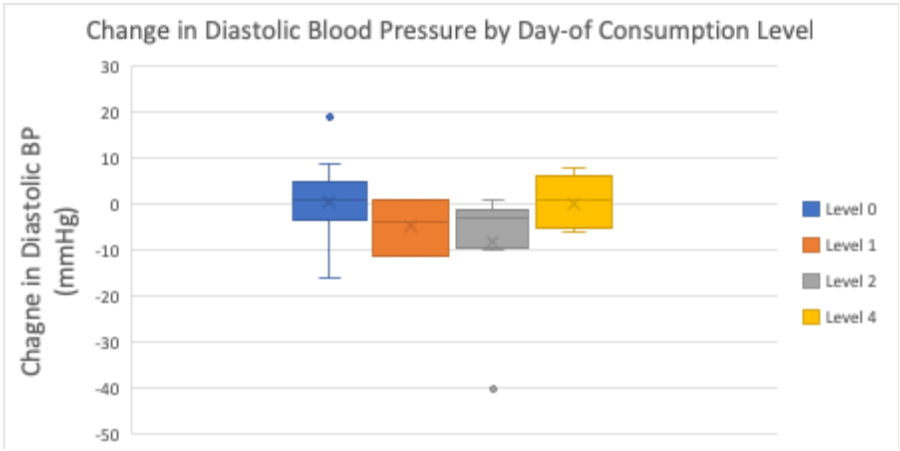

Figure 6: A box and whisker plot showing the mean and distribution from each of the treatment groups' change in diastolic blood pressure data. No statistically significant difference was found between any of these groups. (Groups 3 and 5 did not have enough participants to do valid statistical analysis.)

Figure 7.

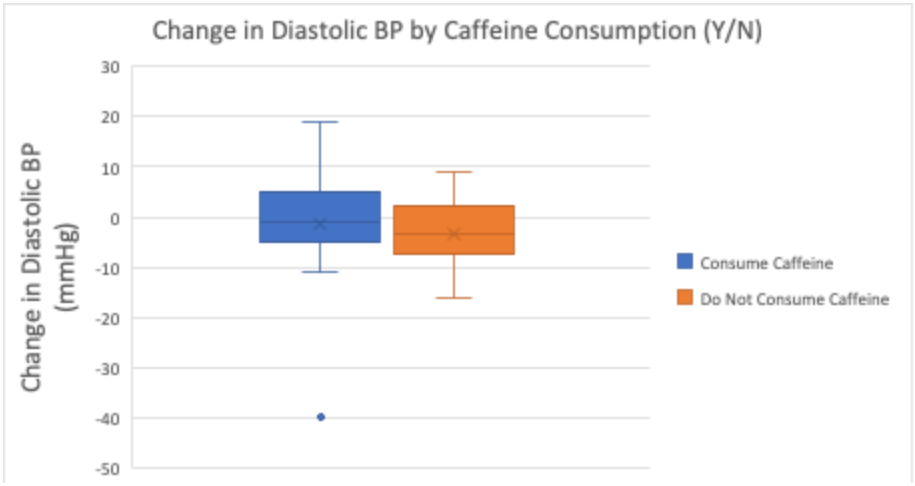

Figure 7: A box and whisker plot showing the mean and distribution from each of the treatment groups' change in diastolic blood pressure data. No statistically significant difference was found between these groups.

Figure 8.

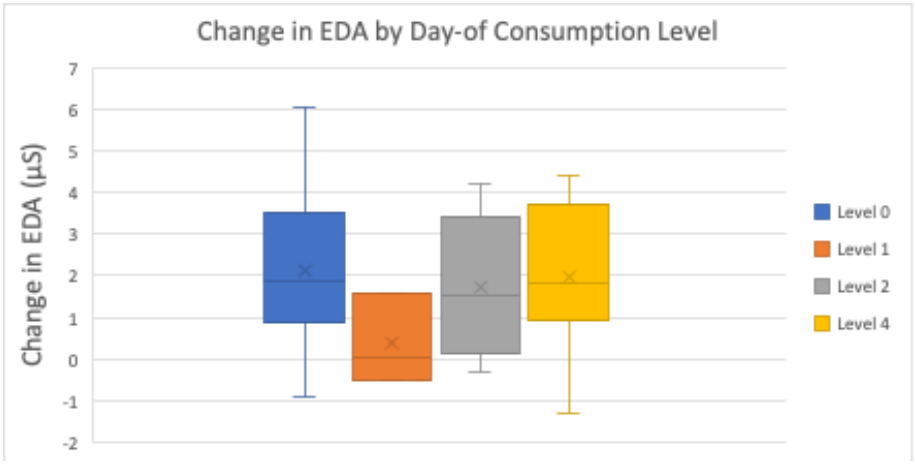

Figure 8: A box and whisker plot showing the mean and distribution from each of the treatment groups' change in electrodermal activity data. No statistically significant difference was found between any of these groups. (Groups 3 and 5 did not have enough participants to do valid statistical analysis.)

Figure 9.

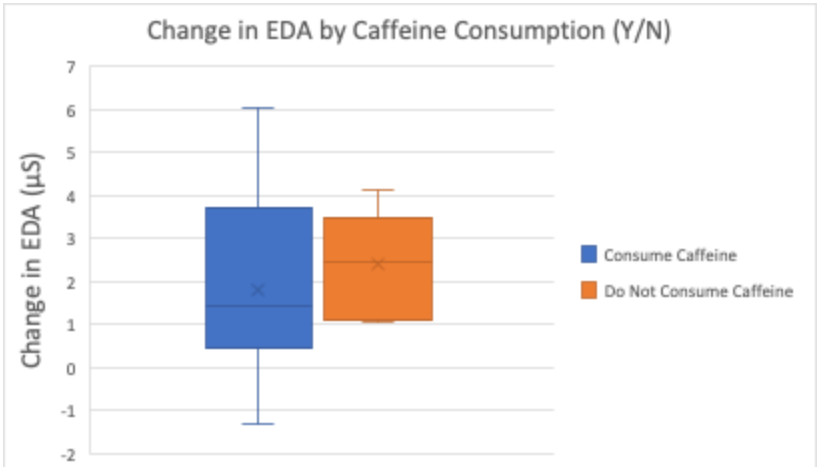

Figure 9: A box and whisker plot showing the mean and distribution from each of the treatment groups' change in electrodermal activity data. No statistically significant difference was found between these groups.

Figure 10.

Figure 10: This is an example of the BIOPAC data collected throughout the experiment. The red arrow denotes the time at which the participant is informed that they will be taking a math test. In this graph, the vertical axis is in microsiemens and the horizontal axis is time recorded in seconds. The average values for EDA were calculated by the BIOPAC software for both the baseline and post-stressor measurements.