Effects of Caffeine on Reactive Hyperemia

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Abstract

The purpose of this experiment was to evaluate the degree of reactive hyperemia and the amount of work output produced in both conditions: control versus caffeine. Dominant arm volume was measured before and after maximal resistance exercise in both trials. The degree of reactive hyperemia was determined based on the volumetric values yielded. Work output was calculated after the resistance exercise was completed. It was hypothesized that the addition of caffeine would increase the arm volume and work output more than the control trial. This study was approved by the Hanover College Institutional Review Board with regard to the use of human participants.

Methods

Ten male participants consented to partake in two randomized trials on separate days. Each participant was to drink 12 ounces of orange juice an hour before each trial. The caffeine dosage was 3 milligrams per kilogram of body weight. Dominant arm volume was measured utilized water displacement before and after maximal resistance exercise. The exercises performed consisted of a dumbbell drop set of single arm curls and single arm triceps extension using set weights of 25, 20, 15, and 10 pounds. Each weight was performed to failure. Dominant arm volume was measured immediately after the completion of each exercise.

Results and Discussion

The results suggest that consuming caffeine prior to performing resistance exercise would yield a higher degree of reactive hyperemia and a higher work output. This suggest that the addition of caffeine prior to exercising would allow a higher volume of blood to enter the occluded muscles and a higher amount of work produced.

Chapter One

Introduction

Reactive hyperemia is a phenomenon in which skeletal muscle blood flow over reacts due to the occlusion that occurs when a muscle contracts to at least 60% of maximum capacity (1). The occlusion to a muscle can take the form of any outside force, like when the doctors take blood pressure readings or within the first second after a muscle contraction (2). The purpose of reactive hyperemia is to reduce the damage to the occluded muscles. The arteries that feed the contracting muscles are completely turned off while the muscle is contracting. When the arteries are shut off this creates a deprivation of oxygen and metabolic waste begins to buildup. This waste is washed away by the constant blood flow that the muscle has. The high surge of blood that enter the muscle after the conclusion of the resistance exercise ensures that all cells within the muscle receive oxygen and waste is flushed. In order to prepare for this high surge of blood entering the muscle, the body secretes vasodilators that in turn increase the dilation of the arteries allowing a higher volume of blood to enter.

It is important to understand the metabolic activity that occurs within the muscle during muscular contractions in order to justify the high surge of blood that enters the muscle after concluding resistance exercise training. Carbon dioxide and lactic acid begin to accumulate due to the muscle using oxygen at an increased rate. Additionally, the muscle’s temperature will increase due to the work begin done while performing the resistance exercise training. Therefore, this increase in metabolic activity creates a hostile environment for the working muscles. The working muscles are demanding oxygen and the removal of waste that is accumulating. The only way to meet the demands of the working muscles is by having the high surge of blood. Furthermore, the metabolic activity that occurs within the muscles become a negative feedback mechanism. The increased metabolic activity that occurs within the muscles begin to work on autoregulation, arterioles, and the precapillary sphincter muscles.

In turn, the sympathetic nervous system is stimulated by the chemoreceptors and thermoreceptors within the working muscles, increasing the heart rate and the amount of blood being delivered. In addition, the sympathetic nervous system aids in the recruitment of motor units. The amount of motor units recruited will yield a stronger muscular contraction. This is due to the all or none principle, which states that when a muscle contracts, it does so at its maximal potential or not at all. With the addition of caffeine, the sympathetic nervous system should be able to recruit more motor units, yielding more strength. More strength yields a higher work output, resulting in a higher degree of reactive hyperemia.

The redistribution of blood flow is an important mechanism the body utilizes in order to effectively deliver blood to the working muscles. The body wants blood flow to go to the working muscles. The arteries and veins have the capability to either constrict or dilate which aid in the redistribution of flow in order to meet the demands of the working muscles during resistant exercise training. To aid in vasodilation, the body secretes nitric oxide. Nitric oxide is produced in the inner layer of blood vessels, the endothelial lining (3). This vasodilator is released in the smooth muscles cells causing them to relax. In turn, the relaxation causes the walls of blood vessels to dilate which prepares the vessels for the high surge of blood that is to come after the conclusion of the resistance exercise training. In contrast, vasoconstriction occurs in the vessels that are not needing blood flow. Therefore, blood flow is directed to the vessels of the working muscles, as they are more readily available. This localized increase in blood flow allows the working muscle to meet the high metabolic demand from exercise, and thus continue the exercise. (4).

Caffeine is the number one most socially accepted and frequently used drug (5). Caffeine occurs naturally in seeds and plants, the most common being coffee beans and cacao beans (6). However, manufacturers have started producing synthetic forms of caffeine and adding it to numerous drinks, foods, and pre-workout supplements for its physical and psychological boost (5). Therefore, randomized controlled trials must be performed in order to evaluate the effects that caffeine has on resistance exercise.

Statement of Purpose

To determine the effects of caffeine on reactive hyperemia after resistance exercise.

Significance of Study

Professional Significance

Resistance training affects the working skeletal muscle. The disruption that occurs leads to the muscle feeling very uncomfortable and making it change in order to prevent disruption in future situation where it is being worked out. Whenever a muscle is worked out and disrupted, there is a systematic need to support the changing environment and improvement in overall function. This support will come from the increased blood flow and the redistribution of blood flow that occurs in order to effectively deliver oxygen and nutrients to the working muscles through reactive hyperemia.

The reactive hyperemic effect following an intense workout can be used to indicate the effectiveness of that workout. This is crucial for athletes or regular weight lifters to be aware of if they are attempting to build muscle successfully. When comparing the amounts of immediate blood flow, it would then be logical that an exercise providing more blood flow is disrupting the muscles more as well. The consumption of caffeine and the effectiveness of the workout can be directly linked to the level of disruption that occurs in the working muscles and the redistribution of blood flow across the body. Thus, a harder working muscle is a more effective muscle. Therefore, this study has a professional significance for the athletes and regular weight lifters who consume a pre-workout supplement because of the large amount of reactive hyperemia that occurs in a workout.

Personal Significance

This study has a substantial personal significance because I have a high interest in exercise, resistance training, and the methods the body uses to recover after putting it through an intense workout. I am interested in evaluating the effects of caffeine on resistance training because many athletes or regular weight lifters tend to take a pre-workout supplement that contain a large amount of caffeine. Recently, I have been aware of these supplements but have always chosen to go in and perform the resistance training naturally. I am intrigued to discover the effects of caffeine on not only reactive hyperemia but also the total amount of work output produced after consuming caffeine and performing exercises that challenge the biceps and triceps.

Delimitations

This study was performed on ten male participants who had prior knowledge to resistance exercise training and had an athletic background. The independent variable was caffeine and the dosage that was given to each participant prior to the testing. Caffeine was consumed an hour prior to the testing in order to maximize the effects of caffeine on reactive hyperemia. The participants would complete resistance exercises by way of a drop set of single arm dumbbell bicep curls and standing single arm dumbbell triceps extension. The dependent variables included the amount of reactive hyperemia that occurred in the dominant arm after each exercise was completed and the total amount of work output produced by the dominant arm. Prior to commencing each exercise trial, the dominant arm volume was measured by way of water displacement using the graduated cylinder. The resistance exercises were performed to the best of the participants' ability. After concluding the exercise, the dominant arm volume was immediately measured in order to access the change in arm volume. Furthermore, work output was calculated by multiplying the amount of weight by the amount of reps performed. The work output values were compared between the control trial and the caffeome trial in order to evaluate the effects of caffeine on work output.

The purpose of this study was to determine if the addition of caffeine would hinder or enhance reactive hyperemia. This comparison between the control and caffeine trial is an interesting study because many athletes who lift weights nowadays consume some sort of pre-workout supplement(s) that contain a large amount of caffeine. It is important to understand if the addition of caffeine would have any sort of effect on the amount of blood flow that is redistributed to the working muscles after the conclusion of resistance training.

Limitations

1. External validity → small sample size
2. Only testing two muscle groups
3. Only testing the dominant arm
4. Not a diverse sample of subjects → only male participants
5. The subject was assumed to follow the instructions of not exercising prior to the testing
6. Only one trial for each exercise per subject

Assumptions

It was assumed that:

1. The subject did not exercise prior to the testing.
2. The subject did not consume a heavy meal prior to the testing.
3. The subject consumed the caffeine pill an hour prior to the testing.
4. The subject inserted the arm in the graduated cylinder to the same level for each trial that was conducted.
5. The subject performed the exercises to maximal exhaustion (another repetition was not physically possible).
6. The graduated cylinder was calibrated correctly and yielded accurate measurement for each trial.
7. The subject performed all exercises with perfectly strict form without cheating.
8. An increase in arm volume after resistance exercise indicated an effect from reactive hyperemia.

Hypotheses

It was hypothesized that:

1. Arm volume will increase with exercise.
2. Arm volume will increase with exercise and the addition of caffeine.
3. Arm volume will increase more with exercise and the addition of caffeine.
4. Work output will increase more with exercise and the addition of caffeine.

Term Definitions

1. Isotonic contraction- muscular contraction where the length of the muscle changes with varying energy and speed throughout the range of motion
2. Range of motion- the measurement of movement around a specific joint or body part in regards to flexion and extension
3. Vasomotor tone- amount of tension in the smooth muscle inside the walls of blood vessels, particularly in arteries
4. Capillaries- network of small blood vessels that connect the smallest arteries with the smallest veins and exchange oxygen, metabolic waste products, and carbon dioxide between blood and tissue cells
5. Arterioles- a small branch of an artery leading into the capillaries
6. Autoregulation- the intrinsic ability of an organ to maintain a constant blood flow
7. Precapillary sphincter muscle- band of smooth muscle that adjusts blood flow into the capillaries
8. Vasoconstriction- the narrowing of blood vessels
9. Vasodilation- the expansion of blood vessels
10. Neuron- a specialized cell transmitting nerve impulses; a nerve cell
11. Ganglia- a structure containing a number of nerve cell bodies, typically linked by synapses, and often forming a swelling on a nerve fiber
12. Synapse- a junction between two nerve cells, consisting of a gap which impulses pass by diffusion of a neurotransmitter
13. Nitric oxide- gas that reacts with oxygen to form nitrogen dioxide, causes smooth muscle in the vessel to relax, creating vasodilation
14. Chemoreceptors- neural receptors that respond to some local chemical change
15. Thermoreceptors- non-specialized sense receptor that codes absolute and relative changes in temperature

Operationally Defined Terms

1. Drop set: a technique where the participant performed an exercise and then dropped the weight and continued for more reps until failure was reached.
2. Water displacement: method that involved putting the participants' arm into water and carefully recording how much the water rose.
3. Dominant arm volume: the amount of space that is taken up by the dominant arm of the participant (in regards to water displacement.
4. Graduated cylinder: known as the pipette apparatus cleaner. For the purpose of this experiment, this piece of equipment was used to measure the volume of the dominant arm. Each marked line on the graduated cylinder represents the amount of liquid that has been measured.

Chapter Two

Background Research

Caffeine

Caffeine is found in many foods and beverages, and over 90% of the US population consume it on a regular basis (8). Unlike most substances and supplements, caffeine can affect cells throughout the body, including muscle cells and the brain (9). In recent years, manufacturers have started producing caffeine synthetically and adding it to drinks and foods for its physical and psychological boost (5). Those who are regular weight lifters have become fond of this drug that has yet to be regulated. A caffeine dose of 200-600 mg may help an athlete with anaerobic exercise which includes sprinting and resistance exercise. This ergogenic effect is small and fades with frequent intake. Once the tolerance in the body rises, more caffeine must be consumed in order to gain the same levels of focus and power.

Caffeine and Exercise

When it comes to exercise, caffeine may have the possibility to increase the use of fat as fuel. This is important because it can help the glucose stored in muscles last longer, which could ultimately delay the time it take the muscles to reach exhaustion (11). Caffeine may also improve muscular contractions and increase tolerance to fatigue. Recent research has observed that doses (5 mg/kg) of body weight improved endurance performance by up to 5%, when consumed once hour before the exercise (12). In relation to this study, research notes state that doses as low as (3 mg/kg) of body weight may be sufficient to reap the benefits that caffeine brings (13).

Summary

Caffeine is a drug that is being consumed at a higher rate among athletes, regular weight lifters, and every day people whether that would be in forms of food, beverages, or supplements.

Blood Flow

General Blood Circulation

Blood is effectively moved throughout the body in order to deliver nutrients and oxygen to tissues and muscles. In order to circulate and distribute blood to all areas of the body, the action of a muscular pump is required. This muscular pump is known as the heart and creates a pressure head that initiates the movement of blood throughout the body.

Blood Flow Within the Heart

Blood enter the heart on the right side through two large veins: the inferior and superior vena cava. The right side of the heart is where deoxygenated blood enters, as it has just been circulated throughout the body. The blood specifically enters the right atrium where it will await the atrial contraction that sends the blood through the tricuspid valve intro the right ventricle. This action is known as diastole, which is defined as the ventricle refilling with blood. Once the ventricle has accumulated enough blood, the tricuspid valve is closed off in order to prevent backflow of blood during ventricle contraction (7). This stage is known as systole. In this stage, the right ventricle contracts, allowing the blood to exit the heart and enter the pulmonary circuit. In turn, the pulmonary circuit transports this blood to the lungs where re-oxygenation occurs. The re-oxygenation occurs due to the gas exchange in the alveoli where oxygen is picked up and carbon dioxide is left behind to expire. The newly oxygenated blood flows through the pulmonary veins back to the heart, into the left atrium (7). Similarly, the atrium will contract during diastole in which the blood is now moved into the left ventricle through the bicuspid valve. Once the blood has filled the left ventricle, the bicuspid valve closes in order to prevent backflow into the left atrium. From here, systole occurs once again, where the contraction will force the oxygenated blood into the aorta via the aortic valve. Finally, the blood exits the heart and is passed to the necessary muscles and tissues throughout the body.

Blood Flow Throughout the Body

Once the blood enters the circulatory system, these vessels are responsible for directing the blood to where it needs to go. As these vessels get further away from the heart, they become smaller. Eventually, the size of the arteries is reduced to the point where they are known as arterioles. These vessels are extremely flexible as they contain elastic properties which allows them to have their unique feature of vasomotor tone. This allows the arterioles to vasoconstrict or vasodilate, depending on what the circumstances are within the body at any given time. Arterioles additionally regulate the flow of blood into the capillaries via the pre-capillary sphincter muscles. The capillaries are even smaller and positioned in “beds” in which the oxygen and nutrients from blood can be diffused into tissues, known as the capillary exchange (7). Once the essential nutrients and oxygen are removed from the oxygenated blood, it becomes deoxygenated once again.

Blood Flow Back to the Heart

Once the blood is diffused into the capillaries, it begins to make its way path to the heart in order to begin the cycle once more. Blood travels from the capillaries into the vessels known as venules. As these move closer to the heart, they in turn increase in size and become veins (7). Unlike blood exiting the heart, blood returning to the heart must deal with gravity working against it. Thankfully, the body is equipped with mechanism and specialized traits that effectively allow the blood to move back to the heart. Traits such as pressure gradients, one-way valves, muscle pumps, respiratory pumps, and gravity all provide assistance in forcing blood through the small vessels into the larger veins (7). Once the deoxygenated blood reaches the superior and inferior vena cava, the process commences once again.

Redistribution of Blood Flow during Exercise

During intense exercise the metabolic demand for oxygen is significantly increased within the skeletal muscle. To meet this high demand, it is of high importance to increase the amount of skeletal muscle blood flow (14). This occurs through the increase in cardiac output where the components are heart rate and stroke volume (7). The increase in heart rate allows for more frequent contractions that send oxygenated blood into the body. Increasing stroke volume increase the amount of that is forced from the heart with each contraction. As this process is important to increase blood flow, it alone does not satisfy the need for extra blood in the working skeletal muscles (15).

To continue, the redistribution of blood flow to areas of the body that are not working must be lowered. During exercise, the working muscles increase their metabolic activity. The environment that the muscle is surrounded by becomes hostile. The muscle begins to increase carbon dioxide and lactic acid concentration while decreasing oxygen and pH concentration. Muscle and body tissues possess the ability to auto regulate itself. The body is able to do this because of chemoreceptors that signal the brain to remove the sympathetic stimulation from the blood vessels feeding the working muscles (7). When this occurs, the vessels will be able to vasodilate, decreasing the vascular resistance. Simultaneously, the same message is being relayed to the vessels of the inactive tissues in order to increase the sympathetic stimulation (7).

When this occurs, the arterioles leading up to the working muscle will be vasocontricted, allowing the blood flow to be directed to that specific muscle.

Reactive Hyperemia

Reactive hyperemia is a phenomenon in which skeletal muscle blood flow overcompensates due to the occlusion that occurs when a muscle contracts to at least 60% of maximum capacity (1). Vigorous exercise occludes the blood vessels leading up to the working muscles. This creates a high demand for oxygen, nutrients, and the removal of waste products. When the muscular contractions stop, a high surge of blood enters the muscle in order to meet the metabolic demands. This results in a hyperemic response in the skeletal muscle, thus the term, reactive hyperemia.

Summary

Having general knowledge of the blood flow throughout the body, how it effectively distributes and redistributes blood during vigorous exercise is significant to this study. Being able to understand the changes that occur within the muscle during resistance exercise is also pertinent to this study. As metabolic demand increases from vigorous exercise, the body effectively redistributes the blood to the needed muscles. However, this does not occur until the muscular contractions stop. Upon cessation, the blood vessels feeding the working muscles will no longer be occluded and the body has prepared for the high surge of blood that is about to enter the capillary beds.

Sympathetic Nervous System

Heart rate is controlled by two branches of the autonomic nervous system: the sympathetic nervous system and the parasympathetic nervous system. The sympathetic nervous system releases the hormones epinephrine and norepinephrine to accelerate the heart rate. In contrast, the parasympathetic nervous system releases the hormone acetylcholine to slow the heart rate. Exercising for any period of time will increase your heart rate and will remain elevated for as long as the exercise is continued. At the beginning of exercise, the body effectively removes the parasympathetic nervous system which enables the heart rate to gradually increase. As the exercise duration or intensity becomes strenuous, the sympathetic nervous system begins to have a greater influence on the heart, causing it to beat faster. This is also known as the reflex action. The brain gets feedback from receptors all over the body to determine how much it should raise the heart rate. The proper receptors provide the body with sufficient heart beats in order to satisfy the increased demand for blood, oxygen, and nutrients. This feedback mechanism typically overreacts, usually 10-15 bpm in order to make sure the body is getting enough of what it needs.

Caffeine and the Sympathetic Nervous System

When caffeine is consumed, it further stimulates the sympathetic nervous system to recruit more motor units. The all or none principle states that when a muscle contracts, it does so at its maximal potential or not at all. Therefore, the greater amount of motor units recruited grants greater strength. With more strength to work with and the addition of caffeine allowed the participants to produce a higher work output than the control trial where no caffeine was consumed. In turn, with more work being produced, a higher degree of reactive hyperemia would occur because there would be more disruption within the working muscles. More disruption means that there is a higher demand for oxygen and a higher concentration of lactic acid and carbon dioxide. Therefore, the body overcompensates by providing the muscle with a higher degree of blood volume to the working muscle, hence reactive hyperemia.

Summary

It is essential to understand how the sympathetic nervous system works as it allows your body to effectively respond to resistance exercise. In addition, the physiological changes that occur when the sympathetic nervous system is stimulated have an effect on how fast and how hard your heart beats. The addition of caffeine further stimulates the sympathetic nervous system by aiding in the recruitment of motor units. The more motor units recruited, the stronger the contraction and more repetitions, ultimately leading to a higher degree of reactive hyperemia.

Related Literature

Hypothesis 1: Arm volume will increase with exercise.

Hypothesis 2: Arm volume will increase with exercise and the addition of caffeine.

Supportive Research

1. Boushel, Robert. Metabolic control of muscle blood flow during exercise in humans. Canadian Journal of Applied Physiology. 28 (5), 757-773. 2003.

The first item of supportive research was a review article that discussed published studies that involved strong muscular contractions from resistance exercise and the blood flow response. "At the beginning of a strong contraction, a certain amount of blood is thrown back into the muscle artery against the high arterial blood pressure." This suggests that the body is sensitive to muscular contractions and will respond immediately. In this article, reactive hyperemia is termed as the "hyperemic effect." This effect was broken down into four separate stages. The first stage is the initial blood flow response when a strong contraction begin. Next, there is an arrest of inflow. This inflow is hindered due to the occlusion of the blood vessels feeding the muscle. The muscular contractions squeeze the vessels and force them to take up less space, causing issues with blood flow to the muscle. The third stage was described as the "overshoot" effect during muscular contraction. When these muscular contractions came to a complete stop, the blood vessels were able to open up completely, allowing the high surge of blood to enter the muscle. The final stage is the hyperemic effect, or the pumping of the worked muscle via these vessels (7). It is clear from the article that the hypothesis would support hypothesis one and two due to the blood flow reaction to muscular contractions.

Hypothesis 3: Arm volume will increase more with exercise and the addition of caffeine.

Hypothesis 4: Work output will increase more with exercise and the addition of caffeine.

Supportive Research

1. Graham, Terry E. "Caffeine and Exercise Performance." *Gatorade Sports Science Institute*, June 2006
2. Collins, P. Blaise, et al. "Short-Term Effects of a Ready-to-Drink Pre-Workout Beverage on Exercise Performance and Recovery." *Nutrients*, MDPI, 1 Aug. 2017.

The first item of supportive literature is a review article that discussed the effect of caffeine on exercise performance. Caffeine is a "controlled or restricted drug" in the athletic world because urinary levels greater than 12 ug/mL following competition are considered illegal by the International Olympic Committee.

It was suggested that caffeine appeared to have been taken up by all tissues. The article highlighted the three major theories for the ergogenic effect of caffeine during exercise. The first theory suggest that caffeine has a direct effect of the central nervous system that affects the perception of effort and/or the neural activation of muscle contraction. The second theory proposes a direct effect of caffeine on skeletal muscle performance. The third theory is the classic or "metabolic" explanation that involves an increase in fat oxidation and a reduction in carbohydrate oxidation.

The second item of supportive literature was study that examined the effects of pre-workout beverage on exercise performance and recovery. The study asks its participants to consume a placebo beverage and the experimental beverage that contained 200 mg of caffeine. This study was pertinent to the processes involved with reactive hyperemia as the participants were to perform “10 repetitions on the bench press and leg press at 70% of 1-RM on day 1 and day 6.” Participants were then randomized to initiate the study with the respective treatment. After waiting for 30 minutes, the participants performed the same resistance exercise with aims of getting a better score than their first bout without a pre-workout beverage.

It was found that on both test days, the participants were able to successfully recover after their first bout of exercise and still be to produce more repetitions after they had consumed their drink with caffeine. These result suggest that the caffeine dosage that was given to the participants was able to recruit enough motor units to not only perform the same amount of work, but surpass it on the same day! Therefore, it can be predicted that the work output will increase more with exercise and the addition of caffeine in this study since the participants will be given a whole week to recover.

Chapter Three

The purpose of this study was to determine the effects of caffeine on reactive hyperemia after resistance exercise. This chapter will provide a thorough description of the participant selection, research design, instrumentation, protocol, procedures, and pilot data involved in this study.

Participant Selection

Ten male participants were recruited for this study. All the participants involved were students who attended Hanover College. The age range for the participants was from 18-22 years old. Each participant was either a Hanover College student athlete or a student who was knowledgeable in resistance exercise. A major key to this study was that the participants were regular weight lifters (at least two times per week) to ensure that they had the ability to perform maximally exerting resistance exercises. The participants were selected based on their willingness to partake in the study.

Research Design

This study consisted of two randomized controlled trials: one with the addition of caffeine and one without. The amount of caffeine that was given to each participant was 3 mg per kg of body weight. The bicep and triceps of the dominant arm were tested in this study and changes in arm volume were measured via water displacement before and after the resistance exercises were completed. Each trial was conducted at least one week apart from one another in order to ensure that there would be no possible soreness within the muscles being tested.

Resistance Exercise Protocol

Each participant was asked to complete an isotonic resistance exercise protocol in which a drop-set of dumbbell curls was performed with five different weighted dumbbells. The dumbbell weights consisted of 25, 20, 15, and 10 pounds. The participants were asked to perform each repetition of standing single arm dumbbell curls and standing single arm dumbbell triceps extension until failure. The participants would start with the standing single arm dumbbell curls with the 25 pound dumbbell until failure and immediately transition into the standing single arm dumbbell triceps extension until failure. This was performed with each dumbbell weight in that order until the participant reached the 10 pound dumbbell until failure. All sets were to be performed with strict form and maximal exertion.

Instrumentation

Arm Volume Measurements

Before and after the resistance exercise was completed, dominant arm volume was measured using water displacement in a graduated cylinder. A black permanent marker was used to draw a line across the middle deltoid so the participant would insert his arm to the same degree during all trials. McCormick's red food coloring was added to the water in order to make the arm volume easier to read. This graduated cylinder was calibrated in such a way that allowed the calculation of arm volume to be effectively measured.

Resistance Exercises

Four weighted dumbbells were used as part of the resistance exercise protocol for both trials. These weighted dumbbells were standard, free weight dumbbells with separate weights that ranged from 10-25 pounds.

Caffeine Dosage

Each participant was given 3mg of caffeine per kilogram of body weight. Vivarin was the brand of caffeine pills that was utilized for this study. A pill cutter was used in order to give the correct dosage to each participant. The caffeine pills were then grinded and mixed with 12 ounces of orange juice. The acidity within the orange juice will eliminate the taste that the caffeine pill contains. This adds creditability to the randomization within the two trials. If the caffeine pill were to be consumed with water, it would be evident which trial would be which and this could skew the data.

Pre-Test Protocol

1. Each participant was asked not to exercise prior to the test. This was done in order to ensure they were able to maximally exert the working muscles. Additionally, it ensured that the baseline level for the pre-exercise measurement was accurate. Any exercise prior to the testing session has the potential to disrupt the accuracy of arm volume measurements.
2. Each participant was asked not consume a heavy meal at least three hours prior to the testing. This was because of previous research showing potential for diet affecting reactive hyperemia and blood flow distribution.
3. Each participant was asked not to consume any caffeine the day of the test. This was done in order to ensure that the only caffeine within their system was the caffeine that was being given to them. Any consumption of caffeine could affect the degree of reactive hyperemia.
4. Each participant was asked to consume the prepared beverage one hour prior to the test (with or without caffeine). This was done to ensure randomization but also previous research suggested that the peak level of caffeine is one hour after consumption.
5. Each participant was asked not ingest any form of inflammatory drugs or prescribed drugs. This was to avoid any unwanted vasodilation or vasoconstriction that could arise from these drugs.
6. Each participant was asked to wear appropriate clothing for the exercise test and water displacement measurements. This clothing consisted of a sleeveless shirt or no shirt if desired. This was done in order to provide comfortability during the resistance exercise and allow the arm volume measurements to be made accurately. Because the participant was supposed to insert his arm up to his middle deltoid, a sleeveless shirt would avoid his clothing from getting wet.
7. Each participant was asked to bring a towel in order to dry off after each arm volume measurement.
8. If any or all of these criteria were not met, the test was rescheduled to a different date.

Dominant Arm Volume Protocol

Before and after the resistance exercise was completed, the dominant arm volume was to be measured. Using the modified graduated cylinder, the dominant arm volume was measured utilizing the following procedure:

1. The graduated cylinder was filled with 10 liters of water. This volume of water was even with the 230mm mark on the strip of measuring tape located on the graduated cylinder near the top.
2. McCormick's red food coloring was added to the water in the graduated cylinder to ensure the accuracy of the readings. Three drops of food coloring were added to the water.
3. Prior to submerging the dominant arm, the participant was marked with a black line utilizing a permanent marker across the middle deltoid. This was a marker so the participant knew how far to insert his arm.
4. The participant was to insert their dominant arm with the marking into the water, until the water reached the black line. This was to ensure that only the working muscles were being measured.
5. The water level within the graduated cylinder was then recorded. Water displacement was measured before and after the resistance exercise in order to compare the difference in arm volume.
6. All measurements were to begin with the water level at 230mm on the strip of measuring tape. This was to provide a baseline starting point and ensure reliability and from measurement to measurement. Water level was recorded to the nearest millimeter on the calibration tape.

Procedure

1. Gather in the exercise physiology laboratory in Science Center at Hanover College
2. Gather and calibrate equipment need as described
3. Resting dominant arm volume measurements were collected using water displacement.
4. The participant was to dry arm off.
5. The participant was asked to complete the resistance exercise protocol as described.
6. After completing the resistance exercise protocol, the dominant arm volume was immediately measured
7. The subject was then to dry off once again.
8. Difference in arm volume were observed and noted.
9. Work output was calculated.
10. Session complete.

Post-Test Protocol

Once the participant was completed with each trial of the exercise resistance, they were asked to report any form of discomfort or symptoms that could have been related to the study. It was made clear that the participant should replenish the fluids lost during the exercises in order to rehydrate. It was also recommended that they should rest their arms after the conclusion of the exercises. Prior to leaving, the graduated cylinder was to be drained and stored. Any other materials that were used during the test session were to be stored in their proper location and the lights in the lab were to be turned off prior to leaving the lab.

Dominant Arm Volume Calculations

Near the top of the graduated cylinder was a piece of measuring tape that read in millimeters. The millimeter values began at 230mm and ended at 0mm. The graduated cylinder was calibrated in a way were 1 centimeter of water level increase was equal to 200 milliliters of water volume increase. This calibration allows the units to be converted from millimeters to centimeters for ease of calculation. The converted amount of centimeters was then multiplied by 200 to provide the total amount of water volume increase, hence water displacement (7). The equation used for this calculation is shown below:

[**(initial volume (mm)) – (final volume (mm))] ÷ (10) x (200) ÷ 1000 = final arm volume (7)**

In order to interpret the results in an easier way, the changes in arm volume were converted from milliliters to liters.

Dominant Arm Work Output Calculation

Work output is the total amount of work produced after completing a resistance exercise workout. The amount of repetitions is multiplied by the amount of weight being lifted. An example of this is provided below:

25 pounds x 22 repetitions = 550 pounds of work produced

Work output is an important aspect of the results because it provides an accurate value to interpret the amount of work that was produced during the resistance exercise training.

Mann-Whitney U Test

The Mann-Whitney U test is a non-parametric alternative test. This test is used to compare two sample means that come from the same population, and used to test whether two sample means are equal or not.

Assumptions of the Mann-Whitney:

The following assumptions are assumed when performing the Mann-Whitney U test:

1. The sample drawn from the population is random.
2. Independence within the samples and mutual independence is assumed. Meaning that an observation is in one group or the other (it cannot be both).
3. Ordinal measurement scale is assumed.

Calculation of the Mann-Whitney U:

$$U = n_1 n_2 + \frac{n_2(n_2+1)}{2} - \sum_{i=n_1+1}^{n_2} R_i$$

Symbols and meaning:
U=Mann-Whitney U test
N_1 = sample size one
N2= Sample size two
R_i = Rank of the sample size

Data Treatment

Once the data was collected in each session, the results were organized into categories of resting dominant arm value, dominant arm value after exercise, change in dominant arm volume, and the total amount of work produced. These values were analyzed in order to determine how much the dominant arm volume changed in the caffeine and controlled trail. From this, a conclusion of how caffeine effects reactive hyperemia can be determined.

Pilot Data Development

The collection of pilot data for this study has significantly developed. To begin the pilot data collection, it was of very high importance to ensure that the graduated cylinder yielded accurate and reliable measurements using the water displacement technique. Therefore, the purpose of the first pilot data session was to measure both the dominant and non-dominant arms in the graduated cylinder, and see if it did in fact yield accurate and reliable measurements. This was performed on one participant five separate times on each arm. The data collected from the graduated cylinder yielded accurate and reliable measurements when measuring arm volume.

The next step in the pilot data process was to ensure that there was a change in arm volume after resistance exercise, and that this change can be observed, and measured. Therefore, the next pilot data session involved the control trial where the participant was not to consume caffeine prior to the test. A measurement of the dominant and non-dominant arm volumes were recorded before and after the resistance exercise. This was also done to ensure that it was reliable and accurate. It was found that arm volume does increase after resistance exercise, and this was able to be measured. Work output was also observed and measured in this step. It was found that work output does increase after performing exercise.

The purpose of this study was to determine the effects of caffeine on reactive hyperemia. This leads into the next step of the pilot data which begins to compare the caffeine trial to the controlled trial. This data collection would determine whether or not the hypotheses were to be supported or rejected. This data collection involved measuring the dominant and non-dominant arm volume before and after resistance exercise. It was determined that the dominant arm volume in the caffeine trial did not increase arm volume more than the control trial. Work output was also observed and calculated in this step. It was also found that work output did not increase more in the caffeine trial in comparison to the controlled trial.

Although the pilot data collected refuted the main hypotheses in this study (3 and 4), it was deduced that this could be the case for this participant. Each participant is their own control and caffeine could have had a lesser effect on this participant in comparison to other participants to come. The must be more data in order to make conclusive statements regarding the effect of caffeine on reactive hyperemia.

Pilot Data

Figure 1

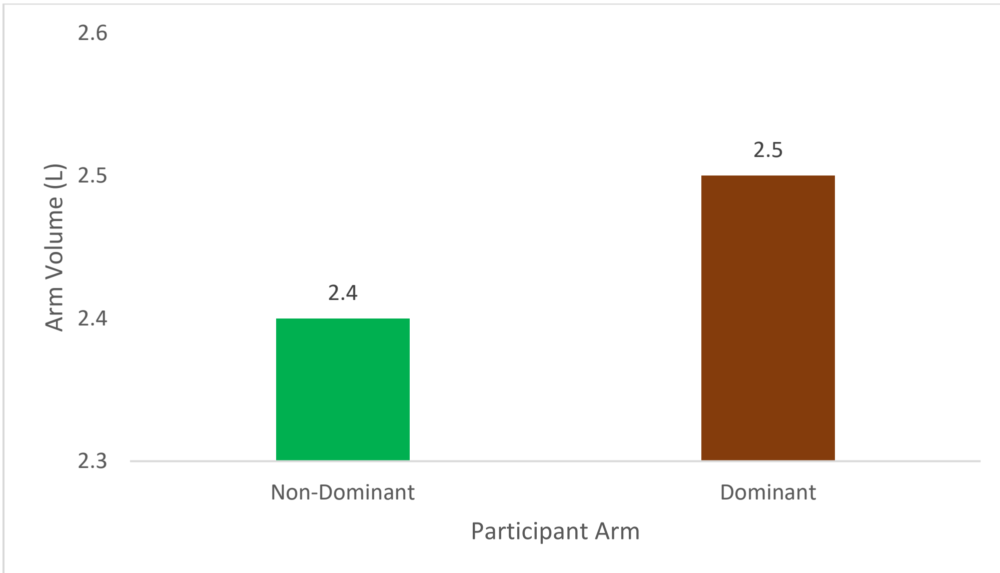

Figure 1. Average Non-Dominant and Dominant Arm Volume at Rest

Figure 1 contains the pilot data that was gathered in order to determine if the non-dominant and dominant arm volume measurements could be accurately and reliably taken using the graduated cylinder via water displacement. Both arms were measured on five separate trials on the same day. The goal of these measurements was to record similar values for all five trials. As seen in the figure, the dominant arm volume (2.4 L) is slightly higher than the non-dominant arm (2.5 L). This was expected because the dominant arm in most people is slightly bigger than their non-dominant arm due to it being used more frequently. These results were concluded to be similar, therefore making the graduated cylinder an accurate and reliable method of measuring arm volume.

Figure 2

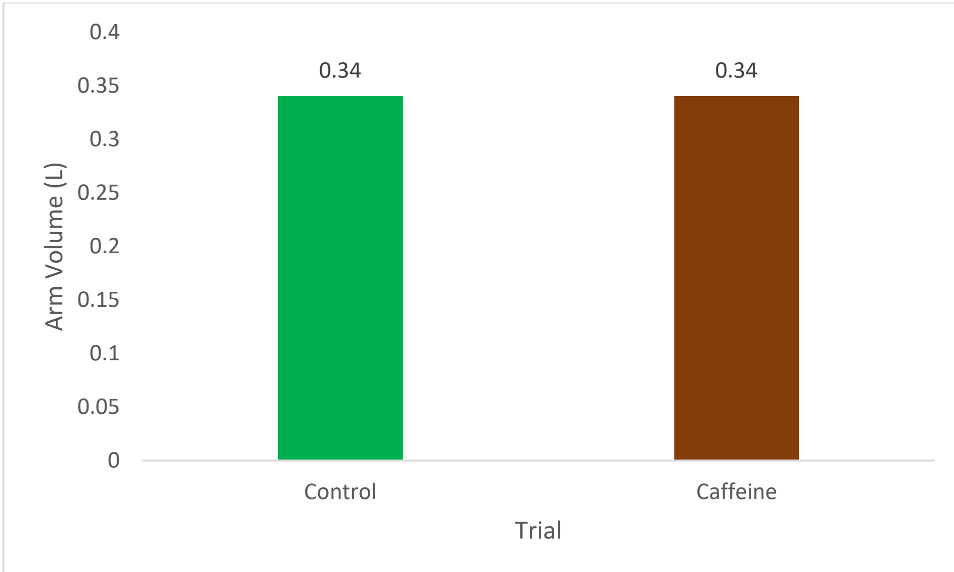

Figure 2. Average Dominant Arm Volume Displacement in the Control and Caffeine Trial

Figure 2 illustrates the pilot data that was collected in order to compare the difference in arm volume displacement in the control and caffeine trial. In both conditions, the arm volume was measured before and after the resistance exercises were completed. This was done to test hypothesis 3: arm volume will increase more with exercise and the addition of caffeine. It was expected to see the dominant arm volume displace more in the caffeine trial. However, when the dominant arm volume calculations were performed, both trials resulted in the same volume displacement (0.34 L) after the exercises were completed, thus rejecting hypothesis 3. In addition, this figure supports hypothesis 1: arm volume will increase with exercise and hypothesis 2: arm volume will increase with exercise and the addition of caffeine. This hypothesis could be accepted once more participants are tested and the study progresses.

Figure 3

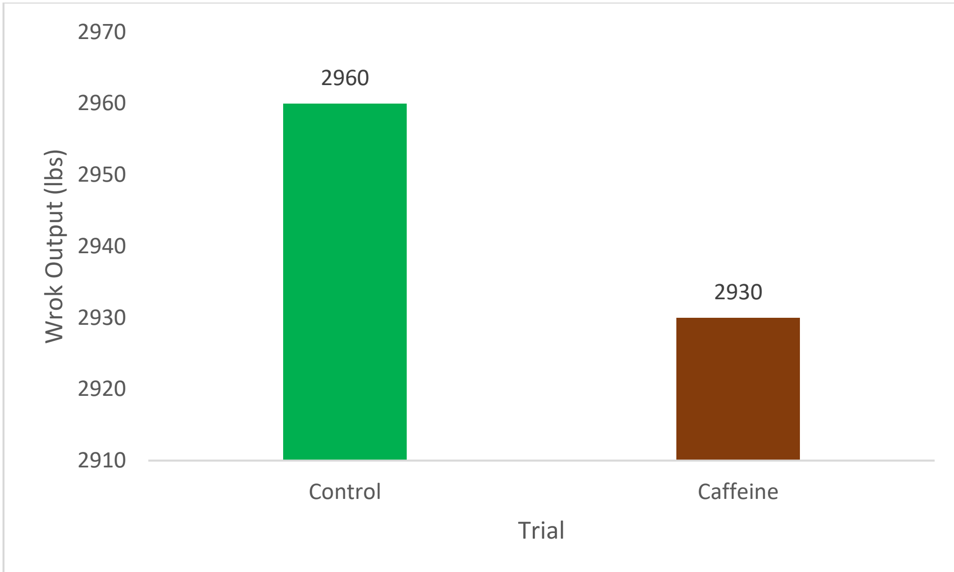

Figure 3. Dominant Arm Work Output in the Control and Caffeine Trial

Figure 3 is a representation of the total dominant work output in both the control and caffeine trial. The data collected was to determine if there is a difference in the amount of work produced when the participant does not consume caffeine and when the participant does consume caffeine. Additionally, this serves to test hypothesis 4: work output will increase more with exercise and the addition of caffeine. As seen in figure 6, there was total work output of 2,960 pounds produced in the dominant arm of the control trial. In contrast, the dominant arm in the caffeine trial only produced a total of 2,920 pounds of work output. Therefore, hypothesis 4 was rejected. It was concluded that the dominant arm in the control trial produced more work output than the dominant arm in the caffeine trial. However, this hypothesis could be accepted as the study progresses and more participants are tested.

Chapter Four

Data and Results

Figure 1

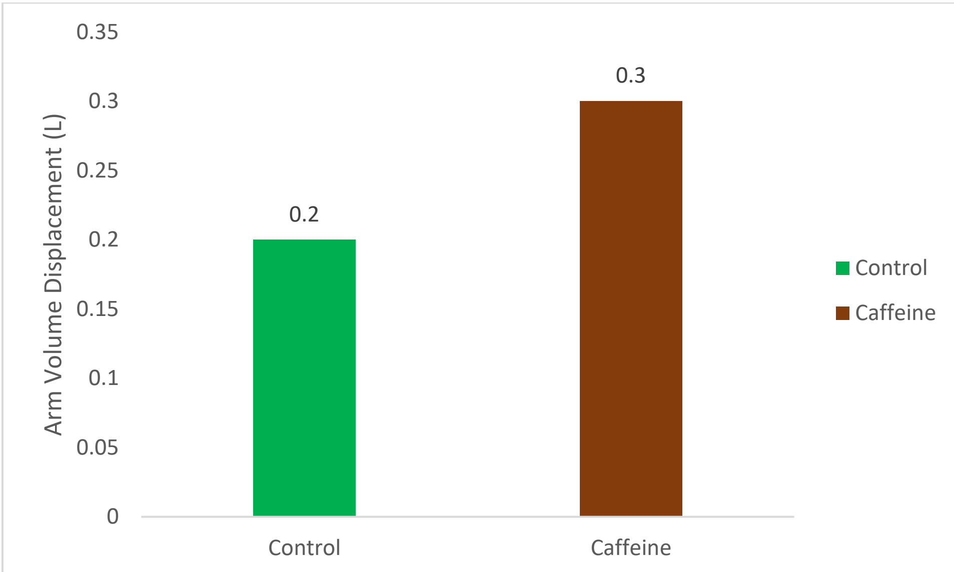

Figure 1: Depiction of Average Dominant Arm Volume Displacement in the Control and Caffeine Trial

The figure above is an illustration of the average arm volume displacement in both trials. It is clear from the figure that on average for all ten participants, their average arm volume displacement was higher for the caffeine trial in comparison to the control trial. On average, the dominant arm volume displacement for the control trial was 0.2 L while 0.3 L for the caffeine trial. These values were concluded to be significant ($P < 0.05$), utilizing the Mann-Whitney U test. In addition, the following hypotheses were accepted due to the results found in the figure above:

1. Arm volume will increase with exercise. **Accepted.**
2. Arm volume will increase with exercise and the addition of caffeine. **Accepted.**
3. Arm volume will increase more with exercise and the addition of caffeine. **Accepted.**

Figure 2

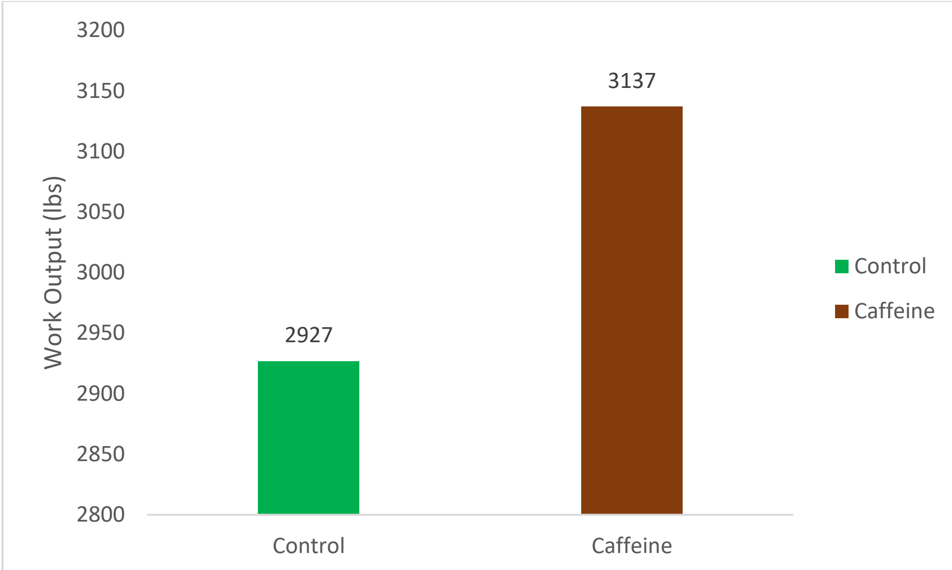

Figure 2: Average Dominant Arm Work Output Produced in Both Trials

Figure 2 is a representation of the total average work output produced by the dominant arm in both trials. As seen above, the control trial on average had a work output of 2927 pounds while the caffeine trial had an average of 3137 pounds. These values were concluded to be significant ($P < 0.05$), utilizing the Mann-Whitney U test. In addition, the following hypothesis was accepted due to the results found in the figure above:

4. Work output will increase more with exercise and the addition of caffeine. **Accepted.**

Chapter Five

Discussion

Why does arm volume increase?

At rest, there is a steady flow of blood throughout the body. This constant flow allows nutrients and oxygen to reach the tissues and muscles. When vigorous muscular contractions occur, this causes a disruption in the working muscles. A strong muscular contraction causes the vessels that innervate the muscle to become slightly occluded, or compressed, temporarily cutting off the blood flow that is needed to compensate for the disruption that is occurring (14). While occluded, the muscle continues to contract. With each muscular contraction, the muscle is utilizing oxygen, producing CO_2, and increasing heat production within the muscle. This creates a hostile environment at the site. This causes a tremendous demand for O_2 and a desire to get rid of the waste products being produced. This high demand forces the arterioles and the pre-capillary sphincter muscles to prepare for the high surge of blood that is to come at the end of the muscular contractions. When the muscular contractions stop, blood rushes into the muscle and overcompensates by providing a high surge of blood, otherwise known as reactive hyperemia.

How does the body manage to yield a high surge of blood?

The human body is a network of arteries and veins responsible for delivering nutrients and oxygen to the working muscles. Arteries carry oxygenated blood while the veins carry the deoxygenated blood back to the heart and ultimately back to the lungs for the gas exchange. The redistribution of blood flow is an essential characteristic that the human body utilizes in order to overcompensate efficiently and effectively. Arterioles are the smallest arteries in our circulatory system and are primarily responsible for peripheral resistance. When the arterioles are open, the resistance is low and the blood flows smoothly. In contrast, when they are constricted, the resistance is high and the blood flood is impeded. Resistance exercise stimulates the sympathetic nervous system forcing it to release hormones such as epinephrine and norepinephrine to accelerate the heart rate. In order to ensure the working muscles that are being used get the proper amount of blood, the body will vasoconstrict the arterioles in areas that are not needing blood flow. Similarly, the autoregulation will vasodilate in the areas that are needing blood flow, such as the biceps, triceps, and all the other arm muscles used in this experiment. Furthermore, the amount of vasodilation will depend on factors that influence autoregulation. Factors such as a decrease in oxygen tension, increased carbon dioxide and nitric oxide. The recruitment of capillaries also assists autoregulation and are a key component regarding reactive hyperemia. The arterioles and pre-capillary sphincter muscles leading up to the muscles being used should be wide open. The pre-capillary sphincter muscles are the gatekeepers of the capillary beds. At rest, only about 5-10 percent of capillaries are open within a muscle. During exercise, the pre-capillary sphincter grant access to almost all the capillary beds in the working muscle, allowing the high surge of blood to reach the entire muscle.

Does caffeine have an effect on reactive hyperemia and work output?

Results of the study made it evident that the addition of caffeine during resistance exercise did have a moderate effect on the degree of reactive hyperemia and the amount of work output produced. This can be explained by describing motor units. The amount of motor units recruited during resistance exercise yield the strength of any muscular contraction. When caffeine is consumed, it further stimulates the sympathetic nervous system to recruit more motor units. The all or none principle states that when a muscle contracts, it does so at its maximal potential or not at all. Therefore, the greater amount of motor units recruited grants greater strength. With more strength to work with and the addition of caffeine allowed the participants to produce a higher work output than the control trial where no caffeine was consumed. In turn, with more work being produced, a higher degree of reactive hyperemia would occur because there would be more disruption within the working muscles. More disruption means that there is a higher demand for oxygen and a higher concentration of lactic acid and carbon dioxide. Therefore, the body overcompensates by providing the muscle with a higher degree of blood volume to the working muscle, hence reactive hyperemia.

Did work output increase because of an increased reactive hyperemia? Or, did reactive hyperemia increase because of a higher work output?

The data in this experiment raises the questions listed above. The answer to this would be that reactive hyperemia increased because of a higher work output. To explain this, the sympathetic nervous system was stimulated and forced to recruit more motor units with the addition of caffeine. The addition of more motor units produced a stronger force, allowing the participants to perform more repetitions, thus more work output. To continue, more work output means that there is more disruption occurring within the muscle. In the caffeine trial, the muscle was demanding more oxygen to be delivered due to the increased amount of work being produced. If this were true, then more blood flow would rush into the muscle to appease it.

Why does overcompensation occur?

During muscular contractions, the two contractile filaments engage with one another inside the muscle cell (7). These filaments, or proteins are called actin and myosin. When signaled to contracted repetitively, the muscle is torn down due to the amount of work being done. When a muscle is broken down, the body rebuilds each muscle fiber. Not only does the body rebuild the broken muscle, but it also overcompensates and adds additional muscle so that if the exercise was performed again, the muscle does not have to go through the same trauma. Similarly, the body performs the same protective mechanism in terms of reactive hyperemia. Reactive hyperemia is the immediate overcompensation to the broken down muscle. The high surge of blood into the exhausted muscle allows the body to commence the rebuilding process by providing nutrients, oxygen, and removing the waste products that were accumulated during the resistance exercises.

Conclusion

To conclude, the resistance exercise performed highlighted the blood flow response. The change in dominant arm volume was observed immediately after resistance exercise was performed. The high surge of blood was measured and increased the volume of the working muscles. It was concluded that the addition of caffeine prior to performing the same resistance exercises caused a higher degree of reactive hyperemia and a greater amount of work output. Therefore, the athletes who consume caffeine prior to resistance training will perform more repetitions and produce more work output. Additionally, they will experience a higher surge of blood to their muscles after completing said training which allows their body to overcompensate and begin the rebuilding process more effectively.

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Appendix

Consent Form

You will be asked to take part in a research study of the effects of caffeine on reactive hyperemia. I am asking you to take part in this study because I have asked you personally to be a participant. Please read this form carefully and ask me any questions you may have before agreeing to take part in the study.

What the study is about: The purpose of this study is to determine the effects of caffeine on reactive hyperemia.

What will you do: If you agree to be in this study, we will conduct two resistance exercise trials. These trials will be focusing on your bicep and triceps muscles in order to determine the difference in arm volume pre and post exercise. The work output will also be determined. The difference in the trials is as followed: The trials will simply have you come in an hour prior to the test in order to weigh you in. You will consume 12 ounces of orange juice and you will not know whether or not this drink contains caffeine or not. The amount of caffeine you will be given depends on your body weight. The caffeine dosage will be 3 mg per kg. Each individual exercise session will take roughly 45 minutes to complete.

Risks and benefits:

There is a risk that you may find after the workout has concluded. You may experience soreness but this is common after completing a workout session to failure.

There are no benefits to you other than improving your bicep and triceps strength.

Compensation:

There is no compensation for volunteering for this study.

Your answers will be confidential. The records of this study will be kept private between you and I. In the written report, I will not include any information that will make it possible to identify you. Research records will be kept with me at all times during the exercise trials and I will be the only one who has access to this.

Taking part is voluntary: Taking part in this study is completely voluntary. You may choose to stop the exercise session at any point if you have maximally exerted yourself or are no longer able to complete the task at hand.

If you have any questions: The researcher that will be conducting this study is Cristian Barron Lopez. Please ask if you have any questions now. If you have questions later, you may contact me at barronlopezc18@hanover.edu or at 1-260-350-7686. If I am not available, you may contact my instructor via email at stamford@hanover.edu.

You will be given a copy of this form to keep for your records.

Statement of Consent: I have read the information above, and have received or will receive answers to any of my questions I asked. I consent to take part in the study.

Your Signature ____________________________________ Date _________________________

Your Name (printed) ____

In addition to agreeing to participate, I also consent to having the interview tape-recorded.

Your Signature ____________________________________ Date __________________________

Signature of person obtaining consent _______________________________ Date ______________________

Printed name of person obtaining consent _______________________________ Date ______________________

Hanover College Institutional Review Board Approval

Study 2017124 approved by faculty sponsor

IRB-do-not-reply@hanover.edu
Thu 12/7/2017 12:54 PM
To: Cristian Barron Lopez

Reply

Inbox

This is a confirmation email letting you know about a change in the status of a human subjects application submitted to the Hanover College Institutional Review Board (IRB).

Study number 2017124, titled **Effects of Caffeine on Reactive Hyperemia**, listed you as

author

and the change in status is that the application has now been

approved by faculty sponsor

If you believe this is an error, please contact the webmaster for the IRB, Bill Altermatt, at altermattw@hanover.edu. The application has been submitted to the IRB. When the IRB review is complete, you will be notified by email. To review the content of the application and check on its status, you may log in at irb.hanover.edu. Remember not to begin data collection until you have received approval from the IRB. If you have any questions, please contac t the chair of the Hanover IRB, Dean Jacks, at jacks@hanover.edu.