THE EFFECT OF CAFFEINE GUM ADMINISTRATION ON BLOOD GLUCOSE AND BLOOD LACTATE DURING CYCLING TO EXHAUSTION

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THE EFFECT OF CAFFEINE GUM ADMINISTRATION ON BLOOD GLUCOSE AND BLOOD LACTATE DURING CYCLING TO EXHAUSTION

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Thesis

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ABSTRACT

The use of caffeine as a performance-enhancing supplement is on the rise due to caffeine's known ergogenic effect. Caffeine is often administered as a liquid or in a capsule, but new research has shown that caffeine gum has a faster absorption rate and a higher relative bioavailability. The purpose of this study is to determine if caffeine gum elicits the same ergogenic effect as a liquid or a capsule by examining blood glucose and blood lactate levels during cycling to exhaustion. Eight (n=8) recreationally active males volunteered to participate in the study. Two hundred milligrams of caffeine gum or a placebo was administered 35 minutes prior to exercise, 5 minutes prior to exercise, or 15 minutes into exercise. Participants cycled at 85% of their VO_{2max} until volitional fatigue. Blood glucose and blood lactate were taken via a finger prick immediately upon entering the laboratory, after the first gum administration, and every 10 minutes during exercise. The results indicated that blood glucose (p=.0432) and blood lactate (p=..934) levels over time with the caffeine versus the placebo were not significant. Also, there was no significance in blood glucose (p=.736) or blood lactate (p=.758) levels when comparing the time administered caffeine trials. This was the first exercise study conducted that administered caffeine gum and should be used as a starting point for future research on caffeinated gum and exercise.

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INTRODUCTION

Endurance athletes are continually searching for a training method and/or supplement to enhance their ability to train and compete. Some of these training methods and/or ergogenic aids include altitude training, carbohydrate loading, blood doping, growth hormone, and synthetic erythropoietin. While these are all effective ergogenic aids, not all of them are legal.

For many years caffeine has been one of the most widely consumed drugs in the world.[18] In moderate doses (<300 mg) caffeine is well tolerated and few significant side effects have been reported.[45,46] Caffeine is becoming increasingly popular among athletes due to its ergogenic effect during exercise. Caffeine has been added to many sports drinks and gels, which makes it easily accessible during athletic competition. While many supplements that enhance exercise are banned, the International Olympic Committee (IOC) considers caffeine a "controlled or restricted" substance. The IOC allows limits of 12 μg of caffeine per milliliter of urine. The National Collegiate Athletic Association (NCAA) sets their upper limit at 15 μg/ml. These are relatively high limits and will allow athletes to consume caffeine before competition within the legal limits.

STATEMENT OF THE PROBLEM

The focus of caffeine research has been on the ergogenic effect of the liquid and capsule delivery methods. In order to determine the mechanism by which caffeine elicits this ergogenic effect, the metabolic effects are often studied during exercise. Cox et al. studied the effects of 6 mg/kg of caffeine in capsule form and Coca-Cola on metabolism and endurance performance in twelve highly trained male cyclists or triathletes.[17] Researchers found that the 6 mg/kg of caffeine enhanced performance during a time trial at the end of a prolonged cycling bout.[17] Researchers also found that the Coca-Cola enhanced exercise performance despite having a low level of caffeine.[17] A study by Van Soeren and Graham examined the effects of caffeine on exercise metabolism after withdrawal. This study delivered caffeine in the form of a capsule to recreational athletes. The results indicated that caffeine increased exercise time in all exercise trials when compared to the placebo.[55] These two studies are very similar to the majority of caffeine research which studies the metabolic and performance effects of caffeine in liquid or capsule form.[4,6,7,16,20,24-27,30,31,41,49,52]

The problem with administering caffeine as a liquid or in a capsule is the timing. Once ingested, caffeine must travel through the gastrointestinal system and be completely absorbed before entering the bloodstream. While caffeine is a rapidly absorbed drug, it does take time to peak in the blood. Therefore, caffeine must be ingested 45 to 60 minutes prior to exercise in order to elicit an ergogenic effect.

Gum is one of the more recent items caffeine has been added to. While no exercise studies have been performed using caffeinated gum, there have been studies that

examined the catecholamine effects on sleep deprivation.[34,35,50] Caffeine gum has proven to be a successful aid for those who need to stay awake especially for a prolonged period.[34]

The major difference between the gum and a liquid/capsule is the amount of time it takes to be absorbed. Caffeine gum has a much quicker absorption time and a higher relative bioavailability.[35] Eighty-five percent of the caffeine in the gum is absorbed sublingually within five minutes.[38] Absorption via the buccal cavity will allow caffeine to bypass the gastrointestinal system and be released directly into the bloodstream. This fast absorption rate and relative bioavailability could mean more of an ergogenic effect during exercise.

STATEMENT OF PURPOSE

Since this is the first study exercise study being performed using caffeinated gum, the intent of this study is to determine if caffeine gum elicits more of an ergogenic effect during submaximal, exhaustive exercise than other forms of caffeine. The purpose of this study is to determine if caffeine gum will have more of an effect on blood glucose and blood lactate levels during cycling to exhaustion. Based on previous research, the researcher hypothesizes that blood glucose and blood lactate will increase with caffeine versus the placebo. It is also hypothesized that there will be a differential effect in blood glucose and blood lactate. Levels will increase depending on if the caffeine was administered at -35 minutes, -5 minutes, or at +15 minutes.

The results of this study could prove to be beneficial for athletes who want to ingest a legal supplement before exercise or competition. Athletes often have tight

schedules, and since caffeine gum is quickly absorbed and readily available, there would be no need to worry about ingesting the caffeine 45 to 60 minutes prior to exercise. This would greatly benefit athletes.

CHAPTER II

REVIEW OF LITERATURE

CAFFEINE

Caffeine is a substance that is found naturally in many plants and is often found in many of the drinks and foods we consume.[3] The widespread use of caffeine is continually on the rise as we can easily find caffeine in coffee, tea, soda, chocolate, energy drinks and gels, gum, and pharmaceutical drugs. Drug companies are mixing caffeine with stimulants, pain relievers, diuretics, cold remedies, diet pills, bronchial and cardiac stimulants, and acne and other skin disorder medicines.[3] For this reason, caffeine is the most widely consumed substance in the world.[23]

Pharmacology

The pharmokinetics of a substance refers to the biological fate in a living organism, and is a composite of the processes of absorption, distribution, metabolism, and excretion.[32,33] Pharmokinetics describes the time course of these processes, including the initial compound and its various metabolites in the fluids, tissues, and excreta of the organism.[32] Once ingested, caffeine is rapidly and completely absorbed from the gastrointestinal tract into the bloodstream.[3,32,33] Peak plasma concentration can be reached in 40 to 60 minutes but the presence of food in the intestine or ingesting large amounts of the drug can slow down the rate of absorption.[3,39,43]

Absorption and Distribution

Once ingested, caffeine is readily distributed throughout the entire body.[42] The hydrophobic properties of caffeine allow its passage through all biological membranes.[23] Human studies have shown that there are no physiological barriers limiting the passage of the drug through tissues and there is no build up of the drug in body tissues.[10,57]

Elimination

The elimination half-life of caffeine varies between three and seven hours with an average of five hours.[28,42] There are a variety of factors that can affect the rate of elimination, such as age, gender, pregnancy, disease, and use of other substances. Gender differences have been observed by researchers who found that the half-life of caffeine for females was 20-30 percent shorter than for males.[14] It was also noted that the half-life of caffeine in contraceptive users was almost twice as long as ovulating females.[14] A slightly shorter plasma caffeine half-life in older men (66 to 78 years) compared to younger men (19 to 24 years) has also been observed.[9] The clearance rate for caffeine decreases during the course of pregnancy, resulting in an almost threefold increase in the plasma half-life during the third trimester.[1] Enzymes in the liver primarily metabolize caffeine; therefore, liver disease can affect the rate of caffeine clearance.[15]

Metabolism

Caffeine is metabolized by the liver and is regulated by the cytochrome P-450 enzyme system. The process of biotransformation, which is the chemical conversion of substances by living organisms or enzyme preparations, includes the demethylation of

caffeine (1,3,7 dimethylxanthine), resulting in three dimethlyxanthine metabolites, paraxanthine (1,7 dimethylxanthine), theobromine (3,7 dimethylxanthine), and theophylline (1,7 dimethylxanthine).[33]

Each of the three dimethylxanthines: paraxanthine, theobromine, and theophylline have their own specific effect on the body. Paraxanthine, the major metabolite of caffeine in humans, is known to increase lipolysis, leading to elevated glycerol and free fatty acid levels in blood plasma. Theobromine dilates blood vessels and increases urine volume and theophylline relaxes smooth muscles of the bronchi. One study reported that paraxanthine accounted for 84% of the demethylations, theobromine 12%, and theophylline 4%.[36]

Multiple pathways are responsible for metabolizing caffeine. These alternative pathways have been established from the metabolites identified in urine.[2] Caffeine is metabolized through a series of reactions. It is completely transformed, with less than 2% of ingested caffeine being recoverable in urine unchanged.[3] The dimethylxanthines are metabolized to dimethyluric acid or monomethylxanthines. The monomethyl-xanthines are then converted to monomethyluric acids.

Mechanisms of Action

There are various reasons that people choose to consume caffeine including staying alert and increasing energy levels. Since the drug is consumed by a large number of people, it is important to note the actions the drug has on the body. Many researchers have sought to understand the cellular mechanisms of caffeine but have only been able to hypothesize the mechanisms of action. The three major hypothesized mechanisms are:

translocation of intracellular calcium, mediation by increased accumulation of cyclic adenosine monophosphate (cAMP) due to inhibition of phosphodiesterase, and competitive blockade of adenosine receptors.[32]

Research has found that caffeine increases the release of calcium from stores in the sarcoplasmic reticulum but may also impair calcium re-uptake into the stores.[40,42] The biochemical mechanism that underlies the actions of caffeine at doses achieved in normal human consumption must be activated at concentrations between the extremes (between barely effective doses and doses that produce toxic effects).[23] This means that the direct release of intracellular calcium, which occurs only at millimolar concentrations, cannot explain the general effects of dietary caffeine.[23]

Caffeine has been found to indirectly facilitate the accumulation of cyclic AMP (cAMP). Cyclic AMP is an adenosine 3', 5'-monophosphate in which the phosphate group forms an ester bond with OH groups attached to both the 3' and 5' carbon atoms of sugar ribose. It has been suggested that caffeine stimulates brain norepinephrine by inhibiting phosphodiesterase, the enzyme that catalyzes the breakdown of cAMP .[13] After the release of epinephrine from the adrenal medulla, the concentration of cAMP increases, but once epinephrine is removed cAMP falls. Caffeine inhibits phosphodiesterase by increasing the persistence of cAMP in cells stimulated by epinephrine, thereby prolonging or intensifying the activity of epinephrine.[48] However, the effect of caffeine on cAMP and the inhibition of cyclic nucleotide phosphodiesterase occurs at rather higher concentrations than those attained during human caffeine consumption and henceforth, have limited importance.[23]

Of the three hypothetical mechanisms, the effect of caffeine on adenosine receptors is the most plausible mechanism for dose levels achieved during typical usage of the drug.[23] Adenosine is a widely present compound in purinergic receptors that regulates biological processes throughout the body, including the cardiovascular and nervous systems.[12,56] Caffeine has a stimulatory effect on blood pressure, rennin release, catecholamine release, lipolysis, respiration, intestinal peristalsis, and urine output while adenosine has an antagonistic effect on these mechanisms. There are two main types of adenosine receptors: the A_1 or high-affinity receptor and the A_2 or low-affinity receptor.[54] Adenosine inhibits adenyl cyclase through the A_1 receptor, while showing an excitatory effect through the A_2 receptor.[19] Caffeine is a competitive inhibitor of adenosine because it binds to the adenosine receptor but does not trigger the chemical cascade that inhibits neurotransmitter release. It blocks the site so adenosine cannot bind and get its message across the synapse. By inhibiting adenosine, caffeine excites the central nervous system and allows for continued stimulation of neurons that otherwise would not fire or would not release neurotransmitters into the synapse.[21,22,37,44] The reduction in adenosine activity results in increased activity of the neurotransmitter dopamine, largely accounting for its stimulatory effects. This inhibition of adenosine is the only known biochemical effect that caffeine has in humans at the concentrations achieved during normal human consumption of the drug.

CAFFEINE GUM

Caffeine is readily available in many food, drinks, and capsules but one does not always have access to one of these forms. Recently, Amurol Confectioners (Yorkville,

IL) developed *Stay Alert*, a caffeinated chewing gum. This gum was developed to provide a quick and convenient source of caffeine in a portable way.[35]

Rate of Absorption

The formulation of a drug can directly influence its dissolution, and the rate and extent of absorption after it has been orally ingested. A gum formulation has many advantages over any other method of delivery. The caffeine from gum is released through mastication and is absorbed through the buccal cavity. Absorption through the buccal cavity is considered faster, especially for lipophilic agents such as caffeine, due to its rich vascular supply.[47] The actions of a drug, once it has been ingested, is usually dependent on speed of delivery; therefore, a faster absorption results in a shorter duration for a dynamic response.[35] Also, absorption of a drug through the buccal cavity will bypass intestinal and hepatic first pass metabolism, which potentially increases the extent of absorption.[47]

GUM VERSUS CAPSULES

The focus of previous research studies has been on the comparison of caffeine doses and modes of delivery (capsule, oral solution, gum) to determine which is the safest, most reliable, and most rapidly absorbed. A study conducted by Novum of Amurol Confectioners concluded that the absorption rate of caffeine was faster when it was administered in the chewing gum as compared with a standard tablet.[38] Research has also shown that approximately 85% of the caffeine is delivered by five minutes of chewing.[38] This would lead to a quicker onset of caffeine's stimulatory effects.[35] A study by Kamimori et al. observed the rate of absorption and relative bioavailability of caffeine in chewing gum versus capsules.[35] Eighty-four male participants were

administered a placebo, 50, 100, or 200 mg of caffeine as chewing gum, or 50, 100, 200 mg of caffeine in capsule form. The results indicated that both the gum and capsule released a comparable amount of caffeine into the system, but the absorption rate for the gum was significantly faster.[35] Another study by Syed et al. examined the pharmokinetics of *Stay Alert* chewing gum in multiple doses.[50] Forty-eight participants were administered a placebo or 50, 100, or 200 mg of *Stay Alert* chewing gum. The results demonstrated that maximum caffeine concentration was nearly the same as the study comparing caffeinated chewing gum to capsules. Also, the rate of absorption was similar to the chewing gum in the previous study, which indicates that the faster rate and extent of absorption with a single dose is maintained across repeated doses.[50]

CAFFEINE AND EXERCISE

Caffeine has become a popular supplement for athletes because of its ergogenic effect. A supplement that has an ergogenic effect is known to enhance athletic performance. Many studies have observed caffeine's ergogenic effect on submaximal endurance exercise. Endurance exercise is frequently used because power can be kept constant and exercise time can be quantified.[26]

Ergogenic Aid

Many researchers have sought to determine the exact mechanism of caffeine's ergogenic effect. Early research has shown that caffeine increases plasma epinephrine, which in turn increases serum free fatty acids (FFAs) and spares muscle glycogen.[16,20,30,49] The focus of research has been on trying to establish the exact mechanism using the hypothesis that caffeine ingestion results in a rise in circulating catecholamines, which mobilizes FFAs from adipocytes, thus increasing the amount of

fat available for active muscle.[16,20,30] The ergogenic effect was thought to be a result of utilization of fat, which would result in the sparing of muscle glycogen and a lowering of the respiratory exchange ratio (RER).[16,20] Since fat provides the most energy, utilizing more fat and less glycogen during exercise, would allow one to exercise longer. But recent research has not been able to prove this hypothesis.

Endurance Exercise

Researchers have performed a variety of studies supplementing caffeine during endurance exercise to try and determine the best time to administer caffeine and to determine the effects caffeine has on substrate metabolism during exercise.[4,7,16,24-27,31,49,55] A study by Cox et al. observed the effects of different protocols of caffeine intake on competitive athletes during endurance exercise.[17] Twenty competitive athletes cycled at steady state exercise (70% VO_{2max}) for two hours. The results showed caffeine enhanced performance by approximately 3% compared to the placebo.[17] In another study by Van Soeren and Graham the effect of caffeine on metabolism, exercise endurance, and catecholamine response in recreational male athletes was examined.[55] Participants in this study were required to cycle to exhaustion at 80-85% VO_{2max}. The results indicated that time to exhaustion was significantly increased in all of the caffeine trials versus the placebo.[55]

BLOOD GLUCOSE

Blood glucose, which is also referred to as blood sugar, is the body's primary source of energy. It is released from the intestines and taken into the body's cells via the bloodstream. Glucose levels are strictly regulated in the body. Normal blood glucose

concentration ("euglycemia") is around 80-120 mg/dl. After eating this level can climb to around 140 mg/dl. Normal fasting glucose concentration is around 70-90 mg/dl.

Effect During Exercise

During exercise, glucose homeostasis can be harder to maintain because skeletal muscle suddenly switches from a situation of little glucose uptake to a situation of greatly increased uptake.[11] The liver, which is the main organ of glucose production, must increase the amount of glucose it is releasing. At the onset of exercise there is a rise in glucose. As exercise continues, levels will decrease, but remain within 10% of normative value.[11]

Glucose Transporters

The ability of our cells to take in glucose depends on several factors, including the type of tissue, the levels of glucose in the blood and tissue, the presence of the hormone insulin, and the physiological status of the tissue.[11] In order for glucose to enter a cell, a transporter is needed. A transporter is a protein molecule that will allow glucose entry across the cell membrane.[29] When a substance is transported down its concentration gradient and across a membrane, it is called facilitated diffusion. There are specific membrane transporters that exist for glucose. There are five glucose transporters (GLUT) that are tissue specific.[29]

Glucose must enter the cell through a transporter in order to be metabolized. Entry into some cells is regulated while other is unregulated.[29] Cells, such as brain, kidney and blood, have unregulated glucose entry because they rely upon glucose as their main source of fuel. Other cells, such as liver, heart, adipose, and muscle have regulated uptake and rely on the GLUT-4 transporter, which is regulated by insulin.[29]

LACTATE

Lactate is a byproduct of metabolism and is often used as a marker of exercise intensity. During aerobic metabolism, glycogen is broken down (glycolysis) and pyruvate is formed. Pyruvate has the ability to enter the mitochondria, which is the powerhouse of the cell, or be turned into lactate via the enzyme lactate dehydrogenase (LDH). During normal exercise, the rate of lactate production and clearance is equal. During moderate intensity exercise, lactate production can rise while lactate clearance may decrease. As exercise intensity increases linearly beyond 50% to 60% of VO_{2max}, the rate of glycolysis, expressed as the rate of pyruvate formation, increases at an even faster rate (Houston, 2006). Resting blood lactate levels are around 1-2 mmol/L but can reach up to 20 mmol/L during intense exercise.

Red blood cells also have the ability to produce lactate. So, blood lactate reflects erythrocyte metabolism and lactate generated through glycolysis by skeletal muscle.[29] As lactate circulates in the blood it can diffuse down a concentration gradient and enter cells or be used by the heart, liver, kidney, and nonworking skeletal muscle.[29] Blood lactate levels during exercise reflect the balance between lactate-producing cells (predominantly active muscle fibers) and those tissues and cells that consume lactate as a fuel (heart and nonworking muscle) or convert it to glucose through the liver and kidney.[29]

CHAPTER III

METHODS

PARTICIPANTS

Eight (n=8) apparently healthy, physically active males were recruited to participate in this study. Participant profile is located in table 2.1. Participants were non-to-moderate caffeine users (<300 mg/day) who were not currently taking medication or utilizing supplements of any kind. Participants exhibited a body fat percentage below 30% as measured via a Dual Energy X-ray Absorptiometry (DEXA) scan (Hologic QDR 4500 Elite-Acclaim Series, Hologic, INC., Bedford, MA). The DEXA was administered at the Kent State University Health Center by a licensed radiation technician. Participants were excluded from the study if they had a history of smoking, signs or symptoms of cardiovascular, metabolic, or respiratory disease, or if they are known to have any cardiovascular, metabolic, or respiratory disease as determined via a health history questionnaire. All participants were fully informed of the nature and possible risks of the investigation before giving their written consent. The Internal Review Board at The University of Akron and Kent State University approved the protocol.

Table 3.1 Participant Profile

Participant	Age (yrs)	Height (cm)	Weight (kg)	VO2max (ml/kg/min)
1	30	175	82.3	47.5
2	21	188	110.0	44.5
3	32	175	66.8	52.0
4	22	175	106.8	44.5
5	28	178	78.5	43.4
6	23	178	75.7	52.2
7	24	178	72.8	46.0
8	24	175	94.0	34.1
Average	25.5	177.7	85.8	45.5

PRE-EXPERIMENTAL PROCEDURE

On the first visit to the exercise science laboratory participants height and weight were measured and an incremental maximum oxygen uptake (VO_{2max}) test was performed. The test consisted of cycling on an electronically braked cycle ergometer (Excalibur 1300W) until volitional fatigue at which time expired air samples were analyzed for oxygen and carbon dioxide concentrations via an automated open circuit system (PARVO, Metabolic Cart, Sandy, Utah) to determine maximal oxygen consumption. A Hans Rudolph three-liter syringe (Hans Rudolph Model 5530, Hands Rudolph, Inc., Kansas City, MO) and known concentration calibration gases were used to calibrate the automated open circuit system prior to each VO_{2max} test. During the graded exercise test, heart rate was continuously measured via a Polar heart rate monitor (Accurex Plus, Polar Electro, Inc., Woodbury, NY) and heart rate (HR) values were recorded via telemetry every 30 seconds. The two highest 30-second oxygen

consumption values were averaged to determine maximal oxygen consumption. Each participants seat and handlebar measurements were recorded to obtain consistency throughout all trials.

Before the cycling trials began, participants came into the laboratory for a trial to determine power output for their cycling trials based on the results of their VO_{2max} test. Participants expired air samples were analyzed for oxygen and carbon dioxide concentrations via a metabolic cart while cycling. Participants cycled until they reached 85% of their maximum oxygen consumption. The participant's power output at this time was recorded and used during the cycling trials.

EXPERIMENTAL PROTOCOL

The next four visits to the laboratory consisted of participants cycling to volitional fatigue, that is, until they fail to maintain the required work output. Each trial was separated by a one-week washout period. Prior to reporting to the laboratory, participants were requested to refrain from eating anything for at least eight hours. Upon arriving at the laboratory, the subject put on a heart rate monitor and rested in a chair in an environmental chamber that was set at 23^{o} Celsius for 50 minutes. The chamber remained at 23^{o} C for all exercise trials. Forty minutes and 10 minutes prior to exercise resting measurements were taken. Blood glucose was measured via a glucometer (Roche Diagnostics, Accu Chek Active, Indianapolis, IN) and blood lactate was measured using a lactate pro analyzer (Arkray, Inc., Lactate Pro Analyzer, Tokyo, Japan). Both blood glucose and blood lactate measurements were taken using a finger prick. Resting blood pressure (BP), HR, and oxygen consumption were also recorded. Ratings of Perceived Exertion (RPE) measurements were recorded via Borg's RPE 6-20 scale.

Two pieces (200 mg of caffeine) of Stay Alert chewing gum (Amurol confectioners, Specialty gum subsidiary of Wrigley's, Yorkville, IL) or a placebo were administered at three time points (35 minutes pre-exercise, 5 minutes pre-exercise, and 15 minutes into exercise). The participants were instructed to chew for five minutes. The gum was administered in a randomized, counterbalanced, double blind manner.

Five minutes pre-exercise, the participant began a warm-up on the electronically braked cycle ergometer with a power output of 50 Watts. During this time, the participant's power output was continually ramped until he reached the appropriate wattage. The participants were instructed to cycle between 60-80 rpms. They were told to not cycle above 120 rpms and the test would stop if they cycled below 40 rpms. At 5, 20 and 25 minutes and 30 and 35 minutes if needed, participants expired air samples were analyzed for oxygen and carbon dioxide concentrations via the metabolic cart to determine oxygen consumption. Heart rate was measured and recorded every minute during the first 5 minutes of exercise to ensure a steady increase. Heart rate was then recorded every 5 minutes during exercise. Every 10 minutes during exercise RPE, BP, blood glucose, and blood lactate measurements were taken and recorded.

Participants cycled until they reached exhaustion. Exhaustion was defined when the participants could no longer cycle or keep above 40 rpms.

STATISTICAL ANALYSES

Since this was a repeated measures study in which the subjects were not all measured at all time points a Repeated Measures ANOVA was not applicable. Therefore, to analyze this study a Mixed Model analysis was conducted. Mixed Models i

also referred to as HLM and Individual Growth Models allows one to model the appropriate covariance matrix while allowing for different test time for each subject.

LIMITATIONS OF THE STUDY

The limitations of this study were the dose of caffeine, the number of participants, the fitness level of the participants, and the use of finger pricks to obtain blood glucose and blood lactate measurements.

For this study participants were given a 200 mg dose of caffeine. There could be a few limitations to this dose. First, this is a fairly small dose considering caffeine is usually dosed by participant's body weight. Since every subject was given the same dose but varied in body weight, the caffeine may have more of an effect on one subject than another. Secondly, our exclusion criterion for caffeine intake was less than 300 mg a day. If participants were able to ingest at least 300 mg then 200 mg would not have much of an effect on them due to habituation.

Another limitation is the number of participants. This study was only able to test eight participants due to schedules and time. While many exercise studies do not use a large number of participants, testing a few more people may have varied the results.

The fitness level of the participants could also be considered a limitation. Caffeine is known to have the greatest ergogenic effect on trained athletes. Our participants were recreationally active males. A few participants were cyclists/endurance athletes, while others were weight lifters. Cycling at 85% of VO2max is not an easy task especially if you train anaerobically. Hence, the caffeine may have not even had enough of an effect on some participants.

The last limitation involved the use of the finger prick to obtain blood glucose and blood lactate measurements during cycling. Cycling at 85% VO2max required participants to maximize the use of the handlebars. Since measurements were taken every ten minutes during exercise, near the end of exercise it was hard for the participants to not hold on to the handlebars in order to obtain a finger prick. Inserting a catheter into an antecubital vein would be much easier but is more invasive, expensive, and requires the expertise of a nurse.

CHAPTER IV

RESULTS AND DISCUSSION

BLOOD GLUCOSE

The first hypothesis tested stated that blood glucose would increase with caffeine when compared to the placebo. This hypothesis was not statistically significant ($p=0.432$) with an $F=0.622$ and a $R^2=0.003$. As can be seen in figure 4.1, the blood glucose levels were very similar.

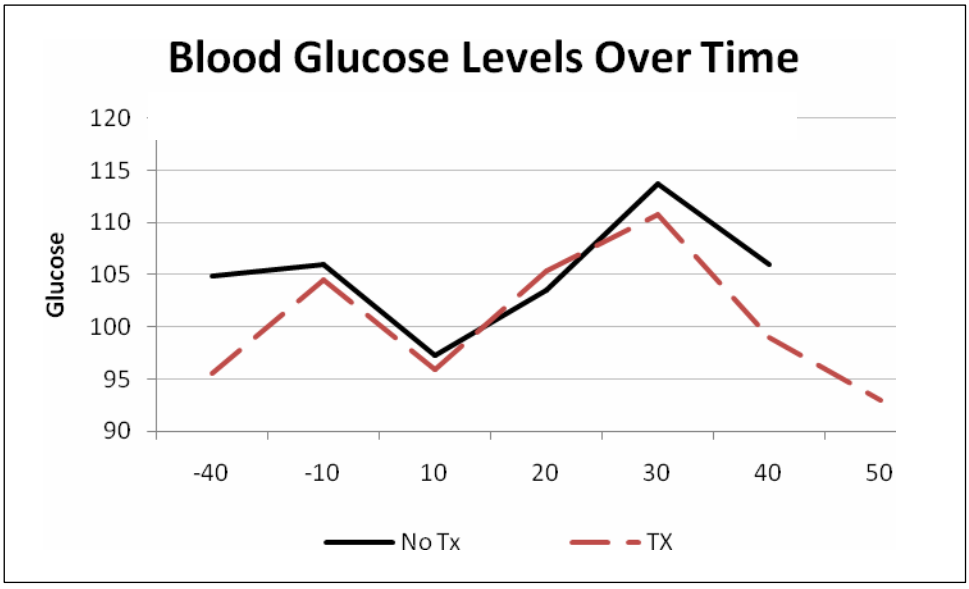

Figure 4.1 Blood Glucose Levels with Caffeine versus the Placebo

The second hypothesis was analyzed to see if there was a differential effect in blood glucose. The hypothesis stated that levels would increase depending on if the caffeine was administered at -35 minutes, -5 minutes, or at +15 minutes.

This hypothesis was not statistically significant (p=0.736) with an F=0.425 and a R^2=.006. As can be seen in figure 4.2, the blood glucose levels were very similar.

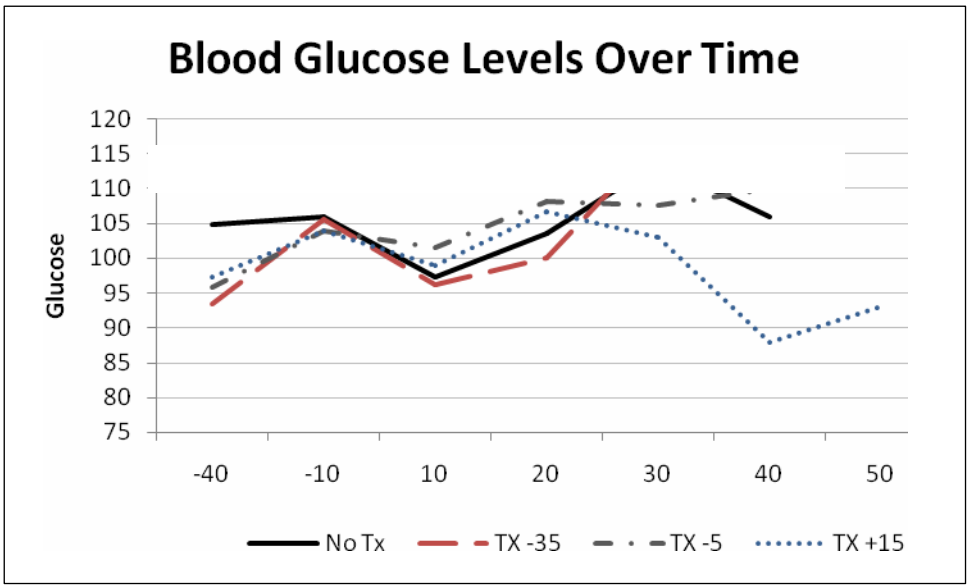

Figure 4.2 Blood Glucose Levels with Time Administered Caffeine

Table 4.1 Blood Glucose Levels in mg/dl with Placebo

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	97	104	77	80	116	-	-
P2	93	100	89	87	99	106	-
P3	110	110	105	131	162	-	-
P4	103	102	115	122	-	-	-
P5	117	117	113	124	-	-	-
P6	91	95	65	77	78	-	-
P7	123	114	117	-	-	-	-
P8	90	104	90	97	103	-	-
Average	103.0	105.7	96.3	102.5	111.6	106.0	-

Table 4.2 Blood Glucose Levels in mg/dl with Caffeine at –35

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	80	88	77	82	93	-	-
P2	85	91	99	100	108	-	-
P3	98	110	122	133	139	-	-
P4	104	112	106	-	-	-	-
P5	101	110	87	98	-	-	-
P6	100	114	92	102	153	-	-
P7	92	113	91	-	-	-	-
P8	88	106	96	85	92	-	-
Average	93.5	105.5	96.2	100.0	117.0	-	-

Table 4.3 Blood Glucose Levels in mg/dl with Caffeine at –5

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	93	97	89	94	90	-	-
P2	96	99	99	119	122	-	-
P3	100	97	124	108	-	-	-
P4	93	98	99	127	-	-	-
P5	100	110	93	102	107	110	-
P6	99	111	58	89	107	-	-
P7	89	105	95	118	-	-	-
P8	96	115	155	109	112	-	-
Average	95.7	104.0	101.5	108.2	107.6	110.0	-

Table 4.4 Blood Glucose Levels in mg/dl with Caffeine at +15

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	88	82	71	84	93	88	93
P2	91	93	92	104	113	-	-
P3	109	109	137	191	-	-	-
P4	100	109	89	104	-	-	-
P5	100	125	127	103	-	-	-
P6	77	95	67	68	-	-	-
P7	112	112	113	-	-	-	-
P8	101	106	95	93	-	-	-
Average	97.2	103.8	98.8	106.7	103.0	88.0	93.0

Caffeine and Blood Glucose during Exercise

Caffeine is known to have an effect on blood glucose levels during exercise. There has been much debate about the exact mechanism of caffeine on glucose during exercise. The finding that caffeine ingestion does not change the RER during exercise means that the combined amount of muscle glycogen and blood glucose oxidized is unaltered.[24,27,51]

The lack of statistical significance with blood glucose levels over time and groups in the present study is similar to results found in previous research. Van Soeren and Graham studied six recreational athletes who were habitual caffeine users. The participants were required to abstain from caffeine prior to the study. The researchers found that caffeine had no effect on blood glucose levels after withdrawal during exercise to exhaustion.[55] Another study by Battram et al. observed the effect of caffeine on glucose kinetics in twelve recreationally active males and found that caffeine did not have an effect on endogenous glucose production.[6] Bangsbo et al. observed the acute and chronic

responses to caffeine and exercise in healthy adults. Twelve active long-distance runners were given 500mg of caffeine before a training session. When compared to the control group, researchers found that blood glucose levels were not altered with the caffeine.[4] These results are similar to two other studies that found caffeine did not alter blood glucose levels when given various doses of caffeine.[24,25]

While many studies have found blood glucose levels to remain unchanged there have been studies that have found caffeine to cause a rise in blood glucose.[17,41,53] Graham and Spriet examined the exercise responses to various doses of caffeine in well-trained endurance athletes. The athletes were given 3, 6, or 9 mg/kg of caffeine. The results indicated that at rest blood glucose concentration did not change between treatments. During exercise the concentration rose and was greater at 15 and 30 minute in the 6 mg/kg trial than in the placebo and the 9 mg/kg trial was higher at 15 minutes than in the placebo.[25]

The reason blood glucose did not show any statistical significance with the gum in the present study could be due to the small dose of caffeine the participants received. The dose given may not have been large enough to cause a significant change in blood glucose during cycling, even though a larger amount of caffeine may have been absorbed through mastication of the gum. The study by Graham and Spriet, which found an increase in blood glucose levels, used larger doses of caffeine than the present study. While there is much debate on caffeine's effect on blood glucose levels, one must consider that all theses previous studies have used caffeine in capsule or liquid form.

BLOOD LACTATE

The third hypothesis stated that blood lactate levels would increase with caffeine when compared to the placebo. This hypothesis was not statistically significant (p=0.934) with an F=0.007 and a R^2=0.000. As can be seen in figure 4.3, the blood lactate levels increase almost identically regardless of caffeine treatment.

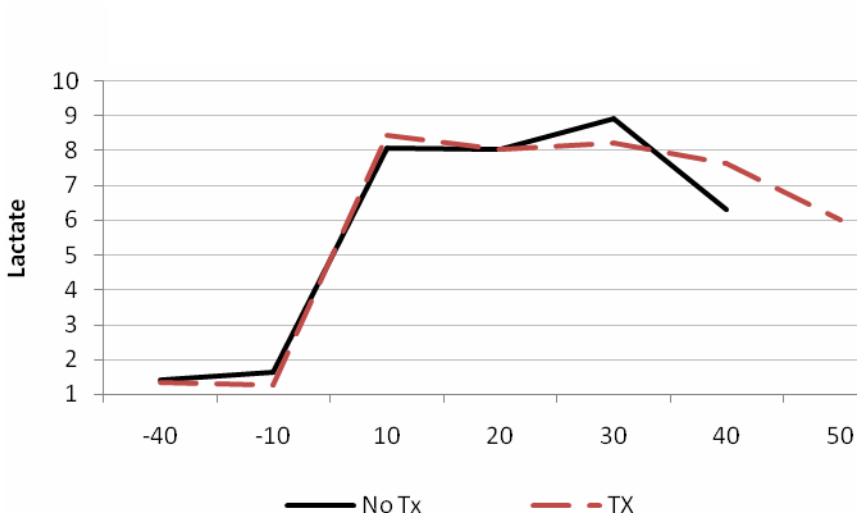

Figure 4.3 Blood Lactate Levels with Caffeine versus Placebo

The final hypothesis was analyzed to see if there was a differential effect in blood glucose. The hypothesis stated that levels would increase depending on if the caffeine was administered at -35 minutes, -5 minutes, or at +15 minutes. This hypotheses was not statistically significant (p=0.758) with an F=0.394 and a R^2=0.004. As can be seen in figure 4.4, blood lactate levels increase almost identically regardless of caffeine treatment.

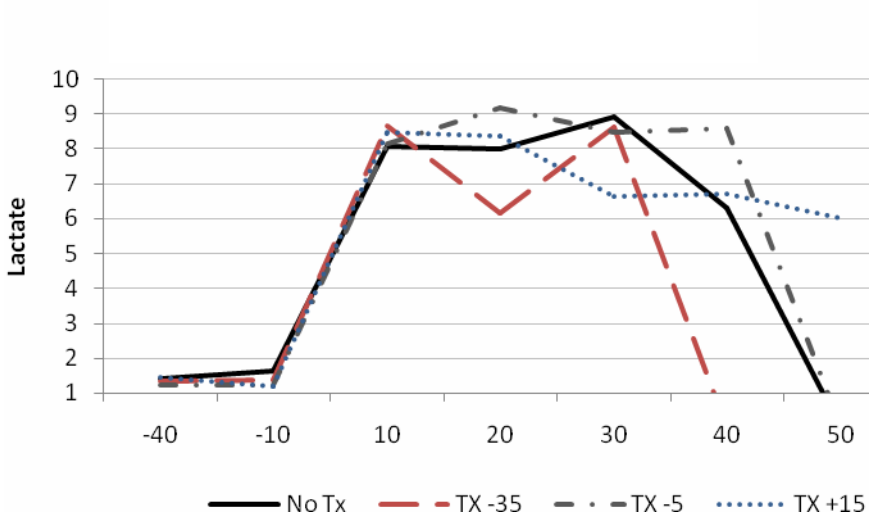

Figure 4.4 Blood Lactate Levels with Time Administered Caffeine

Table 4.5 Blood Lactate Levels in mmol/L with Placebo

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	0.9	0.9	3.3	9.0	12.6	-	-
P2	1.4	1.2	4.6	5.4	6.4	6.3	-
P3	0.8	1.1	7.8	9.0	10.8	-	-
P4	0.8	0.8	14.7	7.7	-	-	-
P5	1.6	2.7	9.1	12.4	-	-	-
P6	2.6	2.3	6.7	4.6	5.9	-	-
P7	1.8	2.4	10.3	-	-	-	-
P8	1.6	1.0	5.0	6.1	6.1	-	-
Average	1.4	1.5	7.6	7.7	8.3	6.3	-

Table 4.6 Blood Lactate Levels in mmol/L with Caffeine at -35

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	0.9	1.2	2.2	3.7	8	-	-
P2	1.0	1.2	4.6	2.3	8.2	-	-
P3	1.3	1.2	11.1	7.6	8.7	-	-
P4	1.8	0.9	20.6	-	-	-	-
P5	1.2	1.2	7.3	8.1	-	-	-
P6	1.2	2.8	8.7	8.2	10.3	-	-
P7	1.0	1.0	9.2	-	-	-	-
P8	2.4	1.4	5.6	7.0	8.0	-	-
Average	1.3	1.3	8.6	6.1	8.6	-	-

Table 4.7 Blood Lactate Levels in mmol/L with Caffeine at -5

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	1.0	1.0	7.0	7.3	13.0	-	-
P2	1.8	1.1	4.7	6.6	6.8	-	-
P3	1.1	0.9	9.1	9.9	-	-	-
P4	0.9	2.2	12	8.6	-	-	-
P5	0.8	0.8	6.1	8.1	7.2	8.6	-
P6	1.2	1.7	6.6	7.1	8.1	-	-
P7	1.1	1.0	11.9	20.7	-	-	-
P8	1.9	1.0	7.9	5.0	7.2	-	-
Average	1.2	1.2	8.1	9.1	8.4	8.6	-

Table 4.8 Blood Lactate Levels in mmol/L with Caffeine at +15

	Time						
Participant	**-40**	**-10**	**10**	**20**	**30**	**40**	**50**
P1	1.0	1.2	6.6	5.6	6.9	6.7	6.0
P2	1.8	1.8	5.0	7.1	6.4	-	-
P3	2.8	1.6	10.8	17.9	-	-	-
P4	1.0	0.8	9.9	7.8	-	-	-
P5	1.3	1.1	4.6	4.9	-	-	-
P6	1.6	1.0	9.9	6.4	-	-	-
P7	1.2	1.0	13.1	-	-	-	-
P8	1.0	1.1	7.8	8.9	-	-	-
Average	1.4	1.2	8.4	8.3	6.6	6.7	6.0

Caffeine and Blood Lactate during Exercise

Research on caffeine's effect on blood lactate during exercise is not often discussed and when it is, the findings are conflicting with the theory that caffeine is glycogen sparing. Most research shows that blood lactate concentration increases after the ingestion of caffeine.[7,25,31,49] An increase in blood lactate is often seen more than an increase in FAA levels and a decrease in RER after caffeine ingestion.[26] An increase in lactate could indicate that there was an increased production by the active muscle or a decreased blood clearance.[31] Van Soeren and Graham observed the metabolic effects of caffeine after withdrawal reported that blood lactate increased in response to exercise. After 2 days of caffeine withdrawal, blood lactate concentration increased at exhaustion with caffeine when compared to the placebo. After 4 days of withdrawal, blood lactate increased at both twenty minutes and exhaustion.[55] A study by Jackman et al. examined caffeine's effect during brief, intense exercise and found that blood lactate concentrations increased during exercise in which caffeine was ingested. Despite the increase, the effect

was not significant.[31] A study conducted by Bell and McLellan investigated caffeine's effect on exercise endurance 1, 3, and 6 hours after ingestion in users and nonusers. Twenty-one subjects cycled to exhaustion at 80% VO2max after ingesting a placebo or 5 mg/kg of caffeine. The results indicated that caffeine increased lactate concentrations before exercise, with the greatest increase after one hour of caffeine ingestion. Lactate levels also increase during exercise for the caffeine users and during the one-hour trial for nonusers. At exhaustion, levels were higher with caffeine ingestion than with the placebo for all trials.[7] Graham and Spriet examined the metabolic and exercise responses to various doses of caffeine. Researchers found that during exercise blood lactate rose moderately with exercise. Participants were given a placebo, 3, 6, or 9 mg/kg of caffeine. Researchers observed an increase in lactate concentration with each dose of caffeine and found significance with 9 mg/kg. Although, lactate showed a significant increase, researchers could not conclude that the increase was due to increased production, as changes in release and/or clearance could have affected the data.[55]

The studies that report an increase in blood lactate conflict with the theory that caffeine is glycogen sparing. If glycogen sparing occurs, then lactate concentrations should not increase due to FFAs being the source of fuel during exercise.

The results of the previous studies conflict with the results of the present study. The present study found lactate to be similar with the caffeine versus the placebo and across all the time points. This could be due to a few reasons. The first is the small dose of caffeine. The second is the fitness level of the participants. Cycling at 85% VO2max is not an easy task, especially if one does not train aerobically. The intensity of the

exercise is going to automatically increase blood lactate regardless of caffeine ingestion. Therefore, research on blood lactate concentration and caffeine ingestion needs to be further studied.

CHAPTER V

SUMMARY

STATEMENT OF THE PROBLEM

The focus of caffeine research has been on the ergogenic effect of the liquid and capsule delivery methods. In order to determine the mechanism by which caffeine elicits this ergogenic effect, the metabolic effects are often studied during exercise. Cox et al. studied the effects of 6 mg/kg of caffeine in capsule form and Coca-Cola on metabolism and endurance performance in twelve highly trained male cyclists or triathletes.[17] Researchers found that the 6 mg/kg of caffeine enhanced performance during a time trial at the end of a prolonged cycling bout.[17] Researchers also found that the Coca-Cola enhanced exercise performance despite having a low level of caffeine.[17] A study by Van Soeren and Graham examined the effects of caffeine on exercise metabolism after withdrawal. This study delivered caffeine in the form of a capsule to recreational athletes. The results indicated that caffeine increased exercise time in all exercise trials when compared to the placebo.[55] These two studies are very similar to the majority of caffeine research which studies the metabolic and performance effects of caffeine in liquid or capsule form.[4,6,7,16,20,24-27,30,31,41,49,52]

The problem with administering caffeine as a liquid or in a capsule is the timing. Once ingested, caffeine must travel through the gastrointestinal system and be completely

absorbed before entering the bloodstream. Although caffeine is rapidly absorbed it does take time to peak in the bloodstream. Therefore, caffeine must be ingested 45 to 60 minutes prior to exercise in order to elicit an ergogenic effect.

Gum is one of the more recent items caffeine has been added to. While no exercise studies have been performed using caffeinated gum, there have been studies that examined the catecholamine effects on sleep deprivation.[34,35,50] Caffeine gum has proven to be a successful aid for those who need to stay awake especially for a prolonged period.[34]

The major difference between the gum and a liquid/capsule is the amount of time it takes to be absorbed. Caffeine gum has a much quicker absorption time and a higher relative bioavailability.[35] Eighty-five percent of the caffeine in the gum is absorbed sublingually within five minutes.[38] Absorption via the buccal cavity will allow caffeine to bypass the gastrointestinal system and be released directly into the bloodstream. This fast absorption rate and relative bioavailability could mean more of an ergogenic effect during exercise.

STATEMENT OF PURPOSE

Since this is the first study exercise study being performed using caffeinated gum, the intent of this study is to determine if caffeine gum elicits more of an ergogenic effect during submaximal, exhaustive exercise than other forms of caffeine. The purpose of this study is to determine if caffeine gum will have more of an effect on blood glucose and blood lactate levels during cycling to exhaustion. Based on previous research, the researcher hypothesizes that blood glucose and blood lactate levels will increase during exercise due to the administration of caffeine gum

SUMMARY OF RESULTS

The first hypothesis tested stated that blood glucose would increase with caffeine versus the placebo. This hypothesis was not statistically significant ($p=0.432$) with an $F=0.622$ and a $R^2=0.003$.

The second hypothesis was analyzed to see if there was a differential effect in blood glucose. The hypothesis stated that levels would increase depending on if the caffeine was administered at -35 minutes, -5 minutes, or at +15 minutes. This hypothesis was not statistically significant ($p=0.736$) with an $F=0.425$ and a $R^2=.006$.

The third hypothesis stated that blood lactate levels would increase with caffeine versus the placebo. This hypothesis was not statistically significant ($p=0.934$) with an $F=0.007$ and a $R^2=0.000$.

The fourth and final hypothesis was analyzed to see if there was a differential effect in blood glucose. The hypothesis stated that levels would increase depending on if the caffeine was administered at -35 minutes, -5 minutes, or at +15 minutes. This hypotheses was not statistically significant ($p=0.758$) with an $F=0.394$ and a $R^2=0.004$.

IMPLICATIONS

The results of this investigation need to be further studied since this is the first exercise study using caffeinated gum. This investigation is a starting point for future research to be conducted. While the results of this study did not show any significance, this study provides researchers with the opportunity to repeat the study using more controlled variables, such as caffeine dose, number of subjects, and the fitness level of the subjects.

POSSIBILITIES FOR FUTURE RESEARCH

Based on the findings of this investigation, more research needs to be done to determine if caffeine gum elicits more of an ergogenic effect than a liquid or a capsule. Future research should increase the dose of caffeine given to the participants. This study used 200 mg of caffeine for all of the participants. The caffeine should be dosed by each participant's body weight. This study used recreationally active males, therefore; future studies should examine the effects on trained participants. Also, future research should look into inserting a catheter into an antecubital vein to retrieve blood glucose and blood lactate measurements. This would allow researchers to get samples every couple of minutes instead of every ten minutes during exercise.

REFERENCES

1. Aldridge, A., Bailey, J., & Neims, A. H. (1981). The disposition of caffeine during and after pregnancy. *Seminars in Perinatology*, 5, 310-314.

2. Arnaud, M. J., Thelin-Doerner, A., Ravussin, E., & Acheson, K. J. (1980). Study of the demethylation of [1,3,7-Me-13C] caffeine in man using respiratory exchange measurements. *Biomedical Mass Spectrometry*, 7, 521-524.

3. Arnaud, M. J. (1987). The pharmacology of caffeine. *Progress in Drug Research*, 31, 273-313.

4. Bangsbo, J., Jacobsen, K., Nordberg, N., Christensen, N. J., & Graham, T. E. (1992). Acute and habitual caffeine ingestion and metabolic responses to steady-state exercise. *Journal of Applied Physiology*, 72(4), 1297-1303.

5. Battig, K. (1985). The physiological effects of caffeine consumption. In M. N. Cllifford & K. C. Willson (Eds), *Coffee: Botany, biochemistry and production of beans and beverages* (pp. 394-439). London: Croom-Helm.

6. Battram, D. S., Graham, T. E., Richter, E. A., & Dela, F. (2005). The effect of caffeine on glucose kinetics in humans-influence of adrenaline. *Journal of Physiology*, 569.1, 347-355.

7. Bell, D. & McMellan, T. (2002). Exercise endurance 1, 3, and 6 H after caffeine ingestion in users and nonusers. *Journal of Applied Physiology*, 93, 1127-1234.

8. Blanchard J., & Sawyers, S. J. A. (1983a). Comparative pharmacokinetics of caffeine in young and elderly men. *Journal of Pharmacokinetics and Biopharmaceutics*, 11, 109.

9. Blanchard J., & Sawyers, S. J. A. (1983b). Relationship between urine flow-rate and renal clearance of caffeine in man. *Journal of Clinical Pharmacology*, 23, 134-138.

10. Bonati, M., & Garattini, S. (1984). *Interspecies comparison of caffeine disposition.* Berlin: Springer-Verlag.

11. Brooks, G. A., Fahey, T. D., & Baldwin, K. M. (2005). Glycogenolysis and glycolysis in muscle: the cellular degradation of sugar and carbohydrate to pyruvate and lactate. *Exercicse Physiology: Human Bioenergetics and Its Applications*, 4th ed, (pp. 59-65). McGraw-Hill Companies, Inc, New York, NY.

12. Bush, A., Busst, C. M., Clark, B., & Barnes, P. J. (1989). Effect of infused adenosine on cardiac output and systemic resistance in normal subjects. *British Journal of Clinical Pharmacology*, 27, 165-171.

13. Butcher, R. W., & Sutherland, E. W. (1962). Adenosine 3'5'-phosphate in biological materials. *Journal of Biological Chemistry*, 237, 1244-1250.

14. Callahan, M. M., Robertson, R. S., Branfman, A. R., McComish, M. F., & Yesair, D. W. (1983). Comparison of caffeine metabolism in three nonsmoking populations after oral administration of radiolabelled caffeine. *Drug Metabolism and Disposition*, 11, 211-217.

15. Carillo, J. A., & Benitez, J. (1994). Caffeine metabolism in a healthy Spanish population: N-acetylator phenotype and oxidation pathways. *Clinical Pharmacology and Therapeutics*, 55, 293-304.

16. Costill, D. L., Dalasky, G. P., Fink, W. J. (1978). Effects of caffeine ingestion on metabolism and exercise performance. *Medicine and Science in Sports and Exercise*, 10, 155-158.

17. Cox, G., Desbrow B., Montgomery, P., Anderson, M., Bruce, C., Macrides, T., Martin, D., Moquin, A., Roberts, A., Hawley, J., & Burke, L. (2002). Effect of different protocols of caffeine intake on metabolism and endurance performance. *Journal of Applied Physiology*, 93, 990-999.

18. Dews, P. B. (1984). Metabolism and Kinetics. In*: Dews PB (ed) Caffeine*. Springer-Verlag, New York.

19. Dunwiddie, T. V. (1985). The physiological role of adenosine in the central nervous system. *International Review of Neurobiology*, 27, 63-139.

20. Essig, D., Costill, D. L., & Van Handel, P. J. (1980). Effects of caffeine ingestion on utilization of muscle glycogen and lipid during leg ergometer cycling. *International Journal of Sports Medicine*, 1, 86-90.

21. Fisone, G. G., Borgkvist, A., & Usiello, A. (2004). Caffeine as a psychomotor stimulant: mechanism of action. *Cellular Molecular Life Science*, 61(7-8), 857-872.

22. Fredholm, B. B. (1995). Adenosine, adenosine receptors and the actions of caffeine. *Pharmaceutic Toxicology*, 76, 93-101.

23. Fredholm, B. B., Battig, K., Holmen, J., Nehlig, A., & Zvartau, E. E. (1999). Actions of caffeine in the brain with special reference to factors that contribute to its widespread use. *Pharmacological Reviews*, 51, 81-125.

24. Graham, T. E., & Spriet, L. L. (1991). Performance and metabolic responses to a high caffeine dose during prolonged exercise. *Canadian Journal of Applied Physiology*, 19, 111-138.

25. Graham, T. E., & Spriet, L. L. (1995). Metabolic, catecholamine, and exercise performance responses to various doses of caffeine. Journal of Applied Physiology, 78 (3), 867-874.

26. Graham, T. E. (2001). Caffeine and exercise: metabolism, endurance and performance. *Sports Medicine*, 31, 785-807.

27. Graham, T. E., Helge, J. W., MacLean, D. A., Kiens, B., & Richter, E. A. (2000). Caffeine ingestion does not alter carbohydrate or fat metabolism in human skeletal muscle during exercise. *Journal of Physiology*, 529.3, 837-847.

28. Hashiguchi, M., Fujimura, A., Ohashi, K., & Ebihara, A. (1992). Diurnal effect on caffeine clearance. *Journal of Clinical Pharmacology*, 32, 184-187.

29. Houston, M. E. (2006). Carbohydrate metabolism. *Biochemistry Primer For Exercise Science*, 3rd ed, (pp. 99-143). Human Kinetics, Champaign, IL.

30. Ivy, J. L., Costill, D. L., Fink, W. J., & Lower, R. W. (1979). Influence of caffeine and carbohydrate feedings on endurance performance. *Medicine and Science in Sports and Exercise*, 11, 6-11.

31. Jackman, M., Wendling, P., Friars, D., & Graham, T. W. (1996). Metabolic, catecholamine, and endurance responses to caffeine during intense exercise. *Journal of Applied Physiology*, 81(4), 1658-1663.

32. James, J. E. (1991). Pharmacology of caffeine. *Caffeine and Health* (pp. 19-33). Academic Press Inc., San Diego, CA.

33. James, J. E. (1997). Pharmacology and toxicology of caffeine. *Understanding Caffeine: A Biobehavioral Analysis* (pp. 14-34). Sage Publications, Thousand Oaks, CA.

34. Kamimori, G. H., Penetar, D. M., Headley, D. B., Thorne, D. R., Oterstetter, R., & Belenky, G. (2000). Effect of three caffeine doses on plasma catecholamines and alertness during prolonged wakefulness. European Journal of Clinical Pharmacology, 56, 537-544.

35. Kamimori, G. H., Karyekar, C. S., Otterstetter, R., Cox, D. S., Balkin, T. J., Belenky, G. L., & Eddington, N. D. (2002). The rate of absorption and relative bioavailability of caffeine administered in chewing gum versus capsules to normal healthy volunteers. *International Journal of Pharmaceutics*, 234, 159-167.

36. Lelo, A., Miners, J. O., Robson, R. A., & Birkett, D. J. (1986b). Quantitative assessment of caffeine partial clearances in man. *British Journal of Clinical Pharmacology*, 22, 183-186.

37. Mitchell, T. B., Lupica, C. R., & Dunwiddie, T. V. (1993). Activity-dependent release of endogenous adenosine modulates synaptic responses in the rat hippocampus. *Neuroscience*, 13, 3439-3447.

38. Novum. (1998). *Relative Bioavailability of Caffeine Chewing Gum Pieces vs. No-Doz Tablets*. Amurol Confections Company: USA.

39. Passmore, A. P., Kondowe, G. B., & Johnston, G. D. (1987). Renal and cardiovascular effects of caffeine: A dose-response study. *Clinical Science*, 72, 749-756.

40. Poledna, J., & Morad, M. (1983). Effects of caffeine on the birefringence signal in single skeletal muscle fibers and mammalian heart. Possible mechanisms of actions. *Pflugers Archiv*, 397, 184-189.

41. Raguso, C. A, Coggan, A. R., Sidossis, L. S, et al. (1996). Effect of theophylline on substrate metabolism during exercise. *Metabolism*, 45(9), 1153-1160.

42. Rall, T. W. (1985). Central nervous system stimulants. In L. S. Goodman & A. Gilman (Eds), *Pharmacological basis of therapeutics* (pp. 589-603). New York: Macmillan.

43. Rall, T. W. (1990a). Drugs used in the treatment of asthma: The methylxanthines, cromolyn, sodium, and other agents. In A. G. Gilman, T. W. Rall, A. S. Nies, & P. Taylor (Eds), *Goodman and Gilman's the pharmacological basis of therapeutics* (pp. 618-673). New York: Pergamon.

44. Rathbone, M. P. et al. (1999). Trophic effects of purines in neurons and glial cells. *Progress in Neurobiology*, 59, 663-690.

45. Robertson, D. & Curatolo, P. (1984). The cardiovascular effects of caffeine. In: *Dews PB (ed) Caffeine*. Springer-Verlag, New York.

46. Serafin, W. (1996). Drugs used in the treatment of asthma: methylxanthines. *The Pharmocological Basis of Therapeutics*, 9th ed. MacMillan Publishing Company, New york, NY.

47. Shargel, L., & Yu, A. (1999). *Applied biopharmaceutics and pharmokinetics*, 4th ed, (p. 768). Appleton & Lange: Stamford, CT.

48. Somani, S. M., & Gupta, P. (1988). Caffeine: A new look at an age-old drug. *International Journal of Clinical Pharmacology, Therapy, and Toxicology*, 26, 521-533.

49. Spriet, L. L., MacLean, D. A., Dyck, D. J., Hultman, E., Cederblad, G., and Graham, T. E. (1992). Caffeine ingestion and muscle metabolism during prolonged exercise in humans. *International Journal of Physiology*, 262, E891-E898.

50. Syed, S. A., Kamimori, G. H., Kelly, W., & Eddington, N. D. (2005). Multiple dose pharmokinetics of caffeine administered in chewing gum to normal healthy volunteers. *Biopharmaceutics and Drug Disposition*, 26, 403-409.

51. Tarnopolsky, M. A., Atkinson, S. A., MacDougall, J. D., Sale, D. G., & Sutton, J. R. (1989). Physiological responses to caffeine during endurance running in habitual caffeine users. *Medicine and Science in Sports and Exercise*, 21, 418-424.

52. Tarnopolsky, M. A. (1994). Caffeine and endurance performance. *Sports Medicine*, 18, 109-125.

53. Trice, I., & Haymes, E. M. (1995). Effects of caffeine ingestion on exercise-induced changes during high-intensity, intermittent exercise. *International Journal of Sports Medicine*, 5, 37-44.

54. Van Calker, D., Muller, M., & Hamprecht, B. (1979). Adenosine regulates via two different types of receptors: The accumulation of cyclic AMP in cultured brain cells. *Journal of Neurochemistry*, 33, 999-1005.

55. Van Soeren, M. H., & Graham, T. E. (1998). Effect of caffeine on metabolism, exercise endurance, and catecholamine responses after withdrawal. *Journal of Applied Physiology*, 85(4), 1493-1501.

56. Watt, A. H., Bayer, A., Routledge, P. A., & Swift, C. G. (1989). Adenosine-induced respiratory and heart rate changes in young and elderly adults. *British Journal of Clinical Pharmacology*, 27, 265-267.

57. Yesair, D. W., Branfman, A. R., & Callahan, M. M. (1984). Human disposition and some biochemical aspects of methylxanthines. In G. A. Spiller (Ed.), *The methylxanthine beverages and foods: Chemistry, consumption, and health effects* (pp. 215-234). New York: Alan R. Liss.

APPENDICES

APPENDIX A

INFORMED CONSENT

Title of Study: *"The Effects of Caffeine Administration Timing on Cycling to Exhaustion"*

Introduction: You are invited to participate in a research study designed and conducted by Andrea Jankowski-Wilkinson and Morgan Russell, Masters' students enrolled in the Exercise Physiology program at The University of Akron in the Department of Sport Science and Wellness Education and Edward Ryan, Doctoral student enrolled in the Exercise, Leisure and Sport program at Kent State University, under the advisement of Dr. Ronald Otterstetter, faculty member at The University of Akron in the Department of Sport Science and Wellness Education.

Purpose: The main objective for this investigation is to determine the best time to administer caffeinated chewing gum in order to see how it affects exercise performance on a bicycle. This study will specifically look at the effect caffeine has on the body at rest and during exercise, how fast fatigue occurs, and mental alertness.

Procedure: Ten subjects will individually participate in the research. If you volunteer for this study you will be required to take part in a preliminary visit and four testing trials. The preliminary visit will last approximately sixty minutes and each of the four testing trials will last approximately ninety minutes. During the preliminary visit, the testing protocol will be explained and you will complete a health history questionnaire as well as a Dual Energy X-ray Absorptiometry (DEXA) scan to determine baseline body composition. You will undergo a graded exercise test on a bicycle until you cannot exercise any longer to determine your highest level of fitness. The bicycle will be set to increase in resistance every minute. Throughout the test, you will be equipped with a mouthpiece similar to a scuba-diving snorkel, which you will breathe through for the entire exercise session. You will also be equipped with a heart rate monitor and a blood pressure cuff for the entire exercise session.

The four testing trials will be separated by seven days, in which data will be collected. You will chew either two pieces of caffeinated gum containing 100 milligrams of caffeine each, or a placebo, 35 minutes pre-exercise, 5 minutes pre-exercise, and 10 minutes following the initiation of exercise. You will be required to complete a cardiovascular endurance capacity test.

The cardiovascular endurance test will be performed on a bicycle. You will cycle at a set intensity that will be predetermined until you cannot cycle any longer. During this test, the amount of oxygen that you breathe in will be measured using the mouthpiece every 10 minutes, heart rate will be measured, the effort you are cycling at will be determined using a chart, blood glucose and lactate levels will be measured by a finger prick, perceived leg pain will be determined by you determining your level of leg pain according to numbers on a chart, and reaction time will be determined using a Palm-held psychomotor vigilance task. The task will require you to react to a stimulus (a bulls-eye), which appears in the middle of a Personal Data Assistant (PDA). Your task will be to press a designated key as soon as possible after the stimulus appears, while on the bicycle.

Two blood specimens of 5 ml each will be drawn three times during the trial. A total of 30 ml will be drawn, which is approximately one tablespoon per trial. The blood specimens will be taken 40 minutes pre-exercise, immediately pre-exercise, and immediately post-exercise.

Blood glucose and blood lactate levels will be measured by a glucometer and lactate analyzer. Your finger will be pricked using stick that looks like a pen and then the blood will immediately be analyzed. You will be pricked 40 minutes pre-exercise, immediately pre-exercise, and every ten minutes during exercise.

Prior to the four testing trials, you will be asked to not eat for 8 hours before each trial and to not consume any alcohol or caffeine containing drinks for 24 hours before each trial.

Inclusion: Healthy males between the ages of 18 and 32 years.

Exclusion: A Health History Questionnaire will be used to determine exclusion criteria. If you answer yes to any question on the questionnaire and do not fall within acceptable quantified limits, you will be disqualified from the study. The three quantified items on the health history questionnaire are alcohol consumption (< 3 drinks per day), caffeine consumption (< 300 mg per day), and if you have had mononucleosis within six months of first trial date. You will be excluded if you use caffeine supplements, smoke, show signs or symptoms of cardiovascular, metabolic, or respiratory diseases or you have a known cardiovascular, metabolic, or respiratory disease. You will also be excluded if you are >30% body fat, due to an increased chance of a cardiac event.

Risk and Discomfort as Related to Caffeine: Risks associated with this investigation are related to the ingestion of caffeine. Although the caffeine doses used in this study are similar to those encountered in everyday life, over the counter supplements have the potential for minor side effects. Side effects for caffeine may include the possibility of headache, dizziness, nausea, and muscle tremor. Individuals who do not consume caffeine on a regular basis may be more prone to side effects than those who are habitual caffeine users. In addition, chewing gum may cause dental fillings to become loose.

Risk and Discomfort as Related to Blood Draws: Mild discomfort, or bruising due to venipuncture is possible. There is a slight risk of infection similar to any puncture in the skin, which will be minimized by using a sterile technique.

The universal precautions to avoid transmission of blood borne pathogens between tester and subject will be observed. Venipuncture will be performed by trained, experienced personnel. Phlebotomy competency training will be completed and documented for all individuals performing blood draws. In the event of a medical emergency, all researchers are CPR, First Aid, and AED trained and certified. Should an emergency occur, the researchers will activate EMS.

Risk and Discomfort as Related to Dual Energy X-Ray Absorptiometry: There is minimal radiation exposure through the DEXA scans. The amount of radiation in this investigation is less than $1/1000^{th}$ of the exposure associated with an average dental X-ray (DEXA=1mR/18 sec vs. Dental X-ray=1138mR).

Risk and Discomfort as Related to Exercise Tests: Mild and localized discomfort, not exceeding that incurred during a normal exercise session, associated with delayed onset muscle soreness due to exercise can be expected. You may experience fatigue and lightheadedness due to exercise. Throughout the test your nose will be clamped and you will breathe through a tube, which you may find uncomfortable.

A health history will be taken to screen out anyone for who strenuous exercise may pose a higher than expected risk. It is important that you provide truthful and accurate information so as to not put yourself at unnecessary risk.

Investigators have completed over 100 VO2max tests without a subject injury or critical event. An investigator will be monitoring the computer, as to stop the test at any point that the subject requests or if the researchers feels it is unsafe for you to continue based on guidelines set forth by the American College of Sports Medicine.

The risk of serious injury is no greater than that which you may experience with a very intense, physical workout. There is an extremely small chance of a serious medical condition occurring, and according to National statistics, 4 out of every 10,000 people may experience a heart attack and 1 out of every 10,000 people may experience sudden death when engaging in intense physical exercise/exertion. <u>You should inform the researchers immediately if you start to have pains in your chest, shoulder or legs, feel dizzy or weak, and experience any shortness of breath, difficulty breathing, or other distressing symptoms during the testing procedure.</u>

Risk and Discomfort as Related to Finger Pricks: Mild discomfort or soreness may occur due to finger pricks. There is a slight risk of infection similar to any puncture in the skin, which will be minimized by using a sterile technique. The universal precautions to

avoid transmission of blood borne pathogens between tester and subject will be observed. Finger pricks will be performed by the researchers.

Benefits: Participating in this study will allow you to learn more about your fitness level from the bicycle exercise test, as well as your personal body composition and bone density via a DEXA scan.

Payments for Participation: You will be monetarily compensated upon the completion of your individual participation in the study. You will be compensated $200, broken down into the following increments based on the completion of each trial: $25 for successful completion of trial 1, $25 for successful completion of trial 2, $50 for successful completion of trial 3 and $100 for successful completion of trial 4. If you are unable to participate in all four trials, your compensation will be prorated accordingly.

Right to refuse or withdraw: Participation in the research is voluntary. You may withdraw consent and discontinue participation in the study at any time without any consequence to you.

Anonymous and Confidential Data Collection: Any identifying information collected will be kept in a locked file cabinet, and only the researchers will have access to the data. As a participant, you will not be individually identified in any publication or presentation of the research results. Only comprehensive data will be used. To insure your privacy, the information found in his study will be subject to the confidentiality and privacy regulations of The University of Akron and Kent State University.

Confidentiality of records: The project director will store all of your information in a locked research file and will identify you only by a number. The project director will keep the number key connecting your name to your number in a separate secure file. The data will be kept in two secure locations; The University of Akron, in Memorial Hall room 60E, and at Kent State University in the gym annex room 163. The data will be kept for three years and then shredded.

Who to contact with questions: If you have any questions at any time, you may contact either of the researchers at (330) 972-7747 or our advisor, Dr. Ronald Otterstetter in the Department of Sport Science and Wellness Education at (330) 972-7738. This project has been reviewed and approved by The University of Akron Institutional Review Board. If you have any questions about your rights as a research participant, you may call Sharon McWhorter, the Associate Director of Research Services & Sponsored Programs at The University of Akron, at (330) 972-8311 of 1-888-232-8790.

Thank you for your willingness to participate in this study.

Andrea Jankowski-Wilksinson, B.S.
Researcher

Morgan Russell, B.S.
Researcher

Edward Ryan, M.S.
Researcher

Ron Otterstetter, PhD
Researcher, Advisor

Acceptance & Signature: I have read the information provided above and all of my questions have been answered. I voluntarily agree to participate in this study. I will receive a copy of this consent form for my information.

_____ Date:_____
Participant Signature

_____ Date:_____
Signature of Witness

APPENDIX B

HEALTH HISTORY QUESTIONNAIRE

The University of Akron
EXERCISE PHYSIOLOGY RESEARCH LAB

HEALTH HISTORY

Thank you for volunteering to be a participant for a study to be conducted in the Exercise Physiology Research Laboratory. Some of the tests used in our experiments require that you perform very strenuous exercise. Consequently, it is important that we have an accurate assessment of your past and present health status to assure that you have no medical conditions that would make the tests especially dangerous for you. Please complete the health history as accurately as you can.

THIS MEDICAL HISTORY IS CONFIDENTIAL AND WILL BE SEEN ONLY BY THE INVESTIGATORS

Name_____ Date____/____/____

Date of Birth____/____/____ Present Age_____yrs

Ethnic Group: ____White
____ African American
____ Hispanic
____ Asian
____ Pacific Islands
____ American Indian
____ Other_____

HOSPITALIZATIONS AND SURGERIES
If you have ever been hospitalized for an illness or operation, please complete the chart below. Do not include childhood tonsillectomy or broken bones.

YEAR_____
OPERATIONS OR LLNESS
_____
_____

YEAR_____
OPERATIONS OR ILLNESS
_____
_____

YEAR_____
OPERATIONS OR ILLNESS
_____
_____

Are you under long-term treatment for a protracted disease, even if presently not taking medication? [ ] Yes [ ] No
If Yes,
explain:_____
_____

MEDICATIONS
Please list all medications that you have taken within the past 8 weeks: (Include prescriptions, vitamins, over-the-counter drugs, nasal sprays, aspirins, etc.)
Check this box [ ] if you have not taken any medication.

MEDICATION_____
REASON FOR TAKING THIS
_____
_____
MEDICATION_____
REASON FOR TAKING THIS
_____
_____
MEDICATION_____
REASON FOR TAKING THIS
_____
_____

ALLERGIES
Please list all allergies you have (include pollen, drugs, alcohol, food, animals, etc.)
Check this box [ ] if you have no allergies.

1._____
2._____
3._____
4._____

When was the last time you were “sick”? (e.g. common cold, flu, fever, etc.)
_____

PROBLEMS AND SYMPTOMS
Place an X in the box next to any of the following problems or symptoms that you have had:

General

[ ] Mononucleosis
If yes, when_____
[ ] Excessive fatigue
[ ] Recent weight loss while not on a diet
[ ] Recent weight gain
[ ] Thyroid disease
[ ] Fever, chills, night sweats
[ ] Diabetes
[ ] Arthritis
[ ] Sickle Cell Anemia
[ ] Heat exhaustion or heat stroke
[ ] Recent sunburn

PROBLEMS AND SYMPTOMS, continued

Heart and Lungs

[ ] Abnormal chest x-ray
[ ] Pain in chest (persistent and/or exercise related)
[ ] Heart attack
[ ] Coronary artery disease
[ ] High blood pressure
[ ] Rheumatic fever
[ ] Peripheral vascular disease
[ ] Blood clots, inflammation of veins (phelebitis)
[ ] Asthma, emphysema, bronchitis
[ ] Shortness of breath
[ ] At rest
[ ] On mild exertion
[ ] Discomfort in chest on exertion
[ ] Palpitation of the heart; skipped or extra beats
[ ] Heart murmur, click
[ ] Other heart trouble
[ ] Lightheadedness or fainting
[ ] Pain in legs when walking
[ ] Swelling of the ankles
[ ] Need to sleep in an elevated position with several pillows

G-U SYSTEM

[ ] Get up at night to urinate frequently
[ ] Frequent thirst
[ ] History of kidney stones, kidney disease

G.I. TRACT

[ ] Eating disorder (e.g. anorexia, bulimia)
[ ] Yellow jaundice
If yes, when_____
[ ] Hepatitis
If yes, when_____
[ ] Poor appetite
[ ] Frequent indigestion or heartburn
[ ] Tarry (black) stool
[ ] Frequent nausea or vomiting
[ ] Intolerance of fatty foods
[ ] Changes in bowel habits
[ ] Persistent constipation
[ ] Frequent diarrhea
[ ] Rectal bleeding
[ ] Unusually foul smelling or floating stools
[ ] Pancreatitis

Nervous System

[ ] Alcohol problem
[ ] Alcohol use
If yes, how many drinks ingested per week? _____
[ ] Frequent or severe headaches
[ ] Stroke
[ ] Attacks of staggering, loss of balance, dizziness
[ ] Persistent or recurrent numbness or tingling of hands or feet
[ ] Episode of difficulty in talking
[ ] Prolonged periods of feeling depressed or “blue”
[ ] Difficulty in concentrating
[ ] Suicidal thoughts
[ ] Have had psychiatric help

Explain any items checked (when, severity, treatment)

_____
_____
_____

Have you ever passed out during or after exertion? YES NO
Do you have a family history of coronary artery disease YES NO
If yes, Who? (Grandparents, parents, siblings, uncles, and aunts)

_____
_____
_____
_____

Are there any other reasons not mentioned above that you feel you should not participate in this research study? YES
NO

Do you currently smoke cigarettes? YES NO

Do you currently use any smokeless tobacco products? YES NO

Do you currently consume caffeine? YES NO
If so, how much?

_____ cans of soda per day
_____ cups of coffee per day
_____ other beverages containing caffeine

APPENDIX C

HUMAN SUBJECTS APPROVAL

November 26, 2007

Andrea Jankowski-Wilkinson
582 Patterson Avenue
Akron, Ohio 44310

Ms. Jankowski-Wilkinson :

The University of Akron's Institutional Review Board for the Protection of Human Subjects (IRB) processed your Application for Review of the research project entitled: "*The Effects of Caffeine Administration Timing on Cycling to Exhaustion*". After initial review, it was determined that your project required a convened meeting held on November 14, 2007. The IRB application number assigned to this project is 20071013.

Your research is now **approved without further qualifications for one year from the convened meeting date.** Per federal guidelines, if you wish to continue the project beyond one year, you must submit a request for continuing review to the IRB. **Any changes in the original research protocol must be approved by the IRB prior to implementation.**

Enclosed are the informed consent documents, which the IRB has approved for your use in this research. Copies of these documents are to be submitted with any application for continuation of this project.

Please note that within two months of the expiration date of this approval, the IRB will forward an annual review reminder notice to you by email as a courtesy. Nevertheless, please note that **it is your responsibility as principal investigator to remember the renewal date of your protocol's review.**

If your project terminates prior to the annual renewal date, please complete the Final Report Form in order to close your IRB file.

Please retain this letter for your files. If this research is being conducted for a master's thesis or doctoral dissertation, you must file a copy of this letter with the thesis or dissertation. If you should have any questions, please do not hesitate to contact me.

Good luck with your research!

Sincerely,

Rosalie Hall

Rosalie Hall, Ph.D.
Chair, Institutional Review Board

Cc: Morgan Russell, Co PI
Ronald Otterstetter, Advisor
Rosalie Hall, IRB Chair

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