

THE EFFECTS OF BEETROOT JUICE SUPPLEMENTATION ON THE OXYGEN COST  
OF BREATHING IN OBESE MALES

A Thesis  
by  
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## **Abstract**

### **THE EFFECTS OF BEETROOT JUICE SUPPLEMENTATION ON THE OXYGEN COST OF BREATHING IN OBESE MALES**

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Introduction: Obese adults exhibit increased oxygen ( $O_2$ ) cost of breathing, whole body  $O_2$  utilization ( $\dot{V}O_2$ ), and ratings of perceived breathlessness when compared with normal weight adults. Beetroot juice supplementation (BRJ) has been reported to reduce whole body  $\dot{V}O_2$  at a given work rate. Purpose: The purpose of this study was to measure the  $O_2$  cost of breathing during a eucapnic voluntary hyperventilation test, and whole body  $\dot{V}O_2$  and ratings of perceived breathlessness during constant work rate cycling in obese males following BRJ supplementation. Methods: Six ( $n=6$ ) obese males performed spirometry measures and a  $\dot{V}O_{2peak}$  test on a cycle ergometer during the initial visit. During the remaining two subsequent visits, subjects mimicked the ventilation and breathing pattern corresponding to 50%, 70%, and 90%  $\dot{V}O_{2peak}$ , to measure the  $O_2$  cost of breathing. After a passive recovery period, subjects cycled at a moderate intensity for 6 minutes. Subjects supplemented with either the placebo or BRJ for ten days in a double-blind, randomized, cross-over design. A 10-day washout period separated the two

supplementation periods. Data was compared between supplements using paired t-tests. Significance was set at  $\alpha = 0.05$ . Results: The O<sub>2</sub> cost of breathing was  $2.32 \pm 1.39$  mL O<sub>2</sub>/L during BRJ conditions and  $2.78 \pm 0.78$  mL O<sub>2</sub>/L during placebo conditions ( $t(3)=0.808$ ,  $p = 0.478$ ). VT was reduced during moderate intensity cycling during BRJ conditions when compared to placebo conditions, but no other physiological or perceptual measures were improved. Conclusion: These findings suggest that BRJ does not lower the O<sub>2</sub> cost of breathing during eucapnic voluntary hyperventilation, nor does BRJ lower  $\dot{V}O_2$  during moderate intensity cycling in young obese males.

Key Words: hyperventilation, moderate intensity exercise, spirometry, breathlessness

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## **Foreword**

The manuscript resulting from this thesis will be submitted to *Medicine and Science in Sports and Exercise*, an international peer-reviewed journal published by the American College of Sports Medicine; it has been formatted according to the style guide for that journal.

## List of Abbreviations and Symbols

<b>ATP</b>	adenosine triphosphate
<b>bpm</b>	beats per minute
<b>br·min<sup>-1</sup></b>	breaths per minute
<b>BRJ</b>	beetroot juice
<b>bts·min<sup>-1</sup></b>	beats per minute
<b>BTPS</b>	body temperature (37°C), ambient pressure, saturated with water vapor
<b>CO<sub>2</sub></b>	carbon dioxide
<b>COB</b>	oxygen cost of breathing
<b><i>f<sub>B</sub></i></b>	breathing frequency
<b>EELV</b>	end expiratory lung volume
<b>FEV<sub>1</sub></b>	forced expiratory volume in one second
<b>FRC</b>	functional residual capacity
<b>FVC</b>	forced vital capacity
<b>HR</b>	heart rate
<b>IC</b>	Inspiratory capacity
<b>kg</b>	kilogram
<b>L</b>	liters
<b>L·min<sup>-1</sup></b>	liters per minute
<b>L·s<sup>-1</sup></b>	liters per second
<b>min</b>	minutes

<b><math>\text{mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}</math></b>	milliliters per kilogram per minute
<b>mm Hg</b>	millimeters of mercury
<b>MVV</b>	maximum voluntary ventilation
<b><math>\text{O}_2</math></b>	oxygen
<b>PCr</b>	phosphocreatine
<b>PEFR</b>	peak expiratory flow rate
<b>RER</b>	respiratory exchange ratio
<b>RPB</b>	rating of perceived breathlessness
<b>RPE</b>	rating of perceived exertion
<b>RPU</b>	rating of perceived unpleasantness
<b>RV</b>	residual volume
<b>s</b>	seconds
<b>SD</b>	standard deviation
<b>SEM</b>	standard error of the mean
<b>STPD</b>	standard temperature ( $0^\circ\text{C}$ ), pressure (760 mmHg) and dry
<b>TLC</b>	total lung capacity
<b>VC</b>	vital capacity
<b><math>\dot{V}_E</math></b>	minute ventilation
<b><math>\dot{V}_{E\text{max}}</math></b>	maximum minute ventilation
<b><math>\dot{V}\text{O}_2</math></b>	oxygen consumption
<b><math>\dot{V}\text{O}_{2\text{peak}}</math></b>	maximal oxygen consumption

$\dot{V}O_{2RM}$	respiratory muscle oxygen consumption
$V_T$	tidal volume
$W$	Watts
$W_{max}$	maximum work capacity

# Chapter 1

## Introduction

### Background

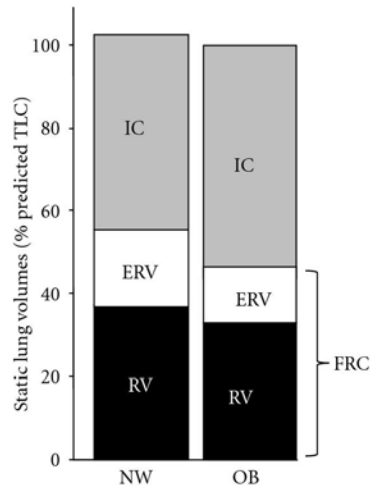
Obesity is a growing epidemic that is causing a national crisis not only on health outcomes but also on economics and sociocultural aspects. Obesity is defined as having a body mass index (BMI)  $\geq 30 \text{ kg}\cdot\text{m}^{-2}$ . The Centers for Disease Control and Prevention reported that in 2016 the prevalence of obesity among American adults was 39.8%, which equates to 93.3 million people (1). In 2008, the medical costs of obesity reportedly totaled \$147 billion; furthermore, obese individuals spend over \$1,400 more per person per year on healthcare costs compared with normal weight individuals (2).

The World Health Organization reported that obesity was the fifth leading risk factor of death in the world (3). Further, three of the top four risk factors are associated with obesity: hypertension, hyperglycemia, and sedentary behavior. Obesity is associated with increased prevalence of type II diabetes, heart disease, stroke, certain types of cancers, and several other preventable chronic diseases. The increased body mass in obesity typically results from years of caloric excess combined with the lack of adequate physical activity. Additionally, obesity can lead to social and psychological issues. A meta-analysis by Luppino (4) concluded that obese adults had a higher prevalence of clinical depression than normal weight adults and that depression was predictive of developing obesity.

Obesity presents challenges to the respiratory and cardiovascular systems at rest and during exercise. It is well-established that additional fat deposits on the chest wall and within the abdomen affect pulmonary function. Specifically, functional residual capacity (FRC) and expiratory reserve volume (ERV) are reduced, while common spirometric indices largely are

maintained similar to that of normal weight individuals (5). These changes are the manifestation of a decrease in compliance of the respiratory system (6). Due to the compliance curve of the chest wall in obesity being shifted to the right, the respiratory compliance curve is shifted to the right, with a less intense slope, making one have to generate greater pressure differentials in order to obtain the desired change in lung volume. These noted effects continue to be evident even during exercise. The result of ERV being diminished compared to normal weight individuals causes the operating lung volume during quiet breathing to be similar to be closer residual (RV) (Figure 1) (7). Thus, end-expiratory lung volume (EELV) is decreased, which requires greater respiratory pressure generation, and tidal volume ( $V_T$ ) becomes positioned closer to the lower nonlinear and less compliant end of the pulmonary system's sigmoid-shaped pressure-volume relation (8).

As a result of the additional stress placed on the respiratory and cardiovascular systems, exercise capacity during submaximal and peak intensities is reduced (9, 10). Furthermore, many obese individuals experience dyspnea during exercise. Sjostrom (11) reported that 80% of obese men felt shortness of breath after walking up two flights of stairs, while only 16% of normal weight males felt shortness of breath. It is estimated that approximately one third of obese adults experience dyspnea during exercise (12). Thus, the prevalence of dyspnea during exercise represents a serious clinical concern to many obese adults. The underlying mechanism of exertional dyspnea in obese adults is poorly understood but it is not believed to be entirely due to alterations in pulmonary function or to the additional metabolic stress placed on the cardiopulmonary system during exercise.



**Figure 1: The alterations in lung volumes caused by obesity (13).**

The  $\text{O}_2$  cost of breathing represents the amount of  $\text{O}_2$  consumed by the respiratory muscles ( $\dot{V}\text{O}_{2\text{RM}}$ ) during the ventilatory cycle. The  $\dot{V}\text{O}_{2\text{RM}}$  is a reflection of the total work accomplished by the respiratory muscles. Typically, only 1-2% of whole body  $\dot{V}\text{O}_2$  at rest is attributed to  $\dot{V}\text{O}_{2\text{RM}}$  (14) but this value will increase exponentially as ventilation meets the metabolic demands of exercise. The  $\dot{V}\text{O}_{2\text{RM}}$  can increase to around 10% of whole body  $\dot{V}\text{O}_2$  during intensive exercise in healthy untrained individuals (15, 16) and can approach 15% of maximal aerobic capacity ( $\dot{V}\text{O}_{2\text{max}}$ ) in highly trained aerobic athletes (16). Yet, obese individuals display a greater  $\dot{V}\text{O}_{2\text{RM}}$  when compared to normal weight individuals at rest (17) and during exercise (18), in part, because the respiratory muscles must complete more work due to the aforementioned greater ventilatory demand and reduced compliance. Therefore, the increased  $\dot{V}\text{O}_{2\text{RM}}$  in obese individuals is attributed to the increased amount of fat within the thorax.

The increased  $\dot{V}\text{O}_{2\text{RM}}$  in obesity may have important clinical considerations. It was suggested that exertional dyspnea might originate from increased  $\text{O}_2$  demand of the respiratory muscles along with their decreases in pulmonary compliance and increases in

airway resistance. Increases in airway resistance can be attributed to breathing at lower lung volumes (5). Babb (19) observed that otherwise-healthy obese adults who experience exertional dyspnea have a  $\dot{V}O_{2RM}$  that is 38%-70% more than obese adults who do not experience exertional dyspnea. Furthermore,  $\dot{V}O_{2RM}$  was positively associated with ratings of perceived breathlessness (RPB), suggesting that the amount of work performed by the respiratory muscles plays an important role in the sensation of respiratory discomfort during exercise. In contrast, weight loss has the ability to decrease  $\dot{V}O_{2RM}$  in obese adults (20); an 8% reduction in body mass reduced  $\dot{V}O_{2RM}$  by 16%. Further, the effect of weight loss on  $\dot{V}O_{2RM}$  accounted for 46% of the decrease in whole body  $\dot{V}O_2$  when cycling at a moderate intensity. These findings suggest that a decrease in  $\dot{V}O_{2RM}$  may improve exercise tolerance via a reduction in whole body  $\dot{V}O_2$  at relative workloads.

Beetroot juice (BRJ) supplementation has been reported to affect resting blood pressure, muscle blood flow, vascular function, and muscle fiber contractile characteristics (21, 22, 23). The marked physiological alterations are largely attributed to nitrate ( $NO_3^-$ ) via nitric oxide synthase dependent and independent pathways (24). Importantly, BRJ also has been reported to decrease whole body  $\dot{V}O_2$  during low- (21) and moderate-intensity exercise in young men who are recreationally trained or athletes (22, 23, 25, 26). Furthermore, BRJ can increase exercise duration and the total amount of work completed during an exhaustive exercise bout in healthy young men who are recreationally active or athletes (21, 22). The decreased energy utilization at a given workload indicates that BRJ improves skeletal muscle efficiency. Thus, BRJ appears to have ergogenic properties that may benefit exercise performance in obese individuals. An improvement in the metabolic efficiency of the respiratory muscles would lower the  $O_2$  cost of breathing.



The potential use of BRJ to improve the metabolic efficiency of skeletal muscles, and specifically the respiratory muscles, may have important implications during exercise in obese adults. Exercise tolerance could be increased in obese men due to the decreased metabolic demands of the respiratory muscles, allowing for more O<sub>2</sub> to be delivered to active skeletal muscles. The majority of studies to date have utilized normal weight adults, though some studies also have examined the effects of BRJ in patients with COPD and heart failure. However, these diseased populations have other dysfunctions, including skeletal muscle dysfunctions. We are interested in examining the effects of chronic BRJ in obese males who are otherwise healthy. To the author's knowledge, only one investigation has examined BRJ and its effects on  $\dot{V}O_2$  in obese subjects and they were adolescents (27). Following supplementation,  $\dot{V}O_2$  at moderate exercise intensities was unchanged, but exercise tolerance was increased. However, the adolescents received a dosage of 5 mmol of nitrate per day, which is below that used in the investigations noting reductions in submaximal exercise  $\dot{V}O_2$  of normal weight individuals (6.2 – 26 mmol·day<sup>-1</sup>); (21, 23, 25, 28). Thus, it is possible that the dosage was insufficient to observe an effect on  $\dot{V}O_2$  during moderate intensity exercise in the obese adolescents (27). Therefore, the effect of BRJ on  $\dot{V}O_{2RM}$  and ratings of perceived breathlessness (RPB) during eucapnic voluntary hyperventilation and whole body  $\dot{V}O_2$  and RPB during moderate intensity cycling in otherwise healthy obese males warrants investigation.

## **Summary**

Obesity is an epidemic to health, economics, and sociocultural aspects within America. Obesity leads to exercise intolerance, an increased  $\dot{V}O_{2RM}$ , and increases in RPB;

all of which may contribute to sedentary behavior in those individuals who are already obese. BRJ is an ergogenic aid that has been shown to improve skeletal muscle efficiency. BRJ may reduce  $\dot{V}O_{2RM}$  and dyspnea during exercise in obese individuals.

### **Statement of the purpose**

The purpose of this study was to measure  $\dot{V}O_{2RM}$  during eucapnic voluntary hyperventilation following BRJ supplementation in obese male adults. Additionally, a secondary aim was to assess whole body  $\dot{V}O_2$  and RPB during constant work rate cycling and examine the association between potential changes in RPB and  $\dot{V}O_{2RM}$  following beetroot juice supplementation.

### **Hypotheses**

The following hypotheses were tested:

Hypothesis 1:  $\dot{V}O_{2RM}$  will be decreased at levels of ventilation corresponding to 50%, 70%, and 90% of  $\dot{V}O_{2peak}$  during the eucapnic voluntary hyperventilation test following BRJ supplementation when compared with following placebo supplementation.

Hypothesis 2: RPB will be decreased at levels of ventilation corresponding to 50%, 70%, and 90% of  $\dot{V}O_{2peak}$  during the eucapnic voluntary hyperventilation test following BRJ supplementation when compared with following placebo supplementation.

Hypothesis 3: Whole body  $\dot{V}O_2$  will be decreased during constant work rate cycling following BRJ supplementation when compared with following placebo supplementation.

Hypothesis 4: Ratings of perceived breathlessness will be decreased during constant work rate cycling following beetroot juice supplementation when compared with following placebo supplementation.

Hypothesis 5: The changes in  $\dot{V}O_{2RM}$  and RPB will be significantly correlated.

### **Significance of the Study**

This study will explore the possible benefits of BRJ on  $\dot{V}O_2$ ,  $\dot{V}O_{2RM}$ , and RPB. Obesity is associated with a greater  $\dot{V}O_{2RM}$  and higher ratings of dyspnea, which make exercise seem harder and discourages many obese individuals from exercising. This study could be beneficial to obese people who do not exercise due to increased dyspnea due to an increased  $\dot{V}O_{2RM}$ . BRJ could be a possible ergogenic aid to make exercise seem easier and therefore increase physical activity. Increased physical activity could lead to weight reduction and a lower risk of developing chronic diseases.

### **Delimitations**

The study was delimited to:

1. Males between the ages of 18 and 40 years.
2. All subjects were obese and otherwise healthy.
3. Visit 1 and visit 2 occurred on the tenth day of the supplementation periods.
4. There was at least a 10-day wash out period between supplementation periods.

5. Subjects drank either BRJ or the placebo in 70 mL shots; one in the morning and one in the evening.
6. Subjects were told to eat similarly during the two-supplementation protocols.
7. Subjects were told to refrain from using mouthwash from the completion of visit 1 until they complete visit 3.
8. Subjects were required to stay seated on the cycle ergometer during the  $\dot{V}O_{2peak}$  test and the two subsequent constant intensity bouts and to maintain a cadence of between 60 and 80 revolutions per minute.
9. Gas analyzers were used to measure fractional gas concentrations of oxygen and carbon dioxide.
10. Inspiratory and expiratory airflow rates were measured with pneumotachs.
11. Posture during the eucapnic voluntary hyperventilation test was the same as during the initial  $\dot{V}O_{2peak}$  test on the cycle ergometer.

## **Limitations**

Interpretation of the results should consider the following limitations:

1. All of the subjects were from the same terrestrial area.
2. Due to the fact that all testing was indoors in a laboratory setting, the results may not be representative in the field.
3. The results can only be able to be applied to obese men who are recreationally active and otherwise healthy.

4. Subjects were discouraged from eating foods high in nitrates, but the foods will not be prohibited.
5.  $\dot{V}O_{2\text{peak}}$  testing required the subject to be motivated, so some subjects may not have reached a true  $\dot{V}O_{2\text{peak}}$ .
6. Subjects consumed BRJ and the placebo while away from the lab, so it can only be inferred that they took the assigned dosages.
7. The influence of blood pH, temperature, and lactate on spirometry measures was unknown.
8. The work of breathing was not be directly measured.
9. Ventilatory mechanics are different between exercise and voluntary hyperventilation at rest.
10. Oral nitrate and nitrite concentrations were not measured.

### **Assumptions**

Interpretations of the results will have to apply the following assumptions:

1. The group that was tested is representative of their respected population (obese men, aged 18-40, recreationally active, otherwise healthy).
2. Subjects were motivated to perform spirometry measures and a  $\dot{V}O_{2\text{peak}}$  test.
3. The subjects consumed the required dosages at the times required.
4. The tubing and mouthpiece required for the  $\dot{V}O_{2\text{peak}}$  did not affect ventilation.

5. Any change in  $\dot{V}O_{2RM}$ , RPB, or  $\dot{V}O_2$  was due to BRJ only.
6. It was assumed that subjects did not partake in strenuous exercise for 24 hours and caffeine/alcohol consumption for 12 hours prior to the 3 visits.
7. The 20-minute break between eucapnic voluntary hyperventilation and the constant intensity exercise test was enough time for  $\dot{V}O_2$  to return to baseline values.
8. Any changes in whole body  $\dot{V}O_2$  during eucapnic voluntary hyperventilation were attributed to the increased metabolic efficiency of the respiratory muscles.

### **Definition of Terms**

*Body Mass Index (BMI)* – a person’s weight in kilograms divided by the square of height in meters (28).

*Carbon dioxide production ( $\dot{V}CO_2$ )* – the volume of carbon dioxide produced per minute (29).

*Cost of breathing ( $\dot{V}O_{2RM}$ )* – the oxygen consumed by the respiratory muscles, is an index for the energy required for ventilation (14).

*Dyspnea* – difficult or labored breathing.

*Eucapnic voluntary hyperventilation* – a test designed to mimic the effect that prolonged exercise has on the airway (30).

*Forced expiratory volume in one second ( $FEV_1$ )* – The maximum volume of air that can be forced out in the first second of expiration from a maximal inspiration, an important measure of pulmonary function (31).

*Forced mid-expiratory flow ( $FEF_{25-75\%}$ )* – The average expiratory flow over the middle half, from 25 to 75%, of the FVC (31).

*Forced vital capacity (FVC)* – The volume of air expelled by a forced maximal expiration to residual volume from a maximal inspiration (31).

*Frequency of breathing ( $f_B$ )* – The breathing rate in breaths per minute.

*Functional residual capacity (FRC)* – The volume of air left in the lungs following a normal expiration. Algebraically, the sum of ERV and RV (31).

*Gas exchange threshold (GET)* – has been used as an index of anaerobic threshold because it can be measured noninvasively. GET is estimated from a breakpoint in breath by breath values of carbon dioxide and oxygen uptake obtained during a progressive exercise test (32).

*Maximal aerobic capacity ( $\dot{V}O_{2max}$ )* – The maximal ability of an individual to take up, transport, and deliver oxygen to the working muscles (33).

*Maximal ventilation ( $\dot{V}_{Emax}$ )* – The highest minute ventilation achieved during exhaustive exercise (34).

*Maximum voluntary ventilation (MVV)* – The maximum volume of air that can be breathed over a specified period of time, typically 12 s (31).

*Oxygen consumption ( $\dot{V}O_2$ )* - The volume of oxygen consumed per minute (29).

*Peak expiratory flow rate (PEFR)* – The maximum flow rate achieved during a forced maximal expiration (31).

*Peak inspiratory flow rate (PIFR)* – The peak inspiratory flow rate achieved during a maximal inspiration (31).

*Rating of Perceived Breathlessness (RPB)* – a scale that allows individuals to subjectively rate their level of dyspnea during exercise or exercise testing (35).

*Rating of Perceived Exertion (RPE)* – a scale that allows individuals to subjectively rate their level of exertion during exercise or exercise testing (35).

*Residual volume (RV)* – The volume of air within the lungs at the end of a maximal expiration (31).

*Tidal volume ( $V_T$ )* – The volume of air moved in and out of the lungs during each breath (29).

*Total lung capacity (TLC)* – The total volume of air within the lungs following a maximal inspiration (31).



## Chapter 2

### Review of Literature

#### Introduction

Beetroot juice (BRJ) has been shown in recent years to have beneficial physiological effects on both moderate and severe intensity exercise (21, 22, 23, 25, 36); however, the majority of research has been conducted on young, normal weight males who are recreationally active or athletes. There has been a lack of research on populations of obese males.  $\dot{V}O_{2RM}$  is the amount of oxygen that the respiratory muscles consume and is an indication of the amount of energy required for ventilation. Obese individuals have an increased  $\dot{V}O_{2RM}$  due to the increased adipose tissue surrounding the thorax, which requires the muscles of the respiratory system to work harder (37, 38, 39). BRJ has demonstrated the ability to lower  $O_2$  kinetics during moderate intensity exercise in non-obese subjects (26, 40).

#### What is Beetroot Juice?

BRJ is juice extracted directly from the roots of beet plants. It is a nutrient dense drink that can contain up to 25 carbohydrates per cup, and is an abundant source of folate, potassium, Vitamin C, fiber, antioxidants, and most importantly nitrate. Nitrate ( $NO_3^-$ ) is the substance within BRJ that is responsible for the beneficial physiological effects during exercise.  $NO_3^-$  is reduced to Nitrite ( $NO_2^-$ ) and then to Nitric Oxide (NO).

The inorganic anions, Nitrate and Nitrite, were once viewed as the end products of Nitric Oxide metabolism and unwanted byproducts from food consumption, but they are now understood as storage pools for Nitric Oxide and complement the L-arginine/NO synthase dependent pathway (41). Plants contain Nitrate Reductase Enzymes that reduce  $NO_3^-$  to  $NO_2^-$ ; however, humans do not produce this enzyme. When  $NO_3^-$  is orally consumed, as with

BRJ, it is concentrated in the saliva and reacts with oral commensal bacteria that reduce it to  $\text{NO}_2^-$  (42).  $\text{NO}_2^-$  is then swallowed and absorbed into the gut for further reduction to NO. To a lesser extent,  $\text{NO}_3^-$  can also be reduced to  $\text{NO}_2^-$  in the liver by xanthine oxidoreductase. Hypoxic conditions and during times in which oral commensal bacterial are low are the only occasions in which xanthine oxidoreductase would be utilized (43).

Once  $\text{NO}_2^-$  is in the gut, it represents the largest storage pool of NO in the body.  $\text{NO}_2^-$  is reduced to NO through several means, but most notably through deoxyhemoglobin and deoxymyoglobin (42). Deoxyhemoglobin reacts with  $\text{NO}_2^-$  to form methemoglobin (a molecule that cannot bind to oxygen) and NO:  $\text{Hb}(\text{Fe}^{2+}) + \text{NO}_2^- + \text{H}^+ \rightarrow \text{Hb}(\text{Fe}^{3+}) + \text{NO} + \text{OH}^-$ . The nitrite reductase activity of deoxyhemoglobin takes place under hypoxic conditions (44, 45). Red blood cells (RBCs) experience increased deoxygenation as they pass through the microcirculation of hypoxic tissue, which would be the case in exercising skeletal muscle, and as a result of this, the RBCs increase the amount of NO that they are producing and releasing to increase blood flow (46). In this way, RBCs are not only acting as oxygen transporters, but also as localized oxygen sensors that mediate local blood flow. This mechanism takes occurs in four major steps: 1)  $\text{NO}_2^-$  is transported across the membrane of the RBC, 2)  $\text{NO}_2^-$  reacts with deoxyhemoglobin to form NO, 3) NO leaves the RBC and enters the hypoxic tissue, 4) lastly NO induces localized vasodilation (46).

Deoxymyoglobin is another potent nitrite reductase, which hypoxic conditions regulate, especially when oxygen concentration within the myoglobin falls below p50. The NO produced from deoxymyoglobin has profound effects on the mitochondria in hypoxic conditions. NO binds to cytochrome c oxidase to inhibit respiration in the mitochondria. This inhibition is more potent as oxygen concentrations decrease and is thought to limit hypoxic

tissue damage through the extension of tissue oxygen gradients, most notably in cardiac tissue (47).

Nitric Oxide is a potent vasodilator, and as explained earlier, it is produced from the precursor molecule, nitrate, which is found in abundance in BRJ. Exercising signals NO through shear stress, which is the parallel friction at the surface of the endothelium caused by an increase in blood flow (i.e. exercise). Shear stress signals endothelial nitric oxide synthase to convert L-arginine into L-citrulline and NO. NO then diffuses across the cell membrane of smooth muscle, which causes guanylate cyclase to convert guanosine triphosphate into cyclic guanosine monophosphate. Cyclic guanosine monophosphate activates Protein Kinase G to lower cellular calcium concentrations to increase vasodilation in the arteries. NO bioavailability is limited in individuals who do not exercise, and reduced by aging (48). These decrements can be avoided by lifelong physical activity (Nyberg 2012). Consuming nitrate rich BRJ can cause increased bioavailability of NO without the need for shear stress or endothelial nitric oxide synthase. BRJ's vasodilatory properties were evidenced in studies conducted by Bailey (22) and Lansley (23); healthy young men were able to reduce their systolic blood pressure after BRJ supplementation by 8 mmHg and 4% respectively.

### **Physiological Benefits during Exercise**

BRJ has been a hot bed for research in recent years, with several studies yielding results that show positive physiological benefits during moderate and severe intensity exercise, in particular,  $\dot{V}O_2$ . In a study by Whitfield (25), 13 recreationally active males supplemented  $26 \text{ mmol} \cdot \text{day}^{-1}$  for 5 days before cycling at 50% and 70% of their  $\dot{V}O_{2\text{peak}}$ . While there were no changes between BRJ and placebo supplementation at light intensity (50%), subjects were able to decrease their  $\dot{V}O_2$  by 100 mL during moderate intensity (70%).

Similarly, in another study when recreationally active men consumed  $5.5 \text{ mmol}\cdot\text{day}^{-1}$  per day for 5 days, they were able to reduce their  $\dot{V}\text{O}_2$  amplitude by 19% during moderate intensity cycling (21). Tan (26) showed similar reductions in  $\dot{V}\text{O}_2$  during 120 minutes of moderate intensity cycling in recreationally trained males. Breese (49) showed that  $8 \text{ mmol}\cdot\text{day}^{-1}$  for 6 days could reduce phase II  $\text{VO}_2$  kinetics in recreationally active males. However, not all studies have shown  $\dot{V}\text{O}_2$  deductions during moderate intensity exercise; Thompson (50) had 16 recreationally trained males supplement on  $5 \text{ mmol}\cdot\text{day}^{-1}$  1.5 hours before cycling at 50% and 70% of  $\dot{V}\text{O}_{2\text{peak}}$ , and there were no significant changes in  $\dot{V}\text{O}_2$  when compared to placebo. Betteridge (51) also failed to produce  $\dot{V}\text{O}_2$  reductions at 65% of  $\dot{V}\text{O}_{2\text{peak}}$ . The previous two studies mentioned were acute supplementation studies, while the four before that were chronic supplementations.

Another common experiment type within BRJ research is severe exercise time until exhaustion tests. Time until exhaustion following BRJ supplementation has been shown to increase by 16% (50) and 22% (49) while cycling. While the total time exercising increased significantly,  $\dot{V}\text{O}_2$  remained the same as placebo supplementation, indicating that power output was increased at no metabolic cost. Similarly, another study showed time until exhaustion to increase by 90 seconds, while exhibiting a slower slow  $\text{VO}_2$  component.

Time trial experiments are another means to show to positive effects of BRJ. If one can decrease their time trial time, while maintaining the same  $\dot{V}\text{O}_2$ , this would indicate an increased power output at no metabolic cost. Club level cyclists were able to improve their time trials at 4 km and 16 km by 2.8% and 2.7% respectively after supplementing  $6.2 \text{ mmol}\cdot\text{day}^{-1}$  2.5 hours before exercise (51). These decreases in time were present with no changes in  $\dot{V}\text{O}_2$ . MacLeod (52) found no significant differences while having trained cyclists

perform a 10 km time trial after they had supplemented on  $6 \text{ mmol}\cdot\text{day}^{-1}$  2 hours before exercise.

### **Mode of Action**

The mode of action behind the increased metabolic efficiency exhibited through BRJ supplementation is still not fully understood; however, several theories among experts in the field exist. The first theory is that the reductions in  $\dot{V}O_2$  stem from mitochondrial biogenesis or an increased functioning among mitochondrial parameters. Two particular studies, which have yielded  $\dot{V}O_2$  reductions during exercise, took muscle biopsies from their subjects and no mitochondrial indices were improved (23, 25). This would suggest that mitochondrial enhancement is not the mechanism of action. In contrast, Vaughan (53) found increases in mitochondrial transcription factor A and glucose transporter 4, which would indicate an increase in mitochondrial biogenesis. However, this study used cultured cells in vitro.

Another common theory among researchers is that BRJ supplementation increases muscle contractile functioning. Most of the research within this theory has been conducted on animal models, so human experimentation is needed for interpretations that are more concise. A study by Hernandez (54) was the first to show that  $\text{NO}_3^-$  supplementation in rats had different effects among different muscle fibers. Type II muscle fibers contained significantly more myoplasmic free calcium ( $\text{Ca}^{2+}$ ) than control rats when electrically stimulated between 20 and 150 Hz. At 100 Hz the rate of force development was also 35% greater in the  $\text{NO}_3^-$  group. The concentrations of the  $\text{Ca}^{2+}$  handling protein calsequestrin 1 and the dihydropyridine receptor were also increased. Calsequestrin 1 is responsible for buffering  $\text{Ca}^{2+}$  back into the sarcoplasmic reticulum and increased concentrations would allow a muscle to contract more frequently (55). None of these effects were observed in Type I

muscle fibers. BRJ supplementation in rats has also been shown to increase vascular conductance and muscle blood flow during moderate intensity exercise in Type II muscle fibers (56), suggesting increased oxygen delivery capabilities and increased efficiency in Type II muscle fibers. This may explain the observed benefits of BRJ supplementation at severe intensity exercise, as Type II muscle fibers are recruited more at that intensity.

Type II muscle fibers have higher concentrations of Phosphocreatine (PCr) which is an essential fuel source during high intensity exercise. Depletion of PCr is associated with fatigue and decreased performance. Bailey (57) analyzed the effects of BRJ supplementation on light and severe intensity knee extension exercise. The degree of PCr degradation during both intensities was reduced accompanied by reductions in estimated ATP utilization during both intensities as well. This would suggest that decreases in  $\dot{V}O_2$  stem from decreased energy needs of skeletal muscle.

### **Dosage**

Supplementation can be broken down into two categories: acute and chronic. Acute supplementation is consumed hours before an exercise bout, while chronic supplementation is consumed for multiple days before an exercise bout. In general, acute supplementations require higher concentrations of  $\text{NO}_3^-$  in order to yield positive metabolic results. Acute supplementation had been able to yield positive metabolic results at  $10.9 \text{ mmol}\cdot\text{day}^{-1}$  (50),  $11.2 \text{ mmol}\cdot\text{day}^{-1}$  (58), and  $8 \text{ mmol}\cdot\text{day}^{-1}$  (51). Chronic supplementation studies tend to have subjects consume BRJ for 6-10 days (21, 22, 23, 25, 49). The mmol per day range from 5.5 mmol per day (21) to  $26 \text{ mmol}\cdot\text{day}^{-1}$  (25).

## Oxygen Cost of Breathing

$\dot{V}O_{2RM}$  is defined as the oxygen consumed by the respiratory muscles and is an index for the energy required for ventilation. Muscles that assist in ventilation during exercise include the diaphragm, external and internal intercostals, sternocleidomastoid, rectus abdominis, external and internal obliques, and several other smaller muscles.  $\dot{V}O_{2RM}$  is quantified in mL of O<sub>2</sub> per L of ventilation (mL O<sub>2</sub>/L).  $\dot{V}O_{2RM}$  increases as minute ventilation increases which is associated with exercise or unobstructed hyperventilation and has a curvilinear slope (16, 60, 61). The work of breathing can be understood as a reflection of the work of the respiratory system, and can be quantified by multiplying the change in volume during a breath and the pressure required to generate that volume change (14). Since energy consumption is a reflection of the amount of work done, an increased work of breathing yields an increased  $\dot{V}O_{2RM}$ . One can interpret this as the metabolic efficiency of the respiratory muscles.

$\dot{V}O_{2RM}$  at rest is extremely low, representing only 1-2% of whole body  $\dot{V}O_2$  (P14). However, as exercise intensities increase,  $\dot{V}O_{2RM}$  will increase exponentially. It can increase to around 10% of whole body  $\dot{V}O_2$  during intense exercise in healthy untrained adults (15, 16), and can reach 15% in highly trained athletes (16). During intense exercise or during higher levels of voluntary hyperventilation, expiration is no longer passive, so expiratory muscles must be recruited in order to reach the necessary ventilation. Cherniak (62) reported  $\dot{V}O_{2RM}$  in normal weight men to be between 0.45 and 1.87 mL O<sub>2</sub>/L. The work of breathing and thus the O<sub>2</sub> required for the respiratory muscles yields different energy requirements during exercise and hyperventilation when the ventilation rate is controlled. Coast (61)

reported that at higher ventilation rates, the work required to breath during hyperventilation is around 25% higher than in exercise.

Most normative data in the research is based on the values of normal weight men. Sex, weight, and disease can alter the work of breathing and the  $\dot{V}O_{2RM}$ . Women's  $\dot{V}O_{2RM}$  is larger than that of males, and it represents a larger portion of their whole body  $\dot{V}O_2$  (63, 64). The additional visceral fat that surrounds the thorax in obese individuals also leads to increases in the  $\dot{V}O_{2RM}$  (62, 65). Increased airway resistance leads to an increased  $\dot{V}O_{2RM}$  in individuals with chronic obstructive pulmonary disease (66, 67).

### **Outcomes**

There will be three main outcomes investigated in young, obese males: the  $\dot{V}O_{2RM}$ ,  $\dot{V}O_2$ , and ratings of perceived breathlessness. There has been a lack of research analyzing the effects of BRJ on obese individuals. Currently there is no research analyzing the relationship between  $\dot{V}O_{2RM}$  and BRJ supplementation. Obese people have been shown to have a higher  $\dot{V}O_{2RM}$ , and BRJ supplementation could possibly be a means of lowering it.

### **O<sub>2</sub> Cost of Breathing**

Increased fat tissue surrounding the respiratory muscles requires the muscles to work harder to breathe, as is the case in obese individuals. Cherniak (62) reported that  $\dot{V}O_{2RM}$  was almost 3 times higher in obese individuals when compared to normal weight individuals: 3.45 mL O<sub>2</sub>/L vs. 1.2 mL O<sub>2</sub>/L. Sharp (65) reported that the work of breathing in obese individuals was 1.3 times greater than normal weight individuals, with some subjects having values close to double that of normal weight subjects. Their respiratory compliance was also remarkable lower. Obese individuals tend to breathe at a less steep pulmonary compliance curve, meaning that greater pressures are required to generate volume changes. As mentioned



earlier, if either pressure or volume differentials are increased, work increases. Kress (17) reported that morbidly obese individuals who were put on mechanical ventilators were able to reduce their  $\dot{V}O_{2RM}$  by 16%, while normal weight individuals were only able to decrease by 1%. The 15% difference can be attributed to the increased visceral fat surrounding the thorax. An 8% weight loss in obese females has been shown to decrease to  $\dot{V}O_{2RM}$  by 16%.

There are currently no studies analyzing the effects of BRJ supplementation on  $\dot{V}O_{2RM}$  in any population.  $NO_3^-$  can make skeletal muscle more efficient during exercise, so it can be predicted that it will yield similar results in the respiratory muscles during hyperventilation. The diaphragm and the rectus abdominis are two of the most important respiratory muscles at higher ventilation rates and contain around 45% Type II muscle fibers (68, 69). Obese individuals require a longer time to recover their PCr levels after exercise due to their slow metabolic recovery rate (70), but BRJ can lower the amount of PCr used (57).

## $\dot{V}O_2$

Obese individuals have a decreased exercise capacity both at submaximal and maximal intensities (9). Their  $\dot{V}O_{2peak}$  in absolute terms is normal, but in relative terms, it is low, suggesting poor cardiorespiratory fitness. While there has been a plethora of studies looking at the effects of BRJ supplementation on normal weight recreationally active and trained men, there has only been one study to look at its effects on an obese sample. Rasica (27) supplemented 10 obese adolescents (8 girls, 2 boys)  $5 \text{ mmol}\cdot\text{day}^{-1}$  for 6 days. There were no significant findings during moderate intensity cycling. A 23% increase in time until exhaustion during severe intensity exercise was observed following BRJ supplementation when compared to the placebo. The slow component of their  $\dot{V}O_2$  response was also reduced.

The lack of statistical findings during the moderate intensity exercise may be due to the small dosage of  $\text{NO}_3^-$  they were given.

Reductions in  $\dot{V}\text{O}_2$  are common during moderate intensity exercise. These reductions range from modest 3% reductions (25) to immense reductions of 19% (21). Phase II kinetics also consistently decrease (21, 41, 71, 72). Phase II kinetics are also called slow component, and slow component is the addition of the anaerobic energy systems during aerobic exercise. The more that the anaerobic systems are utilized, the more lactate and  $\text{H}^+$  ions that are produced. This will cause the pH of the blood to increase and bring about metabolic fatigue.

### **Ratings of Perceived Breathlessness (RPB)**

Ratings of perceived breathlessness (RPB) is a self-reported measure of dyspnea, or the difficulty or the amount of labor felt during breathing. It results from multiple signal interactions with receptors in the central nervous system, peripheral receptors, chemoreceptors, and mechanoreceptors in the upper airway, lungs, and chest wall (73). Levels of perceived dyspnea are increased in obese men and women when there are no other health problems present such as asthma or COPD, which would explain it. Babb (19) reported that around one third of obese individuals experience dyspnea during exercise. Dyspnea would directly raise levels of RPB during exercise. Sjostrom (11) compared shortness of breath in normal weight and obese individuals directly after the subjects had walked up two flights of stairs. 80% of the obese individuals experienced shortness of breath, while only 16% of normal weight males did. The exact mechanism for the increased dyspnea in obese individuals is not completely understood, but it cannot be attributed to airflow obstruction (73).

The only study to directly measure the RPB during exercise after BRJ supplementation was conducted by Thompson (50). There were no differences in RPB during exercise at 50%, 70%, or 90% of  $\dot{V}O_{2peak}$ . Previous studies have shown the potential of BRJ to make exercise more efficient particularly by lowering  $\dot{V}O_2$  at relative workloads.

### **Summary**

The purpose of this study was to analyze the effects of BRJ supplementation on  $\dot{V}O_{2RM}$  and RPB during eucapnic voluntary hyperventilation and on the  $\dot{V}O_2$  and RPB during moderate intensity cycling.

It was hypothesized that  $\dot{V}O_{2RM}$  of breathing and RPB would be lowered during eucapnic voluntary hyperventilation and that the  $\dot{V}O_2$  and RPB would be lowered during moderate intensity cycling following BRJ supplementation when compared to placebo supplementation.

## **Chapter 3**

### **Methods**

#### **Participants**

After the study was approved by the Appalachian State University Institutional Review Board (IRB# 19-0021), class 1 ( $30 \leq \text{BMI} \leq 40$ ) obese males aged 18-40 years were recruited to participate. Subjects were initially screened over the telephone to ensure that they did not have any health complications that would prohibit them from taking part in the study. Subjects were excluded if they had known cardiovascular, respiratory, renal, or metabolic diseases, any signs/symptoms of disease, were a current smoker, or if they had a history of cigarette smoking over 0.5 pack-years. Subjects were excluded if they had participated in an exercise training program in the last six months, as they may have developed exercise adaptations that will limit the findings of this study. When subjects arrived at the laboratory for visit 1, they first signed an informed consent after a researcher had explained to them the requirements of the present study. Subjects then completed a medical history form. Prior to each visit, participants were instructed not to engage in strenuous physical activity in the 24 hours and refrain from caffeine and alcohol ingestion in the 12 hours prior to visiting the laboratory. Subjects were encouraged to visit the laboratory in a rested and hydrated state.

#### **Experimental Design**

A randomized, placebo-controlled experimental design was used to examine the effects of BRJ on  $\dot{V}O_{2RM}$  and RPB during moderate intensity exercise in obese male subjects.

All subjects visited the laboratory on three separate occasions. Subjects completed an informed consent document, and a medical history questionnaire. Following this, height and body mass were recorded, and then underwent tests to determine their body composition, pulmonary functioning, and  $\dot{V}O_{2\text{peak}}$ . During subsequent visits, subjects mimicked, at rest, the ventilation and breathing pattern corresponding to 50%, 70%, and 90%  $\dot{V}O_{2\text{peak}}$ , as measured during the initial visit, to measure  $\dot{V}O_{2\text{RM}}$ . Following a 20-min passive recovery period, subjects then completed 6-min of constant work rate exercise at 90% of the gas exchange threshold (GET) on a cycle ergometer. Maximal volitional mouth pressure was measured prior to the first hyperventilation challenge, immediately after the final hyperventilation challenge, and after the 20 minute rest period. At the beginning of all three visits, subjects had their resting blood pressure measured (SunTech Medical, Morrisville, NC); if their blood pressure was above 140/90, they were not allowed to participate that day. Subjects supplemented with either tomato juice (TJ; placebo beverage) or BRJ in the ten days prior to visits two and three. A 10-day washout period to separate the two supplementation periods was utilized. Subjects completed all laboratory visits at similar times of the day.

### **Pulmonary Functioning Test**

All subjects completed a spirometry and lung volume test in a whole body plethysmograph (Carefusion Vmax 62J Auto Box, Yorba Linda, CA) in accordance with ATS/ERS guidelines (74). The subjects were instructed to breathe through their mouth while wearing a nose-clip. The protocol for all spirometry tests included three breaths at a normal resting tidal volume, then a maximal inhalation, followed by a maximal exhalation, and finally another maximal inhalation. Each participant performed this protocol a minimum of

three times. Forced vital capacity (FVC), forced expiratory volume in one second ( $FEV_1$ ), forced mid expiratory flow rate ( $FEV_{25-75\%}$ ), and peak expiratory flow rate (PEFR) were recorded. Maximal voluntary ventilation (MVV) was measured as the maximal amount of air a participant could exhale in 12 seconds and reported as a one-minute value. During the MVV test, subjects were instructed to inspire and expire maximally, as fast as possible in order to expire the largest volume of air possible in the 12 seconds. Lung volumes that were measured (74) and reported parameters including total lung capacity (TLC), functional residual capacity (FRC), and residual volume (RV).

### **Peak Aerobic Capacity**

Subjects performed an incremental exercise test to determine  $\dot{V}O_{2peak}$  on an electronically braked, computer driven cycle ergometer (Lode Corival, Groningen, The Netherlands). Subjects were initially fitted to the cycle ergometer and the measurement settings were recorded to ensure the same position was utilized during the subsequent visits. Subjects were given verbal and written descriptions before testing for the evaluation of RPB, ratings of perceived unpleasantness (RPU), and ratings of perceived exertion (RPE). Subjects were instructed to point to the corresponding RPB, RPU (Borg 10 point scale), and RPE (Borg 6-20 rating scale) values during the test. Following description of the exercise protocol, the test began by sitting quietly on the cycle ergometer to obtain baseline measures. The exercise work rate started at 30 W and increased 30 W every minute thereafter (75). Subjects were instructed to maintain a cadence between 60 and 80 revolutions per minute. The test was terminated when the participant reached volitional exhaustion or if pedal rate fell below 60 revolutions per minute. Subjects breathed through a 2-way, low resistance non-

rebreathing valve (2700 Hans Rudolph, Shawnee, KS). Expired gases were analyzed continuously through an automated metabolic cart (ParvoMedics True One 2400, Sandy, Utah).  $\dot{V}O_2$  and the volume of carbon dioxide production ( $\dot{V}CO_2$ ) were corrected to STPD conditions, and minute ventilation ( $\dot{V}_E$ ) was corrected to BTPS conditions. Heart rate (HR) was recorded telemetrically (Polar, Kempele, Finland). Pulse oximetry was utilized to measure oxygen saturation (SpO<sub>2</sub>) with a pulse oximeter (Nellcor n-595).

### **Determination of the Gas Exchange Threshold**

Following the  $\dot{V}O_{2peak}$  test, the gas exchange threshold (GET) was determined through a cluster of measurements: 1) the first disproportionate increase in  $\dot{V}CO_2$  vs.  $\dot{V}O_2$ ; and 2) an increase in  $\dot{V}_E/\dot{V}O_2$  with no increase in  $\dot{V}_E/\dot{V}CO_2$  (21). Two members of the research team determined the GET independently. Their determinations had to be within 100 mL of each other, and the average of the two values was recorded as the GET. If their determinations were over 100 mL, a third member of the research team evaluated the GET. Once the GET was determined, 90% of that value was used to set the workload for the constant intensity cycling protocol.

### **Flow-Volume Loops**

Inspiratory and expiratory airflow rates were measured using pneumotachs located on both sides of the ventilatory line. Analog output signals from both pneumotachs were converted to airflow, summed (inspiratory flow minus expiratory flow), and integrated to yield volume. Tidal flow-volume loops (tFVL) were collected 1) each minute of the  $\dot{V}O_{2peak}$  test, 2) during the last two minutes of the eucapnic voluntary hyperventilation test, and 3)

during the last two minutes of the 6-min moderate intensity cycling test. Inspiratory capacity (IC) maneuvers were performed by the subject to aid in the placement of the tFVL within the thoracic gas compression-free maximum flow-volume loops (MFVL) obtained during the pulmonary function testing.

### **Supplementation Procedures**

Following the initial  $\dot{V}O_{2\text{peak}}$  test, subjects were randomly assigned to receive BRJ or TJ. Subjects underwent two 10-day supplementation periods separated by a 10-day washout period. Subjects were asked to record their diet during the first 10-day period, and then they were asked to follow that diet as closely as possible for the second 10-day period. The subjects were given a list of foods that are high in nitrates and were asked to refrain from eating these foods. They were also required to refrain from using mouthwash for the entirety of the study. During the experimental 10-day phase, subjects drank a 70 mL container of BRJ (Beet It Sports, James White Drinks, Ipswich, UK) once in the morning and once in the evening. Each serving contained  $6.45 \text{ mmol}\cdot\text{day}^{-1}$  of  $\text{NO}_3^-$  ( $12.9 \text{ mmol}\cdot\text{day}^{-1}$ ). On day 10, before they arrived for testing, they were instructed to drink both containers two hours before their scheduled visit. Tomato juice (Sacramento, Red Gold LLC, Elwood, IN) served as the placebo, as it has negligible natural nitrate levels (76). The tomato juice was also administered in 70 mL containers, and subjects followed the same consumption protocol. During the informed consent process, subjects were informed that both beverages were test beverages and neither was a placebo. When subjects completed the study, they were informed that TJ was in fact a placebo beverage.



## Eucapnic Hyperventilation Protocol

Subjects mimicked at rest the ventilation and breathing pattern corresponding to 50%, 70%, and 90%  $\dot{V}O_{2\text{peak}}$ . The order in which they completed the ventilation rates was randomly determined via a random number generator. Subjects were seated on the same cycle ergometer and at the same height as they were during their  $\dot{V}O_{2\text{peak}}$  test to assure standardization. Before commencing the eucapnic voluntary hyperventilation trials, subjects sat quietly on the cycle ergometer, while four minutes of resting data were collected. Following the resting period, the inspiratory line was connected to a large balloon reservoir, which was filled with moistened room air, from which the subject breathed. Expired  $\text{CO}_2$  and  $\text{O}_2$  was continuously monitored using a second set of analyzers (CD-3A Carbon Dioxide Analyzer and S-3A Oxygen Analyzer, AEI Technologies, Bastrop, Texas) and maintained at approximately 5-6% by the addition of  $\text{CO}_2$  from the balloon reservoir. The  $V_T$  and  $f_B$  were set to match that for the corresponding  $\dot{V}_E$  measured during the  $\dot{V}O_{2\text{peak}}$  test. The subjects were given instantaneous visual feedback of their tidal volume via a computer monitor. A metronome was set at the cadence of the desired breathing frequency. The first three minutes of the hyperventilation test were used to ensure subjects reached steady state and achieved the desired  $\dot{V}_E$ .  $\dot{V}O_2$  was recorded during the 4<sup>th</sup> and 5<sup>th</sup> minutes and the average was reported. The change in  $\dot{V}O_2$  from rest to that measured during hyperventilation, divided by the change in  $\dot{V}_E$  from rest to that measured during hyperventilation was used calculate the  $\dot{V}O_{2\text{RM}}$  ( $\dot{V}O_{2\text{RM}} = \Delta\dot{V}O_2 / \Delta\dot{V}_E$ ). tFVL were measured during the 5<sup>th</sup> minute of each eucapnic voluntary hyperventilation trial as previously described in the Flow-Volume Loops section. Following completion of the 5<sup>th</sup> minute of hyperventilation, subjects reported their RPB and RPU.

### **Maximum Volitional Mouth Pressure**

The maximal pressures that could be generated at the mouth during inspiration ( $PI_{max}$ ) and expiration ( $PE_{max}$ ) were measured according to the joint statement on respiratory muscle testing between the ATS & ERS (77).  $PI_{max}$  and  $PE_{max}$  were measured at baseline prior to hyperventilation, immediately following the third round of hyperventilation, and following the 20-minute passive recovery time prior to the moderate intensity cycling test. Using a mouthpiece connected to linear pressure transducer (Micro Direct Respiratory Pressure Meter, Vacumed, Ventura, California), subjects performed a maximal inspiratory effort a minimum of three times, from residual volume against an occluded airway for the measurement of  $PI_{max}$ . In addition, a maximal expiratory effort was performed from total lung capacity for the measurement of  $PE_{max}$ . Each expiratory maneuver was repeated at least three times. The maneuvers were alternated with approximately 30 seconds separating each effort. All maneuvers were sustained for 3 to 4 seconds (77). The largest values sustained for one second during the inspiratory and expiratory maneuvers were considered to be the subject's  $PI_{max}$  and  $PE_{max}$ .

### **Constant Intensity Cycling**

Following the eucapnic hyperventilation protocol, subjects were given 20 minutes to sit quietly before beginning the constant intensity cycling portion of the experiment. Subjects were seated on the cycle ergometer with the same seat and handle height as the previous  $\dot{V}O_{2peak}$  test. After obtaining three minutes of baseline measures, subjects began cycling at 90% of their aforementioned GET (21). The subjects were required to pedal at a

rate between 60 and 80 rpms. The subjects inspired room air and their expired air was measured using an automated metabolic cart.  $\dot{V}_E$ ,  $\dot{V}O_2$ , and  $\dot{V}CO_2$  were measured during the final three minutes of the test, as the first three minutes were to allow the subjects to reach steady state. Entering the final minute of the test, subjects were asked to point at the values that corresponded to their RPB, RPU, and RPE.  $P_{ET}CO_2$ ,  $SpO_2$ , and HR were continuously monitored.

### **Data Analysis**

Sample size estimates for changes in  $\dot{V}O_2$  following BRJ range from 6 to 10 and our enrollment target was calibrated to yield 6 evaluable obese male subjects. Previous reports showed that the effect sizes for  $\dot{V}O_2$ -related variables are in the range of 1.2 – 1.7, which was determined to be detectable with these sample sizes at a power above 95%. Sample size calculations were performed using G\*Power, and were based on data reported in published research (21). Data were analyzed using SPSS statistical software (SPSS Inc., Chicago, IL). The data were assessed for normality using the Kolmogorov-Smirnov test. Data was not deemed significant if it were not normally distributed. Pairwise comparisons were used to determine if statistically significant differences exist for the  $\dot{V}O_{2RM}$ , whole body  $\dot{V}O_2$ , and RPB during exercise following placebo and BRJ supplementations. Pearson's correlation coefficient was calculated and the relationship tested for significance between changes in  $\dot{V}O_{2RM}$  and RPB during exercise. The overall type-I error rate was set at 5% ( $\alpha \leq 0.05$ ). Data are expressed as mean  $\pm$  standard deviation (SD).

## Chapter 4

### Results

#### Subject Characteristics

A total of seven subjects consented to participate in the study. One subject was excluded from the study for not adhering to study guidelines. Six (n=6) subjects completed all visits of the study and were included in subsequent data analyses. Two subjects data were excluded from the eucapnic voluntary hyperventilation data set due to analyzer difficulties, so four (n=4) subjects are included in the cost of breathing data. Anthropometric data for subjects are displayed in **Table 1**. In addition to being obese, subjects also exhibited a substantial amount of body fat ( $37.6 \pm 5.8\%$ ) which was in agreeance with the subjects they we sought after to participate in this study. All subjects had no history of smoking. Subjects were recreationally active and not involved in any type of organized aerobic or anaerobic activity.

**Table 1.** Characteristics of study participants (n=6).

<b>Age</b> <b>(yr)</b>	<b>Ht</b> <b>(cm)</b>	<b>Wt</b> <b>(kg)</b>	<b>BMI</b> <b>(kg·m<sup>-2</sup>)</b>
23 ± 1	180.8 ± 11.5	114.1 ± 18.8	34.8 ± 3.2

Values are mean ± SD. Ht, height; Wt, weight; BMI, body mass index

#### Pulmonary Function

All subjects presented pulmonary function values that were above the lower limits of normal according to the prediction equations established by ATS/ERS (**Table 2**). However, FRC and RV were substantially lower than predicted (**Table 3**).

**Table 2.** Spirometry results of study subjects (n=6).

	<b>Measured</b>	<b>% pred</b>
<b>FVC (L)</b>	5.65 ± 1.1	97 ± 10
<b>FEV<sub>1</sub> (L)</b>	4.58 ± 0.9	95 ± 10
<b>FEV<sub>1</sub>/FVC (%)</b>	81.3 ± 7.0	94 ± 8
<b>PEF (L·s<sup>-1</sup>)</b>	9.08 ± 1.5	88 ± 18
<b>MVV (L·min<sup>-1</sup>)</b>	182.67 ± 38.2	95 ± 14

Values are mean ± SD. FVC, forced vital capacity; FEV<sub>1</sub>, forced expiratory volume in 1 sec; PEF, peak expiratory flow; MVV, maximal voluntary ventilation

**Table 3.** Lung volumes of study subjects (n=6).

<b>TLC (L)</b>	<b>FRC (L)</b>	<b>FRC (%TLC)</b>	<b>RV (L)</b>	<b>RV (%TLC)</b>
<b>%pred</b>	<b>%pred</b>	<b>%pred</b>	<b>%pred</b>	<b>%pred</b>
6.90 ± 0.9	2.47 ± 0.7	35.84 ± 7.7	0.96 ± 0.4	14.3 ± 6.4
94 ± 7	75 ± 9	73 ± 16	56 ± 29	72 ± 32

Values are mean ± SD. TLC, total lung capacity; FRC, functional residual capacity; RV, residual volume.

### Peak Aerobic Capacity

Subjects displayed  $\dot{V}O_{2peak}$  values which corresponded to 63 ± 12% of age predicted  $\dot{V}O_{2peak}$  values. Their absolute  $\dot{V}O_{2peak}$  values corresponded to 87 ± 12% of age predicted

values. At  $\dot{V}O_{2\text{peak}}$ , power was  $245 \pm 44$  W and  $\dot{V}_E$  was  $120.0 \pm 26.8$  L·min<sup>-1</sup>. **Table 4** displays additional metabolic data from the  $\dot{V}O_{2\text{peak}}$  test.

**Table 4** Metabolic data at maximal exercise (n=6).

<b><math>\dot{V}O_2</math> (L·min<sup>-1</sup>)</b>	$3.04 \pm 0.66$
<b><math>\dot{V}O_2</math> (mL·kg<sup>-1</sup>·min<sup>-1</sup>)</b>	$26.6 \pm 5.2$
<b><math>\dot{V}CO_2</math> (L·min<sup>-1</sup>)</b>	$3.47 \pm 0.66$
<b>HR (beats·min<sup>-1</sup>)</b>	$180 \pm 20$
<b>RER</b>	$1.15 \pm 0.06$
<b><math>\dot{V}_E/\dot{V}O_2</math></b>	$39.7 \pm 4.4$
<b><math>\dot{V}_E/\dot{V}CO_2</math></b>	$34.5 \pm 2.7$
<b><math>f_B</math> (breaths·min<sup>-1</sup>)</b>	$39.6 \pm 5.2$
<b><math>V_T</math> (L)</b>	$3.08 \pm 0.63$

Values are mean  $\pm$  SD.  $\dot{V}O_2$ , volume of oxygen;  $\dot{V}CO_2$ , volume of carbon dioxide; HR, heart rate; RER, respiratory exchange ratio;  $\dot{V}_E/\dot{V}O_2$ , ventilatory equivalent for oxygen;  $\dot{V}_E/\dot{V}CO_2$ , ventilatory equivalent for carbon dioxide;  $f_B$ , breathing frequency;  $V_T$ , tidal volume

## O<sub>2</sub> Cost of Breathing

### Rest

BRJ had a significant effect on lung volumes and respiratory mechanics at rest. IC was higher (BRJ:  $4.05 \pm 0.56$  L, TJ:  $3.58 \pm 0.56$  L;  $t(3)=-7.228$ ,  $p = 0.005$ ), EELV was lower (BRJ:  $2.55 \pm 0.91$  L, TJ:  $3.02 \pm 0.88$ ;  $t(3)=7.252$ ,  $p = 0.005$ ), the proportion of EELV to TLC

(EELV/TLC) was lower ( $t(3)=6.610$ ,  $p = 0.007$ ), and the total time spent inspiring during a respiratory duty cycle ( $T_i/T_{Tot}$ ) was reduced (BRJ:  $39.1 \pm 4\%$ , TJ:  $42.6 \pm 5\%$ ;  $t(3)=-4.645$ ,  $p = 0.017$ ). There were no significant differences within perceptual values. There were no other metabolic or ventilatory differences across conditions.

**Table 5.** Resting data before the EVH challenges (n=4).

	<b>BRJ</b>	<b>TJ</b>
$\dot{V}O_2$ (L·min <sup>-1</sup> )	0.413 ± 0.08	0.416 ± 0.12
$\dot{V}_E$ (L·min <sup>-1</sup> )	14.94 ± 8.73	16.29 ± 11.51
$\dot{V}_T$ (L)	1.69 ± 1.12	1.76 ± 0.38
$f_B$ (breaths·min <sup>-1</sup> )	12 ± 3	11 ± 3
EELV (%TLC)	38.1 ± 9.3*	45.3 ± 8.6*
EILV (%TLC)	62.3 ± 18.7	71.3 ± 12.6
$P_{ET}CO_2$ (mmHg)	32.5 ± 6.4	35.4 ± 11.2
SpO <sub>2</sub> (%)	98.4 ± 0.7	98.7 ± 0.5
RPB	0 ± 0	0 ± 0
RPU	0 ± 0	0 ± 0

Values are mean ± SD.  $\dot{V}O_2$ , volume of oxygen;  $V_E$ , ventilation;  $V_T$ , tidal volume;  $f_B$ , breathing frequency; EELV, end expiratory lung volume; TLC, total lung capacity; EILV, end inspiratory lung volume;  $P_{ET}CO_2$ , end tidal CO<sub>2</sub>; SpO<sub>2</sub>, saturation of O<sub>2</sub>; RPB, rating of perceived breathlessness; RPU, rating of perceived unpleasantness; \* indicates statistical significance ( $p<0.05$ )

## 50% EVH

The desired  $\dot{V}_E$  was  $34.75 \pm 8.46 \text{ L}\cdot\text{min}^{-1}$ .  $V_T$  was larger in TJ ( $t(3)=3.202$ ,  $p = 0.049$ ) while breathing at  $\dot{V}_E$  corresponding to 50%  $\dot{V}O_{2\text{peak}}$ . There were no other significant differences among any variables during the EVH challenge at 50%. Metabolic and ventilatory data are presented in **Table 6**.

**Table 6.** Data from the 50% EVH challenge (n=4).

	<b>BRJ</b>	<b>TJ</b>
$\dot{V}O_2 \text{ (L}\cdot\text{min}^{-1})$	$0.446 \pm 0.09$	$0.449 \pm 0.11$
$\dot{V}_E \text{ (L}\cdot\text{min}^{-1})$	$38.88 \pm 10.61$	$39.61 \pm 11.08$
$\dot{V}_T \text{ (L)}^*$	$1.89 \pm 0.45$ *	$2.20 \pm 0.54$
$f_B \text{ (breaths}\cdot\text{min}^{-1})$	$20 \pm 4$	$20 \pm 3$
<b>EELV (%TLC)</b>	$49.2 \pm 12.1$	$57.9 \pm 8.8$
<b>EILV (%TLC)</b>	$78.6 \pm 13.6$	$89.0 \pm 10.1$
<b>P<sub>ET</sub>CO<sub>2</sub> (mmHg)</b>	$48.2 \pm 3.1$	$44.6 \pm 2.8$
<b>SpO<sub>2</sub> (%)</b>	$98.3 \pm 1.2$	$99.3 \pm 0.5$
<b>RPB</b>	$0.6 \pm 0.9$	$0.5 \pm 1.0$
<b>RPU</b>	$0.6 \pm 0.9$	$0.8 \pm 1.5$

Values are mean  $\pm$  SD.  $\dot{V}O_2$ , volume of oxygen;  $V_E$ , ventilation;  $V_T$ , tidal volume;  $f_B$ , breathing frequency; EELV, end expiratory lung volume; TLC, total lung capacity; EILV, end inspiratory lung volume;  $P_{ET}CO_2$ , end tidal CO<sub>2</sub>; SpO<sub>2</sub>, saturation of O<sub>2</sub>; RPB, rating of perceived breathlessness; RPU, rating of perceived unpleasantness; \* indicates statistical significance ( $p < 0.05$ )



## 70% EVH

The desired  $\dot{V}_E$  was  $61.25 \pm 20.21 \text{ L}\cdot\text{min}^{-1}$ . During the EVH challenge at 70%,  $P_{ETCO_2}$  was significantly elevated in TJ conditions ( $t(3)=8.202$ ,  $p = 0.004$ ). There were no other significant differences among any variables. Metabolic and ventilatory data are presented in **Table 7**.

**Table 7.** Data from the 70% EVH challenge (n=4).

	<b>BRJ</b>	<b>TJ</b>
$\dot{V}O_2 \text{ (L}\cdot\text{min}^{-1}\text{)}$	$0.474 \pm 0.10$	$0.487 \pm 0.12$
$\dot{V}_E \text{ (L}\cdot\text{min}^{-1}\text{)}$	$67.83 \pm 20.89$	$67.76 \pm 21.78$
$\dot{V}_T \text{ (L)}$	$2.55 \pm 0.60$	$2.35 \pm 0.50$
$f_B \text{ (breaths}\cdot\text{min}^{-1}\text{)}$	$26 \pm 5$	$26 \pm 5$
<b>EELV (%TLC)</b>	$48.8 \pm 8.5$	$43.6 \pm 8.3$
<b>EILV (%TLC)</b>	$88.1 \pm 13.6$	$79.6 \pm 16.3$
<b><math>P_{ETCO_2}</math> (mmHg)</b>	$41.2 \pm 2.4^*$	$45.4 \pm 2.0$
<b>SpO<sub>2</sub> (%)</b>	$98.6 \pm 0.7$	$98.8 \pm 0.9$
<b>RPB</b>	$0.6 \pm 0.9$	$0.1 \pm 0.3$
<b>RPU</b>	$0.6 \pm 0.9$	$0.3 \pm .5$

Values are mean  $\pm$  SD.  $\dot{V}O_2$ , volume of oxygen;  $\dot{V}_E$ , ventilation;  $\dot{V}_T$ , tidal volume;  $f_B$ , breathing frequency; EELV, end expiratory lung volume; TLC, total lung capacity; EILV, end inspiratory lung volume;  $P_{ETCO_2}$ , end tidal CO<sub>2</sub>; SpO<sub>2</sub>, saturation of O<sub>2</sub>; RPB, rating of perceived breathlessness; RPU, rating of perceived unpleasantness; \* indicates statistical significance ( $p < 0.05$ )

## 90% EVH

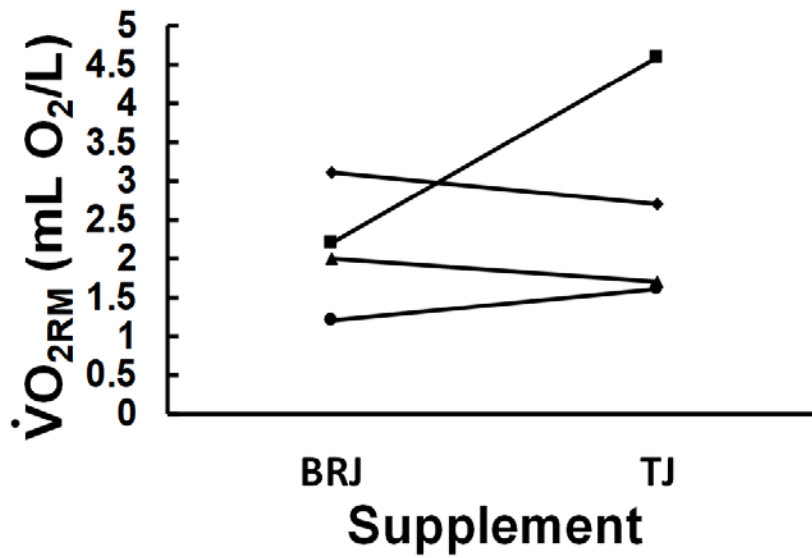
The desired  $\dot{V}_E$  was  $94.75 \pm 30.09 \text{ L}\cdot\text{min}^{-1}$ .  $\dot{V}_E$  was significantly reduced during the EVH challenge at 90% in BRJ when compared with in TJ ( $t(3)=3.240$ ,  $p = 0.048$ ). There were no other significant differences among variables. Metabolic and ventilatory data are presented in **Table 8**.

**Table 8.** Data from the 90% EVH challenge (n=4).

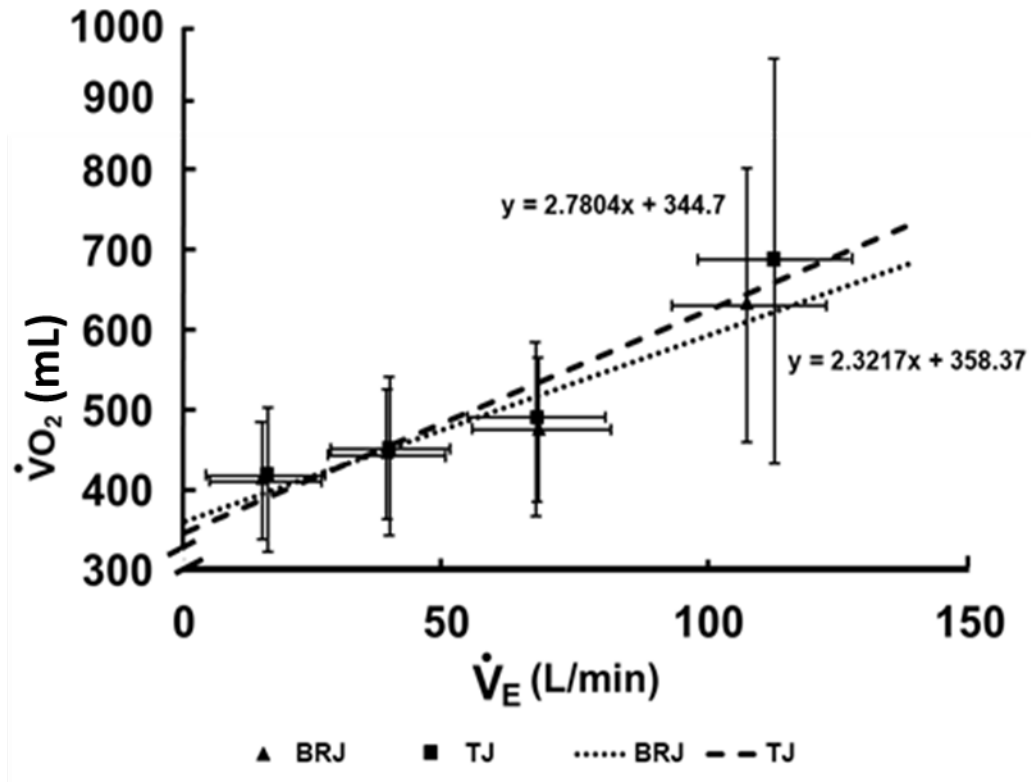
	<b>BRJ</b>	<b>TJ</b>
$\dot{V}O_2 \text{ (L}\cdot\text{min}^{-1})$	$0.633 \pm 0.19$	$0.685 \pm 0.28$
$\dot{V}_E \text{ (L}\cdot\text{min}^{-1})$	$107.72 \pm 30.37 *$	$113.07 \pm 32.46$
$\dot{V}_T \text{ (L)}$	$2.75 \pm 0.51$	$2.74 \pm 0.40$
$f_B \text{ (breaths}\cdot\text{min}^{-1})$	$34 \pm 4$	$35 \pm 4$
<b>EELV (%TLC)</b>	$41.1 \pm 7.0$	$38.1 \pm 4.3$
<b>EILV (%TLC)</b>	$82.9 \pm 5.5$	$79.9 \pm 8.6$
<b>P<sub>ET</sub>CO<sub>2</sub> (mmHg)</b>	$41.3 \pm 2.1$	$43.6 \pm 3.4$
<b>SpO<sub>2</sub> (%)</b>	$98.1 \pm 1.4$	$98.3 \pm 1.2$
<b>RPB</b>	$1.6 \pm 2.3$	$1.1 \pm 1.3$
<b>RPU</b>	$1.4 \pm 1.8$	$1.5 \pm 1.9$

Values are mean  $\pm$  SD.  $\dot{V}O_2$ , volume of oxygen;  $\dot{V}_E$ , ventilation;  $\dot{V}_T$ , tidal volume;  $f_B$ , breathing frequency; EELV, end expiratory lung volume; TLC, total lung capacity; EILV, end inspiratory lung volume; P<sub>ET</sub>CO<sub>2</sub>, end tidal CO<sub>2</sub>; SpO<sub>2</sub>, saturation of O<sub>2</sub>; RPB, rating of perceived breathlessness; RPU, rating of perceived unpleasantness; \* indicates statistical significance ( $p < 0.05$ )

$\dot{V}O_{2RM}$  was  $2.32 \pm 1.39$  mL O<sub>2</sub>/L during BRJ conditions and  $2.78 \pm 0.78$  mL O<sub>2</sub>/L during TJ conditions ( $t(3)=0.808$ ,  $p = 0.478$ ), **Figure 2**.



**Figure 2:** Individual subject responses in resting  $\dot{V}O_{2RM}$  across conditions.



**Figure 3:**  $\dot{V}O_{2RM}$  average trend lines across conditions, accompanied with average  $\dot{V}O_{2RM}$  data points.

### 6 Minute Constant Intensity Cycling

There were no significant differences between supplements at rest for any ventilatory mechanic or metabolic data. Resting metabolic and ventilatory data are presented in **Table 9**.

**Table 9.** Resting data before 6-min cycling (n=6).

	<b>BRJ</b>	<b>TJ</b>
<b>HR (bpm)</b>	80.3 ± 12.8	80.6 ± 10.4
<b><math>\dot{V}O_2</math> (mL·kg<sup>-1</sup>·min<sup>-1</sup>)</b>	4.05 ± 0.65	4.22 ± 0.96
<b><math>\dot{V}O_2</math> (L·min<sup>-1</sup>)</b>	0.473 ± 0.133	0.501 ± 0.173
<b><math>\dot{V}_E</math> (L·min<sup>-1</sup>)</b>	19.21 ± 8.00	19.54 ± 8.94
<b><math>\dot{V}_T</math> (L)</b>	1.39 ± 0.40	1.42 ± 0.51
<b><math>f_B</math> (breaths·min<sup>-1</sup>)</b>	14 ± 3	14 ± 3
<b>EELV (%TLC)</b>	42.8 ± 7.2	38.5 ± 6.2
<b>EILV (%TLC)</b>	65.7 ± 12.0	59.9 ± 19.2

Values are mean ± SD. HR, heart rate;  $\dot{V}O_2$ , volume of oxygen;  $\dot{V}_E$ , ventilation;  $\dot{V}_T$ , tidal volume;  $f_B$ , breathing frequency; EELV, end expiratory lung volume; TLC, total lung capacity; EILV, end inspiratory lung volume; \* indicates statistical difference(p>0.05)

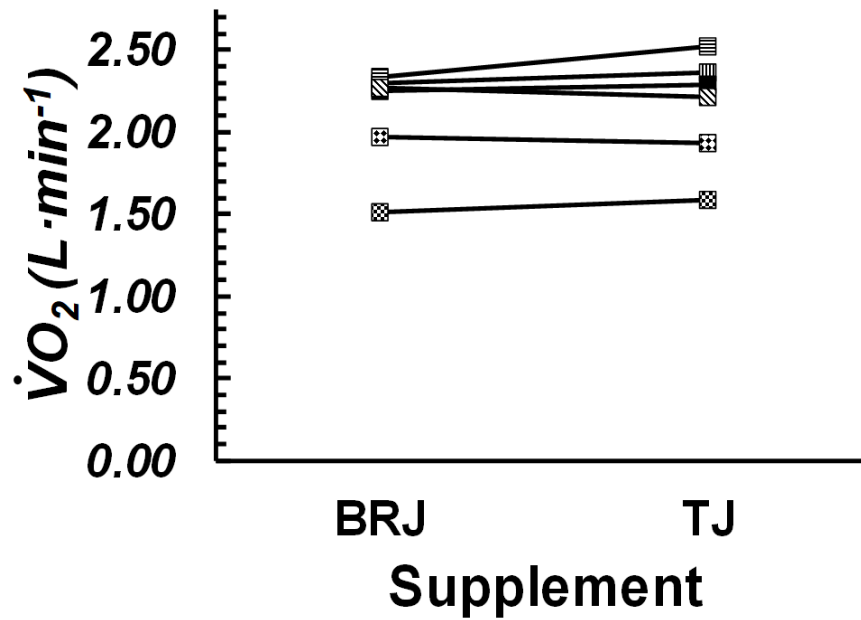
The average  $W_R$  assigned to subjects was  $132 \pm 24$  W ( $54 \pm 8$  %  $W_{Rmax}$ ). Breathing pattern during exercise was altered:  $\dot{V}_T$  was significantly lower in BRJ compared with in TJ ( $t(5)=3.438$ ,  $p = 0.018$ ,  $d = 1.40$ ); however,  $\dot{V}_E$  was unaltered between supplements ( $t(5)=0.442$ ,  $p = 0.677$ ,  $d = 0.18$ ). There were no other significant differences for any perceptual, ventilatory mechanic, and metabolic data between supplements. The  $O_2$  cost of cycling was not significantly different (BRJ:  $14.21 \pm 2.81$  mL·min<sup>-1</sup>·W<sup>-1</sup>, TJ:  $14.59 \pm 2.53$  mL·min<sup>-1</sup>·W<sup>-1</sup>;  $t(5)=1.595$ ,  $p = 0.172$ ).

**Table 10.** 6-min cycling data (n=6).

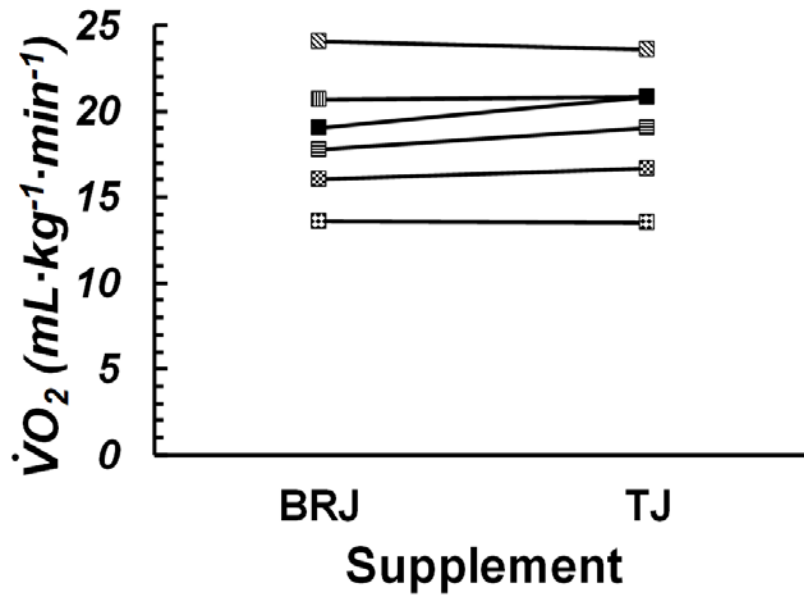
	<b>BRJ</b>	<b>TJ</b>
<b>HR (bpm)</b>	136.7 ± 21.9	137.4 ± 14.1
<b><math>\dot{V}O_2</math> (mL·kg<sup>-1</sup>·min<sup>-1</sup>)</b>	18.52 ± 3.64	19.07 ± 3.56
<b><math>\dot{V}O_2</math> (L·min<sup>-1</sup>)</b>	2.107 ± 0.318	0.2.151 ± 0.338
<b><math>\dot{V}_E</math> (L·min<sup>-1</sup>)</b>	66.16 ± 9.43	67.68 ± 13.35
<b><math>\dot{V}_T</math> (L)</b>	2.56 ± 0.55*	2.83 ± 0.65
<b><math>f_B</math> (breaths·min<sup>-1</sup>)</b>	27 ± 4	24 ± 4
<b>EELV (%TLC)</b>	39.2 ± 13.9	31.1 ± 17.2
<b>EILV (%TLC)</b>	77.5 ± 13.1	68.9 ± 19.7
<b>P<sub>ET</sub>CO<sub>2</sub> (mmHg)</b>	38.3 ± 4.6	39.8 ± 2.9
<b>SpO<sub>2</sub> (%)</b>	96.0 ± 1.3	96.5 ± 1.5
<b>RPB</b>	2.9 ± 2.6	3.4 ± 2.3
<b>RPU</b>	2.7 ± 2.4	3.1 ± 2.5
<b>RPE</b>	12.3 ± 3.7	11.8 ± 3.3

Values are mean ± SD. HR, heart rate;  $\dot{V}O_2$ , volume of oxygen;  $\dot{V}_E$ , ventilation;  $\dot{V}_T$ , tidal volume;  $f_B$ , breathing frequency; EELV, end expiratory lung volume; TLC, total lung capacity; EILV, end inspiratory lung volume; P<sub>ET</sub>CO<sub>2</sub>, end tidal CO<sub>2</sub>; SpO<sub>2</sub>, saturation of O<sub>2</sub>; RPB, rating of perceived breathlessness; RPU, rating of perceived unpleasantness; RPE, rating of perceived exertion; \* indicates statistical significance (p<0.05)

RER (BRJ:  $0.979 \pm 0.062$ , TJ:  $1.018 \pm 0.070$ ;  $t(5)=1.531$ ,  $p = 0.186$ ,  $d = 0.63$ ) and  $\dot{V}CO_2$  (BRJ:  $2.064 \pm 0.358$ , TJ:  $2.193 \pm 0.384$ ;  $t(5)=2.295$ ,  $p = 0.070$ ,  $d = 0.94$ ) were unaltered during exercise, suggesting no change in substrate utilization.

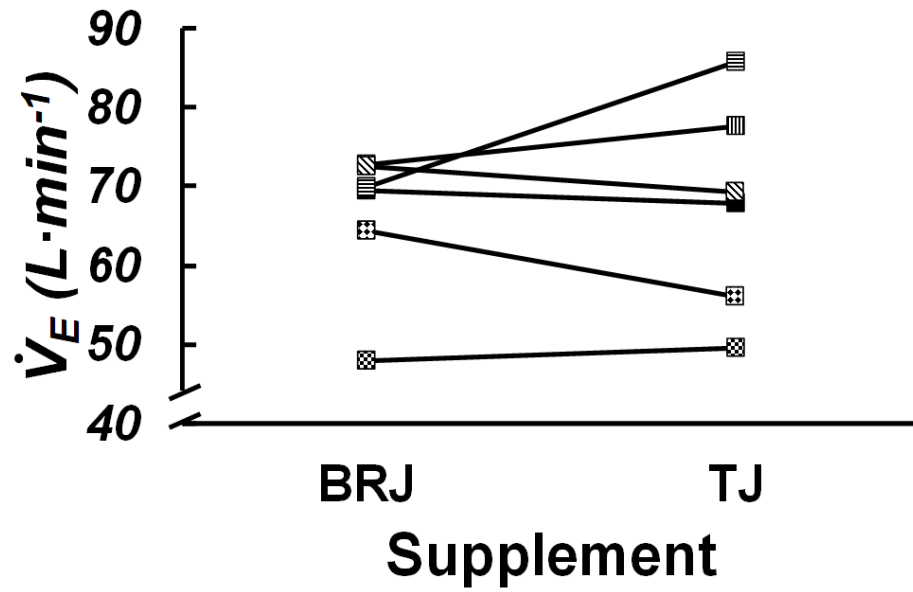


**Figure 4:** Individual subject responses in  $\dot{V}O_2$  ( $L \cdot \text{min}^{-1}$ ) between supplements during constant-load cycling. There was no main effect ( $t(5)=1.210$ ,  $p = 0.280$ ,  $d = 0.49$ ).

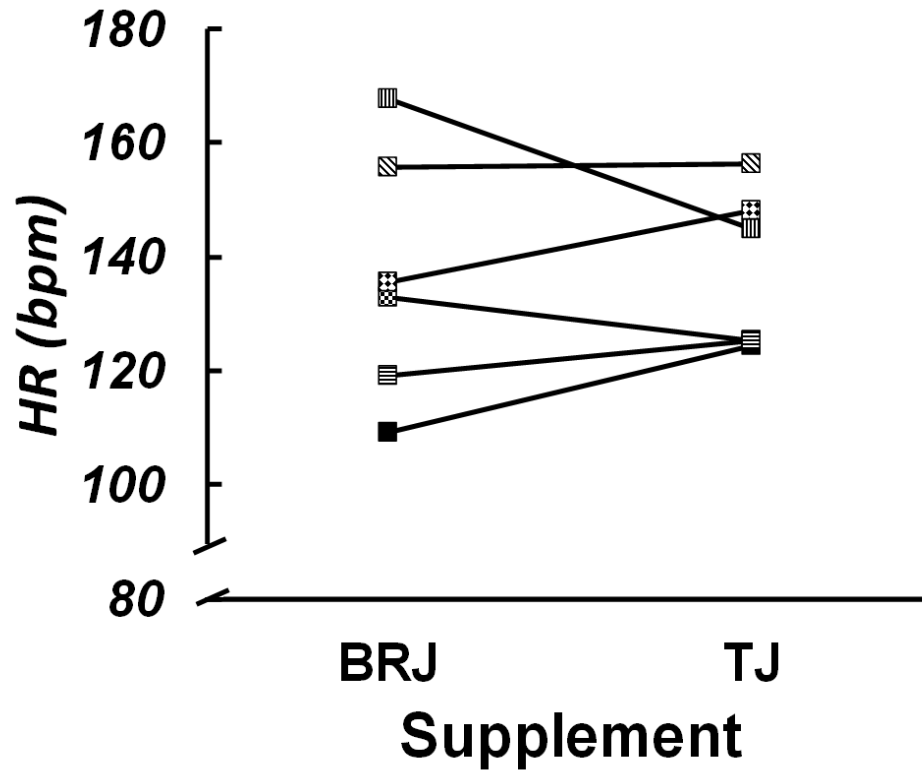


**Figure 5:** Individual subject responses in  $\dot{V}O_2$  (mL·kg<sup>-1</sup>·min<sup>-1</sup>) during constant-load cycling between supplements. There was no main effect ( $t(5)=1.600$ ,  $p = 0.170$ ,  $d = 0.65$ ).





**Figure 6:** Individual subject responses in  $\dot{V}_E$  during constant-load cycling between supplements. There was no main effect ( $t(5)=0.442$ ,  $p = 0.677$ ).



**Figure 7:** Individual subject responses in HR during constant-load cycling between supplements. There was no main effect ( $t(5)=0.121$ ,  $p = 0.908$ ).

There were no significant differences among perceptual values during exercise (RPB ( $t(5) = 1.225$ ,  $p = 0.275$ ), RPU ( $t(5)=1.536$ ,  $p = 0.185$ ), and RPE ( $t(5)=-1.464$ ,  $p = 0.203$ )).

The  $\dot{V}O_{2RM}$  during cycling was not significantly different between supplements (BRJ:  $155.46 \pm 83.21$  mL, TJ:  $180.46 \pm 106.70$  mL;  $t(3)=0.798$ ,  $p = 0.640$ ). This equates to 7.38% of total  $\dot{V}O_2$  in BRJ conditions and 8.39% in TJ conditions.

### Correlations of the Changes in $\dot{V}O_{2RM}$ and RPB

The changes between in  $\dot{V}O_{2RM}$  and RPB between supplements was not significantly correlated ( $p = 0.886$ ).

### Maximal Volitional Mouth Pressure

There were no differences for  $PI_{max}$  at baseline, following the third bout of the EVH challenge, following the 20-minute recovery period, or the percent change from any of the time points. The inspiratory data is in **Table 11**.

**Table 11.** Inspiratory values from the maximal volitional mouth pressure measurements (n=6).

	<b>BRJ</b>	<b>TJ</b>
<b>Baseline (cm H<sub>2</sub>O)</b>	136 ± 22	133 ± 15
<b>Post EVH (cm H<sub>2</sub>O)</b>	129 ± 16	136 ± 18
<b>Change from baseline (%)</b>	-4 ± 12	2 ± 11
<b>Post Rest (cm H<sub>2</sub>O)</b>	128 ± 12	128 ± 17
<b>Post Rest Change from Baseline %</b>	-3 ± 13	-5 ± 17
<b>Post Rest Change from Post EVH (%)</b>	-1 ± 11	-5 ± 10

Values are mean ± SD.; cm H<sub>2</sub>O, centimeters of water.

There were no differences for  $PE_{max}$  at baseline, following the third bout of the EVH challenge, following the 20-minute recovery period, or the percent change from any of the time points. The expiratory data is in **Table 12**.

**Table 12.** Expiratory values from the maximal volitional mouth pressure measurements (n=6).

	<b>BRJ</b>	<b>TJ</b>
<b>Baseline (cm H<sub>2</sub>O)</b>	166 ± 45	148 ± 33
<b>Post EVH (cm H<sub>2</sub>O)</b>	163 ± 44	158 ± 36
<b>Change from baseline (%)</b>	-2 ± 11	7 ± 10
<b>Post Rest (cm H<sub>2</sub>O)</b>	168 ± 32	155 ± 34
<b>Post Rest Change from Baseline %</b>	-3 ± 12	-6 ± 17
<b>Post Rest Change from Post EVH (%)</b>	5 ± 11	-1 ± 10

Values are mean ± SD.; cm H<sub>2</sub>O, centimeters of water.

## Chapter 5

### Discussion

The purpose of this study was to examine the  $\dot{V}O_{2RM}$  and ratings of perceived breathlessness (RPB) during eucapnic voluntary hyperventilation and to examine  $\dot{V}O_2$  and RPB during moderate intensity cycling in young, obese males following beetroot juice (BRJ) supplementation. To our knowledge, this is the first to study to examine the efficacy of BRJ in relation to  $\dot{V}O_{2RM}$  during exercise in obese but otherwise healthy men.

These data demonstrate that BRJ did not alter  $\dot{V}O_{2RM}$  or any perceptual values during the eucapnic voluntary hyperventilation tests compared with those following TJ supplementation. Further, during moderate intensity cycling, BRJ did not have an effect on any metabolic or perceptual values. The changes in  $\dot{V}O_{2RM}$  and RPB from TJ to BRJ also did not correlate. Thus, our findings in obese men indicate that BRJ does not alter skeletal respiratory or locomotor muscle efficiency in obese men, and does not provide an ergogenic effect during moderate intensity exercise nor during eucapnic voluntary hyperventilation.

All subjects who participated were young and obese but otherwise healthy. Their medical histories were absent of any known cardiovascular, metabolic, respiratory, or renal diseases that would alter their performance. All subjects were recreationally active and did not partake in any type of organized aerobic or anaerobic training that would potentially give them exercise related physiological adaptations. The American College of Sports and Medicine recommends that adults participate in cardiorespiratory exercise training for at least  $150 \text{ min} \cdot \text{wk}^{-1}$  at a moderate intensity, or for  $75 \text{ min} \cdot \text{wk}^{-1}$  at a vigorous intensity. In addition, they recommend that adults weight train 2-3 days per week with a focus on large muscle groups (78). Our subjects did not meet any of these recommendations, but they were not

sedentary. Subjects in our study engaged in activities 2-3 times per week that did not meet the criteria of cardiorespiratory training. Activities that our subjects participated in included light walking, hiking, and unstructured weight lifting a few times a month.

### **Pulmonary Function**

All subjects had normal spirometry measures based on their age, height, and sex. However, subjects did exhibit reduced lung volumes and capacities when compared to their predicted values, with notable reductions in FRC, ERV, and RV, which are consistent with other literature on obese populations (79, 80). One particular study reported that when obese men decreased their body weight by 11% and fat mass by 21%, that their FRC increased by 18% (80). These changes were observed with the absence of changes in FEV<sub>1</sub> and FEV<sub>1</sub>/FVC. Lung volumes decrease due to the increased fat surrounding the thorax in obesity, but lung function is often not altered until morbid obesity (BMI  $\geq$  40 kg·m<sup>-2</sup>). All pulmonary function data were collected according to ATS/ERS guidelines with well-established and well-maintained equipment to ensure precise measurements.

### **Peak Aerobic Capacity**

Subjects exhibited normal cardiorespiratory functioning by achieving normal values for absolute  $\dot{V}O_2$  ( $87 \pm 12\%$ ), but displayed poor physical fitness as shown by their relative  $\dot{V}O_2$  only reaching  $63 \pm 12\%$  of their age predicted maximums. This is consistent with the work of Lorenzo and Babb, in which they reported that there was no statistical differences between the absolute  $\dot{V}O_{2peak}$  between normal weight and obese individuals, but normal weight individuals had a significantly higher relative  $\dot{V}O_{2peak}$  when compared with obese individuals. The average  $W_R$  only reached  $245 \pm 44$  W, which is also low compared to their predicted values. ( $64 \pm 10\%$ ). These results confirm that we recruited individuals without

exercise based physiological adaptations. However, these results are similar to previous studies; there are negative correlations between BMI and  $W_{Rmax}$  in young males ( $r = -0.87$ ,  $p = 0.0001$ ) (81), and BMI negatively correlates with relative  $\dot{V}O_{2max}$  ( $r = -0.48$ ,  $p < 0.01$ ) (82).

### **O<sub>2</sub> Cost of Breathing**

During resting conditions before the start of the EVH challenges,  $\dot{V}O_2$  and  $\dot{V}_E$  were not different between conditions. However, respiratory mechanics were altered; EELV was elevated in TJ when compared to BRJ, as was EELV/TLC. IC was elevated in BRJ when compared to TJ, and the respiratory duty cycle was reduced after BRJ supplementation when compared to TJ. EELV/TLC at rest is at the point where FRC falls, which is approximately 35% of TLC in normal, healthy-weight individuals. BRJ brought FRC to a point that was closer to normal (i.e., non-obese) when compared to TJ (BRJ:  $38.1 \pm 9.3$ , TJ:  $45.3 \pm 8.6$ ;  $p = 0.005$ ). An elevation in EELV/TLC at rest is associated with expiratory flow limitation (EFL), which is where the expired portion of a tFVL touches the maximal FVL (83). This means that expiratory flow cannot be further increased by increasing expiratory muscle effort because it is maximum at that tidal volume. It is typical for EELV to increase as exercise increases, but an increase at rest is associated with negative health outcomes. For example, COPD is associated with EFL, and BRJ could be a means of decreasing this. Only one study has analyzed the effects of BRJ on COPD patients, but they only analyzed its effects on exercise tolerance, in which there was no benefit following BRJ supplementation on moderate intensity exercise, and did not report any pulmonary function or respiratory mechanics data (84). In the current study, IC was lower in TJ conditions simply as a function of the elevated EELV. Neither condition has the ability to alter TLC, so IC was increased in BRJ conditions.

$T_i/T_{Tot}$  or the respiratory duty cycle was decreased at rest following BRJ supplementation when compared to TJ. Obese individuals express an elevated respiratory duty cycle at rest (85), which, since inspiration requires active respiratory muscle contraction, means that they spend more time with their muscles contracted during resting breathing. At rest, expiration is passive and does not require muscle contraction. The more time that musculature is contracting, the more energy that is used per respiratory cycle. BRJ may have the potential to lower the time the respiratory muscles spend contracting per respiratory cycle. However, in the current study we did not see this manifest as a meaningful reduction in oxygen consumption.

$\dot{V}O_{2RM}$  was unaffected by BRJ supplementation, contrary to our initial hypothesis. Not only were whole body  $\dot{V}O_2$  values while breathing at 50%, 70%, and 90%  $\dot{V}O_{2peak}$  not different between the two conditions, but the slopes relating  $\dot{V}O_2$  to  $\dot{V}_E$  were not different;  $\dot{V}O_{2RM}$  was  $2.8 \pm 0.78 \text{ mL O}_2 \cdot \text{L}^{-1}$  and  $2.3 \pm 1.39 \text{ mL O}_2 \cdot \text{L}^{-1}$  in TJ and BRJ, respectively. These values are smaller than the values reported by Cherniak (62). Cherniak reported that the average  $\dot{V}O_{2RM}$  in obese individuals was  $3.45 \text{ mL O}_2 \cdot \text{L}^{-1}$ , which was roughly 3 times more than his value ( $1.2 \pm 0.6 \text{ mL O}_2 \cdot \text{L}^{-1}$ ) of normal weight individuals. While our  $\dot{V}O_{2RM}$  data are around double the values that have been reported in normal weight men, they still are reduced when compared to that in obese subjects of previous studies. It is well known that  $\dot{V}O_{2RM}$  increases with age (64, 86). This has been attributed to a number of factors such as increased airway resistance with age. Thus, differences in the ages of study participants may partially, at least, explain why our values are lower. The data of Cherniak were collected from middle aged participants ( $46 \pm 10$ ), which included some women, who have a higher  $\dot{V}O_{2RM}$  (55). Our data were collected in young obese males ( $23 \pm 1$ ). There were also glaring



methodological differences between the studies, as we had subjects breath at a predetermined  $V_T$  and  $f_b$ , while Cherniak allowed for spontaneous breathing. We also used the addition of  $CO_2$  to inspired air to increase the drive to breathe, while he used an increased dead space to achieve this.

In summary, a cluster of factors could explain why are values were substantially lower than those reported by Cherniak. His inclusion of women and a middle aged population would yield significant elevations in  $\dot{V}O_{2RM}$ . His use of spontaneous breathing was much different from our protocol. Our subjects had to be consciously aware of their breathing, which causes conscious centers of the brain such as the motor cortex to become involved. Spontaneous breathing is more natural as well, whereas our  $f_B$  and  $V_T$  values were assigned based off of their  $\dot{V}O_{2peak}$  data.

During the EVH challenge at 90%, subjects who had consumed BRJ had a significantly lower  $\dot{V}_E$  than when they had consumed TJ. The target  $\dot{V}_E$  was  $94.75 \pm 30.09$   $L \cdot \text{min}^{-1}$ , so both groups overshot the desired  $\dot{V}_E$ , but the TJ group overshot it significantly more. This could not be attributed to familiarization, as out of our four subjects, two began with BRJ and two began with TJ. We had subjects hyperventilate at each rate for five minutes, but we did not sample data until the last two minutes. It is possible that subjects became fatigued during those last two minutes during BRJ supplementation and were unable to reach the same  $\dot{V}_E$ . However, elevations in perceptual responses did not accompany this. Due to two subjects being omitted from statistical analysis, and we could only analyze four subjects, this could produce a type I error. It is unlikely that respiratory fatigue was to blame for this, as Aaron et al. (16) had subjects hyperventilate at 100% of their  $\dot{V}O_{2max}$ , and five out of the six subjects were able to do this for over 15 minutes. As stated above, there was also

no increases in perceptual responses accompanying the reduced  $\dot{V}_E$  following BRJ conditions. Perceptual values, in particular, are multi-faceted, and are not all physiological based, with implications being tied in with gender, age, emotional state, and previous experiences (87). These influences accompanied with BRJ's inability to lower  $\dot{V}O_{2RM}$ , all account for the lack of statistical significance among perceptual values. While  $\dot{V}_E$  was elevated at 90% in TJ conditions,  $\dot{V}O_{2RM}$  was not, indicating that there was no increased metabolic efficiency in the respiratory muscles. The failure to increase metabolic efficiency is consistent with the failure to lower perceptual values in BRJ conditions.

### **Moderate Intensity Cycling**

BRJ supplementation did not affect metabolic parameters during moderate intensity cycling. However, it did decrease  $V_T$ , while  $f_B$  and  $\dot{V}_E$  were unaltered. One function of an increased  $V_T$  is to remove  $CO_2$  from the body.  $\dot{V}CO_2$  was elevated in TJ conditions, but the difference between  $\dot{V}CO_2$  in BRJ and TJ failed to reach statistical significance ( $p = 0.07$ ,  $d = 0.94$ ). However, there was a large effect size for  $\dot{V}CO_2$ . If BRJ was able to reduce the amount of  $CO_2$  in the blood, less  $CO_2$  would be expired which could reduce  $V_T$ . I believe with more subjects,  $\dot{V}CO_2$  would have reached statistical significance and would have been able to explain our reduction in  $V_T$ .

Our subjects displayed an elevated  $O_2$  cost of cycling compared to normal weight individuals. In normal weight individuals values of  $9-10 \text{ mL}\cdot\text{min}^{-1}\cdot\text{W}^{-1}$  are considered typical (88), while our subjects had values of  $14.21 \pm 2.81 \text{ mL}\cdot\text{min}^{-1}\cdot\text{W}^{-1}$  in BRJ and  $14.59 \pm 2.53 \text{ mL}\cdot\text{min}^{-1}\cdot\text{W}^{-1}$ . These findings are similar to the only other BRJ study that has been conducted within an obese population (27). Obese adolescents in that study displayed an  $O_2$  cost of exercise that was  $12.9 \pm 1.1 \text{ mL}\cdot\text{min}^{-1}\cdot\text{W}^{-1}$  in BRJ and  $13.3 \pm 1.7 \text{ mL}\cdot\text{min}^{-1}\cdot\text{W}^{-1}$  in

their placebo. Both studies used a moderate intensity exercise; however, the previous study also examined the O<sub>2</sub> cost during severe intensity exercise until exhaustion. In agreement with our study, they also reported that BRJ did not lower  $\dot{V}O_2$  during moderate intensity exercise; however, subjects were able to exercise longer during severe intensity exercise. Breese (49) and Thompson (50) also recorded results that showed that BRJ did not improve exercise efficiency at moderate intensities in normal weight men, but did specifically during vigorous intensity. If BRJ does preserve PCr more efficiently during exercise when compared to baseline (57), we would expect to see greater effects at higher exercise intensities, as more Type II muscle fibers would be recruited, and it is well known that Type II muscle fibers have higher concentrations of PCr. When cycling at 75% of  $\dot{V}O_{2max}$ , normal weight men and women recruit 55% of type II muscle fibers when estimated via a PCr/Cr ratio method (89). Our subjects exercised at 69.3% of  $\dot{V}O_{2peak}$  during BRJ conditions and 70.8% during TJ conditions. From this it can be interpreted that our subjects recruited at least half of their type II muscle fibers. More research is needed in the field of BRJ supplementation to obese individuals. The additional adipose tissue could slow down the metabolic rate to the point where the NO<sub>3</sub><sup>-</sup> supplementation is blunted.

### **Dosage**

In the present study we had subjects consume 12.9 mmol·day<sup>-1</sup> NO<sub>3</sub><sup>-</sup>. This amount was chosen because we were giving the supplement to obese men. BRJ supplementation is under-researched in obesity, so the dosage was chosen to ensure that there was enough NO<sub>3</sub><sup>-</sup> being consumed to be metabolized. Our dosage was approximately double of that of normal weight subjects (21, 23), but our subjects did not weigh double compared to those subjects. This would suggest that our subjects were not under dosed and received a large enough dose

to possibly exhibit physiological effects. Rasica et al. supplemented obese adolescent subjects with only  $5 \text{ mmol}\cdot\text{day}^{-1}$  of  $\text{NO}_3^-$ , and they did not find significant effects at moderate intensity exercise (27). Other studies have yielded positive physiological responses to BRJ at moderate intensity exercise including dosages of  $5.5 \text{ mmol}\cdot\text{day}^{-1}$  and  $6.2 \text{ mmol}\cdot\text{day}^{-1}$  of  $\text{NO}_3^-$  for 5 and 6 days respectively (21, 23). Based on previous studies, our dosage and supplementation period was sufficient to elicit changes. Furthermore, subjects were given a list of foods that were high in  $\text{NO}_3^-$  and asked not to eat them, to ensure that diet would not have an effect on our results. Thus, subjects consumed more than enough  $\text{NO}_3^-$  in order for it to be at elevated concentrations in the body and have the potential to elicit physiological changes in the body.

### **Limitations**

The small sample size is the main limitation of this study. Many of the variables in the present study have moderate to large effects sizes but no statistically significant differences. A larger sample size may yield statistical significance for the variables studied. Our analyzers that measured expired gases malfunctioned for two subjects which resulted in the omission of their  $\dot{V}\text{O}_{2\text{RM}}$  data, further reducing our sample size. If those other two subjects had been included with viable data, we may have seen a significant effect. Additionally, we did not directly measure plasma  $\text{NO}_2^-$  levels orally, so it is unknown if subjects had elevated levels of  $\text{NO}_2^-$ , and therefore  $\text{NO}_3^-$ . However, previous studies have reported elevated  $\text{NO}_3^-$  and  $\text{NO}_2^-$  using an identical supplementation protocol in normal weight subjects (21, 22, 23, 25). If subjects followed the supplementation procedures, we expect that their values would have been similar.

## Conclusion

In conclusion, the consumption of  $12.9 \text{ mmol}\cdot\text{day}^{-1}$  of  $\text{NO}_3^-$  in the form of BRJ does not lower  $\dot{V}\text{O}_{2\text{RM}}$  at rest or during exercise, nor does it lower whole-body  $\dot{V}\text{O}_2$  during moderate intensity cycling in obese young men. During moderate intensity exercise,  $V_T$  was reduced following BRJ conditions and  $\dot{V}\text{CO}_2$  was reduced as well but the reduction in  $\dot{V}\text{CO}_2$  was not significant ( $p = 0.07$ ,  $d = 0.94$ ). A larger sample size may have made this value significant indicating less  $\text{CO}_2$  production during moderate intensity exercise in obese young men following BRJ supplementation.

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## Appendices

### Appendix A.a: Subject Information

ID	STATUS	RACE	AGE	DOB	SEX
001	Complete	Caucasian	23	06/11/95	M
002	Complete	Caucasian	24	03/25/94	M
003	Complete	Caucasian	23	09/17/95	M
004	Complete	Caucasian	24	11/02/94	M
005	Complete	Caucasian	22	08/05/96	M
007	Complete	Caucasian	21	07/25/97	M

ID	HT (cm)	WT (kg)	WT:HT	BMI	%BF	FM (kg)	FFM (kg)	Body Density
001	185.0	116.0	0.627	33.9	34.70	40.25	75.74343	1.0212
002	191.0	113.3	0.593	31.1	29.20	33.09	80.24047	1.0329
003	161.5	93.3	0.577	35.8	40.50	37.77	55.4897	1.0092
004	189.0	142.2	0.753	39.8	46.40	65.99	76.23206	0.9971
005	186.0	125.7	0.676	36.3	35.80	45.01	80.7116	1.0190
007	172.0	93.8	0.546	31.7	38.80	36.41	57.43069	1.0127

ID	HxDOE	snore-subjective	snore-objective	Hx Asth	Asthma Info	Smoke HX	Pks/day	Years Sm	Pks/yr
001	N	Y	Y	N	NA	N	0	0	0
002	N	N	N	N	NA	N	0	0	0
003	N	N	N	N	NA	N	0	0	0
004	N	N	N	N	NA	N	0	0	0
005	N	Y	Y	N	NA	N	0	0	0
007	N	Y	Y	N	NA	N	0	0	0

ID	Ex	Type Ex	ExFreq (per/wk)	ExDuration (min)
001	Y	Hiking	2-3	60-120
002	Y	Hiking, some weights at house	2-3	120.0
003	Y	some resistance training	3	60.0
004	Y	some resistance training	2-3	60.0
005	Y	walking	2-3	20.0
007	Y	Run	1	15.0

**Appendix A.a: Subject Information continued**

<b>ID</b>	<b>Meds</b>
<b>001</b>	None
<b>002</b>	None
<b>003</b>	Xyzal
<b>004</b>	None
<b>005</b>	Terbinafine, Doxycycline
<b>007</b>	None

**Appendix A.b: Pulmonary Function**

<b>ID</b>	<b>SPIRO</b>	<b>FVC</b>	<b>FVC PP NHANES</b>	<b>FVC P Knudson</b>	<b>FVC PP Knudson</b>	<b>FEV1</b>	<b>FEV1 PP NHANES</b>
<b>001</b>	Spiro	6.11	101	5.728	107	5.60	112
<b>002</b>	Spiro	7.37	114	6.156	120	5.26	99
<b>003</b>	Spiro	4.37	97	4.342	101	3.39	89
<b>004</b>	Spiro	5.91	93	6.038	98	5.07	98
<b>005</b>	Spiro	5.58	91	5.713	98	4.57	90
<b>007</b>	Spiro	4.54	87	4.813	94	3.61	82

<b>ID</b>	<b>FEV1 P Knudson</b>	<b>FEV1 PP Knudson</b>	<b>FEV1/FVC</b>	<b>FEV1/FVC PP Knudson</b>	<b>25-75%</b>	<b>25-75% PP</b>
<b>001</b>	4.946	113	91.653028	10614.348	7.55	148
<b>002</b>	5.321	99	71.370421	8256.849	4.68	89
<b>003</b>	3.727	91	77.574371	9037.9747	2.86	67
<b>004</b>	5.217	97	85.786802	9928.1115	6.19	119
<b>005</b>	4.935	93	81.899642	9482.5661	4.76	92
<b>007</b>	4.144	87	79.515419	9235.3236	3.25	69

<b>ID</b>	<b>PEF</b>	<b>PEF PP</b>	<b>FET100%</b>	<b>MVV</b>	<b>MVV PP</b>
<b>001</b>	12.36	114	7.15	245	123
<b>002</b>	6.88	60	7.35	197	93
<b>003</b>	8.14	93	13.76	133	87
<b>004</b>	8.65	77	7.40	180	87
<b>005</b>	9.09	84	9.05	185	91
<b>007</b>	9.35	97	7.43	156	89

**Appendix A.b: Pulmonary Function continued**

ID	LUNG VOL	TLC	TLC P G/B	TLC PP G/B	TLC P ATS/ERS	TLC PP ATS/ERS
001	LungVol	6.55	7.88	83	7.70	85
002	LungVol	8.11	8.43	96	8.18	99
003	LungVol	5.93	5.67	105	5.82	102
004	LungVol	7.50	8.24	91	8.02	94
005	LungVol	7.49	7.99	94	7.78	96
007	LungVol	5.80	6.69	87	6.66	87

ID	Vtg	FRC PL	FRC %TLC	Pred FRC %TLC	PP FRC %TLC	FRC Pred G/B	FRC PP G/B
001	3.25	2.74	41.83	48.63	86.02	3.50	78
002	4.98	3.46	42.66	48.84	87.35	3.97	87
003	2.55	1.54	25.97	48.63	53.40	2.31	67
004	2.18	2.00	26.67	48.84	54.60	3.30	61
005	3.12	2.70	36.05	48.42	74.45	3.39	80
007	3.70	2.40	41.38	48.21	85.83	3.03	79

ID	FRC P ATS/ERS	FRC PP ATS/ERS	ERV	RV	RV P G/B	RV PP G/B
001	3.45	80	2.21	0.37	1.94	19
002	3.60	96	2.61	0.58	2.12	27
003	2.90	53	0.25	1.26	1.30	97
004	3.55	56	0.72	1.09	2.06	53
005	3.46	78	1.07	1.36	1.95	70
007	3.12	77	1.20	1.10	1.55	71

ID	RV P ATS/ERS	RV PP ATS/ERS	RV/TLC	VC	VC PP
001	1.70	22	6	6.18	102
002	1.80	32	7	7.53	116
003	1.39	91	21	4.67	104
004	1.77	61	15	6.41	101
005	1.69	80	18	6.13	100
007	1.49	74	19	4.80	92

**Appendix A.b: Pulmonary Function continued**

<b>ID</b>	<b>IC</b>	<b>IC P G/B</b>	<b>IC PP G/B</b>	<b>IC P ATS/ERS</b>	<b>IC PP ATS/ERS</b>	<b>Raw</b>	<b>Raw PP</b>
<b>001</b>	3.81	4.38	87	4.26	90	1.96	158
<b>002</b>	4.65	4.46	104	4.59	101	1.69	157
<b>003</b>	4.39	3.36	131	2.93	150	3.56	167
<b>004</b>	5.49	4.94	111	4.47	123	1.97	153
<b>005</b>	4.79	4.60	104	4.32	111	2.22	173
<b>007</b>	3.40	3.66	93	3.54	96	2.30	155

<b>ID</b>	<b>Gaw</b>	<b>Gaw PP</b>	<b>sRaw</b>	<b>sRaw PP</b>	<b>sGaw</b>	<b>sGaw PP</b>
<b>001</b>	0.509	59	6.38	143	0.157	70
<b>002</b>	0.593	61	8.39	193	0.119	52
<b>003</b>	0.281	49	9.10	177	0.110	56
<b>004</b>	0.507	61	4.29	95	0.233	105
<b>005</b>	0.451	53	6.93	154	0.144	65
<b>007</b>	0.430	58	8.60	185	0.120	54

### Appendix A.c: $\dot{V}O_{2peak}$

ID	Stage	IETWatts	IETMax Time	P <sub>B</sub>	V <sub>E</sub> IET	V <sub>T</sub> IET	f <sub>B</sub> IET
001	.	30	1	682.0	20.16	1.24	16.21
001		60	2	682.0	23.61	1.59	16.76
001	.	90	3	682.0	31.41	1.55	20.24
001		120	4	682.0	40.66	2.00	20.37
001	.	150	5	682.0	51.83	2.11	24.54
001	.	180	6	682.0	68.38	2.70	25.30
001	.	210	7	682.0	84.11	2.95	28.51
001	.	240	8	682.0	112.08	3.05	36.71
001	.	270	9	682.0	150.19	3.07	48.98
001	<b>Max</b>	.	.	682.0	150.19	3.07	48.98
002	.	30	1	677.0	21.33	1.16	18.35
002		60	2	677.0	28.96	1.28	22.60
002	.	90	3	677.0	32.62	1.47	22.24
002		120	4	677.0	41.92	1.76	23.86
002	.	150	5	677.0	54.65	2.08	26.27
002	.	180	6	677.0	64.94	2.39	27.13
002	.	210	7	677.0	77.41	2.40	32.21
002	.	240	8	677.0	95.40	2.83	33.72
002	.	270	9	677.0	113.23	3.20	35.37
002	.	300	10	677.0	140.63	3.50	40.13
002	<b>Max</b>	.	.	677.0	140.63	3.50	40.13
003	.	30	1	676.0	19.84	1.59	12.49
003		60	2	676.0	25.04	1.43	17.56
003	.	90	3	676.0	32.27	1.56	20.70
003		120	4	676.0	42.14	1.79	23.53
003	.	150	5	676.0	57.99	2.09	27.74
003	.	180	6	676.0	84.29	2.30	36.59
003	<b>Max</b>	.	.	676.0	84.29	2.30	36.59

### Appendix A.c: $\dot{V}O_{2peak}$ continued

ID	Stage	IETWatts	IETMax Time	P <sub>B</sub>	V <sub>E</sub> IET	V <sub>T</sub> IET	f <sub>B</sub> IET
004	.	30	1	680.0	30.35	1.52	19.91
004		60	2	680.0	32.22	1.72	18.77
004	.	90	3	680.0	36.26	1.81	20.00
004		120	4	680.0	45.63	2.37	19.24
004	.	150	5	680.0	54.73	2.72	20.12
004	.	180	6	680.0	70.75	3.04	23.29
004	.	210	7	680.0	89.99	3.45	26.11
004	.	240	8	680.0	115.56	3.98	29.01
004	.	270	9	680.0	139.48	3.68	37.92
004	<b>Max</b>	.	.	680.0	139.48	3.98	37.92
005	.	30	1	687.0	30.16	1.65	18.25
005		60	2	687.0	33.62	2.12	15.88
005	.	90	3	687.0	37.31	2.34	15.94
005		120	4	687.0	42.72	2.22	19.20
005	.	150	5	687.0	45.76	2.52	18.13
005	.	180	6	687.0	56.38	2.89	19.53
005	.	210	7	687.0	72.70	3.17	22.92
005	.	240	8	687.0	104.84	3.11	33.73
005	<b>Max</b>	.	.	687.0	104.84	3.17	33.73
007	.	30	1	685.0	22.18	1.48	14.96
007		60	2	685.0	29.17	2.20	13.25
007	.	90	3	685.0	32.91	1.88	17.47
007		120	4	685.0	42.75	1.72	24.91
007	.	150	5	685.0	54.72	2.39	22.88
007	.	180	6	685.0	71.48	2.33	30.73
007	.	210	7	685.0	100.64	2.48	40.52
007	<b>Max</b>	.	.	685.0	100.64	2.48	40.52

**Appendix A.c:  $\dot{V}O_{2peak}$  continued**

ID	Stage	IETWatts	VO <sub>2</sub> IET	VO <sub>2</sub> ml/kg IET	Peak VO <sub>2</sub> IET	PerVO <sub>2</sub> IET	VCO <sub>2</sub> IET	RER IET
001	.	30	0.818	7.0	3.35	24.38	0.55	0.67
001		60	1.155	9.9	3.35	34.44	0.81	0.70
001	.	90	1.318	11.3	3.35	39.30	0.98	0.74
001		120	1.649	14.1	3.35	49.17	1.34	0.81
001	.	150	1.855	15.9	3.35	55.31	1.67	0.90
001	.	180	2.276	19.5	3.35	67.86	2.2	0.97
001	.	210	2.513	21.5	3.35	74.93	2.65	1.05
001	.	240	2.970	25.4	3.35	88.55	3.34	1.12
001	.	270	3.354	28.7	3.35	100.00	3.94	1.17
001	<b>Max</b>	.	3.354	28.7	3.35	100.00	3.94	1.17
002	.	30	0.749	6.6	3.84	19.51	0.45	0.60
002		60	1.215	10.7	3.84	31.67	0.71	0.58
002	.	90	1.456	12.8	3.84	37.96	0.898328	0.62
002		120	1.854	16.3	3.84	48.31	1.229556	0.66
002	.	150	2.166	19.1	3.84	56.44	1.663023	0.77
002	.	180	2.355	20.7	3.84	61.38	2.04385	0.87
002	.	210	2.614	23.0	3.84	68.13	2.472945	0.95
002	.	240	2.998	26.4	3.84	78.13	3.026569	1.01
002	.	270	3.306	29.1	3.84	86.16	3.527342	1.07
002	.	300	3.873	34.1	3.84	100.94	4.212956	1.09
002	<b>Max</b>	.	3.873	34.1	3.84	100.94	4.212956	1.09
003	.	30	0.661	7.0	1.91	34.58	0.59	0.89
003		60	0.962	10.2	1.91	50.34	0.77	0.80
003	.	90	1.162	12.3	1.91	60.78	1.01623	0.87
003		120	1.434	15.1	1.91	75.02	1.400026	0.98
003	.	150	1.644	17.3	1.91	85.99	1.838763	1.12
003	.	180	1.912	20.2	1.91	99.98	2.373558	1.24
003	<b>Max</b>	.	1.912	20.2	1.91	100.00	2.37356	1.24

### Appendix A.c: $\dot{V}O_{2peak}$ continued

ID	Stage	IETWatts	VO <sub>2</sub> IET	VO <sub>2</sub> ml/kg IET	Peak VO <sub>2</sub> IET	PerVO <sub>2</sub> IET	VCO <sub>2</sub> IET	RER IET
004	.	30	0.856	6.0	3.30	25.90	0.81	0.95
004		60	1.126	7.9	3.30	34.09	0.95	0.85
004	.	90	1.388	9.8	3.30	42.02	1.149158	0.83
004		120	1.679	11.8	3.30	50.84	1.489439	0.89
004	.	150	1.993	14.0	3.30	60.33	1.873099	0.94
004	.	180	2.296	16.2	3.30	69.50	2.342919	1.02
004	.	210	2.643	18.6	3.30	80.02	2.829495	1.07
004	.	240	3.035	21.4	3.30	91.89	3.408691	1.12
004	.	270	3.303	23.3	3.30	99.99	3.822419	1.16
004	<b>Max</b>	.	3.303	23.3	3.30	100.09	3.82242	1.16
005	.	30	1.138	9.0	2.95	38.61	0.81	0.72
005		60	1.333	10.5	2.95	45.22	1.01	0.76
005	.	90	1.532	12.1	2.95	51.99	1.172745	0.77
005		120	1.767	13.9	2.95	59.97	1.409288	0.80
005	.	150	2.031	16.0	2.95	68.91	1.615465	0.80
005	.	180	2.400	18.9	2.95	81.43	2.084781	0.87
005	.	210	2.836	22.3	2.95	96.23	2.684601	0.95
005	.	240	2.947	23.2	2.95	100.00	3.224397	1.09
005	<b>Max</b>	.	2.947	23.2	2.95	100.00	3.224397	1.09
007	.	30	0.846	9.0	3.01	28.11	0.74	0.87
007		60	1.265	13.4	3.01	42.02	1.04	0.82
007	.	90	1.466	15.6	3.01	48.72	1.219888	0.83
007		120	1.755	18.6	3.01	58.29	1.603364	0.91
007	.	150	2.028	21.5	3.01	67.37	2.066415	1.02
007	.	180	2.403	25.5	3.01	79.84	2.589382	1.08
007	.	210	2.835	30.1	3.01	94.19	3.2536	1.15
007	<b>Max</b>	.	2.835	30.1	3.01	94.19	3.2536	1.15



**Appendix A.c:  $\dot{V}O_{2peak}$  continued**

ID	Stage	IETWatts	$V_E/VO_2$	$V_E/VCO_2$	Pred WT IET	$PVO_2$	$PPVO_2$
			IET	IET		PWT	IET
001	.	30	24.65	36.65	85.45	4.932	16.578
001		60	20.44	29.22	85.45	4.932	23.416
001	.	90	23.83	32.05	85.45	4.932	26.721
001		120	24.66	30.34	85.45	4.932	33.431
001	.	150	27.94	31.04	85.45	4.932	37.608
001	.	180	30.04	31.08	85.45	4.932	46.143
001	.	210	33.47	31.74	85.45	4.932	50.948
001	.	240	37.74	33.56	85.45	4.932	60.213
001	.	270	44.78	38.12	85.45	4.932	67.998
001	<b>Max</b>	.	44.78	38.12	85.45	4.932	67.998
002	.	30	28.50	47.60	90.19	4.751	15.758
002		60	23.83	40.83	90.19	4.751	25.576
002	.	90	22.40	36.31	90.19	4.751	30.656
002		120	22.61	34.09	90.19	4.751	39.015
002	.	150	25.23	32.86	90.19	4.751	45.581
002	.	180	27.57	31.77	90.19	4.751	49.570
002	.	210	29.61	31.30	90.19	4.751	55.020
002	.	240	31.82	31.52	90.19	4.751	63.102
002	.	270	34.25	32.10	90.19	4.751	69.588
002	.	300	36.31	33.38	90.19	4.751	81.518
002	<b>Max</b>	.	36.31	33.38	90.19	4.751	81.520
003	.	30	30.00	33.80	66.89	4.000	16.528
003		60	26.02	32.47	66.89	4.000	24.059
003	.	90	27.77	31.75	66.89	4.000	29.053
003		120	29.38	30.10	66.89	4.000	35.856
003	.	150	35.27	31.54	66.89	4.000	41.098
003	.	180	44.09	35.51	66.89	4.000	47.788
003	<b>Max</b>	.	44.08	35.51	66.89	4.000	47.796

### Appendix A.c: $\dot{V}O_{2peak}$ continued

ID	Stage	IETWatts	$V_E/VO_2$	$V_E/VCO_2$	Pred WT IET	$PVO_2$	$PPVO_2$
			IET	IET		PWT IET	PWT IET
004	.	30	35.48	37.25	88.61	5.939	14.407
004		60	28.62	33.77	88.61	5.939	18.959
004	.	90	26.13	31.56	88.61	5.939	23.368
004		120	27.17	30.64	88.61	5.939	28.279
004	.	150	27.46	29.22	88.61	5.939	33.555
004	.	180	30.82	30.20	88.61	5.939	38.653
004	.	210	34.05	31.80	88.61	5.939	44.505
004	.	240	38.07	33.90	88.61	5.939	51.106
004	.	270	42.23	36.49	88.61	5.939	55.613
004	<b>Max</b>	.	42.23	36.49	88.61	5.939	55.618
005	.	30	26.50	37.05	86.24	5.406	21.050
005		60	25.23	33.30	86.24	5.406	24.651
005	.	90	24.35	31.82	86.24	5.406	28.343
005		120	24.18	30.31	86.24	5.406	32.690
005	.	150	22.53	28.32	86.24	5.406	37.565
005	.	180	23.49	27.04	86.24	5.406	44.391
005	.	210	25.64	27.08	86.24	5.406	52.457
005	.	240	35.58	32.52	86.24	5.406	54.516
005	<b>Max</b>	.	35.58	32.51	86.24	5.406	54.515
007	.	30	26.22	30.10	75.18	4.049	20.894
007		60	23.06	27.97	75.18	4.049	31.235
007	.	90	22.44	26.98	75.18	4.049	36.216
007		120	24.37	26.66	75.18	4.049	43.332
007	.	150	26.99	26.48	75.18	4.049	50.084
007	.	180	29.74	27.61	75.18	4.049	59.354
007	.	210	35.50	30.93	75.18	4.049	70.016
007	<b>Max</b>	.	35.50	30.93	75.18	4.049	70.016

### Appendix A.c: $\dot{V}O_{2peak}$ continued

ID	Stage	IETWatts	PVO <sub>2</sub> +C PWT IET	PPVO <sub>2</sub> +C PWT IET	HR IET	PHR IET	HR PP IET	O <sub>2</sub> Pul IET	PetCO <sub>2</sub> IET
001	.	30	5.121	15.967	110	202	0.54	7.468	26
001		60	5.121	22.553	109	202	0.54	10.596	29
001	.	90	5.121	25.736	118	202	0.58	11.169	29
001		120	5.121	32.200	129	202	0.64	12.783	31
001	.	150	5.121	36.222	134	202	0.66	13.843	30
001	.	180	5.121	44.443	158	202	0.78	14.405	30
001	.	210	5.121	49.071	171	202	0.84	14.696	30
001	.	240	5.121	57.994	181	202	0.89	16.409	28
001	.	270	5.121	65.493	192	202	0.95	17.469	25
001	<b>Max</b>	.	5.121	65.493	192	202	0.95	17.469	31
002	.	30	4.891	15.305	86	201	0.43	8.705	20
002		60	4.891	24.841	93	201	0.46	13.066	23
002	.	90	4.891	29.775	98	201	0.49	14.862	26
002		120	4.891	37.894	106	201	0.53	17.487	28
002	.	150	4.891	44.272	115	201	0.57	18.831	29
002	.	180	4.891	48.146	126	201	0.63	18.691	30
002	.	210	4.891	53.440	136	201	0.68	19.220	30
002	.	240	4.891	61.290	151	201	0.75	19.920	30
002	.	270	4.891	67.590	161	201	0.80	20.535	29
002	.	300	4.891	79.178	171	201	0.85	22.649	28
002	<b>Max</b>	.	4.891	79.179	171	201	0.85	22.649	30
003	.	30	4.168	15.864	125	202	0.62	5.289	28
003		60	4.168	23.092	127	202	0.63	7.578	29
003	.	90	4.168	27.885	143	202	0.71	8.127	30
003		120	4.168	34.415	154	202	0.76	9.314	31
003	.	150	4.168	39.446	170	202	0.84	9.671	30
003	.	180	4.168	45.867	177	202	0.87	10.800	26
003	<b>Max</b>	.	4.168	45.875	177	202	0.87	10.802	31

### Appendix A.c: $\dot{V}O_{2peak}$ continued

ID	Stage	IETWatts	PVO <sub>2</sub> +C PWT IET	PPVO <sub>2</sub> +C PWT IET	HR IET	PHR IET	HR PP IET	O <sub>2</sub> Pul IET	PetCO <sub>2</sub> IET
004	.	30	6.259	13.669	129	201	0.64	6.632	25
004		60	6.259	17.988	135	201	0.67	8.340	28
004	.	90	6.259	22.172	150	201	0.74	9.252	30
004		120	6.259	26.831	158	201	0.78	10.629	31
004	.	150	6.259	31.837	173	201	0.86	11.552	32
004	.	180	6.259	36.675	182	201	0.90	12.613	31
004	.	210	6.259	42.227	196	201	0.97	13.519	30
004	.	240	6.259	48.490	203	201	1.01	14.951	28
004	.	270	6.259	52.767	208	201	1.03	15.878	26
004	<b>Max</b>	.	6.259	52.771	208	201	1.03	15.880	32
005	.	30	5.650	20.139	95	204	0.47	11.978	25
005		60	5.650	23.584	96	204	0.47	13.881	28
005	.	90	5.650	27.117	100	204	0.49	15.322	29
005		120	5.650	31.275	107	204	0.53	16.516	31
005	.	150	5.650	35.939	115	204	0.57	17.658	33
005	.	180	5.650	42.469	127	204	0.62	18.895	34
005	.	210	5.650	50.186	137	204	0.67	20.775	35
005	.	240	5.650	52.156	148	204	0.73	19.913	29
005	<b>Max</b>	.	5.650	52.155	148	204	0.73	19.912	35
007	.	30	4.164	20.318	131	205	0.64	6.458	45
007		60	4.164	30.374	136	205	0.66	9.299	42
007	.	90	4.164	35.218	139	205	0.68	10.550	45
007		120	4.164	42.138	146	205	0.71	12.017	44
007	.	150	4.164	48.704	157	205	0.77	12.917	47
007	.	180	4.164	57.719	166	205	0.81	14.478	44
007	.	210	4.164	68.087	181	205	0.88	15.663	37
007	<b>Max</b>	.	4.164	68.087	181	205	0.88	15.663	47

**Appendix A.c:  $\dot{V}O_{2peak}$  continued**

<b>ID</b>	<b>Stage</b>	<b>IETWatts</b>	<b>RPB IET</b>	<b>RPU IET</b>	<b>RPE IET</b>
001	.	30	0	0	7
001		60	0.5	0	7
001	.	90	0.5	0	7
001		120	0.5	0	8
001	.	150	0.5	0	8
001	.	180	1	0.5	9
001	.	210	3	3	11
001	.	240	4	4	13
001	.	270	7	7	17
001	<b>Max</b>	.	7	7	17
002	.	30	0	0	7
002		60	0	0	7
002	.	90	0	0	7
002		120	0	0	7
002	.	150	0.5	0	7
002	.	180	0.5	0.5	7
002	.	210	0.5	0.5	7
002	.	240	1	1	8
002	.	270	2	1	11
002	.	300	3	4	15
002	<b>Max</b>	.	3	4	15
003	.	30	0.5	0.5	7
003		60	1	1	8
003	.	90	4	3	10
003		120	4	3	12
003	.	150	5	5	14
003	.	180	8	7	15
003	<b>Max</b>	.	8	7	15

**Appendix A.c:  $\dot{V}O_{2peak}$  continued**

ID	Stage	IETWatts	RPB IET	RPU IET	RPE IET
004	.	30	0	0	7
004		60	0	0	7
004	.	90	0.5	0.5	7
004		120	0.5	0.5	8
004	.	150	1	0.5	8
004	.	180	2	2	10
004	.	210	3	3	13
004	.	240	5	5	15
004	.	270	8	7	17
004	<b>Max</b>	.	8	7	17
005	.	30	3	0	9
005		60	3	0	9
005	.	90	3	0.5	9
005		120	3	1	11
005	.	150	4	3	13
005	.	180	5	4	15
005	.	210	7	5	15
005	.	240	7	7	15
005	<b>Max</b>	.	7	7	15
007	.	30	2	2	9
007		60	2	2	9
007	.	90	2	3	11
007		120	4	3	13
007	.	150	4	4	14
007	.	180	6	5	14
007	.	210	8	9	16
007	<b>Max</b>	.	8	9	16

**Appendix A.d: Eucapnic Voluntary Hyperventilation Beetroot Juice**

ID	Slope from rest	Y intercept	Stage	P <sub>B</sub>	VE	VE/MVV 6MIN
1	.	.	RestMin1	686.0	11.44700	4.672
1	0.00047	0.451	50	686.0	48.72900	19.889
1	0.00083	0.442	70	686.0	85.51900	34.906
1	0.00306	0.358	90	686.0	133.43800	54.464
2	.	.	RestMin1	687.0	27.82000	14.122
2	0.0008	0.468	50	687.0	44.98400	22.835
2	0.00118	0.455	70	687.0	83.43200	42.351
2	0.00225	0.408	90	687.0	124.14500	63.018
3	.	.	RestMin1	689.0	8.41200	6.325
3	0.00061	0.303	50	689.0	24.72600	18.591
3	0.00062	0.303	70	689.0	41.21500	30.989
3	0.00123	0.291	90	689.0	64.87700	48.780
7	.	.	RestMin1	684.0	12.06900	7.737
7	0.00358	0.353	50	684.0	37.06300	23.758
7	0.00197	0.386	70	684.0	61.15700	39.203
7	0.00199	0.385	90	684.0	108.42800	69.505

ID	VO <sub>2</sub> COB	VO <sub>2</sub> ml/kg COB	VE/VO <sub>2</sub> COB	RPB COB	RPU COB	MinVi Mech COB	MinVe Mech COB
1	0.4560	0.004	25.103	0.000	0.000	-13.989	14.134
1	0.4740	0.004	102.804	0.000	0.000	-46.568	44.910
1	0.5180	0.005	165.095	0.000	0.000	-80.612	83.101
1	0.8390	0.008	159.044	0.500	0.500	-116.158	116.566
2	0.4910	0.004	56.660	0.000	0.000	-33.470	35.248
2	0.5050	0.004	89.077	0.000	0.000	-44.941	41.131
2	0.5550	0.005	150.328	0.000	0.000	-78.486	77.384
2	0.7110	0.006	174.606	0.000	0.500	-106.208	107.156
3	0.3090	0.003	27.223	0.000	0.000	-10.445	10.288
3	0.3190	0.003	77.511	0.500	0.500	-23.920	23.779
3	0.3290	0.003	125.274	0.500	0.500	-38.616	40.166
3	0.3790	0.004	171.179	1.000	0.500	-58.375	58.207
7	0.3960	0.004	30.477	0.000	0.000	-11.676	11.176
7	0.4860	0.005	76.261	2.000	2.000	-39.270	39.940
7	0.4920	0.005	124.303	2.000	2.000	-60.673	61.366
7	0.6010	0.006	180.413	5.000	4.000	-94.130	94.600

**Appendix A.d: Eucapnic Voluntary Hyperventilation Beetroot Juice continued**

ID	VE/MVV Mech COB	Vt Mech COB	Vt/FVC Mech COB	fB Mech COB	Avg IC COB	Typ IC COB	EELVavg Mech COB	EELVtyp Mech COB
1	5.769	1.894	30.996	7.534	4.215	4.268	2.336	2.28
1	18.331	2.240	36.665	20.035	4.359	4.086	2.191	2.46
1	33.919	3.193	52.257	26.046	3.218	3.271	3.332	3.28
1	47.578	3.166	51.811	37.032	4.414	5.087	2.136	1.46
<hr/>								
2	17.892	3.204	43.477	12.684	4.379	4.327	3.731	3.78
2	20.879	1.623	22.025	25.401	3.260	2.500	4.850	5.61
2	39.281	2.344	31.806	33.059	4.897	5.385	3.213	2.72
2	54.394	3.137	42.564	34.295	4.999	4.964	3.111	3.15
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3	7.735	0.835	19.101	12.972	4.391	4.394	1.539	1.54
3	17.879	1.396	31.949	17.034	3.201	3.283	2.729	2.65
3	30.200	1.819	41.633	22.100	2.403	2.953	3.527	2.98
3	43.765	2.086	47.734	27.937	3.067	2.954	2.863	2.98
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7	7.164	0.836	18.409	13.877	3.212	3.017	2.588	2.78
7	25.603	2.316	51.010	17.271	2.472	2.617	3.328	3.18
7	39.337	2.828	62.292	21.707	3.176	2.600	2.624	3.20
7	60.641	2.609	57.469	36.251	3.183	2.898	2.617	2.90

ID	EELVavg %TLC COB	EELVtyp %TLC COB	EILVavg Mech COB	EILVtyp Mech COB	EILVavg %TLC COB	EILVtyp %TLC COB	Ti Mech COB	Te Mech COB
1	35.65649	34.83969	4.229338	4.175838	64.57005	63.75325	3.48645	4.528713
1	33.44602	37.61985	4.43095	4.704336	67.64809	71.82192	1.3351	1.669157
1	50.86591	50.06565	6.524609	6.472192	99.61235	98.81209	1.168642	1.140092
1	32.61545	22.33588	5.301953	4.628641	80.94585	70.66627	0.844218	0.732082
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2	46.00049	46.64242	6.93486	6.98692	85.50999	86.15191	2.06001	3.29719
2	59.80222	69.16893	6.4732	7.23284	79.81751	89.18422	0.92864	1.36089
2	39.61977	33.59556	5.557231	5.068668	68.52319	62.49899	0.8506	0.9687
2	38.36609	38.79162	6.24849	6.283	77.04673	77.47226	0.797925	0.9597
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3	25.94793	25.91062	2.373412	2.3712	40.02381	39.98651	1.761713	3.522525
3	46.02602	44.63406	4.1255	4.042957	69.56998	68.17803	1.604764	1.934764
3	59.47519	50.20573	5.34625	4.796571	90.15599	80.88653	1.247986	1.470971
3	48.27187	50.18718	4.948516	5.062094	83.44884	85.36415	0.974194	1.176622
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7	44.61495	47.99138	3.423442	3.619275	59.02486	62.40129	1.818383	2.901383
7	57.38459	54.87241	5.644165	5.498459	97.31319	94.80102	1.722588	1.769894
7	45.23459	55.16552	5.451684	6.027678	93.99455	103.9255	1.495261	1.323444
7	45.12517	50.04138	5.226335	5.511475	90.10922	95.02543	0.85402	0.811275



**Appendix A.d: Eucapnic Voluntary Hyperventilation Beetroot Juice continued**

<b>ID</b>	<b>Tl/Ttot Mech COB</b>	<b>Vt/Ti Mech COB</b>	<b>Vt/Te Mech COB</b>	<b>PetCO2 Mech COB</b>	<b>Max Pi Mech COB</b>	<b>Max Pe Mech COB</b>	<b>P to P Mech COB</b>
1	43.62684	0.5432	0.418185	32.21295	-1.23563	1.603538	2.839168
1	44.44999	1.677954	1.342136	41.09651	-3.57271	3.015264	6.587974
1	50.59287	2.732139	2.800556	38.5034	-5.61303	8.09265	13.70568
1	51.94679	3.749791	4.324162	39.76576	-7.96451	15.02786	22.99237
2	38.10324	1.555439	0.971803	23.7278	-2.08644	2.81875	4.90519
2	38.70657	1.747976	1.192778	45.83608	-3.14384	2.79541	5.93925
2	46.74832	2.755782	2.419808	40.30351	-4.20832	4.895095	9.103415
2	45.4219	3.931447	3.26873	41.22527	-7.50541	6.773485	14.2789
3	34.20705	0.4738	0.236961	38.60786	-1.09069	1.056713	2.147403
3	45.27634	0.870008	0.721616	44.00782	-1.5505	1.460286	3.010786
3	45.92601	1.457846	1.23685	44.28624	-2.20429	2.256143	4.460433
3	45.28784	2.141251	1.772867	44.28539	-3.96403	4.232539	8.196569
7	40.39304	0.459625	0.288061	35.52933	-1.25498	1.100742	2.355722
7	49.26685	1.344407	1.308473	47.55831	-3.84285	4.219324	8.062174
7	52.63119	1.891361	2.136908	41.72172	-4.19679	5.156106	9.352896
7	51.03046	3.055051	3.216018	39.7848	-7.09735	7.629535	14.72689

**Appendix A.e: Eucapnic Voluntary Hyperventilation Tomato Juice**

ID	Slope from rest	Y intercept	Stage	P <sub>B</sub>	VE	VE/MVV 6MIN
1	.	.	RestMin1	687.0	14.07600	5.745
1	0.00085	0.489	50	687.0	50.95800	20.799
1	0.00081	0.49	70	687.0	82.52900	33.685
1	0.00267	0.415	90	687.0	138.02800	56.338
2	.	.	RestMin1	688.0	32.90700	16.704
2	0.00032	0.499	50	688.0	44.65000	22.665
2	0.00152	0.453	70	688.0	86.73200	44.026
2	0.0047	0.306	90	688.0	134.25800	68.151
3	.	.	RestMin1	685.0	6.56700	4.938
3	0.00242	0.23	50	685.0	25.08300	18.859
3	0.00211	0.234	70	685.0	39.13200	29.423
3	0.00159	0.244	90	685.0	67.36400	50.650
7	.	.	RestMin1	684.0	11.61200	7.444
7	0.00187	0.386	50	684.0	37.76500	24.208
7	0.00161	0.391	70	684.0	62.65200	40.162
7	0.00169	0.389	90	684.0	112.64400	72.208

ID	VO <sub>2</sub> COB	VO <sub>2</sub> ml/kg COB	VE/VO <sub>2</sub> COB	RPB COB	RPU COB	MinVi Mech COB	MinVe Mech COB
1	0.5010	0.004	28.096	0.000	0.000	-17.063	17.007
1	0.5330	0.005	95.606	0.000	0.000	-46.753	47.567
1	0.5570	0.005	148.167	0.000	0.000	-72.753	73.337
1	0.8340	0.007	165.501	1.000	2.000	-105.190	106.659
2	0.5090	0.004	64.650	0.000	0.000	-31.609	33.238
2	0.5130	0.004	87.037	0.000	0.000	-47.727	48.759
2	0.5870	0.005	147.755	0.000	0.000	-82.017	80.354
2	0.9820	0.008	136.719	0.000	0.000	-113.551	115.159
3	0.2460	0.003	26.695	0.000	0.000	-10.638	10.837
3	0.2910	0.003	86.196	0.000	0.000	-26.018	26.826
3	0.3150	0.003	124.229	0.000	0.000	-37.440	36.121
3	0.3450	0.004	195.258	0.500	0.000	-64.611	63.361
7	0.4080	0.004	28.461	0.000	0.000	-12.607	12.630
7	0.4570	0.005	82.637	2.000	3.000	-36.624	36.189
7	0.4900	0.005	127.861	0.500	1.000	-60.096	59.801
7	0.5800	0.006	194.214	3.000	4.000	-101.672	102.582

**Appendix A.e: Eucapnic Voluntary Hyperventilation Tomato Juice continued**

ID	VE/MVV Mech COB	Vt Mech COB	Vt/FVC Mech COB	fB Mech COB	Avg IC COB	Typ IC COB	EELVavg Mech COB	EELVtyp Mech COB
1	6.942	2.284	37.388	7.641	3.549	3.633	3.001	2.92
1	19.415	2.369	38.770	20.131	2.502	2.426	4.048	4.12
1	29.934	2.820	46.149	26.010	3.314	3.445	3.236	3.11
1	43.534	2.788	45.630	38.113	4.162	4.130	2.388	2.42
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2	16.872	2.500	33.920	13.538	3.991	3.957	4.119	4.15
2	24.751	2.026	27.485	24.319	2.586	1.992	5.524	6.12
2	40.789	2.423	32.878	33.229	4.700	4.122	3.410	3.99
2	58.456	3.125	42.395	36.940	4.719	4.888	3.391	3.22
<hr/>								
3	8.148	1.283	29.364	9.155	3.968	3.914	1.962	2.02
3	20.170	1.566	35.846	17.121	3.067	2.925	2.863	3.00
3	27.159	1.645	37.637	22.040	4.002	4.024	1.928	1.91
3	47.640	2.179	49.858	29.086	3.984	4.181	1.946	1.75
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7	8.096	0.954	21.012	13.479	2.803	2.806	2.997	2.99
7	23.198	2.138	47.087	17.027	2.696	2.880	3.104	2.92
7	38.334	2.503	55.128	23.934	2.866	3.091	2.934	2.71
7	65.758	2.852	62.822	36.005	3.404	3.178	2.396	2.62

ID	EELVavg %TLC COB	EELVtyp %TLC COB	EILVavg Mech COB	EILVtyp Mech COB	EILVavg %TLC COB	EILVtyp %TLC COB	Ti Mech COB	Te Mech COB
1	45.82012	44.53588	5.285609	5.201491	80.69632	79.41208	3.784118	4.317727
1	61.80356	62.95573	6.41698	6.492447	97.96916	99.12133	1.60046	1.39426
1	49.40063	47.40916	6.055429	5.924988	92.4493	90.45783	1.0762	1.2314
1	36.46342	36.95115	5.176346	5.208292	79.02818	79.51591	0.823623	0.754908
<hr/>								
2	50.78915	51.20838	6.61887	6.65287	81.61369	82.03292	1.93807	2.75515
2	68.11905	75.43403	7.55011	8.143355	93.0963	100.4113	1.066882	4.433064
2	42.05015	49.17263	5.833407	6.41104	71.92857	79.05105	0.865227	0.947087
2	41.81856	39.72503	6.516	6.346215	80.34525	78.25173	0.742985	0.885946
<hr/>								
3	33.09157	33.99831	3.24554	3.29931	54.73086	55.63761	2.66603	4.9542
3	48.27474	50.67285	4.429154	4.571362	74.69062	77.08874	1.487477	2.023592
3	32.50872	32.13828	3.572517	3.55055	60.24481	59.87437	1.211133	1.519058
3	32.81034	29.49747	4.124446	3.927993	69.55221	66.23934	0.929287	1.135793
<hr/>								
7	51.67384	51.61552	3.951041	3.947658	68.1214	68.06307	2.072833	2.51945
7	53.52188	50.34655	5.242019	5.05785	90.37964	87.20431	1.88165	1.6643
7	50.58595	46.70345	5.436785	5.2116	93.73767	89.85517	1.392245	1.151175
7	41.30517	45.2069	5.247818	5.474118	90.47962	94.38134	0.828455	0.839314

**Appendix A.e: Eucapnic Voluntary Hyperventilation Tomato Juice continued**

<b>ID</b>	<b>TI/Ttot Mech COB</b>	<b>Vt/Ti Mech COB</b>	<b>Vt/Te Mech COB</b>	<b>PetCO2 Mech COB</b>	<b>Max Pi Mech COB</b>	<b>Max Pe Mech COB</b>	<b>P to P Mech COB</b>
1	46.57683	0.603679	0.529073	34.56196	-1.40454	1.825936	3.230476
1	53.41075	1.480104	1.698999	47.31385	-3.26193	3.538487	6.800417
1	46.63728	2.620041	2.289823	43.84544	-5.82556	5.578453	11.40401
1	52.25595	3.385034	3.693155	43.4724	-6.78911	10.69372	17.48283
2	42.42583	1.289876	0.907344	20.26014	-2.04716	4.67208	6.71924
2	42.95927	1.898668	0.456942	44.62446	-2.87552	2.361764	5.237284
2	47.78448	2.800583	2.558519	43.60093	-4.33079	4.73274	9.06353
2	45.64239	4.205354	3.526756	43.45972	-8.42559	7.079869	15.50546
3	35.8376	0.481319	0.259015	46.78002	-1.14938	1.31964	2.46902
3	42.36257	1.0531	0.7741	52.12438	-1.75581	1.737115	3.492925
3	44.32728	1.358026	1.082743	47.65117	-2.12198	2.701042	4.823022
3	44.99091	2.344586	1.918301	47.93096	-3.80007	4.944087	8.744157
7	45.39123	0.460219	0.378637	39.8945	-0.96207	1.335733	2.297803
7	53.07749	1.136104	1.284474	48.73433	-1.96338	3.0299	4.99328
7	54.37868	1.797672	2.174126	46.48693	-3.60329	5.841455	9.444745
7	49.6698	3.442695	3.398154	39.67271	-7.59001	9.081895	16.67191

### Appendix A.f: 6-Minute Cycling Beetroot Juice

ID	Stage	P <sub>B</sub>	Watts 6MIN	VE 6MIN	VE/MVV 6MIN	VT 6MIN	fB 6MIN
001	RestMin1	686.0	0.0	21	8.608	2	12.1
001	RestMin2	686.0	0.0	21	8.378	1	14.1
001	RestMin3	686.0	0.0	25	10.095	2	12.0
001	MeanRest	686	0.0	22.11645	9.027124	1.746128	12.75277
001	Min1	686.0	155.0	34	13.824	2	19.6
001	Min2	686.0	155.0	60	24.394	3	21.6
001	Min3	686.0	155.0	60	24.540	3	21.3
001	Min4	686.0	155.0	68	27.695	3	23.9
001	Min5	686.0	155.0	73	29.738	3	25.4
001	Min6	686.0	155.0	78	31.665	3	25.6
001	MeanEx	686	155.0	72.76305	29.6992	2.910547	24.98483
002	RestMin1	687.0	0.0	34	17.256	2	20.5
002	RestMin2	687.0	0.0	21	10.528	2	12.3
002	RestMin3	687.0	0.0	27	13.509	2	12.2
002	MeanRest	687	0.0	27.1158	13.76437	1.837349	15.03397
002	Min1	687.0	150.0	29	14.613	1	23.1
002	Min2	687.0	150.0	47	23.655	2	30.4
002	Min3	687.0	150.0	59	29.766	2	26.2
002	Min4	687.0	150.0	64	32.578	2	29.5
002	Min5	687.0	150.0	69	35.123	2	32.9
002	Min6	687.0	150.0	75	38.213	2	32.8
002	MeanEx	687	150.0	69.55004	35.30459	2.191341	31.73305
003	RestMin1	689.0	0.0	8	5.869	1	5.5
003	RestMin2	689.0	0.0	7	5.131	1	11.7
003	RestMin3	689.0	0.0	9	6.941	1	14.0
003	MeanRest	689	0.0	7.953934	5.980401	0.88826	10.40076
003	Min1	689.0	102.0	20	14.840	1	15.4
003	Min2	689.0	102.0	33	24.524	2	20.0
003	Min3	689.0	102.0	41	30.555	2	23.1
003	Min4	689.0	102.0	46	34.290	2	24.0
003	Min5	689.0	102.0	47	35.544	2	25.1
003	Min6	689.0	102.0	51	38.290	2	27.7
003	MeanEx	689	102.0	47.93515	36.04147	1.875154	25.58639

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>P<sub>B</sub></b>	<b>Watts 6MIN</b>	<b>VE 6MIN</b>	<b>VE/MVV 6MIN</b>	<b>VT 6MIN</b>	<b>fB 6MIN</b>
004	RestMin1	682.0	0.0	25	13.685	1	18.5
004	RestMin2	682.0	0.0	30	16.700	2	18.2
004	RestMin3	682.0	0.0	28	15.642	1	20.9
004	MeanRest	682	0.0	27.61582	15.34212	1.441899	19.22105
004	Min1	682.0	108.0	45	25.139	2	19.2
004	Min2	682.0	108.0	49	27.061	2	19.5
004	Min3	682.0	108.0	62	34.419	3	19.5
004	Min4	682.0	108.0	63	35.227	3	24.1
004	Min5	682.0	108.0	64	35.538	3	23.4
004	Min6	682.0	108.0	66	36.502	3	23.4
004	MeanEx	682	108.0	64.36035	35.75575	2.723534	23.63854
005	RestMin1	686.0	0.0	19	10.482	2	12.4
005	RestMin2	686.0	0.0	21	11.398	2	12.6
005	RestMin3	686.0	0.0	15	8.021	1	11.5
005	MeanRest	686	0.0	18.43902	9.967037	1.509174	12.1651
005	Min1	686.0	152.0	35	18.764	2	16.5
005	Min2	686.0	152.0	45	24.380	3	15.1
005	Min3	686.0	152.0	54	29.161	3	15.7
005	Min4	686.0	152.0	59	32.099	4	16.6
005	Min5	686.0	152.0	72	38.717	4	19.2
005	Min6	686.0	152.0	78	42.348	3	27.7
005	MeanEx	686	152.0	69.78412	37.72115	3.382657	21.15302
007	RestMin1	684.0	0.0	10	6.541	1	10.2
007	RestMin2	684.0	0.0	16	10.458	1	13.9
007	RestMin3	684.0	0.0	10	6.169	1	11.8
007	MeanRest	684	0.0	12.04709	7.722493	0.92283	11.99014
007	Min1	684.0	125.0	28	17.831	2	16.5
007	Min2	684.0	125.0	48	30.574	2	19.5
007	Min3	684.0	125.0	58	37.064	2	26.9
007	Min4	684.0	125.0	66	42.151	2	28.6
007	Min5	684.0	125.0	72	45.901	2	33.1
007	Min6	684.0	125.0	80	51.541	2	35.3
007	MeanEx	684	125.0	72.58819	46.53089	2.248601	32.30639

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>VO2 6MIN</b>	<b>VO2 ml/kg 6MIN</b>	<b>Peak VO 6MIN</b>	<b>PerVO 6MIN</b>	<b>VCO2 6MIN</b>	<b>RER 6MIN</b>
001	RestMin1	0.4411	0.004	2.391	18.448	0.464	1.052
001	RestMin2	0.4824	0.004	2.391	20.176	0.445	0.922
001	RestMin3	0.5603	0.005	2.391	23.434	0.527	0.940
001	MeanRest	0.494574	0.00444	2.390871	20.68595	0.478476	0.9713074
001	Min1	1.3817	0.012	2.391	57.792	0.874	0.632
001	Min2	1.8714	0.017	2.391	78.271	1.662	0.888
001	Min3	2.1114	0.019	2.391	88.311	1.847	0.875
001	Min4	2.2243	0.020	2.391	93.033	2.044	0.919
001	Min5	2.2811	0.020	2.391	95.408	2.134	0.936
001	Min6	2.3909	0.021	2.391	100.000	2.251	0.941
001	MeanEx	2.298751	0.020635	2.390871	96.14701	2.142991	0.93198217
002	RestMin1	0.5445	0.005	2.355	23.124	0.593	1.088
002	RestMin2	0.3936	0.003	2.355	16.714	0.379	0.962
002	RestMin3	0.6622	0.006	2.355	28.123	0.527	0.796
002	MeanRest	0.533409	0.00452	2.3546	22.6539	0.49948	0.948934
002	Min1	0.9535	0.008	2.355	40.494	0.673	0.706
002	Min2	2.0565	0.017	2.355	87.341	1.394	0.678
002	Min3	2.2286	0.019	2.355	94.650	1.961	0.880
002	Min4	2.1985	0.019	2.355	93.371	2.121	0.965
002	Min5	2.1910	0.019	2.355	93.053	2.159	0.986
002	Min6	2.3546	0.020	2.355	100.002	2.321	0.986
002	MeanEx	2.248069	0.019051	2.3546	95.47564	2.200461	0.97866302
003	RestMin1	0.3013	0.003	1.564	19.267	0.204	0.677
003	RestMin2	0.2555	0.003	1.564	16.338	0.188	0.734
003	RestMin3	0.3254	0.003	1.564	20.809	0.234	0.719
003	MeanRest	0.294047	0.003118	1.5637	18.80456	0.208397	0.70974524
003	Min1	0.6745	0.007	1.564	43.137	0.567	0.840
003	Min2	1.3466	0.014	1.564	86.115	1.104	0.820
003	Min3	1.3857	0.015	1.564	88.615	1.330	0.960
003	Min4	1.4796	0.016	1.564	94.621	1.484	1.003
003	Min5	1.4947	0.016	1.564	95.584	1.507	1.008
003	Min6	1.5637	0.017	1.564	99.998	1.576	1.008
003	MeanEx	1.512637	0.016041	1.5637	96.73448	1.522192	1.00628154

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>VO2 6MIN</b>	<b>VO2 ml/kg 6MIN</b>	<b>Peak VO 6MIN</b>	<b>PerVO 6MIN</b>	<b>VCO2 6MIN</b>	<b>RER 6MIN</b>
004	RestMin1	0.6707	0.005	2.041	32.864	0.559	0.834
004	RestMin2	0.7619	0.005	2.041	37.330	0.682	0.895
004	RestMin3	0.6162	0.004	2.041	30.195	0.579	0.940
004	MeanRest	0.682949	0.00471	2.040923	33.46277	0.606683	0.88941827
004	Min1	1.2203	0.008	2.041	59.793	1.059	0.867
004	Min2	1.5923	0.011	2.041	78.018	1.321	0.830
004	Min3	1.9511	0.013	2.041	95.599	1.714	0.878
004	Min4	1.8896	0.013	2.041	92.583	1.701	0.900
004	Min5	2.0409	0.014	2.041	100.000	1.773	0.869
004	Min6	1.9915	0.014	2.041	97.577	1.760	0.884
004	MeanEx	1.973979	0.013614	2.040923	96.71992	1.744726	0.88426232
005	RestMin1	0.4349	0.003	2.553	17.037	0.457	1.050
005	RestMin2	0.5375	0.004	2.553	21.055	0.505	0.940
005	RestMin3	0.3617	0.003	2.553	14.170	0.356	0.983
005	MeanRest	0.444712	0.003382	2.552802	17.42055	0.439085	0.99085983
005	Min1	1.2316	0.009	2.553	48.245	1.045	0.849
005	Min2	1.8825	0.014	2.553	73.744	1.683	0.894
005	Min3	2.1022	0.016	2.553	82.348	2.111	1.004
005	Min4	2.2336	0.017	2.553	87.495	2.307	1.033
005	Min5	2.5528	0.019	2.553	100.000	2.641	1.035
005	Min6	2.2179	0.017	2.553	86.882	2.452	1.106
005	MeanEx	2.334773	0.017755	2.552802	91.45924	2.466691	1.05765562
007	RestMin1	0.3213	0.003	3.354	9.580	0.300	0.935
007	RestMin2	0.4998	0.005	3.354	14.902	0.387	0.773
007	RestMin3	0.3463	0.004	3.354	10.326	0.280	0.808
007	MeanRest	0.389148	0.004118	3.354	11.6025	0.322244	0.83870802
007	Min1	1.1219	0.012	3.354	33.449	0.866	0.772
007	Min2	1.8192	0.019	3.354	54.241	1.722	0.947
007	Min3	1.9485	0.021	3.354	58.095	2.011	1.032
007	Min4	2.1772	0.023	3.354	64.912	2.234	1.026
007	Min5	2.2747	0.024	3.354	67.820	2.296	1.009
007	Min6	2.3626	0.025	3.354	70.440	2.397	1.014
007	MeanEx	2.271467	0.024037	3.354	67.72411	2.309018	1.01669725



**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

ID	Stage	VE/VO2 6MIN	VE/VCO2 6MIN	Vent Resp Ex 6MIN	HR 6MIN	PHR 6MIN	HR PP 6MIN	O2Pul 6MIN
001	RestMin1	47.813	45.46	.	102.07	202.46	50.417	4.321
001	RestMin2	42.553	46.16	.	98.85	202.46	48.823	4.880
001	RestMin3	44.146	46.94	.	99.45	202.46	49.121	5.634
001	MeanRest	44.83728	46.18927	.	100.12	202.459	49.45375	4.944961
001	Min1	24.511	38.76	.	124.93	202.46	61.705	11.060
001	Min2	31.937	35.96	.	147.23	202.46	72.721	12.710
001	Min3	28.475	32.56	.	157.60	202.46	77.845	13.397
001	Min4	30.506	33.20	.	164.33	202.46	81.166	13.536
001	Min5	31.940	34.14	.	167.49	202.46	82.728	13.619
001	Min6	32.448	34.46	.	171.28	202.46	84.601	13.959
001	MeanEx	31.63102	33.934	30.42723	167.7006	202.459	82.83187	13.7045
002	RestMin1	62.435	57.36	.	65.18	202.46	32.194	8.353
002	RestMin2	52.700	54.75	.	64.77	202.46	31.993	6.076
002	RestMin3	40.188	50.50	.	62.69	202.46	30.965	10.563
002	MeanRest	51.77448	54.20402	.	64.21	202.459	31.71741	8.330682
002	Min1	30.192	42.78	.	83.72	202.46	41.350	11.389
002	Min2	22.660	33.44	.	96.90	202.46	47.863	21.223
002	Min3	26.312	29.90	.	103.35	202.46	51.050	21.563
002	Min4	29.191	30.27	.	105.67	202.46	52.194	20.805
002	Min5	31.580	32.04	.	110.67	202.46	54.663	19.798
002	Min6	31.971	32.43	.	111.51	202.46	55.080	21.115
002	MeanEx	30.91396	31.57844	24.94692	109.2853	202.459	53.97897	20.5728
003	RestMin1	25.911	38.30	.	65.54	202.46	32.374	4.596
003	RestMin2	26.710	36.38	.	67.81	202.46	33.495	3.768
003	RestMin3	28.371	39.48	.	77.30	202.46	38.183	4.209
003	MeanRest	26.99742	38.05526	.	70.22033	202.459	34.68373	4.191039
003	Min1	29.261	34.84	.	101.88	202.46	50.323	6.621
003	Min2	24.222	29.56	.	110.27	202.46	54.467	12.211
003	Min3	29.328	30.56	.	119.04	202.46	58.797	11.640
003	Min4	30.823	30.74	.	127.22	202.46	62.838	11.630
003	Min5	31.628	31.37	.	133.88	202.46	66.126	11.164
003	Min6	32.569	32.32	.	137.53	202.46	67.930	11.370
003	MeanEx	31.67335	31.47442	30.43186	132.8759	202.459	65.63103	11.38804

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

ID	Stage	VE/VO2 6MIN	VE/VCO2 6MIN	Vent Resp Ex 6MIN	HR 6MIN	PHR 6MIN	HR PP 6MIN	O2Pul 6MIN
004	RestMin1	36.725	44.04	.	83.34	202.46	41.163	8.048
004	RestMin2	39.454	44.09	.	83.99	202.46	41.483	9.071
004	RestMin3	45.689	48.63	.	87.71	202.46	43.323	7.026
004	MeanRest	40.6228	45.58716	.	85.01154	202.459	41.98951	8.048543
004	Min1	37.081	42.75	.	112.69	202.46	55.662	10.829
004	Min2	30.591	36.87	.	129.63	202.46	64.026	12.284
004	Min3	31.754	36.15	.	134.70	202.46	66.531	14.485
004	Min4	33.558	37.28	.	132.24	202.46	65.315	14.289
004	Min5	31.343	36.07	.	136.46	202.46	67.401	14.956
004	Min6	32.992	37.33	.	138.01	202.46	68.167	14.430
004	MeanEx	32.63108	36.89504	32.28744	135.5685	202.459	66.96098	14.55844
005	RestMin1	44.585	42.46	.	77.74	202.46	38.398	5.595
005	RestMin2	39.231	41.76	.	75.80	202.46	37.439	7.091
005	RestMin3	41.025	41.74	.	76.46	202.46	37.765	4.731
005	MeanRest	41.61365	41.98347	.	76.66539	202.459	37.86712	5.805558
005	Min1	28.186	33.21	.	94.74	202.46	46.796	13.000
005	Min2	23.958	26.80	.	102.57	202.46	50.663	18.353
005	Min3	25.663	25.55	.	110.03	202.46	54.349	19.105
005	Min4	26.586	25.74	.	115.56	202.46	57.078	19.328
005	Min5	28.058	27.12	.	118.91	202.46	58.733	21.468
005	Min6	35.323	31.95	.	122.85	202.46	60.678	18.054
005	MeanEx	29.98897	28.27046	25.32302	119.1065	202.459	58.82995	19.61691
007	RestMin1	31.755	33.97	.	82.73	202.46	40.865	3.884
007	RestMin2	32.642	42.20	.	86.88	202.46	42.911	5.753
007	RestMin3	27.786	34.39	.	87.18	202.46	43.059	3.973
007	MeanRest	30.72795	36.85576	.	85.59562	202.459	42.278	4.536485
007	Min1	24.795	32.12	.	120.31	202.46	59.427	9.325
007	Min2	26.218	27.70	.	140.49	202.46	69.394	12.949
007	Min3	29.674	28.75	.	146.41	202.46	72.315	13.309
007	Min4	30.202	29.43	.	150.28	202.46	74.225	14.488
007	Min5	31.480	31.19	.	155.28	202.46	76.699	14.648
007	Min6	34.032	33.55	.	161.24	202.46	79.641	14.652
007	MeanEx	31.90473	31.388	30.47207	155.6006	202.459	76.85534	14.59618

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>PetCO2 6MIN</b>	<b>PulseOx 6MIN</b>	<b>RPB 6MIN</b>	<b>RPU 6MIN</b>	<b>RPE 6MIN</b>	<b>FECO2 6MIN</b>	<b>PmeCO2 calc 6MIN</b>
001	RestMin1			.	.	.	0.030	19.237
001	RestMin2			.	.	.	0.030	18.952
001	RestMin3			0.000	0.000	0.000	0.029	18.640
001	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.029645	18.94311
001	Min1	30.900	91.600	.	.	.	0.035	22.523
001	Min2	34.400	95.600	0.500	0.000	8.000	0.038	24.255
001	Min3	37.300	92.700	.	.	.	0.042	26.766
001	Min4	35.500	92.700	0.500	0.500	9.000	0.041	26.251
001	Min5	35.500	95.000	.	.	.	0.040	25.536
001	Min6	35.500	95.000	0.500	0.500	11.000	0.040	25.297
001	MeanEx	35.5	94.23333	0.5	0.5	10	0.040211	25.69458
002	RestMin1			.	.	.	0.024	15.301
002	RestMin2			.	.	.	0.025	16.018
002	RestMin3			0.000	0.000	0.000	0.027	17.348
002	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.025347	16.2221
002	Min1	32.900	95.630	.	.	.	0.032	20.434
002	Min2	43.700	95.630	0.000	0.000	7.000	0.041	26.070
002	Min3	40.300	95.630	.	.	.	0.046	29.123
002	Min4	39.700	96.810	0.000	0.000	7.000	0.045	28.771
002	Min5	36.900	96.810	.	.	.	0.042	27.190
002	Min6	36.900	95.630	1.000	0.500	7.000	0.042	26.869
002	MeanEx	37.83333	96.41667	0.5	0.25	7	0.043141	27.60992
003	RestMin1			.	.	.	0.036	22.794
003	RestMin2			.	.	.	0.037	23.980
003	RestMin3			0.000	0.000	0.000	0.034	22.118
003	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.035769	22.964
003	Min1	40.500	98.750	.	.	.	0.039	25.029
003	Min2	43.900	96.810	0.500	0.500	9.000	0.046	29.458
003	Min3	39.800	97.500	.	.	.	0.044	28.494
003	Min4	39.800	97.500	1.000	1.000	10.000	0.044	28.331
003	Min5	39.800	97.500	.	.	.	0.043	27.770
003	Min6	40.500	97.500	1.000	1.000	12.000	0.042	26.961
003	MeanEx	40.03333	97.5	1	1	11	0.043127	27.68752

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

ID	Stage	PetCO2 6MIN	PulseOx 6MIN	HR Mech 6MIN	RPB 6MIN	RPU 6MIN	RPE 6MIN	FECO2 6MIN	PmeCO2 calc 6MIN
004	RestMin1		.	.	.	.	.	0.031	19.851
004	RestMin2		.	.	.	.	.	0.031	19.828
004	RestMin3		.	.	0.000	0.000	0.000	0.028	18.001
004	MeanRest	#DIV/0!	#DIV/0!	#DIV/0!	0	0	0	0.030278	19.22674
004	Min1	30.480	98.830	.	.	.	.	0.032	20.444
004	Min2	32.490	98.830	.	2.000	2.000	11.000	0.037	23.664
004	Min3	34.510	97.660	.	.	.	.	0.038	24.129
004	Min4	30.480	97.660	.	3.000	3.000	11.000	0.037	23.402
004	Min5	34.510	97.010	.	.	.	.	0.038	24.181
004	Min6	33.160	91.680	.	3.000	3.000	11.000	0.037	23.373
004	MeanEx	32.71667	95.45	#DIV/0!	3	3	11	0.037247	23.65194
005	RestMin1		.	.	.	.	.	0.032	20.581
005	RestMin2		.	.	.	.	.	0.033	20.924
005	RestMin3		.	.	1.000	0.000	0.000	0.033	20.934
005	MeanRest	#DIV/0!	#DIV/0!	#DIV/0!	1	0	0	0.032571	20.8128
005	Min1	37.000	99.900	.	.	.	.	0.041	26.244
005	Min2	44.300	97.400	.	3.000	2.000	13.000	0.051	32.455
005	Min3	46.200	99.000	.	.	.	.	0.053	34.029
005	Min4	46.200	97.600	.	4.000	2.000	14.000	0.053	33.779
005	Min5	46.200	96.700	.	.	.	.	0.050	32.080
005	Min6	46.200	97.600	.	5.000	5.000	17.000	0.043	27.265
005	MeanEx	46.2	97.3	#DIV/0!	4.5	3.5	15.5	0.048578	31.04123
007	RestMin1	.	.	.	.	.	.	0.043	27.273
007	RestMin2	.	.	.	.	.	.	0.040	25.315
007	RestMin3	.	.	.	.	.	.	0.040	25.348
007	MeanRest	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	0.041	25.97895
007	Min1	43.700	95.630	.	.	.	.	0.043	27.128
007	Min2	43.100	95.630	.	3.000	4.000	13.000	0.049	31.415
007	Min3	36.900	95.630	.	.	.	.	0.048	30.273
007	Min4	39.100	94.800	.	5.000	6.000	15.000	0.046	29.579
007	Min5	36.300	95.510	.	.	.	.	0.044	27.927
007	Min6	38.000	95.510	.	7.000	6.000	16.000	0.041	25.980
007	MeanEx	37.8	95.27333	#DIV/0!	6	6	15.5	0.045	27.82833

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

ID	Stage	Vd/Vt calc 6MIN	MinVi Mech 6MIN	MinVe Mech 6MIN	VE/MVV Mech 6MIN	Vt Mech 6MIN	Vt/FVC Mech 6MIN	fB Mech 6MIN	Avg IC 6MIN	Typ IC 6MIN
001	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	RestMin3	0.253	-24.116	24.410	9.963	2.005	32.811	12.300	3.217	3.431
001	MeanRest	#VALUE!	-24.1157	24.4103	#VALUE!	2.004725	#VALUE!	12.29978	3.217319	3.4312
001	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min6	0.253	-75.255	77.649	31.693	2.711	44.368	28.778	2.618	2.598
001	MeanEx	#VALUE!	-75.2554	77.64885	#VALUE!	2.710879	#VALUE!	28.77849	2.618171	2.5975
002	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	RestMin3	0.325	-31.717	31.602	16.041	2.344	31.803	17.389	4.206	3.896
002	MeanRest	#VALUE!	-31.7165	31.60172	#VALUE!	2.34391	#VALUE!	17.38881	4.206227	3.8963
002	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min6	0.266	-77.040	77.378	39.278	2.315	31.414	33.520	3.752	3.779
002	MeanEx	#VALUE!	-77.0395	77.37843	#VALUE!	2.315233	#VALUE!	33.51958	3.752306	3.7792
003	RestMin1	0.402	.	.	8.128	.	18.479	.	.	.
003	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	RestMin3	#REF!	-10.536	10.810	#REF!	0.808	#REF!	13.414	3.745	3.707
003	MeanRest	#VALUE!	-10.5359	10.8097	#VALUE!	0.80754	#VALUE!	13.41405	3.74451	3.7066
003	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min6	0.287	-55.011	55.281	41.564	1.986	45.455	28.302	3.995	3.975
003	MeanEx	#VALUE!	-55.0105	55.28061	#VALUE!	1.986371	#VALUE!	28.30174	3.994996	3.9745

### Appendix A.f: 6-Minute Cycling Beetroot Juice continued

ID	Stage	Vd/Vt calc 6MIN	MinVi Mech 6MIN	MinVe Mech 6MIN	VE/MVV Mech 6MIN	Vt Mech 6MIN	Vt/FVC Mech 6MIN	fB Mech 6MIN	Avg IC 6MIN	Typ IC 6MIN
004	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	RestMin3	0.327	-32.985	34.076	18.931	1.928	32.627	18.063	4.571	4.657
004	MeanRest	#VALUE!	-32.9854	34.07603	#VALUE!	1.928267	#VALUE!	18.06302	4.571175	4.6565
004	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min6	0.284	-69.610	70.356	39.087	3.182	53.833	25.761	5.191	5.331
004	MeanEx	#VALUE!	-69.6095	70.35574	#VALUE!	3.181544	#VALUE!	25.76069	5.191269	5.3305
005	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	RestMin3	0.316	-17.639	17.397	9.404	1.623	29.093	11.168	4.962	4.795
005	MeanRest	#VALUE!	-17.6394	17.39671	#VALUE!	1.6234	#VALUE!	11.168	4.961782	4.7946
005	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min6	0.321	-78.522	80.158	43.329	2.939	52.671	35.428	5.402	5.659
005	MeanEx	#VALUE!	-78.5217	80.15794	#VALUE!	2.939021	#VALUE!	35.42796	5.4018	5.6586
007	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	RestMin3	0.266	-9.167	11.358	7.281	0.951	20.954	16.554	2.994	3.257
007	MeanRest	#VALUE!	-9.16703	11.35762	#VALUE!	0.95132	#VALUE!	16.5535	2.99418	3.2566
007	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min6	0.249	-93.512	93.581	59.988	2.574	56.692	36.847	4.047	3.868
007	MeanEx	#VALUE!	-93.5115	93.5813	#VALUE!	2.573817	#VALUE!	36.84669	4.047256	3.8683

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EELVavg Mech 6MIN</b>	<b>EELVtyp Mech 6MIN</b>	<b>EELVavg %TLC 6MIN</b>	<b>EELVTyp %TLC 6MIN</b>	<b>EILVavg Mech 6MIN</b>	<b>EILVtyp Mech 6MIN</b>
001	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	RestMin3	3.333	3.12	50.88063	47.61527	5.337406	5.123525
001	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min6	3.932	3.95	60.02792	60.34351	6.642708	6.663379
001	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	RestMin3	3.904	4.21	48.1353	51.95684	6.247683	6.55761
002	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min6	4.358	4.33	53.73236	53.40074	6.672927	6.646033
002	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	RestMin1	2.185	2.22	36.85481	37.4941	2.99303	3.03094
003	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	RestMin3	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!
003	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min6	1.935	1.96	32.63076	32.97639	3.921375	3.941871
003	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EELVavg Mech 6MIN</b>	<b>EELVtyp Mech 6MIN</b>	<b>EELVavg %TLC 6MIN</b>	<b>EELVTyp %TLC 6MIN</b>	<b>EILVavg Mech 6MIN</b>	<b>EILVtyp Mech 6MIN</b>
004	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	RestMin3	2.929	2.84	39.051	37.91333	4.857092	4.771767
004	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min6	2.309	2.17	30.78308	28.92667	5.490275	5.351044
004	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	RestMin3	2.528	2.70	33.75458	35.98665	4.151618	4.3188
005	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min6	2.088	1.83	27.87984	24.45127	5.027221	4.770421
005	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	RestMin3	2.806	2.54	48.37621	43.85172	3.75714	3.49472
007	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min6	1.753	1.93	30.21972	33.30517	4.326561	4.505517
007	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!



**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EILVavg %TLC 6MIN</b>	<b>EILVtyp %TLC 6MIN</b>	<b>Ti Mech 6MIN</b>	<b>Te Mech 6MIN</b>	<b>TI/Ttot Mech 6MIN</b>	<b>Vt/Ti Mech 6MIN</b>
001	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	RestMin3	81.48711	78.22176	2.260363	2.770531	44.56443	0.886904
001	MeanRest	#VALUE!	#VALUE!	2.260363	2.770531	44.56443	#VALUE!
001	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min6	101.4154	101.731	0.90935	1.201543	43.57531	2.981117
001	MeanEx	#VALUE!	#VALUE!	0.90935	1.201543	43.57531	#VALUE!
002	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	RestMin3	77.03678	80.85832	1.968445	2.330527	44.27231	1.190742
002	MeanRest	#VALUE!	#VALUE!	1.968445	2.330527	44.27231	#VALUE!
002	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min6	82.28023	81.94862	0.809278	0.991317	44.89003	2.860862
002	MeanEx	#VALUE!	#VALUE!	0.809278	0.991317	44.89003	#VALUE!
003	RestMin1	50.47268	51.11197				0.489231
003	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	RestMin3	#REF!	#REF!	1.65063	2.91815	35.5194	#REF!
003	MeanRest	#VALUE!	#VALUE!	1.65063	2.91815	35.5194	#VALUE!
003	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min6	66.12774	66.47337	1.016313	1.1507	46.34649	1.954487
003	MeanEx	#VALUE!	#VALUE!	1.016313	1.1507	46.34649	#VALUE!

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EILVavg %TLC 6MIN</b>	<b>EILVtyp %TLC 6MIN</b>	<b>Ti Mech 6MIN</b>	<b>Te Mech 6MIN</b>	<b>TI/Ttot Mech 6MIN</b>	<b>Vt/Ti Mech 6MIN</b>
004	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	RestMin3	64.76123	63.62356	1.575183	1.9682	44.48399	1.224154
004	MeanRest	#VALUE!	#VALUE!	1.575183	1.9682	44.48399	#VALUE!
004	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min6	73.20367	71.34725	1.292163	1.298288	50.70133	2.462185
004	MeanEx	#VALUE!	#VALUE!	1.292163	1.298288	50.70133	#VALUE!
005	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	RestMin3	55.42881	57.66088	2.376073	3.250536	42.5571	0.683228
005	MeanRest	#VALUE!	#VALUE!	2.376073	3.250536	42.5571	#VALUE!
005	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min6	67.11911	63.69053	1.151629	1.089421	49.55026	2.552055
005	MeanEx	#VALUE!	#VALUE!	1.151629	1.089421	49.55026	#VALUE!
007	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	RestMin3	64.77828	60.25379	2.23283	2.67616	46.03638	0.42606
007	MeanRest	#VALUE!	#VALUE!	2.23283	2.67616	46.03638	#VALUE!
007	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min6	74.59588	77.68133	0.881906	0.799239	51.27189	2.918471
007	MeanEx	#VALUE!	#VALUE!	0.881906	0.799239	51.27189	#VALUE!

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>Vt/Te Mech 6MIN</b>	<b>PetCO2 Mech 6MIN</b>	<b>expBTPS Mech 6MIN</b>	<b>Max Pi Mech 6MIN</b>	<b>Max Pe Mech 6MIN</b>	<b>P to P Mech 6MIN</b>
001	RestMin1	#VALUE!	.	.	.	.	.
001	RestMin2	#VALUE!	.	.	.	.	.
001	RestMin3	0.723589	24.96956	.	-1.71813	2.075931	3.79406
001	MeanRest	#VALUE!	24.96956	#DIV/0!	-1.71813	2.075931	3.79406
001	Min1	#VALUE!	.	.	.	.	.
001	Min2	#VALUE!	.	.	.	.	.
001	Min3	#VALUE!	.	.	.	.	.
001	Min4	#VALUE!	.	.	.	.	.
001	Min5	#VALUE!	.	.	.	.	.
001	Min6	2.256165	33.87436	.	-3.53713	5.121136	8.65827
001	MeanEx	#VALUE!	33.87436	#DIV/0!	-3.53713	5.121136	8.65827
002	RestMin1	#VALUE!	.	.	.	.	.
002	RestMin2	#VALUE!	.	.	.	.	.
002	RestMin3	1.005742	25.7057	.	-1.67063	3.311418	4.982048
002	MeanRest	#VALUE!	25.7057	#DIV/0!	-1.67063	3.311418	4.982048
002	Min1	#VALUE!	.	.	.	.	.
002	Min2	#VALUE!	.	.	.	.	.
002	Min3	#VALUE!	.	.	.	.	.
002	Min4	#VALUE!	.	.	.	.	.
002	Min5	#VALUE!	.	.	.	.	.
002	Min6	2.335512	36.59469	.	-2.80608	4.484633	7.290713
002	MeanEx	#VALUE!	36.59469	#DIV/0!	-2.80608	4.484633	7.290713
003	RestMin1	0.27673		.			
003	RestMin2	#VALUE!	.	.	.	.	.
003	RestMin3	#REF!	38.12225	.	-1.10509	1.15501	2.2601
003	MeanRest	#VALUE!	38.12225	#DIV/0!	-1.10509	1.15501	2.2601
003	Min1	#VALUE!	.	.	.	.	.
003	Min2	#VALUE!	.	.	.	.	.
003	Min3	#VALUE!	.	.	.	.	.
003	Min4	#VALUE!	.	.	.	.	.
003	Min5	#VALUE!	.	.	.	.	.
003	Min6	1.726228	37.82935	.	-2.31877	3.713683	6.032453
003	MeanEx	#VALUE!	37.82935	#DIV/0!	-2.31877	3.713683	6.032453

**Appendix A.f: 6-Minute Cycling Beetroot Juice continued**

<b>ID</b>	<b>Stage</b>	<b>Vt/Te Mech 6MIN</b>	<b>PetCO2 Mech 6MIN</b>	<b>expBTPS Mech 6MIN</b>	<b>Max Pi Mech 6MIN</b>	<b>Max Pe Mech 6MIN</b>	<b>P to P Mech 6MIN</b>
004	RestMin1	#VALUE!	.	.	.	.	.
004	RestMin2	#VALUE!	.	.	.	.	.
004	RestMin3	0.979711	26.74448	.	-1.78473	2.452342	4.237072
004	MeanRest	#VALUE!	26.74448	#DIV/0!	-1.78473	2.452342	4.237072
004	Min1	#VALUE!	.	.	.	.	.
004	Min2	#VALUE!	.	.	.	.	.
004	Min3	#VALUE!	.	.	.	.	.
004	Min4	#VALUE!	.	.	.	.	.
004	Min5	#VALUE!	.	.	.	.	.
004	Min6	2.450569	32.65206	.	-2.56997	4.503369	7.073339
004	MeanEx	#VALUE!	32.65206	#DIV/0!	-2.56997	4.503369	7.073339
005	RestMin1	#VALUE!	.	.	.	.	.
005	RestMin2	#VALUE!	.	.	.	.	.
005	RestMin3	0.499425	30.58471	.	-1.36693	1.848527	3.215457
005	MeanRest	#VALUE!	30.58471	#DIV/0!	-1.36693	1.848527	3.215457
005	Min1	#VALUE!	.	.	.	.	.
005	Min2	#VALUE!	.	.	.	.	.
005	Min3	#VALUE!	.	.	.	.	.
005	Min4	#VALUE!	.	.	.	.	.
005	Min5	#VALUE!	.	.	.	.	.
005	Min6	2.697783	40.13618	.	-3.11614	5.885436	9.001576
005	MeanEx	#VALUE!	40.13618	#DIV/0!	-3.11614	5.885436	9.001576
007	RestMin1	#VALUE!	.	.	.	.	.
007	RestMin2	#VALUE!	.	.	.	.	.
007	RestMin3	0.355479	34.54452	.	-0.99667	0.84569	1.84236
007	MeanRest	#VALUE!	34.54452	#DIV/0!	-0.99667	0.84569	1.84236
007	Min1	#VALUE!	.	.	.	.	.
007	Min2	#VALUE!	.	.	.	.	.
007	Min3	#VALUE!	.	.	.	.	.
007	Min4	#VALUE!	.	.	.	.	.
007	Min5	#VALUE!	.	.	.	.	.
007	Min6	3.220335	34.59155	.	-3.72741	6.728739	10.45615
007	MeanEx	#VALUE!	34.59155	#DIV/0!	-3.72741	6.728739	10.45615

### Appendix A.g: 6-Minute Cycling Tomato Juice

ID	Stage	P <sub>B</sub>	Watts 6MIN	VE 6MIN	VE/MVV 6MIN	VT 6MIN	fB 6MIN
001	RestMin1	687.0	0.0	20	8.176	2	10.4
001	RestMin2	687.0	0.0	18	7.487	2	9.2
001	RestMin3	687.0	0.0	20	8.032	2	12.0
001	MeanRest	687	0.0	19.35113	7.89842	1.858457	10.50832
001	Min1	687.0	155.0	34	13.837	2	14.7
001	Min2	687.0	155.0	48	19.614	3	17.0
001	Min3	687.0	155.0	62	25.306	3	20.6
001	Min4	687.0	155.0	70	28.517	3	20.4
001	Min5	687.0	155.0	77	31.528	3	24.8
001	Min6	687.0	155.0	86	34.936	3	28.0
001	MeanEx	687	155.0	77.56839	31.66057	3.201291	24.38334
002	RestMin1	688.0	0.0	39	20.016	2	18.1
002	RestMin2	688.0	0.0	34	17.370	2	16.8
002	RestMin3	688.0	0.0	24	12.165	1	17.4
002	MeanRest	688	0.0	32.53915	16.51733	1.864704	17.43091
002	Min1	688.0	150.0	44	22.394	2	22.5
002	Min2	688.0	150.0	45	23.061	2	23.5
002	Min3	688.0	150.0	60	30.699	2	24.5
002	Min4	688.0	150.0	63	31.751	3	22.9
002	Min5	688.0	150.0	67	34.217	3	25.7
002	Min6	688.0	150.0	73	37.298	3	26.0
002	MeanEx	688	150.0	67.81152	34.42209	2.724313	24.88754
003	RestMin1	685.0	0.0	8	5.770	1	12.0
003	RestMin2	685.0	0.0	9	6.686	1	13.1
003	RestMin3	685.0	0.0	8	5.727	1	12.1
003	MeanRest	685	0.0	8.06119	6.061045	0.650042	12.3854
003	Min1	685.0	102.0	23	17.384	2	15.4
003	Min2	685.0	102.0	33	24.908	1	22.5
003	Min3	685.0	102.0	41	30.982	2	21.4
003	Min4	685.0	102.0	47	35.642	2	24.6
003	Min5	685.0	102.0	50	37.649	2	26.1
003	Min6	685.0	102.0	51	38.575	2	24.4
003	MeanEx	685	102.0	49.594	37.28872	1.98216	25.04267

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>P<sub>B</sub></b>	<b>Watts 6MIN</b>	<b>VE 6MIN</b>	<b>VE/MVV 6MIN</b>	<b>VT 6MIN</b>	<b>fB 6MIN</b>
004	RestMin1	681.0	0.0	23	12.637	1	15.7
004	RestMin2	681.0	0.0	28	15.287	2	18.1
004	RestMin3	681.0	0.0	24	13.376	1	18.6
004	MeanRest	681	0.0	24.77984	13.76658	1.42125	17.46545
004	Min1	681.0	108.0	36	19.744	2	14.6
004	Min2	681.0	108.0	50	27.653	3	16.0
004	Min3	681.0	108.0	55	30.698	3	17.6
004	Min4	681.0	108.0	58	32.074	3	17.7
004	Min5	681.0	108.0	54	29.834	3	18.6
004	Min6	681.0	108.0	57	31.416	3	18.4
004	MeanEx	681	108.0	55.99475	31.10819	3.074764	18.2314
005	RestMin1	686.0	0.0	18	9.563	1	12.5
005	RestMin2	686.0	0.0	21	11.153	2	10.3
005	RestMin3	686.0	0.0	25	13.688	2	13.7
005	MeanRest	686	0.0	21.21631	11.46828	1.759423	12.13795
005	Min1	686.0	152.0	40	21.651	2	16.8
005	Min2	686.0	152.0	57	30.930	3	17.7
005	Min3	686.0	152.0	59	31.795	3	17.4
005	Min4	686.0	152.0	62	33.536	4	16.8
005	Min5	686.0	152.0	65	34.891	4	17.7
005	Min6	686.0	152.0	131	70.634	4	33.1
005	MeanEx	686	152.0	85.75434	46.3537	3.760245	22.54497
007	RestMin1	684.0	0.0	11	6.897	1	10.0
007	RestMin2	684.0	0.0	11	6.943	1	11.5
007	RestMin3	684.0	0.0	12	7.822	1	12.8
007	MeanRest	684	0.0	11.26385	7.220415	0.991395	11.42296
007	Min1	684.0	125.0	28	17.888	2	17.6
007	Min2	684.0	125.0	49	31.155	2	24.7
007	Min3	684.0	125.0	54	34.878	2	27.0
007	Min4	684.0	125.0	62	39.564	2	26.7
007	Min5	684.0	125.0	70	44.556	2	31.6
007	Min6	684.0	125.0	77	49.218	2	34.9
007	MeanEx	684	125.0	69.33563	44.44592	2.239461	31.03342

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>VO2 6MIN</b>	<b>VO2 ml/kg 6MIN</b>	<b>Peak VO 6MIN</b>	<b>PerVO 6MIN</b>	<b>VCO2 6MIN</b>	<b>RER 6MIN</b>
001	RestMin1	0.4654	0.004	2.398	19.410	0.498	1.071
001	RestMin2	0.5028	0.004	2.398	20.967	0.484	0.963
001	RestMin3	0.4944	0.004	2.398	20.616	0.518	1.048
001	MeanRest	0.48752	0.004295	2.397877	20.33132	0.500238	1.027312
001	Min1	1.0914	0.010	2.398	45.517	0.962	0.881
001	Min2	1.8763	0.017	2.398	78.247	1.687	0.899
001	Min3	2.1953	0.019	2.398	91.552	2.256	1.028
001	Min4	2.3023	0.020	2.398	96.013	2.439	1.059
001	Min5	2.3853	0.021	2.398	99.474	2.505	1.050
001	Min6	2.3979	0.021	2.398	100.000	2.541	1.060
001	MeanEx	2.361807	0.020809	2.397877	98.49577	2.495152	1.05648529
002	RestMin1	0.7951	0.007	2.381	33.388	0.728	0.916
002	RestMin2	0.5423	0.005	2.381	22.771	0.593	1.094
002	RestMin3	0.5588	0.005	2.381	23.465	0.462	0.828
002	MeanRest	0.632035	0.005397	2.38132	26.54138	0.594585	0.94579131
002	Min1	1.4306	0.012	2.381	60.074	1.054	0.737
002	Min2	1.8913	0.016	2.381	79.423	1.406	0.744
002	Min3	2.2660	0.019	2.381	95.157	1.954	0.862
002	Min4	2.1675	0.019	2.381	91.021	2.041	0.942
002	Min5	2.3170	0.020	2.381	97.299	2.161	0.933
002	Min6	2.3813	0.020	2.381	100.000	2.260	0.949
002	MeanEx	2.288607	0.019544	2.38132	96.10663	2.15413	0.94118163
003	RestMin1	0.2724	0.003	1.663	16.385	0.200	0.735
003	RestMin2	0.3040	0.003	1.663	18.285	0.225	0.740
003	RestMin3	0.2557	0.003	1.663	15.381	0.199	0.777
003	MeanRest	0.277407	0.002908	1.662742	16.68371	0.207936	0.75051312
003	Min1	0.7761	0.008	1.663	46.678	0.720	0.927
003	Min2	1.2978	0.014	1.663	78.052	1.152	0.887
003	Min3	1.4262	0.015	1.663	85.777	1.448	1.015
003	Min4	1.5397	0.016	1.663	92.597	1.655	1.075
003	Min5	1.5688	0.016	1.663	94.350	1.688	1.076
003	Min6	1.6627	0.017	1.663	100.000	1.765	1.061
003	MeanEx	1.590394	0.016671	1.662742	95.64887	1.702561	1.07073809

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>VO2 6MIN</b>	<b>VO2 ml/kg 6MIN</b>	<b>Peak VO 6MIN</b>	<b>PerVO 6MIN</b>	<b>VCO2 6MIN</b>	<b>RER 6MIN</b>
004	RestMin1	0.6435	0.004	1.966	32.732	0.571	0.888
004	RestMin2	0.7531	0.005	1.966	38.310	0.686	0.911
004	RestMin3	0.5718	0.004	1.966	29.085	0.567	0.991
004	MeanRest	0.656118	0.004588	1.965859	33.37561	0.608199	0.93023725
004	Min1	1.2617	0.009	1.966	64.178	1.024	0.812
004	Min2	1.7680	0.012	1.966	89.937	1.590	0.899
004	Min3	1.8982	0.013	1.966	96.558	1.771	0.933
004	Min4	1.9142	0.013	1.966	97.370	1.798	0.939
004	Min5	1.9110	0.013	1.966	97.207	1.722	0.901
004	Min6	1.9659	0.014	1.966	100.000	1.814	0.923
004	MeanEx	1.930329	0.013499	1.965859	98.19265	1.77788	0.9209997
005	RestMin1	0.6487	0.005	2.893	22.422	0.527	0.813
005	RestMin2	0.6140	0.005	2.893	21.222	0.551	0.897
005	RestMin3	0.6725	0.005	2.893	23.244	0.650	0.966
005	MeanRest	0.645055	0.004861	2.893164	22.29582	0.576124	0.89232192
005	Min1	1.4589	0.011	2.893	50.426	1.214	0.832
005	Min2	2.0511	0.015	2.893	70.895	1.910	0.931
005	Min3	1.9939	0.015	2.893	68.916	2.025	1.016
005	Min4	2.2586	0.017	2.893	78.068	2.214	0.980
005	Min5	2.4227	0.018	2.893	83.740	2.328	0.961
005	Min6	2.8932	0.022	2.893	100.000	3.328	1.150
005	MeanEx	2.524846	0.019027	2.893164	87.26936	2.623659	1.03061514
007	RestMin1	0.1832	0.002	2.414	7.589	0.329	1.794
007	RestMin2	0.3328	0.004	2.414	13.785	0.343	1.030
007	RestMin3	0.4030	0.004	2.414	16.694	0.365	0.907
007	MeanRest	0.306338	0.003269	2.414086	12.68961	0.345549	1.24333476
007	Min1	1.1117	0.012	2.414	46.051	1.003	0.902
007	Min2	1.9008	0.020	2.414	78.736	1.899	0.999
007	Min3	1.9345	0.021	2.414	80.133	2.074	1.072
007	Min4	2.0215	0.022	2.414	83.736	2.243	1.110
007	Min5	2.2020	0.024	2.414	91.216	2.391	1.086
007	Min6	2.4141	0.026	2.414	100.000	2.580	1.069
007	MeanEx	2.21253	0.023613	2.414086	91.65083	2.404696	1.08806347



**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>VE/VO2 6MIN</b>	<b>VE/VCO2 6MIN</b>	<b>Vent Resp Ex 6MIN</b>	<b>HR 6MIN</b>	<b>PHR 6MIN</b>	<b>HR PP 6MIN</b>	<b>O2Pul 6MIN</b>
001	RestMin1	43.039	40.20	.	85.80	202.46	42.380	5.424
001	RestMin2	36.483	37.89	.	87.69	202.46	43.310	5.734
001	RestMin3	39.807	37.97	.	80.50	202.46	39.759	6.141
001	MeanRest	39.77646	38.68593	.	84.66136	202.459	41.81655	5.766547
001	Min1	31.061	35.26	.	83.32	202.46	41.154	13.099
001	Min2	25.611	28.49	.	120.72	202.46	59.625	15.543
001	Min3	28.242	27.48	.	135.81	202.46	67.080	16.165
001	Min4	30.347	28.65	.	137.55	202.46	67.941	16.737
001	Min5	32.384	30.83	.	146.50	202.46	72.362	16.281
001	Min6	35.695	33.68	.	150.96	202.46	74.562	15.885
001	MeanEx	32.80884	31.05362	29.18285	145.0045	202.459	71.62166	16.30106
002	RestMin1	49.596	54.16	.	72.32	202.46	35.722	10.993
002	RestMin2	63.106	57.68	.	75.74	202.46	37.410	7.160
002	RestMin3	42.889	51.82	.	71.42	202.46	35.278	7.823
002	MeanRest	51.8637	54.55483	.	73.16	202.459	36.13675	8.658743
002	Min1	30.838	41.85	.	94.65	202.46	46.748	15.115
002	Min2	24.020	32.30	.	110.48	202.46	54.571	17.118
002	Min3	26.689	30.95	.	118.25	202.46	58.405	19.163
002	Min4	28.858	30.64	.	120.69	202.46	59.610	17.960
002	Min5	29.093	31.19	.	124.11	202.46	61.303	18.668
002	Min6	30.856	32.52	.	128.62	202.46	63.531	18.514
002	MeanEx	29.60209	31.44853	22.61709	124.4741	202.459	61.48116	18.38073
003	RestMin1	28.169	38.34	.	66.59	202.46	32.890	4.091
003	RestMin2	29.248	39.54	.	64.53	202.46	31.873	4.712
003	RestMin3	29.782	38.32	.	67.22	202.46	33.201	3.805
003	MeanRest	29.06653	38.7348	.	66.11287	202.459	32.65494	4.202509
003	Min1	29.789	32.12	.	102.56	202.46	50.659	7.567
003	Min2	25.526	28.76	.	108.74	202.46	53.712	11.934
003	Min3	28.892	28.45	.	115.61	202.46	57.105	12.336
003	Min4	30.789	28.64	.	122.21	202.46	60.363	12.598
003	Min5	31.918	29.66	.	123.01	202.46	60.757	12.754
003	Min6	30.856	29.07	.	130.39	202.46	64.401	12.752
003	MeanEx	31.18757	29.12676	27.78811	125.2013	202.459	61.84033	12.70147

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>VE/VO2 6MIN</b>	<b>VE/VCO2 6MIN</b>	<b>Vent Resp Ex 6MIN</b>	<b>HR 6MIN</b>	<b>PHR 6MIN</b>	<b>HR PP 6MIN</b>	<b>O2Pul 6MIN</b>
004	RestMin1	35.350	39.80	.	95.29	202.46	47.067	6.753
004	RestMin2	36.537	40.09	.	98.52	202.46	48.660	7.645
004	RestMin3	42.108	42.48	.	95.34	202.46	47.091	5.997
004	MeanRest	37.99846	40.79153	.	96.38279	202.459	47.60608	6.798111
004	Min1	28.169	34.71	.	124.62	202.46	61.554	10.124
004	Min2	28.153	31.30	.	147.28	202.46	72.747	12.004
004	Min3	29.109	31.19	.	148.47	202.46	73.336	12.785
004	Min4	30.161	32.11	.	145.66	202.46	71.944	13.142
004	Min5	28.102	31.18	.	149.38	202.46	73.784	12.792
004	Min6	28.766	31.18	.	149.30	202.46	73.742	13.167
004	MeanEx	29.00955	31.49174	26.6867	148.112	202.459	73.15656	13.03383
005	RestMin1	27.273	33.54	.	81.17	202.46	40.093	7.992
005	RestMin2	33.606	37.44	.	77.10	202.46	38.084	7.963
005	RestMin3	37.657	38.97	.	80.42	202.46	39.721	8.362
005	MeanRest	32.84522	36.65113	.	79.56497	202.459	39.2993	8.105653
005	Min1	27.455	33.00	.	96.33	202.46	47.578	15.145
005	Min2	27.898	29.95	.	107.73	202.46	53.212	19.039
005	Min3	29.501	29.04	.	115.45	202.46	57.021	17.271
005	Min4	27.469	28.02	.	118.25	202.46	58.409	19.100
005	Min5	26.643	27.72	.	123.39	202.46	60.947	19.635
005	Min6	45.166	39.26	.	133.90	202.46	66.139	21.606
005	MeanEx	33.09251	31.66739	31.51986	125.1834	202.459	61.83148	20.11352
007	RestMin1	58.728	32.74	.	83.91	202.46	41.445	2.183
007	RestMin2	32.544	31.61	.	84.42	202.46	41.697	3.942
007	RestMin3	30.276	33.40	.	82.17	202.46	40.585	4.905
007	MeanRest	40.516	32.58124	.	83.49917	202.459	41.24251	3.676748
007	Min1	25.101	27.83	.	128.85	202.46	63.641	8.628
007	Min2	25.570	25.59	.	137.96	202.46	68.143	13.777
007	Min3	28.127	26.23	.	141.09	202.46	69.689	13.711
007	Min4	30.533	27.51	.	149.18	202.46	73.683	13.551
007	Min5	31.565	29.07	.	157.53	202.46	77.807	13.979
007	Min6	31.805	29.76	.	162.35	202.46	80.189	14.870
007	MeanEx	31.30072	28.78184	28.20186	156.3513	202.459	77.22615	14.13308

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>PetCO2 6MIN</b>	<b>PulseOx 6MIN</b>	<b>RPB 6MIN</b>	<b>RPU 6MIN</b>	<b>RPE 6MIN</b>	<b>FECO2 6MIN</b>	<b>PmeCO2 calc 6MIN</b>
001	RestMin1			.	.	.	0.034	21.721
001	RestMin2			.	.	.	0.036	23.035
001	RestMin3			0.000	0.000	0.000	0.036	22.986
001	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.035282	22.58063
001	Min1	43.020	93.700				0.039	24.735
001	Min2	45.670	87.400	0.500	0.500	8.000	0.048	30.554
001	Min3	42.310	93.000				0.049	31.456
001	Min4	40.380	93.700	2.000	2.000	9.000	0.047	30.378
001	Min5	41.830	94.400	.	.	.	0.041	26.472
001	Min6	37.020	96.500	2.000	2.000	11.000	0.040	25.878
001	MeanEx	39.74333	94.86667	2	2	10	0.043088	27.57627
002	RestMin1			.	.	.	0.025	16.190
002	RestMin2			.	.	.	0.024	15.217
002	RestMin3			0.000	0.000	0.000	0.026	16.912
002	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.025127	16.10654
002	Min1	30.300	98.700	.	.	.	0.033	20.881
002	Min2	36.700	98.700	0.000	0.000	7.000	0.042	26.976
002	Min3	40.200	98.700	.	.	.	0.044	28.142
002	Min4	39.000	98.700	0.000	0.000	7.000	0.043	27.819
002	Min5	37.900	98.900	.	.	.	0.044	27.930
002	Min6	36.700	98.900	0.500	0.500	7.000	0.042	26.796
002	MeanEx	37.86667	98.83333	0.25	0.25	7	0.042925	27.51487
003	RestMin1			.	.	.	0.036	22.768
003	RestMin2			.	.	.	0.035	22.085
003	RestMin3			0.000	0.000	0.000	0.036	22.776
003	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.035333	22.54273
003	Min1	44.140	98.850	.	.	.	0.043	27.125
003	Min2	44.140	95.660	1.000	1.000	8.000	0.047	30.261
003	Min3	50.790	96.720	.	.	.	0.048	30.585
003	Min4	47.770	96.720	2.000	1.000	9.000	0.048	30.383
003	Min5	45.350	96.720	.	.	.	0.046	29.346
003	Min6	43.350	95.550	3.000	2.000	11.000	0.047	29.940
003	MeanEx	45.49	96.33	2.5	1.5	10	0.046849	29.88976

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

ID	Stage	PetCO2 6MIN	PulseOx 6MIN	RPB 6MIN	RPU 6MIN	RPE 6MIN	FECO2 6MIN	PmeCO2 calc 6MIN
004	RestMin1		.	.	.	.	0.035	21.937
004	RestMin2		.	.	.	.	0.034	21.781
004	RestMin3		.	0.000	0.000	0.000	0.032	20.568
004	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.033799	21.42873
004	Min1	38.300	98.820	.	.	.	0.040	25.122
004	Min2	38.300	97.550	2.000	2.000	11.000	0.044	27.824
004	Min3	38.300	97.550	.	.	.	0.044	27.922
004	Min4	37.800	96.760	2.000	3.000	11.000	0.043	27.130
004	Min5	41.100	96.760	.	.	.	0.044	27.931
004	Min6	36.900	96.760	3.000	2.000	11.000	0.044	27.931
004	MeanEx	38.6	96.76	2.5	2.5	11	0.043634	27.66392
005	RestMin1		.	.	.	.	0.041	25.985
005	RestMin2		.	.	.	.	0.036	23.305
005	RestMin3		.	2.000	0.000	0.000	0.035	22.404
005	MeanRest	#DIV/0!	#DIV/0!	2	0	0	0.037399	23.89795
005	Min1	36.700	97.700	.	.	.	0.041	26.407
005	Min2	41.900	96.700	3.000	1.000	10.000	0.045	29.068
005	Min3	41.900	96.700	.	.	.	0.047	29.969
005	Min4	41.900	96.700	3.000	3.000	12.000	0.049	31.059
005	Min5	44.200	96.700	.	.	.	0.049	31.388
005	Min6	28.000	98.800	5.000	5.000	15.000	0.035	22.234
005	MeanEx	38.03333	97.4	4	4	13.5	0.044173	28.22678
007	RestMin1	.	.	.	.	.	0.042	26.611
007	RestMin2	.	.	.	.	.	0.043	27.556
007	RestMin3	.	.	0.000	0.000	0.000	0.041	26.098
007	MeanRest	#DIV/0!	#DIV/0!	0	0	0	0.042001	26.7549
007	Min1	44.200	99.770	.	.	.	0.049	31.261
007	Min2	44.200	98.710	2.000	3.000	13.000	0.053	33.978
007	Min3	41.900	98.710	.	.	.	0.052	33.153
007	Min4	39.600	97.540	3.000	4.000	15.000	0.050	31.623
007	Min5	39.600	98.710	.	.	.	0.047	29.936
007	Min6	37.300	100.000	7.000	7.000	16.000	0.046	29.253
007	MeanEx	38.83333	98.75	5	5.5	15.5	0.047521	30.27069

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

ID	Stage	Vd/Vt calc 6MIN	MinVi Mech 6MIN	MinVe Mech 6MIN	VE/MVV Mech 6MIN	Vt Mech 6MIN	Vt/FVC Mech 6MIN	fB Mech 6MIN	Avg IC 6MIN	Typ IC 6MIN
001	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	RestMin3	0.256	-20.187	19.870	8.110	1.842	30.147	11.253	4.077	3.866
001	MeanRest	#VALUE!	-20.1873	19.87023	#VALUE!	1.842	#VALUE!	11.25272	4.07687	3.8656
001	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
001	Min6	0.311	-73.227	73.217	29.885	2.593	42.433	28.074	4.115	4.069
001	MeanEx	#VALUE!	-73.2272	73.21729	#VALUE!	2.592686	#VALUE!	28.07361	4.114621	4.069
002	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	RestMin3	0.326	-24.941	23.925	12.145	1.421	19.282	19.968	4.368	4.100
002	MeanRest	#VALUE!	-24.9412	23.92495	#VALUE!	1.421053	#VALUE!	19.96844	4.36806	4.0996
002	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
002	Min6	0.249	-79.187	78.648	39.923	2.688	36.473	30.903	4.794	4.768
002	MeanEx	#VALUE!	-79.1867	78.64845	#VALUE!	2.68807	#VALUE!	30.90264	4.793686	4.7676
003	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	RestMin3	0.478	-9.099	9.315	7.003	0.770	17.609	12.065	3.894	3.880
003	MeanRest	#VALUE!	-9.09901	9.314513	#VALUE!	0.769525	#VALUE!	12.06484	3.89355	3.8803
003	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
003	Min6	0.362	-54.563	55.397	41.652	2.252	51.537	24.570	3.008	2.856
003	MeanEx	#VALUE!	-54.5627	55.39656	#VALUE!	2.252167	#VALUE!	24.56983	3.007694	2.8555

### Appendix A.g: 6-Minute Cycling Tomato Juice continued

ID	Stage	Vd/Vt calc 6MIN	MinVi Mech 6MIN	MinVe Mech 6MIN	VE/MVV Mech 6MIN	Vt Mech 6MIN	Vt/FVC Mech 6MIN	fB Mech 6MIN	Avg IC 6MIN	Typ IC 6MIN
004	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	RestMin3	0.363	-25.487	24.588	13.660	1.404	23.759	18.191	5.066	4.811
004	MeanRest	#VALUE!	-25.4873	24.58824	#VALUE!	1.404133	#VALUE!	18.19136	5.065647	4.8109
004	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
004	Min6	0.231	-61.273	60.352	33.529	3.321	56.195	18.547	5.655	5.987
004	MeanEx	#VALUE!	-61.2732	60.35234	#VALUE!	3.321144	#VALUE!	18.54681	5.654638	5.9868
005	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	RestMin3	0.249	-31.546	31.354	16.948	2.355	42.213	13.250	4.935	4.701
005	MeanRest	#VALUE!	-31.5457	31.35434	#VALUE!	2.35547	#VALUE!	13.25013	4.93541	4.7006
005	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
005	Min6	0.250	-154.473	155.061	83.817	4.364	78.206	35.536	9.564	9.932
005	MeanEx	#VALUE!	-154.473	155.0611	#VALUE!	4.363878	#VALUE!	35.53581	9.5641	9.9315
007	RestMin1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	RestMin2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	RestMin3	0.319	-13.576	13.120	8.410	1.064	23.426	12.922	3.117	3.369
007	MeanRest	#VALUE!	-13.5764	13.11963	#VALUE!	1.06356	#VALUE!	12.92225	3.11683	3.3688
007	Min1	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min2	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min3	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min4	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min5	#VALUE!	.	.	#VALUE!	.	#VALUE!	.	.	.
007	Min6	0.223	-83.313	83.409	53.467	2.403	52.940	34.723	3.803	3.813
007	MeanEx	#VALUE!	-83.3125	83.40891	#VALUE!	2.403493	#VALUE!	34.72285	3.802536	3.8131

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EELVavg Mech 6MIN</b>	<b>EELVtyp Mech 6MIN</b>	<b>EELVavg %TLC 6MIN</b>	<b>EELVTyp %TLC 6MIN</b>	<b>EILVavg Mech 6MIN</b>	<b>EILVtyp Mech 6MIN</b>
001	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	RestMin3	2.473	2.68	37.75771	40.98321	4.31513	4.5264
001	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
001	Min6	2.435	2.48	37.18136	37.87786	5.028065	5.073686
001	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	RestMin3	3.742	4.01	46.13983	49.45006	5.162993	5.431453
002	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
002	Min6	3.316	3.34	40.89166	41.21332	6.004384	6.03047
002	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	RestMin3	2.036	2.05	34.34148	34.56492	2.805975	2.819225
003	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
003	Min6	2.922	3.07	49.28003	51.84654	5.174473	5.326667
003	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EELVavg Mech 6MIN</b>	<b>EELVtyp Mech 6MIN</b>	<b>EELVavg %TLC 6MIN</b>	<b>EELVTyp %TLC 6MIN</b>	<b>EILVavg Mech 6MIN</b>	<b>EILVtyp Mech 6MIN</b>
004	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	RestMin3	2.434	2.69	32.45804	35.85467	3.838486	4.093233
004	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
004	Min6	1.845	1.51	24.60483	20.176	5.166506	4.834344
004	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	RestMin3	2.555	2.79	34.10668	37.24166	4.91006	5.14487
005	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
005	Min6	0.000	0.00	0	0	2.289778	1.922378
005	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	RestMin1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	RestMin2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	RestMin3	2.683	2.43	46.26155	41.91724	3.74673	3.49476
007	MeanRest	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min1	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min2	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min3	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min4	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min5	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!
007	Min6	1.997	1.99	34.43903	34.2569	4.400957	4.390393
007	MeanEx	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!	#VALUE!



**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EILVavg %TLC 6MIN</b>	<b>EILVtyp %TLC 6MIN</b>	<b>Ti Mech 6MIN</b>	<b>Te Mech 6MIN</b>	<b>TI/Ttot Mech 6MIN</b>	<b>Vt/Ti Mech 6MIN</b>
001	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	RestMin3	65.87985	69.10534	2.47262	3.02405	45.86527	0.744959
001	MeanRest	#VALUE!	#VALUE!	2.47262	3.02405	45.86527	#VALUE!
001	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
001	Min6	76.76435	77.46085	1.03905	1.149107	46.87564	2.495247
001	MeanEx	#VALUE!	#VALUE!	1.03905	1.149107	46.87564	#VALUE!
002	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	RestMin3	63.66206	66.97229	1.28476	2.461607	37.14003	1.106084
002	MeanRest	#VALUE!	#VALUE!	1.28476	2.461607	37.14003	#VALUE!
002	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
002	Min6	74.03679	74.35845	1.1388	1.2766	47.1474	2.360441
002	MeanEx	#VALUE!	#VALUE!	1.1388	1.2766	47.1474	#VALUE!
003	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	RestMin3	47.3183	47.54174	1.504613	3.522175	29.95464	0.511444
003	MeanRest	#VALUE!	#VALUE!	1.504613	3.522175	29.95464	#VALUE!
003	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
003	Min6	87.25924	89.82575	1.15775	1.307244	46.40758	1.945296
003	MeanEx	#VALUE!	#VALUE!	1.15775	1.307244	46.40758	#VALUE!

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>EILVavg %TLC 6MIN</b>	<b>EILVtyp %TLC 6MIN</b>	<b>Ti Mech 6MIN</b>	<b>Te Mech 6MIN</b>	<b>TI/Ttot Mech 6MIN</b>	<b>Vt/Ti Mech 6MIN</b>
004	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	RestMin3	51.17981	54.57644	1.495633	1.898633	44.97419	0.938822
004	MeanRest	#VALUE!	#VALUE!	1.495633	1.898633	44.97419	#VALUE!
004	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
004	Min6	68.88675	64.45792	1.583831	1.698531	48.15909	2.096906
004	MeanEx	#VALUE!	#VALUE!	1.583831	1.698531	48.15909	#VALUE!
005	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	RestMin3	65.55487	68.68985	2.27279	2.44575	49.21096	1.036378
005	MeanRest	#VALUE!	#VALUE!	2.27279	2.44575	49.21096	#VALUE!
005	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
005	Min6	30.57113	25.66593	0.9406	0.8	54.0382	4.639462
005	MeanEx	#VALUE!	#VALUE!	0.9406	0.8	54.0382	#VALUE!
007	RestMin1	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	RestMin2	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	RestMin3	64.59879	60.25448	2.38247	2.49455	47.69781	0.446411
007	MeanRest	#VALUE!	#VALUE!	2.38247	2.49455	47.69781	#VALUE!
007	Min1	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min2	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min3	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min4	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min5	#VALUE!	#VALUE!	.	.	.	#VALUE!
007	Min6	75.87857	75.69643	0.939607	0.836371	51.63681	2.557977
007	MeanEx	#VALUE!	#VALUE!	0.939607	0.836371	51.63681	#VALUE!

**Appendix A.g: 6-Minute Cycling Tomato Juice continued**

<b>ID</b>	<b>Stage</b>	<b>Vt/Te Mech 6MIN</b>	<b>PetCO2 Mech 6MIN</b>	<b>expBTPS Mech 6MIN</b>	<b>Max Pi Mech 6MIN</b>	<b>Max Pe Mech 6MIN</b>	<b>P to P Mech 6MIN</b>
004	RestMin1	#VALUE!	.	.	.	.	.
004	RestMin2	#VALUE!	.	.	.	.	.
004	RestMin3	0.739549	32.27237	.	-1.52099	1.5446	3.06559
004	MeanRest	#VALUE!	32.27237	#DIV/0!	-1.52099	1.5446	3.06559
004	Min1	#VALUE!	.	.	.	.	.
004	Min2	#VALUE!	.	.	.	.	.
004	Min3	#VALUE!	.	.	.	.	.
004	Min4	#VALUE!	.	.	.	.	.
004	Min5	#VALUE!	.	.	.	.	.
004	Min6	1.955304	36.34428	.	-2.43609	3.433369	5.869459
004	MeanEx	#VALUE!	36.34428	#DIV/0!	-2.43609	3.433369	5.869459
005	RestMin1	#VALUE!	.	.	.	.	.
005	RestMin2	#VALUE!	.	.	.	.	.
005	RestMin3	0.963087	29.84333	.	-1.64345	2.87054	4.51399
005	MeanRest	#VALUE!	29.84333	#DIV/0!	-1.64345	2.87054	4.51399
005	Min1	#VALUE!	.	.	.	.	.
005	Min2	#VALUE!	.	.	.	.	.
005	Min3	#VALUE!	.	.	.	.	.
005	Min4	#VALUE!	.	.	.	.	.
005	Min5	#VALUE!	.	.	.	.	.
005	Min6	5.454848	29.6429	.	-5.6721	11.6399	17.312
005	MeanEx	#VALUE!	29.6429	#DIV/0!	-5.6721	11.6399	17.312
007	RestMin1	#VALUE!	.	.	.	.	.
007	RestMin2	#VALUE!	.	.	.	.	.
007	RestMin3	0.426353	38.3033	.	-1.00411	1.464	2.46811
007	MeanRest	#VALUE!	38.3033	#DIV/0!	-1.00411	1.464	2.46811
007	Min1	#VALUE!	.	.	.	.	.
007	Min2	#VALUE!	.	.	.	.	.
007	Min3	#VALUE!	.	.	.	.	.
007	Min4	#VALUE!	.	.	.	.	.
007	Min5	#VALUE!	.	.	.	.	.
007	Min6	2.873716	37.67021	.	-3.10673	5.627714	8.734444
007	MeanEx	#VALUE!	37.67021	#DIV/0!	-3.10673	5.627714	8.734444

## Appendix B: Institutional Review Board Approval



**INSTITUTIONAL REVIEW BOARD**  
Office of Research Protections  
ASU Box 32068  
Boone, NC 28608  
828.262.2692  
Web site: <http://researchprotections.appstate.edu>  
Email: [irb@appstate.edu](mailto:irb@appstate.edu)  
Federalwide Assurance (FWA) #00001076

**To:** Dalton Fletcher  
Health and Exercise Science  
CAMPUS EMAIL

**From:** Dr. Lisa Emery, Deputy IRB Chairperson  
**RE:** Notice of IRB Approval by Full Board Review  
**Agrants #:**  
**Grant Title:**

**STUDY #:** 19-0021

**STUDY TITLE:** The effects of beetroot juice supplementation on the oxygen cost of breathing in young, obese males.

**Submission Type:** Initial

**Approval Date:** 10/12/2018

**Expiration Date of Approval:** 10/11/2019

The Institutional Review Board (IRB) reviewed this study at a convened meeting and approved this study for the period indicated above. IRB approval is limited to the activities described in the IRB approved materials, and extends to the performance of the described activities in the sites identified in the IRB application. In accordance with this approval, IRB findings and approval conditions for the conduct of this research are listed below.

### **Study Regulatory and other findings:**

The IRB determined that this study involves more than minimal risk to participants.

All approved documents for this study, including consent forms, can be accessed by logging into IRBIS. Use the following directions to access approved study documents.

1. Log into IRBIS
2. Click "Home" on the top toolbar
3. Click "My Studies" under the heading "All My Studies"
4. Click on the IRB number for the study you wish to access
5. Click on the reference ID for your submission
6. Click "Attachments" on the left-hand side toolbar
7. Click on the appropriate documents you wish to download

## Appendix C: Informed Consent Statement

### Appalachian State University Informed Consent for Participants in Research Projects Involving Human Subjects

**IRB Study #: 19-0021**

**Principal**

**Investigator:** Dalton Fletcher, B.S. Email: [fletcherds@appstate.edu](mailto:fletcherds@appstate.edu)

Advisor: Jonathon Stickford, Ph.D. Email: [stickfordjl@appstate.edu](mailto:stickfordjl@appstate.edu)

Advisor: R. Andrew Shanely, Ph.D. Email: [shanelyra@appstate.edu](mailto:shanelyra@appstate.edu)

Advisor: Abigail Stickford, Ph.D. Email: [stickfordas@appstate.edu](mailto:stickfordas@appstate.edu)

**Research Assistants:** Jayvaughn Oliver, B.S. Email: [oliverjt@appstate.edu](mailto:oliverjt@appstate.edu)  
Erica Larson, M.S. Email: [larsone@appstate.edu](mailto:larsone@appstate.edu)  
John Cantu Email: [cantujw@appstate.edu](mailto:cantujw@appstate.edu)

This is to certify that I, \_\_\_\_\_ have been given the following information with respect to my participation as a volunteer in a program of investigation under the supervision of Dalton Fletcher B.S. to which Jonathon Stickford, Ph.D., Andrew Shanely, Ph.D., Abigail Stickford, Ph.D., Erica Larson M.S., John Cantu, and/or Jayvaughn Oliver B.S. may be assisting.

**1. Purpose of the study:**

Obesity increases the energy needed to breath. Obesity may make breathing feel tough or hard, making exercise feel tough or hard. Beetroot juice and tomato juice are drinks you can buy. Both drinks have natural ingredients that may lower the energy your breathing muscles need during exercise and make exercise feel easier. We want to know if beetroot juice or tomato juice is better at lowering the energy your breathing muscles need during exercise and makes exercise feel easier. You will be 1 of 10 participants if you decide to be in the study. We estimate that being in the study will take about 5 hours.

**2. Inclusion Criteria: You may participate in the study if the following apply to you:**

- Sex: Males will be included in the study.
- Ethnicity: Any
- Age: 18 – 40 years of age
- Interest in participating in the research study
- Obese ( $30 \text{ kg/m}^2 \leq \text{BMI} \leq 40 \text{ kg/m}^2$ ) and otherwise healthy
- Able to speak and read English

## Appendix C: Informed Consent Statement continued

- Provide informed consent
- Available during times the data collection is offered.
- Current non-smoker
- Smoking history less than ½ pack-years.
- Willing to not use mouthwash during the entire time you are taking part in the study
- Resting blood pressure under 140/90
- Is currently not taking any medications or supplements that are known vasodilators

**Exclusion Criteria: You should not participate in this study if any of the following apply to you:**

- Known cardiovascular, metabolic or kidney disease, or signs/symptoms of cardiovascular, metabolic or kidney disease will exclude you from this study
- Currently or you have been involved in an organized exercise program in the past 6 months
- If lung function is not in the range we are looking for you will not be allowed in the study
- BMI under 30 or above 40 kg/m<sup>2</sup>
- Current smoker
- Smoking history equal to or greater than ½ pack-years
- Unwilling to stop using of mouthwash
- Resting blood pressure above 140/90
- Is currently taking medications or supplements that are known vasodilators

**3. Procedures: Please read the descriptions of each experimental day and write your initials in the space provided.**

**You could be asked to repeat a trial, procedure, or test. This could happen for many reasons such as equipment failure, power outage, inconclusive test results, etc. However, you do not have to repeat a trial, procedure, and/or test if you do not wish to do so. If we ask you to repeat a trial we will not give you more money.**

Below is a timeline showing all visits and experiments which you will complete in this study.

<u>Visit 1</u>	<u>Visits 2 and 3</u>
Informed consent	Resting seated hyperventilation
Lung function testing	Moderate intensity cycling
Body composition	
Maximal exercise test	

\_\_\_\_\_ **initial Prescreen:** You may be telephoned by the Principle Investigator or a Research Assistant (see page 1) and asked screening questions to determine your eligibility for the study.

***Visit 1:***

\_\_\_\_\_ **initial Consent and Questionnaires:** Potential participants who meet inclusion criteria will be invited to a screening interview within the laboratory located on campus (Leon Levine Hall of

## Appendix C: Informed Consent Statement continued

Health Sciences). At this screening visit, the study will be explained in-depth to you by the PI or a trained research assistant. You will be provided time to consider your options and have all questions answered - if you agree to participate, you will then provide your written informed consent.

After you have provided consent (~30 minutes), you will be asked to complete a medical history questionnaire (~10 minutes). You will be asked to bring a list of all current medications and supplements that you are currently taking. If any of these medications or supplements are vasodilators you will not be allowed to participate in the study. You will also have your resting blood pressure measured to ensure that it is under 140/90. If it is not, you will not be allowed to participate in the study.

\_\_\_\_\_ **initial Lung Function Testing:** You will be asked to perform tests of breathing function. The protocol will follow that described by the American Thoracic Society. These tests include measurement of the total amount of air your lungs can hold, the amount of air that you can push out with one maximal breath, the amount of air that you can breathe out as hard as you can in one second, and the maximum amount of air that you can in and out of your lungs in 12 seconds. For all these procedures, you will wear a nose clip and breathe through a disposable mouthpiece. These tests will take about 30 minutes.

\_\_\_\_\_ **initial Body Composition:** Following completion of the questionnaires, we will measure your height, weight, and body composition. Your percent body fat will be estimated using a BodPod. You will sit in the BodPod, an enclosed chamber, and may hear some clicking while air pressure changes to estimate your body volume. This piece of equipment estimates your body's fat and muscle by air movement. This is an accurate method to estimate your body composition. These procedures will take about 15 minutes.

\_\_\_\_\_ **initial Maximal Exercise Test ( $\dot{V}O_{2peak}$ ):** You will be asked to exercise as hard as you can to measure your physical fitness. This test is often described as a  $\dot{V}O_{2peak}$  test. You should be rested, well nourished, and hydrated for the test and avoid caffeine 3 hours before the test, and not drink alcohol for 12 hours before the test. Avoid exercise in the 24 hours before the test and report any medication that you are using to the testing staff before the test. When you are ready for the test, the investigators will help you equipment to assure your comfort. You will be fitted with a rubber mouthpiece and nose clip. Your breathing pattern, exercise energy use, blood pressure, and blood oxygen level will be monitored during the test. This takes about 45 minutes, with exercise lasting about 15 minutes.

### *Cycling Protocol*

You will exercise on a stationary bicycle. Prior to exercise, you will rest sitting on the bike with both hands on the handle bars for 5 minutes. You will breathe through a mouthpiece and wear a nose clip during the test. Your breathing pattern, exercise energy needs, blood pressure, and arterial blood oxygen level will be monitored during the test. A cuff will be placed onto your arm, and a monitor will automatically display your blood pressure values. The amount of oxygen in the blood will be monitored using a sensor placed on the skin (e.g., forehead). Throughout the test, you will be asked about your perceptual responses (e.g., ratings of perceived breathlessness, unpleasantness of breathlessness, and exertion) to the exercise. You will be asked to rate your responses by pointing with your finger to a number on a scale. The number will be repeated out loud in order to confirm your choice. During the exercise you may have an even stronger or greater intensity of breathlessness than you have previously experienced. If this occurs, you should point to the word "maximal" if the severity is greater than 10. After the mouthpiece is removed, you can tell us the number. You may stop exercise when you wish,

## Appendix C: Informed Consent Statement continued

because of personal feelings of fatigue or discomfort, but you will be encouraged to cycle for as long as you can. Following completion of the test, you will perform a light intensity cool-down.

### *Flow-Volume Loops*

During the maximal exercise test, the speed at which you breathe air in and out and the volume of air you breathe will be measured. Approximately once every 60 seconds during the exercise test, you will be prompted to breathe in completely, filling your lungs with air, and then return to normal breathing. Before and after the exercise test, while at rest, you will be prompted to perform 3 breathing maneuvers, where you complete a big breath in (filling your lungs completely with air) followed by a forceful breath out (breathe out all the air you can). The investigators will coach you through these maneuvers. These procedures are performed during the maximal exercise test, and add no additional time.

### *Visit 2-3:*

\_\_\_\_\_ **initial** **Supplementation Procedure:** Following the first visit, you will be given beetroot juice or tomato juice. When taking beetroot juice, you will consume two 70 mL bottles of beetroot juice twice a day, once in the morning and once in the evening, for a total of 140 mL per day. When taking tomato juice, you will consume two 70 mL bottles twice a day, once in the morning and once in the evening, for a total of 140 mL per day. The order in which you consume the beverages will be randomly decided.

\_\_\_\_\_ **initial** **Nutrition Recall:** You will be asked to record all foods, supplements, and medications you consume during the 10-day supplementation procedure. You will be asked to keep your diet as similar as possible during the two different supplementation procedures.

\_\_\_\_\_ **initial** **Food Avoidances:** You will be given a list of foods not eat during the 10 day beetroot juice period or during the 10 day tomato juice period.

\_\_\_\_\_ **initial** **Cessation of Mouthwash Use:** You may not use mouthwash during this study.

\_\_\_\_\_ **initial** **Resting Seated Hyperventilation:** While seated, but not exercising, on a cycle ergometer, you will begin by quietly breathing for 6 minutes. Following this you will be asked to breath at ventilation rates corresponding to 50%, 70%, and 90% of your  $VO_{2peak}$  for 5 minutes each. The order of ventilation rates will be randomly determined. You will be breathing in gas that contains the addition of  $CO_2$  in order to prevent you from feeling dizzy. Additionally, the air will be moistened to minimize throat dryness. You will be given live feedback on a computer screen on the amount of air that you need to breath in and a metronome will signal how often you need to breath. At the end of each ventilation rate, you will be asked to quantify your level of breathlessness. You will be given 5 minutes of rest between each ventilation rate. A small finger cuff will be placed on one of your fingers which will measure how much blood is pumped by your heart. The measurements will take place right before the test at rest, and during the last minute of each hyperventilation phase. This will take about an hour.

\_\_\_\_\_ **initial** **Moderate Intensity Cycling:** You will begin by sitting quietly on the cycle ergometer for 5 minutes. Then you will warm up by cycling with the cycle ergometer unloaded (no



## Appendix C: Informed Consent Statement continued

resistance) for 3 minutes. You will then be asked to cycle for 6-minutes at a predetermined work rate. Entering into the last minute, you will be asked to rate you perceived breathlessness, unpleasantness due to breathlessness, and exertion. A small finger cuff will be placed on one of your fingers which will measure how much blood is pumped by your heart. The measurements will be taken right before the test at rest, and during the last minute of cycling. This will take about 30 minutes.

### *Mouth Pressure*

A mouthpiece with a small air hole in it will be placed into your mouth before hyperventilation, immediately after your third round of hyperventilation, and immediately before the onset of the cycling protocol. You will be asked to inhale as much as air as possible. You will be asked to do this at least 3 times at each setting (at least nine times total on visits 2 and 3). Each attempt will be separated by 1 minute. You will also be asked to exhale as much air as possible. You will be asked to do this at least 3 times at each setting (at least nine times total on visits 2 and 3). Each attempt will be separated by 1 minute.

### *Resting Blood Pressure*

Upon arriving for visits 2 and 3, your resting blood pressure will once again be taken. If it is above 140/90, you will not be allowed to exercise on that day. You will be asked to come back another day.

### *Duration of participation*

The shortest duration possible to complete all three visits is 31 days. Visit 1 will occur on day one. You will then consume one of the beverages for 10 days. On the tenth day, you will come for visit 2. You will then undergo at least a 10 day washout period in which you consume neither beverage. Finally, you will consume the other beverage for 10 days. On the tenth day, you will come for visit 3. The duration may be longer than 31 days if there are scheduling conflicts or if your resting blood pressure is above 140/90 on visits 2 and/or 3.

### **3. Discomforts and risks:**

The risks involved with measuring/monitoring/performing: questionnaires, physical characteristics, lung function testing, flow-volume loops, and breathlessness during exercise.

Body Composition: The risks involved are experiencing slight discomfort due to the small space.

Maximal Exercise Test: The risks involved are abnormal heart beats, abnormal blood pressure responses, muscle cramps, muscle strain and/or joint injury, delayed muscle soreness (1 to 2 days afterwards), light headedness, fatigue, and in rare instances, heart attack.

Beetroot Juice Consumption: The risks involved with consuming beetroot juice may cause urine to be pink.

Tomato Juice Supplementation: There are no known risks in consuming 140 mL/day of tomato juice.

Resting Seated Hyperventilation: The risks involved are that you may become light headed or feel out of breath at higher ventilation rates.

## Appendix C: Informed Consent Statement continued

Loss of Confidentiality: Any time information is collected; there is a potential risk for loss of confidentiality. Every effort will be made to keep your information confidential; however, this cannot be guaranteed.

Other Risks: There may possibly be other side effects that are unknown at this time. If you are concerned about other, unknown side effects, please discuss this with the researchers.

**How you can help reduce some of the risks:** During your participation in this research, the researchers will closely observe your testing to determine whether there are problems that need medical care. It is your responsibility to do the following:

- Ask questions about anything you do not understand.
- Keep appointments.
- Follow the study researchers' instructions.
- Let the researchers know if your telephone number changes.
- Tell the researchers before you take any new medication.
- Tell your regular doctor about your participation in this research.

4. **a. Benefits to you:** You can expect to receive information about your cardiovascular conditioning, and physical fitness. We are performing these tests for research purposes only. The information collected is not intended for diagnostic or therapeutic purposes. Under no circumstance will the investigator or research staff interpret results as normal or abnormal. We are unable to make any medical comments about your results. The results will not be looked at for medical diagnostic or medical treatment purposes.

**b. Potential benefits to society:** The knowledge gained from this study will determine if one of the beverages being tested has the potential to lower the oxygen cost of breathing in obese, young males.

5. **Alternative procedures that could be utilized:** Not participating in the study. The procedures and drinks used in this study are frequently used in research and are the most appropriate methods to accomplish the goals of this research.

### 6. **Time duration of the procedures and study:**

You will need to visit the Leon Levine Hall of Health Sciences (1179 State Farm Road) for the following:

\_\_\_\_\_ initial Visit 1 (about 1.5 - 2 hr)

\_\_\_\_\_ initial Visit 2 (about 1.5 hr).

\_\_\_\_\_ initial Visit 3 (about 1.5 hr).

*Approximately 5.0 hours*

## Appendix C: Informed Consent Statement continued

7. **Statement of confidentiality:** Study participants are coded by an identification number for statistical analyses. All records are kept in a secure location. All records associated with your participation in the study will be subject to the university confidentiality standards and in the event of any publication resulting from the research no personally identifiable information will be disclosed. The Office of Human Research Protections in the U.S. Department of Health and Human Services, the U.S. Food and Drug Administration (FDA), the Office for Research Protections at Appalachian State University and the Institutional Review Board may review records related to this project.
8. **Right to ask questions:** Please contact Dalton Fletcher, B.S. ([fletcherds@appstate.edu](mailto:fletcherds@appstate.edu), c. 336 – 247 -1887) or Jonathon Stickford, Ph.D. ([stickfordjl@appstate.edu](mailto:stickfordjl@appstate.edu) 828-262-7471), with questions, complaints, or concerns about this research. If you have any questions about your rights as a research subject, please contact the IRB Administrator at the Appalachian State University Institutional Review Board Office at (828) 262-2692, [irb@appstate.edu](mailto:irb@appstate.edu). This study has been approved on 2/23/2019 by the Institutional Review Board (IRB) at Appalachian State University. This approval will expire on 10/11/2019 unless the IRB renews the approval of this research.
9. **Compensation:** You will receive a total of \$30 upon completion of this study:  
**Compensation Breakdown:** You will be compensated \$10 for each visit completed to be received following you last visit.

You may be asked to repeat a trial. If you agree to repeat a trial, you will be paid for the repeated trial as stated above.

10. **Injury Clause:** In the unlikely event you become injured as a result of your participation in this study, standard emergency procedures will be followed. If you get hurt or sick when you are not at the research site, you should call your doctor or call 911 in an emergency. If your illness or injury could be related to the research, tell the doctors or emergency room staff about the research study, the name of the Principal Investigator, and provide a copy of this consent form if possible. Please contact the PI as soon as possible (Dalton Fletcher B.S. [fletcherds@appstate.edu](mailto:fletcherds@appstate.edu), c. 336-247-1887) or Jonathon Stickford, Ph.D. ([stickfordjl@appstate.edu](mailto:stickfordjl@appstate.edu), 828-262-7471). It is the policy of this institution to provide neither financial compensation nor free medical treatment for research-related injury. You will be responsible for any costs for medical care not paid by your insurance company. No other compensation is offered by Appalachian State University. By signing this document, you are not waiving any legal rights that you have against Appalachian State University for injury resulting from negligence of the University or its investigators.
11. **Voluntary participation:** Your participation in this study is voluntary. You may withdraw from this study at any time by informing the research personnel. You may decline to answer certain questions and may decide not to comply with certain procedures. However, your being in the study may be contingent upon answering these questions or complying with the procedures. The researcher may end your role in the study without your consent if the researcher deems that your health or behavior adversely affects the study or increases risks to you beyond those approved by the Institutional Review Board and agreed upon by you in this document. You have been given an opportunity to ask any questions you may have, and all such questions or inquiries have been answered to your satisfaction.

**Appendix C: Informed Consent Statement continued**

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Volunteer Date

I, the undersigned, have defined and explained the studies involved to the above volunteer.

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Person Obtaining Consent Date

Appendix D: Telephone Screening Form

**Initial Telephone Screening Form for Oxygen Cost of Breathing Study**

**General Information:**

Name:

\_\_\_\_\_

Email:

\_\_\_\_\_

Address (if no email):

Age: \_\_\_\_\_ Sex: \_\_\_\_\_ Phone: \_\_\_\_\_

Height: \_\_\_\_\_ Weight: \_\_\_\_\_ Allergies (latex?)

**Exclusion Criteria: any question answered “yes” in this section will disqualify the potential subject.**

- |    | <b>Yes</b>               | <b>No</b>                |   |
|----|--------------------------|--------------------------|---|
| 1. | <input type="checkbox"/> | <input type="checkbox"/> | Age – outside the ages of 18 and 40 yr?   |
| 2. | <input type="checkbox"/> | <input type="checkbox"/> | Do you currently smoke tobacco cigarettes? Any history of smoking? If so, how much did you smoke and for how long? (Must be less than 0.5 pack-yrs) |
| 3. | <input type="checkbox"/> | <input type="checkbox"/> | Have you ever been diagnosed with a sleep disorder or use CPAP?   |
| 4. | <input type="checkbox"/> | <input type="checkbox"/> | Do you have a history of asthma, COPD, or any lung issues?  |
| 5. | <input type="checkbox"/> | <input type="checkbox"/> | Do you have a history of an irregular heartbeat or any heart condition? (Have you had an EKG performed?)  |
| 6. | <input type="checkbox"/> | <input type="checkbox"/> | Do you have any known metabolic or renal diseases?  |
| 7. |                          |                          | Do you have any known health conditions?  |
|    | <input type="checkbox"/> | <input type="checkbox"/> | High blood pressure?  |
|    | <input type="checkbox"/> | <input type="checkbox"/> | Diabetes?   |
|    | <input type="checkbox"/> | <input type="checkbox"/> | Thyroid issues?   |

**Appendix D: Telephone Screening Form continued**

8.   Are you currently engaging in an organized exercise program? ( $\geq 90$  min/wk of organized activity)

**Inclusion Criteria: any question answered “yes” in this section will qualify the potential subject**

**Yes No**

1.   Do you have a body mass index  $\geq 30$  kg/m<sup>2</sup>, but  $\leq 40$  kg/m<sup>2</sup>?

## Appendix E: Medical History Form

<b>Appalachian State University – Integrative Human Physiology Laboratories</b>		
1179 State Farm Road, Boone, NC 28607 Phone: (828) 262-7471		
<b>ASU</b>	<b>Medical History Form</b>	<b>Page 1</b>
<b>Subject ID#:</b>	<b>Study:</b>	
Highest Education Achieved:		
Ethnicity:		
<input type="checkbox"/> <b>Hispanic or Latino.</b> A person of Cuban, Mexican, Puerto Rican, South or Central American, or other Spanish culture or origin, regardless of race. The term "Spanish origin" can be used in addition to "Hispanic or Latino."		
<input type="checkbox"/> <b>Not Hispanic or Latino.</b>		
Race: What race do you consider yourself to be?		
<input type="checkbox"/> <b>American Indian or Alaska Native.</b> A person having origins in any of the original peoples of North, South, or Central America, and who maintains a tribal affiliation or community attachment.		
<input type="checkbox"/> <b>Asian.</b> A person having origins in any of the original peoples of the Far East, Southeast Asia, or the Indian subcontinent, including, for example, Cambodia, China, India, Japan, Korea, Malaysia, Pakistan, the Philippine Islands, Thailand, and Vietnam. (Note: Individuals from the <del>Philippine</del> <span style="color: red;">Cebu</span> Islands have been recorded as Pacific Islanders in previous data collection strategies.)		
<input type="checkbox"/> <b>Black or African American.</b> A person having origins in any of the black racial groups of Africa. Terms such as "Haitian" or "Negro" can be used in addition to "Black" or "African American".		
<input type="checkbox"/> <b>Native Hawaiian or Pacific Islander.</b> A person having origins in any of the original peoples of Hawaii, Guam, Samoa, or other Pacific islands.		
<input type="checkbox"/> <b>White.</b> A person having origins in any of the original peoples of Europe, the Middle East, or North Africa.		
<input type="checkbox"/> <b>Check here if you do not wish to disclose any or all of the above information.</b>		
<b>Medications:</b> include over the counter drugs/oral contraceptives/dietary supplements		
Name/Dosage/How often taken:		
<b>Allergies:</b>		
<b>Smoking History:</b>		
Do you smoke? Yes No Cigarettes? Pipe / Cigar? Other? If you quit, what year did you quit _____		
_____ # packs per day for _____ # of years What year did you start smoking? _____		
Have you ever been exposed to second hand smoke? _____ Home _____ Work _____ Other _____ Years		

## Appendix E: Medical History Form continued

ASU		Medical History Form	Page 2
<b>Medical History:</b>			
<b>NO</b>	<b>YES</b>	<b>Please explain any "YES" answers below:</b>	
		high blood pressure	
		swelling	
		chest pain / history of heart attack	
		extra heart beats, racing or fluttering	
		abnormal electrocardiogram (ECG)	
		other heart trouble (e.g. murmur, valve problems)	
		high cholesterol	
		diabetes (e.g. frequent urination and abnormal thirst)	
		seizures	
		stroke	
		fainting or black-out spells, dizziness	
		anxiety (diagnosed)	
		depression (diagnosed)	
		recurrent fatigue (e.g. feeling tired or extreme lack of energy)	
		insomnia or poor sleeping	
		thyroid problems	
		difficulty breathing	
		emphysema/ asthma/ chronic bronchitis	
		cough, sputum (phlegm)	
		tuberculosis	
		chronic infection	
		stomach/GI problems (e.g. heart burn, nausea, vomiting, diarrhea, constipation, abdominal pain, gas pain, black stools, blood in stools)	
		hepatitis	
		bleeding disorder (e.g. bleeding or bruising easily)	
		kidney/ urinary problems (e.g. frequent urination, burning when urinating, urine changing in color)	
		joint injuries/ joint pain, back pain, or leg pain	
		arthritis (rheumatoid or osteoarthritis)	
		hearing problems (e.g. impaired hearing or ringing in the ears)	
		migraine headaches	
		vision problems (exclude corrected near/far sightedness)	
		surgical procedures (e.g. <del>c-sections</del> , appendectomy, augmentations, knee and back surgeries, tonsillectomy, etc)	
<b>Additional Notes:</b>			



## Appendix E: Medical History Form continued

ASU	Medical History Form	Page 3
<b>Exercise History:</b>		
Do you currently exercise aerobically?	How many years?	Duration:
	Types of Exercise:	Frequency:
Do you compete in endurance events?	How many years?	Frequency:
	What events?	Athlete in college? Yes No
Any other types of exercise?	How many years?	Duration:
	Types of Exercise:	Frequency:
If you are currently sedentary, when did you last exercise?	How many years?	Duration:
	Types of Exercise:	Frequency:
<b>Weight History:</b>		
If overweight, how long have you been overweight?	Were you overweight as a child?	By how much?
	How many times has your weight changed?	
Any events that led up to your obesity? (E.g., injury) Yes No		If yes, how many events? 1 2 3 4 5 >5
<b>Sleep History:</b>		
Have you ever been diagnosed with a sleep disorder?	Yes	No
Do you use CPAP/BIPAP at night?	Yes	No
Do you snore at night?	Yes	No
Has someone ever told you that you snore at night?	Yes	No
Do you have daytime sleepiness?	Yes	No
<b>Authorization to Release Information - Please check all that applies and sign/date.</b>		
<input type="checkbox"/>	I authorize Appalachian State University to collect and save the above protected health information on me for purposes of research. I understand that all information is private and confidential.	
<input type="checkbox"/>	I authorize Appalachian State University to keep this information and any information gained from my participation in their studies in a database so that they may contact me.	
<input type="checkbox"/>	The above information is correct and complete to the best of my knowledge.	
<b>Signature</b>		<b>Date</b>

## Appendix F: 24-Hour Health History Form

### 24-HOUR HEALTH HISTORY

Study: \_\_\_\_\_ Age: \_\_\_\_\_ Height: \_\_\_\_\_ Weight: \_\_\_\_\_  
 Subject Number: \_\_\_\_\_ Date: \_\_\_\_\_

Do you have: Head cold <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> Nasal Congestion <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> Headache <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> Sore Throat <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> Digestive Upset <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> Intestinal Disorder <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> General Fatigue <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/> Muscle Soreness <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/>	How do you feel? Good <input type="checkbox"/> Fair <input type="checkbox"/> Not so good <input type="checkbox"/> Bad <input type="checkbox"/>	# of hours sleep _____ How was your sleep? Normal <input type="checkbox"/> Wakeful <input type="checkbox"/> Restless <input type="checkbox"/>	# of hours since eating: _____ What did you eat? _____ _____ _____ _____
Medicine taken in last 24 hours: _____ _____ _____	Physical activity in last 24 hours: _____ _____ _____		Any unusual physical activity in last 24 hours? _____ _____ _____
Any leg cramps Since last activity? Yes <input type="checkbox"/> No <input type="checkbox"/> Yes <input type="checkbox"/> No <input type="checkbox"/>			

\*\* Take weight with each visit.

## Appendix G: Nutrition Log

# Food Recording Form

Subject number \_\_\_\_\_ Age \_\_\_\_\_ Date \_\_\_\_\_  
(mm/dd/yyyy)

### Directions for Using the Food Diary

1. Please record ALL food, beverage, medications and supplements eaten/ingested for the three days that are assigned for you. To reduce error, please follow these directions:
  - a. Keep your food diary current (list foods immediately after they are eaten).
  - b. Whenever possible, MEASURE the volume consumed by using cups and tablespoons. Record amounts in household measures---ounces, tablespoons, cups, and slices.
  - c. Be specific when describing the food item eaten, and the method that was used to prepare the food. Remember to include condiments, sugar, oils, butter, and other visible fats. For example:

<i>Apple, raw, fresh, with peel</i>	<i>1 medium</i>
<i>Bread, whole wheat, fresh</i>	<i>2 slices</i>
<i>Margarine, soft from tub</i>	<i>1 tablespoon</i>
<i>Tylenol</i>	<i>300 milligrams</i>
<i>Sugar, white</i>	<i>1 teaspoon</i>
<i>Milk, non fat</i>	<i>2 cups</i>
<i>Fish, salmon, baked</i>	<i>10 ounces</i>
  - d. This food record will be analyzed with a computerized dietary analysis program, so please provide sufficient detail.

Appendix G: Nutrition Log continued

*Day One*

<b>Time</b> <b>Food/Beverage/Medication</b> <b>Consumed</b>	<b>Food Item and</b> <b>Method of</b> <b>Preparation</b>	<b>Amount</b> <b>Consumed</b> (in cups, tablespoons, ounces, milligrams, etc.)

## Appendix H: Foods to Avoid

### Foods to Avoid Eating (High Nitrate Levels)

- Celery
- Chervil
- Cress
- Lamb's Lettuce
- Lettuce
- Radish
- Red beetroot
- Rocket
- Spinach
- Swiss chard
- Celeriac
- Chinese Cabbage
- Endive
- Escarola
- Fennel
- Kohlrabi
- Leaf chicory
- Leek
- Parsley

**Appendix I: Ratings of Perceived Breathlessness Scale**

Rate Your Breathing

**0 Nothing at all**

**0.5 Very, very weak (Just Noticeable)**

**1 Very Weak**

**2 Weak (Light)**

**3 Moderate**

**4 Somewhat Strong**

**5 Strong (Heavy)**

**6**

**7 Very Strong**

**8**

**9**

**10 Very, very strong (Almost max)**

## **Appendix J: Ratings of Perceived Breathlessness Description**

**This is a scale for rating:**

### **BREATHLESSNESS**

**The number 0 represents no breathlessness. The number 10 represents the strongest or greatest breathlessness that you have ever experienced. Each minute during the exercise test you will be asked to point to a number, with your finger, which represents your perceived level of breathlessness at the time. The number will be repeated out loud in order to confirm your choice. During the exercise test you may have an even stronger or greater intensity of breathlessness than you have previously experienced. You should then point to the word “maximal” if the severity is greater than 10. You can tell us this number after the mouthpiece has been removed.**

**Appendix K: Ratings of Perceived Unpleasantness Scale**

Rate Unpleasantness of your Breathing

**0 Not Unpleasant**

**0.5 Very, very weak (Just Noticeable)**

**1 Very Weak Unpleasantness**

**2 Weak (Light) Unpleasantness**

**3 Moderate Unpleasantness**

**4 Somewhat Strong Unpleasantness**

**5 Strong (Heavy) Unpleasantness**

**6**

**7 Very Unpleasant**

**8**

**9**

**10 Maximal Imaginable Unpleasantness**



## **Appendix L: Ratings of Perceived Unpleasantness Description**

**This is a scale for rating:**

### **UNPLEASANTNESS OF BREATHLESSNESS**

**Unpleasantness expresses the affective evaluation of the sensation regardless of whether the intensity is high or low. Unpleasantness describes how much you affectively dislike something or feel terrified by it. A low unpleasantness indicates that the perceived breathlessness does not feel bad. A high unpleasantness signifies that the breathlessness feels very bad or terrifying regardless of whether the intensity of the sensation is high or low.**

**During the exercise test you will be asked to point to a number, with your finger, which represents your perceived level of the unpleasantness of breathlessness at the time. The number will be repeated out loud in order to confirm your choice.**

## Appendix M: Ratings of Perceived Exertion Scale

Rate Your Exercise

**6**

**7 Very, very light**

**8**

**9 Very light**

**10**

**11 Fairly light**

**12**

**13 Somewhat hard**

**14**

**15 Hard**

**16**

**17 Very hard**

**18**

**Appendix M: Ratings of Perceived Exertion continued**

**19 Very, very hard**

**20**

## **Appendix N: Ratings of Perceived Exertion Description**

**This is a scale for rating:**

### **PERCIEVED EXERTION**

**During the graded exercise test we want you to pay close attention to how hard the work rate is for you. The feeling should be your total amount of exertion and fatigue, combining all sensations and feelings of physical stress, effort and fatigue. Don't concern yourself with any one factor such as leg pain, shortness of breath or exercise intensity, but try to concentrate on your total, inner feeling of exertion. Don't underestimate or overestimate, just be as accurate as possible.**

## **Vita**

Dalton Scott Fletcher was born on November 3<sup>rd</sup>, 1994 to Scott and Lynn Fletcher in Lexington, North Carolina. He graduated from Central Davidson High School in 2013. He attended Appalachian State and received a bachelor's degree in exercise science with a minor in psychology in 2013. He immediately returned to Appalachian State to receive his master's degree in exercise with a focus in research in 2019.

Dalton is plans to obtain a Ph.D. in the future and will be applying to schools in the upcoming year. Until then, he hopes to work as clinical exercise physiologist.