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The Effects of Fitness Level and Sex on EPOC Following High Intensity Interval and Moderate Intensity Aerobic Exercise

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THE EFFECTS OF FITNESS LEVEL AND SEX ON EPOC FOLLOWING HIGH INTENSITY INTERVAL AND MODERATE INTENSITY AEROBIC EXERCISE

By

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ABSTRACT

THE EFFECTS OF FITNESS LEVEL AND SEX ON EPOC FOLLOWING HIGH INTENSITY INTERVAL AND MODERATE INTENSITY AEROBIC EXERCISE

Rachel Lauren Simmons
Old Dominion University, 2016
Chair: Dr. David P. Swain

In the past few years, much attention has been directed toward shorter-duration vigorous intensity aerobic exercise as opposed to longer-duration moderate intensity exercise. There is conflicting evidence as to whether vigorous exercise can result in a greater excess post-exercise oxygen consumption (EPOC) and thus more calories burned when compared to moderate continuous exercise performed for a longer duration. In addition, the literature contains a dearth of information isolating and investigating male and female response to these exercises, as well as fit and unfit subjects. The purpose of this study was to quantify EPOC following moderate intensity aerobic exercise (MOD) and high intensity aerobic intervals (HIAI) of equal energy expenditure. It was hypothesized that (1) EPOC following HIAI would be greater than following MOD, (2) EPOC of males and females would not differ, and (3) more fit subjects would have less of an increase in EPOC from MOD to HIAI than less fit subjects, predicted as a negative correlation between subjects' VO_{2max} and delta EPOC. Eleven subjects (5 male, 6 female; age 25.1 ± 2.0 yr; height 169 ± 3 cm; mass 67.6 ± 3.0 kg; VO_{2max} 41.0 ± 1.9 $mL \cdot min^{-1} \cdot kg^{-1}$) participated in MOD and HIAI trials in a counterbalanced order separated by at least 48 hours. HIAI was ten 1-min intervals at 90% maximal aerobic power (P_{max}), alternated with 1-min intervals at 60% P_{max}. MOD was 30 min at 50% P_{max}. Warm-ups and cool-downs were also matched for total work. All exercise was performed on a cycle

ergometer. Data are reported as mean $\pm$ SE. VO_2 at rest was 4.0 $\pm$ 0.3 $mL \cdot min^{-1} \cdot kg^{-1}$ preceding the MOD trial, and 4.1 $\pm$ 0.4 $mL \cdot min^{-1} \cdot kg^{-1}$ for HIAI. Net VO_2 during exercise (inclusive of warm-ups and cool-downs) was 52.7 $\pm$ 3.6 L MOD, and 52.0 $\pm$ 3.9 L HIAI. There was a weak trend ($p = 0.164$) for EPOC over 41 min of recovery to be greater in the HIAI trial (3.02 $\pm$ 0.48 L) than the MOD trial (1.98 $\pm$ 0.56 L). EPOC during minutes 0-10 post-exercise was significantly greater ($p = 0.009$) following HIAI (2.28 $\pm$ 0.32 L) than MOD (1.37 $\pm$ 0.43 L). Following a one-min water break, EPOC during minutes 11-41 post-exercise was 0.74 $\pm$ 0.28 L HIAI, and 0.56 $\pm$ 0.43 L MOD, which were statistically similar ($p = 0.807$). For EPOC expressed relative to body mass, males had a significantly greater ($p = 0.035$) EPOC over the entire 41-min recovery period than females (HIAI males: 58.0 $\pm$ 8.7, females: 32.5 $\pm$ 4.9 $mL \cdot kg^{-1}$; MOD males: 44.9 $\pm$ 14.0, females: 18.7 $\pm$ 10.6 $mL \cdot kg^{-1}$). For the separate time periods of 0-10 min and 11-41 min, there were trends for males to have higher EPOC than females ($p = 0.086$ and 0.053, respectively). When total EPOC was corrected for fat free mass there was no significant difference between male and female responses to either condition ($p = 0.162$).There was no correlation between VO_{2max} and absolute ΔEPOC (L), whether using the total time frame (0-41 min, $p = 0.958$), or the early (0-10 min, $p = 0.958$) or late (11-41 min, $p = 0.281$) phases of EPOC. In conclusion, EPOC consisted of only 3.7% of the net oxygen consumption during the MOD exercise trial, and only 5.8% of the net oxygen consumption during the HIAI trial. While the HIAI EPOC was significantly greater than the MOD EPOC, neither EPOC contributes substantially to the overall energy cost of the exercise, and is thus unlikely to have significant effects on weight loss or other health outcomes. When prescribing exercise to those who are less fit or are new to exercising, it is important to consider the intensity and time that is most tolerable when the primary goal is weight loss.

This thesis is dedicated to my grandfather Lewis Harris Bridges and my grandmother Nancy Bell Bridges as evidence of ‘holding onto my future’. I love you both so very much.

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TABLE OF CONTENTS

LIST OF TABLES ix

LIST OF FIGURES x

Chapter

I. INTRODUCTION 1
- STATEMENT OF THE PROBLEM 2
- STATEMENT OF PURPOSE 3
- SIGNIFICANCE OF THE STUDY 3
- HYPOTHESES 3
- INDEPENDENT VARIABLE 4
- DEPENDENT VARIABLES 4
- DELIMITATIONS 4
- LIMITATIONS 5
- OPERATIONAL DEFINITIONS 5

II. LITERATURE REVIEW 6
- SCIENTIFIC BACKGROUND 6
- METHODOLOGICAL CONSIDERATIONS 8
- EPOC STUDIES OF MORE THAN ONE INTENSITY 9
- EQUATED WORKLOAD STUDIES 12
- EPOC AND FITNESS LEVEL 16

III. METHODOLOGY 22
- RESEARCH DESIGN 22
- SAMPLE 22
- SUBJECT RECRUITMENT 23
- MAXIMAL EXERCISE TEST 23
- EXERCISE TRIALS 24
- EPOC MEASUREMENT 24
- STATISTICAL ANALYSIS 25

IV. RESULTS 26
- ANTHROPOMETRIC VARIABLES 26
- EPOC RESPONSE: MOD VS. HIAI 26
- EPOC RESPONSE: MALES VS. FEMALES 29
- EPOC RELATIONSHIP TO VO2MAX 31

V. DISCUSSION 33
- EPOC RESPONSE: MOD VS. HIAI 32
- EPOC RESPONSE: MALES VS. FEMALES 33
- EPOC RELATIONSHIP TO VO2MAX 36
- CONCLUSIONS 36

APPENDICES 38

A. SCREENING QUESTIONNAIRE 38
B. INFORMED CONSENT DOCUMENT 39
C. MENSTRUAL CYCLE PHASE QUESTIONNAIRE 43

REFERENCES 44

VITA 48

LIST OF TABLES

Table

1. EPOC quantity following different intensities and durations of aerobic exercise. 18

2. Subject characteristics 26

3. Summary of studies including only males, comparing AEUs to EPOC 34

LIST OF FIGURES

Figure

1. Absolute EPOC minutes 0-41 following MOD and HIAI conditions 27

2. Absolute EPOC minutes 0-10 following MOD and HIAI conditions 28

3. Absolute EPOC minutes 11-41 following MOD and HIAI conditions 28

4. EPOC minutes 1-41, male vs. female response to MOD and HIAI conditions.....29

5. EPOC minutes 0-10, male vs. female response to MOD and HIAI conditions.....30

6. EPOC minutes 11-41, male vs. female response to MOD and HIAI conditions...30

7. ΔEPOC minutes 1-10 vs. VO_{2max}..31

CHAPTER I

INTRODUCTION

In the past few years, much attention has been directed toward shorter-duration vigorous aerobic exercise (VIG; 60-89% oxygen consumption reserve, VO_2R) as opposed to longer-duration moderate intensity exercise (MOD; 40-59% VO_2R). Some authors claim that VIG can result in more calories burned *after* the exercise when compared to MOD exercise performed for a longer duration, creating a potential for greater fat loss (Boutcher, 2011; LaForgia, 1997). The elevated level of metabolism and caloric expenditure after cessation of exercise is known as excess post-exercise oxygen consumption (EPOC). Levels of EPOC can vary depending on the type, intensity, and duration of the activity. EPOC seems to be affected the most by increases in intensity. Several studies have demonstrated a greater level of EPOC following VIG than MOD, with workload controlled in some (LaForgia et al., 1997; Larsen et al., 2014; Phelain et al., 1997) but not all studies (Knuttgen et al., 1970; Mann et al., 2014). Two studies, however, have found no significant difference in EPOC after VIG versus MOD when workload was controlled (McGarvey et al., 2005; Sedlock et al., 1991). One possible reason for this discrepancy is the variation in fitness levels across subjects. Aerobic fitness seems to correlate inversely with EPOC (Matsuo et al., 2012). In fact, just twelve weeks of endurance training reduced EPOC following exercise at a given absolute intensity, though it remained unchanged at the same relative intensity (Sedlock et al., 2010). Previously, Sedlock et al. (1994) found that when performing a weight-supported exercise, there was no difference between EPOCs of fit versus unfit subjects. Unfortunately, there were only five males in each group. The majority of EPOC studies use twelve or fewer subjects (Gore & Withers, 1990; Knuttgen et al., 1970; LaForgia et al., 1997; Larsen et al., 1997; Matsuo et al., 2012; McGarvey et al., 2005; Phelain et al., 1997; Sedlock et al., 1991; Sedlock et al., 1994). Of these studies, only two consisted of both male and female subjects (Knuttgen et al., 1970; Phelain et al., 1997). With inconsistencies in the literature regarding EPOC magnitude in VIG versus MOD conditions, fit versus unfit subjects, and the lack of inclusion of female subjects, further research is needed to understand EPOC at different intensities.

Statement of the Problem

Some studies comparing various intensities did not control for the amount of work performed in each session, making it difficult to compare EPOC values (Gore & Withers, 1990; Knuttgen et al., 1970; Mann et al., 2014). Other studies equated workloads between each condition, but did not address the potential effect of the subjects' fitness level (LaForgia et al., 1997; Larsen et al., 2014; McGarvey et al., 2005; Phelain et al., 1997; Sedlock et al., 1991). With the fact that there were differences found in EPOC between fit and unfit subjects, it is essential that workload be prescribed on a basis of relative intensity when comparing EPOCs between different intensities with the same workload (Sedlock et al., 2010; Matsuo et al., 2012). As later described in the literature review, EPOC following exercise seems to increase exponentially as intensity enters the vigorous range. All of the studies discussed in the first two sections of the literature review utilized subjects with a wide range of fitness levels from $\pm$ 1.5 to $\pm$ 9.3 $mL \cdot min^{-1} \cdot kg^{-1}$ SD, making a comparison between fitness levels essential to providing accurate EPOC data.

Statement of Purpose

The purpose of this study was to quantify EPOC following different intensities of cycling exercise, and to compare these values between fit and unfit subjects, and between men and women. All subjects performed a 20-minute vigorous high intensity aerobic interval exercise (HIAI) with one minute at 90% of maximal aerobic power (P_{max}) followed by one minute at 60% of P_{max}, and a moderate exercise bout at 50% P_{max} (MOD). Both exercise conditions consisted of equal workloads.

Significance of the Study

To our knowledge, only three studies have compared EPOC in fit and unfit subjects. This study allows us to determine a correlation between EPOC magnitude with fitness level and sex. Our study included females, adding to the body of literature on EPOC that currently lacks many female subjects. We are also able to find any differences between fit and unfit subjects with regard to how EPOC responds to various intensities.

Hypotheses

H1: There is an intensity effect on EPOC. The absolute EPOC (L) following a bout of high-intensity aerobic intervals will be greater than the absolute EPOC following a bout of continuous moderate-intensity aerobic exercise of equal work. Also, the relative EPOC (EPOC in L divided by VO_2 in L during the exercise) following a bout of high-intensity aerobic intervals will be greater than the relative EPOC following a bout of continuous moderate-intensity aerobic exercise of equal work.

H2: The intensity effect will be greater in less aerobically fit subjects. The increase in absolute EPOC from MOD to HIAI will be inversely correlated with VO_{2max}. Also, the increase in relative EPOC from MOD to HIAI will be inversely correlated with VO_{2max}.

H3: There will be no difference in EPOC responses between men and women. The relative EPOC following MOD in men will not be different from that in women, and the relative EPOC following HIAI in men will not be different from that in women.

Independent Variable Subjects will exercise at the two prescribed intensities in a randomized, counterbalanced order. MOD exercise will be 30 minutes at 50% of each subject's P_{max}. HIAI exercise will consist of ten 1-min intervals at 90% P_{max} (each work interval followed by one minute of 60% P_{max}).

Dependent Variables

EPOC magnitude is measured in L of oxygen consumption immediately following the bout of exercise for a total of three hours (total VO_2 minus pre-exercise resting VO_2). RER is also measured as VO_2/VCO_2 during and after exercise.

Delimitations

Our subject population consisted of healthy subjects, ages 18-45 years. We excluded subjects who were at moderate or high risk for cardiovascular problems during exercise, according to American College of Sports Medicine guidelines (ACSM, 2010), i.e., with two or more cardiovascular risk factors (hypertension, smoking, age, family history, prediabetes, dyslipidemia, obesity, and sedentary behavior) or with known cardiopulmonary or metabolic disease, or symptoms of such.

Limitations

The subjects were instructed to record their diet 24 hours prior to the first exercise session, and were asked to repeat the exact diet prior to the next exercise testing session to prevent variations in the thermogenic effect of food. They were asked to consume no food for one hour prior to testing. Subjects were also asked to refrain from caffeine and alcohol consumption 24 hours prior to testing, and to refrain from vigorous exercise over the same time period. Female subjects were all tested within the follicular phase of the menstrual cycle as self-reported. While the subjects were asked about compliance prior to testing, we cannot know if they answered truthfully. However, measuring RMR prior to each exercise session allowed us to control the potential effects of diet, caffeine or alcohol intake, physical activity, and menstrual cycle phase.

Operational Definitions

VO_{2max}- Maximum value of VO_2 during the incremental cycle ergometer test where the subject could no longer continue or pedal cadence drops 10 rpm below the designated level. An RER $\geq$1.10 or a plateau in VO_2 will serve as confirmation of attainment of a true maximal value.

Maximal Aerobic Power (P_{max}): The highest power maintained for 3 minutes during the incremental exercise test, prorated for any portion of an uncompleted final stage.

Resting Metabolic Rate – Average VO_2 over the last 10 minutes of a 20-minute seated resting period. Considered as baseline resting oxygen consumption.

EPOC – Net VO_2 during post-exercise period: post-exercise total oxygen consumption minus baseline resting oxygen consumption. Total EPOC will be derived from area under the curve of VO_2 measurements after exercise minus resting level.

Menstrual Cycle Phase – All testing will be performed in the follicular phase beginning with the cessation of the period (day 1) until ovulation (day 14).

Aerobic Exercise Units (AEUs) – Time in minutes multiplied by % VO_{2max}.

CHAPTER II

LITERATURE REVIEW

The purpose of this literature review is to examine the effect of exercise duration, intensity and total work on EPOC, and to discuss conflicting literature in equated workload studies to arrive with a statement of the problem. The first section will discuss the early literature and basic concept of EPOC. The second section will address studies that examined EPOC following more than one intensity of exercise, with special attention on those studies that equated the workload between conditions. Table 1 is a compilation of the results of these studies.

Scientific Background

The discovery and subsequent attempt at a thorough scientific description of EPOC and its causes began in the early 1920s. In order to have a greater understanding of current literature, a history on EPOC and its progression is important to note. EPOC was first discovered by August Krogh and noted by A.V. Hill in 1923 and was termed "oxygen debt." Based on Hill's research with frog muscle contraction, it was believed to be a result of the combustion of lactate following exercise (A.V. Hill, 1923; Bassett, 2002). The exercise period necessitated glycogen breakdown, which formed lactic acid that 'needed to be cleared.' In 1933, however, Margaria, Edwards, and Dill noted that blood lactate decline during recovery was logarithmic in nature, whereas O_2 consumption during recovery was not, i.e., the decline in blood lactate was not as rapid as that in oxygen uptake (Margaria et al., 1933). When one observes the O_2 uptake immediately after the cessation of high intensity exercise, O_2 uptake exponentially decreases, whereas lactic acid declines more slowly. This finding compelled Margaria, Edwards, and Dill to separate the oxygen debt theory into two phases. The first, rapid decline in O_2 uptake was considered the alactacid phase in which O_2 consumption had nothing to do with blood lactate metabolism. The second, slow phase was termed the lactacid phase and was believed to be associated with the resynthesis of glycogen (Brooks et al., 1999, p. 204). Ole Bang challenged this theory by examining various durations of exercise (Bang et al., 1936). He demonstrated that blood lactate reached maximum levels at the ten-minute mark during exercise and began to decline regardless of the duration of exercise, and that oxygen debt after exercise still continued to exist in a predictable manner, regardless of the changes in lactate levels during exercise. This demonstrated that lactic acid could not be the sole cause of oxygen consumption after exercise (Brooks et al., 1999, p. 206). By the 1960s several differences in mammalian skeletal muscle and lactate metabolism were identified when compared to amphibious biology, thus the need arose to reassess the theory on 'oxygen debt' and its relationship to lactate metabolism. In mammals, a process known as the Cori cycle occurs in the liver in which lactate produced by skeletal muscle can be converted to glucose. In 1970, Barnard et al. pharmacologically blocked gluconeogenesis in the liver and determined that only 40% of the 'oxygen debt' could be attributed to the metabolic cost of lactate removal. This posed many questions for researchers to determine the source of the roughly 60% of recovery oxygen consumption not caused by lactate conversion, if the 'oxygen debt' is truly a result of lactate removal at all. These questions led to the research of Brooks et al. in 1980 who traced the pathways of [^{14}C]lactate using radiochromatograms of blood, liver, kidney, heart, and skeletal muscle tissue samples in [^{14}C]lactate-infused rats during rest, intermittent, and continuous exercise conditions (Brooks et al., 1980). It was then discovered that lactate is a major metabolic substrate, and the majority of lactate is oxidized. In fact, less than 20% of the total EPOC was due to resynthesis of glycogen. This became a major turning point in our understanding of the fates and functions of lactate: it does not cause the oxygen debt, rather it aids in the recovery process by serving as a metabolic substrate. Since then, oxygen debt is now referred to as EPOC as termed by Brooks. The true major causes of EPOC are currently understood to be from the need to replenish oxygen bound to hemoglobin and myoglobin (occurs in a few breaths following cessation of exercise), replenishing ATP and CP stores (replenished after a few minutes), recovering the work of the heart and diaphragm from exercise (10-20 minutes). Metabolism is still elevated due to increased body temperature, circulating catecholamines and other hormones (e.g., T_4, T_3) from exercise (30-60 minutes), and the resynthesis of glycogen (can take several hours to a day). In the mitochondria, the temperature increase causes a decrease in energy efficiency within the mitochondria, an increase in sympathetic nervous system activity, and a change in sodium, potassium, and calcium ion levels due to exercise and its effect on hormones (Brooks et al., 1999, p. 208-210).

Methodological Considerations

EPOC is most commonly measured using indirect open circuit calorimetry through expired gas analysis. All of the studies discussed in this literature review use this method to analyze resting metabolic rate (RMR), oxygen consumption (VO_2), and EPOC. While these are very common methods of measuring EPOC, there are several methodological issues to consider. Measurement of baseline RMR on a separate day accounts for diurnal variations and potential pre-exercise anticipation, but is not necessary when studies are not including feeding periods (LaForgia et al., 2005). Also, the thermic effect of food must be considered with meal timing before and after exercise. Several studies that separated fasting and fed control periods demonstrated that there was no significant difference in metabolic rate between fasted and fed states during non-exercise and exercise periods (Broeder et al., 1991; Willms et al., 1991; LaForgia et al., 2005). It is important to ensure subjects eat the same meal and the same times prior to each exercise bout when comparing within-subjects data. The circadian rhythmicity of hormones must also be considered when selecting measurement times for subjects. Cortisol plays a significant role in metabolism, and peaks in the morning hours (Hayes et al., 2010). It is therefore important to measure each subject at the same time of day to minimize these effects. When including female subjects, it is important to test during the same phase of the menstrual cycle to avoid intra-individual RMR variation that can be as large as 11.8% (Henry et al., 2003).

EPOC Studies of More Than One Intensity

Knuttgen et al. (1970) measured EPOC in 12 subjects (5 female) after cycling during various intensities and durations in three series (A, B, C). Subjects were between the ages of 17 and 26 years old and were not endurance trained. Subjects reported the same time each day in a fasted state. Subjects performed a 20-minute resting period and VO_2 was measured using Douglas bags. Subjects then pedaled at 60 rpm in all conditions. In series A, all subjects exercised for a duration of 15 minutes at an average of 50%, 70%, and 90% VO_{2max}, yielding approximate aerobic exercise units (AEU, duration in minutes times % VO_{2max}) of 7.5, 10.5, 13.5, respectively. In series B, the first seven subjects performed steady-state exercise at 60% of aerobic capacity for 15, 35, and 55 minutes (~AEU: 9, 21, 33). In series C, the other five subjects cycled at various intensities (55-83% aerobic capacity) for 15 and 55 minutes; the results were not clearly presented and will not be discussed. Blood lactate was measured at rest, at minute-13 for all trials, at minute-53 in extended trials, and during recovery. EPOC was measured until baseline was reached. In series A, EPOC was 1.5 L at 50%, 1.8 L at 70%, and 3.3 L at 90% VO_{2max}. Thus, the 40% increase in intensity (and total work) from 50% to 70% VO_{2max} resulted in only a 20% increase in EPOC, while the 80% increase in intensity (and total work) from 50% to 90% of VO_{2max} resulted in a more than doubling (120% increase) of EPOC, suggesting an exponential increase with increasing workload. In series B, performed at 60% VO_{2max}, EPOC was 1.6 L for 15 minutes of exercise, 1.9 L for 35 minutes, and 2 L for 55 minutes. When duration (and total work) was more than doubled and then more than tripled from 15 minutes, EPOC increased by only 19% and 25%, respectively. Comparing series A and B reveals that when total work is increased by extending duration, EPOC is only marginally increased. Also, increasing intensity within a modest aerobic range (50-70% VO_{2max}) has little effect on EPOC, but increasing intensity to a level approaching VO_{2max} increases EPOC greatly.

Gore & Withers (1990) also compared various intensities and durations, but using treadmill exercise. Nine physically active males 21.9 $\pm$ 2.2 years (VO_{2max} 63$\pm$ 5.7 $mL \cdot min^{-1} \cdot kg^{-1}$) performed nine separate interventions at 30, 50, and 70% of their VO_{2max} for 20, 50, and 80 minutes at each intensity. RMRs were performed on two separate days after eight hours of bed rest (before and after the experiment). Subjects came on a separate day to determine treadmill speeds to elicit respective VO_{2max} percentages. Skin and core temperatures were measured continuously. Subjects consumed a pre-weighed meal for dinner the night before testing, and a light pre-weighed breakfast four hours before testing. Subjects reported at 0630 and began exercising after one hour of bed rest. Each subject performed the three different durations under the three different intensities in a counterbalanced design. EPOC was measured over 8 hours post-exercise, which included a standardized meal after 3 hours. EPOC following exercise at 30% VO_{2max} was low (1.0-1.4 L) and was not significantly different between the various durations. EPOC was greater at 50% VO_{2max} and did increase with duration of exercise (3.1 L, 5.2 L, 6.1 L, after 20 min, 50 min and 80 min, respectively). EPOC was greater still at 70% VO_{2max} and increased with duration (5.7 L, 10.0 L, 14.6 L after 20 min, 50 min and 80 min, respectively). Comparing the EPOC of 5.2 L after 20 min at 70% VO_{2max} (~14 AEU) and the EPOC of 10.0 L after 50 min at 50% VO_{2max} (~25 AEU), these results indicate that intensity increases EPOC more than total work does.

Mann et al. (2014) sought to investigate the effect of exercise intensity on EPOC in 38 endurance-trained runners (20 male) who had been regularly training for at least three months. During the initial visit, a graded exercise test was performed to assess VO_{2max}, maximum heart rate (HR_{max}) and peak treadmill running speed (PTRS). The following three visits were randomized and consisted of twenty minutes of treadmill running at 60, 70, or 80% VO_{2max}. The three exercise trials were all completed within a seven-day period. Recovery VO_2 was measured during a quiet, seated position for a total of 15 minutes. Results demonstrated that EPOC increased with increasing intensity. The average EPOCs were 1.2 L at 60% VO_{2max}, 1.3 L at 70% VO_{2max}, and 1.6 L at 80% VO_{2max}. Aerobic exercise units were 12, 14, and 16, respectively. This demonstrates that with a 33% increase in intensity and total work from 60 to 80% VO_{2max}, EPOC also increased by 33%. These results contradict the finding of a greater increase in EPOC at high aerobic intensities as demonstrated in previous literature (Knuttgen et al., 1970; Gore & Withers, 1990). None of these studies controlled the total of amount of work performed during the exercise trials.

Equated Workload Studies

LaForgia et al. (1997) attempted to equate workloads comparing HIIE to lower intensity longer duration treadmill running. Subjects were eight male middle-distance runners, 21.1 $\pm$ 3.1 years of age. Subjects lay supine for 50 minutes to gather RMR data. Subjects then randomly performed either running for 30 minutes at 70% VO_{2max} (~21 AEU), or twenty 1-min intervals of 105% VO_{2max} (~ 21 AEU) separated by 2 minutes of rest in-between. Exercise sessions were separated by a few days. The HIIE demonstrated double the amount of EPOC (15 L) compared to the submaximal exercise (6.9 L), supporting an intensity effect on EPOC.

In a study conducted by Sedlock et al. (1991), seven physically active females performed two randomized trials at either 40% VO_{2max} or 60% VO_{2max}, with EPOC measurements following immediately until baseline was reached. After initial VO_{2max} was assessed, subjects returned after at least two days to perform the exercise trials on a cycle ergometer. Each exercise condition was separated by at least two days. Each exercise condition began with a pre-exercise resting VO_2 measurement for 45 minutes with the final 15 minutes collected. The exercise duration was terminated once each condition reached 850 kJ of work on the cycle ergometer. The average duration of the 40% and 60% VO_{2max} trials was 41 minutes and 27 minutes, respectively. EPOC was slightly but not significantly larger at 60% VO_{2max} (36.1 $\pm$ 12.6 kJ) compared to 40% (29.8 $\pm$ 17.2 kJ).

A study conducted by McGarvey et al. (2005), found no significant difference in EPOC during MOD and interval cycling HIIE. Twelve physically active males, 30 $\pm$ 7 years of age with an average VO_{2max} of 49.7 $\pm$ 8.9 ml·kg^{-1}·min^{-1}, participated in the study. Testing consisted of four sessions with at least 72 hours in-between. The first session allowed for cycle familiarization, and an assessment of VO_{2max} and ventilatory threshold (V_T) during a graded exercise test. Subjects came in for the second session for testing to practice the HIIE protocol and to design the workload to fatigue the subject after seven intervals so the eighth could not be completed. The third and fourth sessions (MOD and HIIE treatments) were randomly assigned. Both treatments began with an RMR that consisted of 15 minutes of rest, followed by 20 minutes of collection. Treatments also began with a six-minute warm-up beginning with 50 W. Power was increased by 5 W every minute until it reached 5 W below each subject's 65% VO_{2max} power output. MOD treatment consisted of 30-32 minutes at 65% VO_{2max}. Duration of MOD treatment was adjusted to match the work output of their HIIE condition. The MOD condition consisted of 35 minutes of exercise alternating between two minutes at 90% VO_{2max} and three minutes at 30% VO_{2max} (14 min total of 90% and 21 min total of 30% VO_{2max}). After each exercise condition, the subject was immediately seated, consumed 125 mL of water, and then lay supine for two hours of RMR collection. As expected, baseline RMRs were not different between conditions. Although they attempted to equate total work, it was slightly but significantly greater in the MOD treatment compared to the HIIE by 3.63 kJ. AEUs were 19.5 for the MOD treatment, and 18.9 for the HIIE treatment. EPOC was determined using two methods. The 1-standard deviation method determined the termination time of measuring EPOC as when the values fell within 1-SD of baseline

VO_2 for two consecutive minutes. The 5-min EPOC method determined the termination time of measuring EPOC as when the five-min average recovery VO_2 equaled the baseline VO_2. In either the 1-SD or 5-min EPOC method, there was no significant difference between the MOD (6.2 L, 7.0 L) versus HIIE (6.7 L, 7.6 L) treatments, despite the much greater intensity in the HIIE as compared to MOD condition. However, while the maximum intensity in HIIE was 90% VO_{2max}, the average intensity during the entire 35 minutes of intervals was 54% VO_{2max}, slightly less, rather than substantially more, than the 65% of the MOD condition.

Phelain et al. (1997) examined EPOC responses in eight physically active females (22-31 years old) with a greater difference in cycling intensities than Sedlock et al. (1997). Subjects were randomly assigned to the low intensity exercise (LIE) condition (50% VO_{2max}), vigorous intensity exercise condition (VIG) (75% VO_{2max}) and a control condition (CON) (quiet sitting for one hour). The order of these assignments was counterbalanced. The VIG and LIE conditions were terminated once the subject reached an energy expenditure of 500 kcal. The exercise conditions lasted 65-90 minutes for LIE, and 40-60 minutes for VIG. The average duration was 77.8 minutes for LIE (38.9 AEU) and 50.9 minutes for VIG (38.2 AEU). All testing began at 1200 hours and there were at least three days separating each condition. Before testing, a VO_{2max} test was performed on a cycle ergometer, and on a separate day RMR was measured no later than one week before testing and consisted of a 45-minute supine rest period with data collected during the final 30 minutes. EPOC after exercise was measured every three minutes over a total period of three hours. The first 35 minutes was in a seated position, and then transitioned to supine for the remaining time. Results demonstrated that the mean EPOC of the VIG condition (9 L) was significantly greater than the LIE condition (4.8 L) while AEUs and caloric expenditures were similar between conditions.

In sedentary subjects with metabolic syndrome, VIG elicited a greater EPOC when compared to continuous moderate intensity exercise (Larsen et al., 2014). Seven sedentary men with metabolic syndrome participated in the study. Subjects were between 39-70 years of age (mean = 56.7 $\pm$ 10.8 yr). One week before the experiment, subjects reported to the lab to assess VO_{2max} and HR_{max} as well as anthropometric measures. Subjects reported to the lab for testing at the same time of day after a 12-h overnight fast and were instructed not to perform any vigorous activity the previous 48 hours. After 20 minutes of supine rest, subjects underwent a 20-minute RMR. After the RMR, subjects performed the same warm-up (10 minutes at 50-60% VO_{2max}) followed by either a single 4-min bout at 80-90% VO_{2max} (1-AIT, ~9 AEU including warm-up), four 4-min intervals at 80-90% VO_{2max} interspersed with 3 min at 50-60% VO_{2max} (4-AIT, ~24 AEU), or 47 min of continuous moderate exercise at 50-60% VO_{2max} (CME, ~26 AEU) that were randomly assigned. Each exercise session was separated by at least 48-h. After each exercise session was performed, subjects returned to the canopy within 5 minutes of cessation of exercise, and EPOC was measured until it returned to baseline VO_2. Results demonstrated that the 4-AIT exercise condition had a significantly longer EPOC period (70.4 min) than 1-AIT (35.9 min) and CME (45.0 min). EPOC was significantly greater in the 4-AIT (2.86 L) than the 1-AIT (1.36 L) and the CME (1.44 L) conditions. EPOC for the 4-AIT condition was approximately double that for CME.

When comparing two different intensities in a moderate range (50-75% VO_{2max}), differences in EPOCs were conflicting in the research. Several studies demonstrated a modest increase in EPOC with an increase in intensity in the mid-range (Knuttgen et al., 1970; Phelain et al., 1997; Gore & Withers, 1990) while others found no significant difference (Sedlock et al., 1991; Mann et al., 2014). When comparing moderate intensity to high intensity exercise (≥ 80% VO_{2max}), EPOC seems to be consistently greater in the high intensity interventions and it becomes clear there is an intensity-dependent effect (Knuttgen et al., 1970; LaForgia et al., 1997; Larsen et al., 2014; Mann et al., 2014). Of these studies, two equated the workloads between the different conditions (LaForgia et al., 1997; Larsen et al., 2014). The findings of McGarvey et al. (2005), however, found no significant difference between HIIE at 90% VO_{2max} versus continuous exercise at 65% VO_{2max} when work was equated. The differences in findings of EPOC after various intensities with equated workloads might be due to the large range of fitness levels in their subjects.

EPOC and Fitness Level

Short & Sedlock (1997) investigated EPOC and recovery rate in trained versus untrained subjects during cycling exercise. Twelve trained (5 female) and 10 untrained (6 female) subjects completed the study with an average VO_{2max} of 48.6 $ml \cdot kg^{-1} \cdot min^{-1}$ for trained women, 56.7 $ml \cdot kg^{-1} \cdot min^{-1}$ for trained men, 35.9 $ml \cdot kg^{-1} \cdot min^{-1}$ for untrained women, and 39.8 $ml \cdot kg^{-1} \cdot min^{-1}$ for untrained men. Inclusion criteria for trained group was > 45 $ml \cdot kg^{-1} \cdot min^{-1}$ VO_{2max}, and <40 $ml \cdot kg^{-1} \cdot min^{-1}$ VO_{2max} for the untrained group. Two EPOC trails were then performed in a counterbalanced order separated by at least 72 hours. Work rates were performed at an absolute and relative level on separate days. Work rates were adjusted to elicit 70% VO_{2max} (relative work rate) in one condition, and 1.5 L/min (absolute work rate) each for 30 minutes. Before each exercise session, subjects sat resting for 15 minutes before VO_2 was collected for an additional 10 minutes. After exercise, EPOC was continuously measured in a seated position for 60 minutes, or until VO_2 returned to baseline. In all but two trials, VO_2 returned to baseline within 60 minutes. In the untrained group, there was no significant difference between EPOC magnitudes in relative or absolute intensities (3.5 L versus 2.4 L, respectively). In the trained group, however, EPOC magnitudes were significantly larger in the relative intensity compared to the absolute intensity (3.2 L versus 1.5 L, respectively). In the relative intensity condition, the trained group had significantly quicker recovery rates than untrained up until the 8-min mark post-exercise.

In a study conducted by Sedlock et al. (2010), EPOC was analyzed before and after aerobic exercise training in an attempt to quantify the impact of fitness level on EPOC magnitude. Twenty relatively fit men (VO_{2max} 55.1 $\pm$ 1.2 $ml \cdot kg^{-1} \cdot min^{-1}$) were randomly assigned to either an exercise group (EX) or control group (CON) after pretesting. Pretesting included VO_{2max} testing and an EPOC measurement after 30 minutes of exercise at 70% of VO_{2max}. EX performed 12 weeks of endurance training on a treadmill beginning with three sessions per week for 20 minutes at 60% VO_{2max}. Workload gradually increased so that by the 12th week, subjects were exercising four sessions per week for 40 minutes at 80% VO_{2max}. CON did not participate in an intervention and were instructed to maintain normal activity levels. EPOC was measured once after 12 weeks in the control group, and twice in the exercise group: once at each subject's absolute intensity in the pre-test, and once at a higher intensity that corresponded to 70% of the subject's *new* VO_{2max}. EPOC was measured lying supine in a 120-minute recovery period immediately after exercise. As expected, VO_{2max} increased significantly in the exercise group from pre to post (46.2 ± 1.2, 51.0 ± 1.3 $ml \cdot kg^{-1} \cdot min^{-1}$). EPOC following exercise at the same absolute intensity was significantly lower after exercise training (7.9 L pre, 6.3 L post), whereas there was no significant difference pre to post when exercising at the same relative intensity (7.9 L pre, 8.2 L post). The CON group EPOC did not differ pre to post (7.4 L and 7.7 L, respectively). The significant difference between EPOC magnitudes between absolute and relative intensity demonstrates the need to prescribe exercise intensities relative to each subject's fitness level.

Matsuo et al. (2012) also found differences in EPOC in individuals with different levels of fitness. Ten male subjects with VO_{2max} values ranging between 36.2 and 61.4 $ml \cdot kg^{-1} \cdot min^{-1}$ participated in cycling exercise consisting of sprint interval training (SIT), high-intensity interval aerobic training (HIIE), and continuous aerobic training (MOD), each separated by one week. SIT consisted of seven sets of 30-s cycling at 120% VO_{2max} each followed by 15-s of rest. The HIIE consisted of three sets of 3-min cycling at 80-90% VO2max each followed by 2-min active rest at 50% VO_{2max}. MOD consisted of 40-min cycling at 60-65% VO_{2max}. All subjects arrived for testing at the same time of day. EPOC was measured 180 minutes after cessation of exercise (10 minutes in the face mask immediately after exercise, then 10 minutes at minutes 30, 60, 90, 120, 150, and 180). Prior to each exercise, subjects were seated for twenty minutes, and then RMR was measured for fifteen minutes. Average VO_2 data during the final ten minutes was considered the baseline value. EPOC was significantly greater in SIT (6.8 ± 4.0 L) than MOD (2.9 ± 2.8 L), but was not significantly different from HIIE (4.5 ± 3.3 L). There were also no significant differences between HIIE and MOD. In addition, the relationship between subjects' VO_{2max} and the ratio of EPOC to net exercise VO2 correlated inversely with fitness level in the SIT and HIIE conditions, but no such correlation in the MOD condition. Also, there was a significant negative correlation between absolute EPOC and VO_{2max} for the HIIE condition, but not for the SIT and MOD conditions. Table 1 summarizes EPOC quantities from various studies that compare more than one intensity.

Table 1. EPOC quantity following different intensities and durations of aerobic exercise.

Author	Subjects	Subject VO_{2max} ($mL \cdot kg^{-1} \cdot min^{-1}$) $\pm$ SD	Study Design	EPOC (L)
Gore & Withers, 1990	9 males	63.0 $\pm$ 5.7	Treadmill: 30%, 50%, and 70% each for 20, 50, and 80 min	30%: 1.0 L, 1.4 L, 1.0 L 50%: 3.1 L, 5.2 L, 6.1 L 70%: 5.7 L, 10.0 L, 14.6 L
Knuttgen et al., 1970	12 (5 female)	40.0 $\pm$ 9.3	Cycling: 15 min @ 50, 70, 90%; 60% for 15, 35, 55 min; Between 55 – 80% for 15 or 55 min	50%: 1.5 L; 70%: 1.8 L; 90%: 3.3 L 60%: 1.6 L (15 min), 1.9 L (35 min), 2 L (55 min) 2 L (15 min), 2.1 L (55 min)
LaForgia et al.,	8 males	69.2 $\pm$ 4.0	Treadmill: 30 min	70%: 6.9 L

1997			@ 70%; 20 1-min intervals @ 105% with 2-min rest intervals.	105%: 15 L
Larsen et al., 2014	7 males with metabolic syndrome	33.2 + 8.8	Treadmill: 4x4-min intervals @ 85-95% HRmax with 3-min intervals @ 70%; 1x4 min interval @ 85-95% HRmax with 3-min interval @ 70%; 47 min @ 70%	4x4: 2.86 L 1x4: 1.36 L 70%: 1.44 L
Mann et al., 2014	38 males	57.1 + 6.6	Treadmill: 20 min @ 60%, 70%, and 80%	60%: 1.2 L 70%: 1.3 L 80%: 1.6 L
Matsuo et al., 2012	10 males	52 + 9.2	Cycling: 7 30-s intervals at 120% with 15-s rest intervals; 3 3-min intervals at 80-90% with 2-min intervals at 50%; 40 min at 60-65%	120%: 6.8 L 80-90%: 4.5 L 60-65%: 2.9 L
McGarvey et al., 2005	12 males	49.7 + 8.9	Cycling: 7 2-min intervals at 90% with 3-min intervals at 30%; ~ 30 min @	90%: 7.6 L; 65%: 7.0 L

			65%	
Phelain et al., 1997	8 females	47.4+ 4.2	Cycling: 50% and 75% until 500 kcal expenditure reached (approx. 77.8 min for 50%, 50.9 min for 75%)	50%: 4.8 L 75%: 9 L
Sedlock et al., 1991	7 females	Not Reported	Cycling: 40% (~ 41 min) and 60% (~ 27 min) until 850 kJ reached	40%: 1.4 L, 60%: 1.7 L
Sedlock et al., 2010	20 males, exercise/control	EX:46.2 + 1.2 pre, 51.0 + 1.3 post CON: 45.1 + 1.4 pre, 44.5 + 1.3 post (all reported in SEM)	Treadmill: 30 min at 70% PRE, 30 min post at 70% absolute (abs), 30 min at 70% new VO2max relative intensity (rel)	PRE: 7.8 L fit, 7.4 unfit POST: 6.3 L fit abs, 8.2 L fit rel; 7.7 L unfit
Short & Sedlock, 1997	22 (11 female)	Fit: 56.7 male, 48.6 female; Unfit: 39.8 male, 35.9 female	Cycling: 70% for 30 min (relative); 1.5L/min workload for 30 min (absolute)	Fit: 3.2 L rel, 1.5 L abs Unfit: 3.5 L rel, 2.4 L abs

CHAPTER III

METHODOLOGY

The purpose of this chapter is to describe the detailed procedures involved in this research study including subject recruitment and selection, and the data collection process. All elements of this research study were examined and accepted by Old Dominion University's Institutional Review Board prior to its start. Subjects were asked to provide informed consent, and all personal information or identifiers were kept confidential.

Research Design

This was a correlational study designed to identify the relationship between fitness levels and magnitude of EPOC after exercise of two different intensities. Fit and unfit subjects performed an initial exercise test and then two exercise trials in random order: a moderate intensity exercise bout (MOD) at 50% of each subject's maximal aerobic power, and high intensity aerobic interval exercise (HIAI) at 90% and 60% of each subject's maximal aerobic power. This study aimed to quantify the magnitude of EPOC following these two conditions and assess the effect of aerobic fitness, as well as compare the responses of male and female subjects.

Sample

A total of 27 subjects were recruited (14 male, 13 female). Of these subjects, 11 college-age students between the ages of 20 and 44 years, 5 males and 6 females, completed the study. Of the 16 that dropped, one had gained > 5 lb between initial testing and the first trial, the rest had scheduling conflicts. Subjects all met ACSM criteria for being low risk for cardiovascular events during exercise, as described earlier. Subjects were not on medication that affected metabolic rate (such as prednisone) or heart rate (such as beta blockers). Other exclusionary criteria included any physical inability to complete a maximum exercise test on a cycle ergometer, females with amenorrhea or who may have been pregnant, or subjects with a recent caloric restriction or large self-reported fluctuations in weight loss or gain (> 5 lb in the past month). The first visit included a screening questionnaire to determine subject eligibility (Appendix A).

Subject Recruitment

Subjects were recruited through email and class announcements at Old Dominion University. Interested participants completed a questionnaire. Those who qualified and volunteered signed an informed consent form outlining the nature of the study, its purpose, and the potential risks/benefits of participation (Appendix B). All eligible subjects were instructed to keep a food intake log for the 24 hours prior to each visit using the free MyFitnessPal (MyFitnessPal, Inc.) dietary recording website and phone application., and were asked to repeat the diet of trial day one on trial day two All of the subjects completed their logs on the phone application and each diet (visit 1 versus visit 2) was identical to one another. All female subjects were also given a menstrual cycle questionnaire to determine menstrual cycle phase; their start date began the day after their last period (Appendix C).

Maximal Exercise Test

The first visit occurred in the Human Performance Laboratory of Old Dominion University's Student Recreation Center and consisted of measurement of anthropometric data including height, mass, and body composition; the last using air displacement plethysmography (BodPod *Cosmed*; Illinois). Subjects then rested in a seated position for

10 minutes followed by a 10-minute RMR measurement utilizing a metabolic assessment cart (ParvoMedics TrueOne 2400, Sandy, UT) for open circuit spirometry. Subjects then became familiarized with the Velotron cycle ergometer (RacerMate) and the facemask used in VO_2 assessment. The VO_{2max} test consisted of 3-min stages. Women began with 35 W and increase by 35 W per stage. Men began with 45 W and increased by 45 W per stage. Cadence was maintained at approximately 70 rpm (65-75). The test was terminated when the subject's cadence fell below 60 rpm despite strong verbal encouragement. VO_{2max} was determined as the average of the highest consecutive 3 values (sampled every 15 seconds) of VO_2 achieved during the test with an RER above 1.10.

Exercise Trials

Visits two and three were randomized to either MOD or HIAI exercise condition, which were both preceded with a warm-up pedaling at 70 rpm. For the MOD condition, the warm-up was 3 minutes at 40% P_{max}. The subject then pedaled at 50% of P_{max} for 30 minutes. The warm-up for the HIAI trial consisted of 2 minutes at 30% P_{max} followed by 1 minute at 60% P_{max}, which was equivalent in energy expenditure to the MOD warm-up. The HIAI itself consisted of ten intervals of exercise: one minute at 90% P_{max} followed by one minute of 60% P_{max} (except for the last 60% P_{max} interval, which was followed immediately by the cool-down). The cool-down was the same after each condition and consisted of 2 minutes at 30% P_{max}. Each testing day was separated by at least 48 hours.

EPOC Measurement

After the cool-down, EPOC measurements began and was the same for each exercise condition: the subject remained on the cycle for 5 minutes while resting, then transferred to a seated position on a chair for another 5 minutes with the facemask still intact and VO_2 measurement continuously sampling. The subject was then allowed 250 mL of water prior to donning the facemask for an additional 30 minutes of VO_2 sampling. The water break was timed for one minute. The subject was allowed to read quietly during and in-between VO_2 measurements. After the 30-minute VO_2 measurement, there was a 30-minute seated resting period followed by a final 10-minute VO_2 measurement.

Statistical Analysis

For hypotheses 1 and 3, a two-way ANOVA (intensity and sex; repeated measures on intensity) was used to compare mean values of EPOC (and of relative EPOC) between the two intensity trials and between men and women. For hypothesis 2, a Pearson correlation was used to determine the relationship between delta EPOC (EPOC following HIAI – EPOC following MOD) and VO_{2max}, and for the delta of relative EPOC vs VO_{2max}. Differences between trials in VO_2 during rest and exercise were examined using paired Student-t tests. Differences between the subject characteristics of males and females were examined with unpaired Student-t tests. Statistics were performed using SPSS Macintosh Version 20 (IBM Company Headquarters, Chicago, IL). Alpha level for statistical significance was set at $p \leq 0.05$. All results are given as mean $\pm$ SE.

CHAPTER IV

RESULTS

Characteristics of the 11 subjects who completed the study are provided in Table 2.

Table 2. Subject characteristics (mean + SE).

	Age (yr)	Height (cm)	Mass (kg)	Body Fat (%)	VO_{2max} ($mL \cdot min^{-1} \cdot kg^{-1}$)
All (n = 11)	25.1 + 2.0	169 + 3	67.6 + 3.0	19.0 + 2.1	41.0 + 1.9
Males (n = 5)	27.6 + 4.3	176 + 3	70.8 + 6.2	13.4 + 1.7	44.9 + 2.9
Females (n = 6)	23.0 + 1.0	164 + 2	65.0 + 1.8	23.7 + 2.3	37.7 + 1.8
M vs. F	p = 0.280	p = 0.010	p = 0.354	p = 0.006	p = 0.053

EPOC Response: MOD vs. HIAI

The resting VO_2s prior to each trial were not significantly different from each other (MOD: 4.0 + 0.3 $mL \cdot min^{-1} \cdot kg^{-1}$, HIAI: 4.1 + 0.4 $mL \cdot min^{-1} \cdot kg^{-1}$), p = 0.374. The net VO_2s during each trial were not significantly different (MOD: 53.6 + 3.5 L, HIAI: 52.0 + 3.9 L), p = 0.361. VO_2 during the latest recovery period (71-81 min post-exercise) was not significantly different from resting VO_2, and has been removed from the data analysis. There was a significant EPOC during minutes 0-41, as well as separately during minutes 0-10, and 11-41. Figures 1, 2, and 3 illustrate EPOC magnitude following each condition for the respective time periods. For the overall EPOC during minutes 0-41, there was no significant difference between HIAI (3.02 + 0.48 L) and MOD (1.98 + 0.56 L) conditions, p = 0.164. However, given the weak trend for a difference between trials, separate two-way ANOVAs were performed on the 0-10 and 11-41 time periods. EPOC during the first ten minutes post-exercise was significantly greater following HIAI (2.28 ± 0.32 L) than MOD (1.37 ± 0.43 L), p = 0.009. EPOC during minutes 11-41 was not significantly different between the HIAI (0.74 ± 0.28 L) and MOD (0.56 ± 0.43 L) conditions, p = 0.807. Similar results were found when absolute values were normalized to body weight.

Figure 1. Absolute EPOC minutes 0-41 following MOD and HIAI conditions.

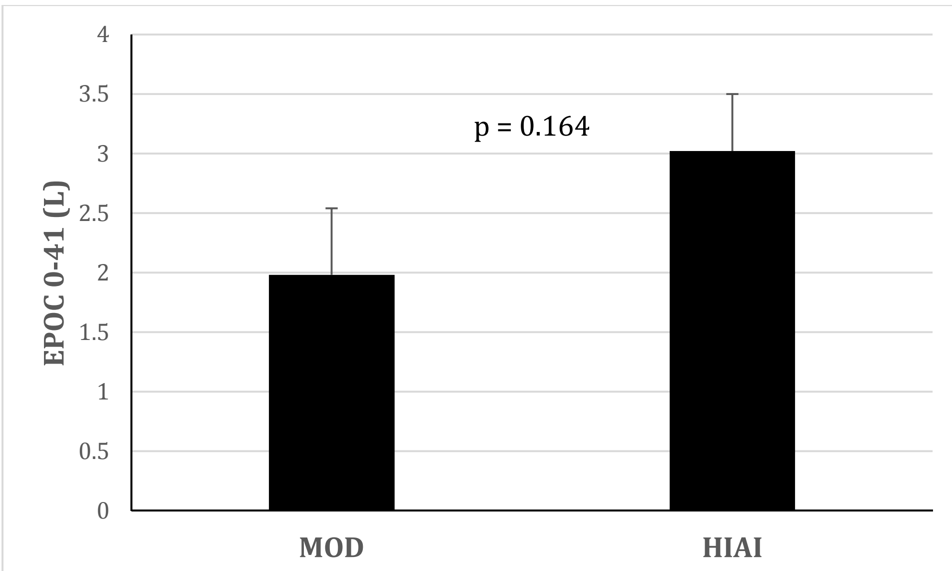


Figure 2. Absolute EPOC minutes 0-10 following MOD and HIAI conditions.

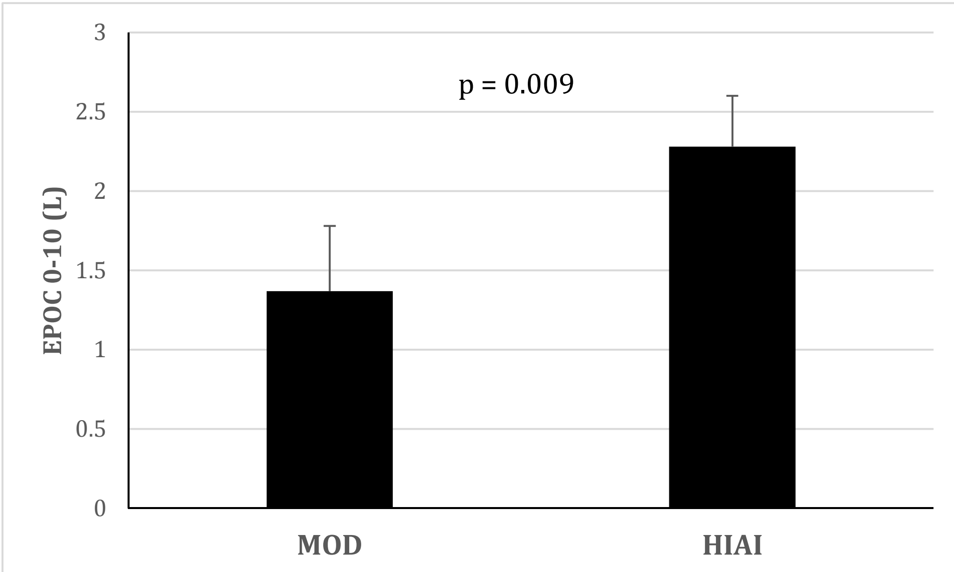
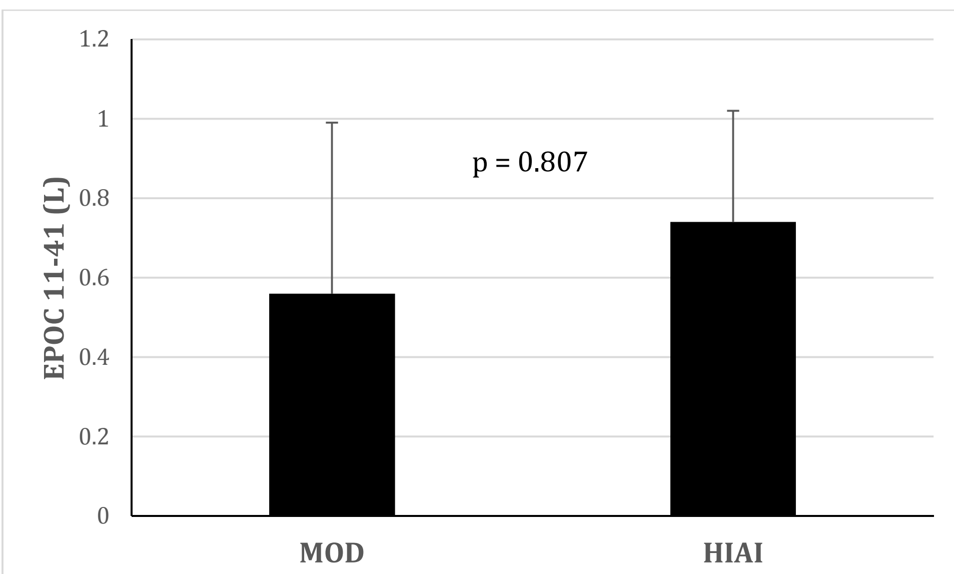


Figure 3. Absolute EPOC minutes 11-41 following MOD and HIAI conditions.



EPOC Response: Males vs. Females

For total relative EPOC minutes 0-41 (Fig. 4), males had a significantly higher response to both HIAI (males: 58.0 ± 8.7 $mL \cdot kg^{-1}$, females: 32.5 ± 4.9 $mL \cdot kg^{-1}$) and MOD (males: 44.9 ± 14.0 $mL \cdot kg^{-1}$, females: 18.7 ± 10.6 $mL \cdot kg^{-1}$), p = 0.035. A trend for a greater EPOC for males occurred during minutes 0-10 following HIAI (males: 43.6 ± 5.3 $mL \cdot kg^{-1}$, females: 25.9 ± 5.0 $mL \cdot kg^{-1}$) and MOD (males: 24.5 ± 6.4 $mL \cdot kg^{-1}$, females: 18.6 ± 3.2 $mL \cdot kg^{-1}$), p = 0.086. The same trend was observed in minutes 11-41, following HIAI (males: 14.4 ± 4.7 $mL \cdot kg^{-1}$, females: 6.6 ± 5.7 $mL \cdot kg^{-1}$) and MOD (males: 20.4 ± 8.5 $mL \cdot kg^{-1}$, females: -2.0 ± 8.7 $mL \cdot kg^{-1}$), p = 0.053. Similar results were found in absolute EPOC values. Figures 5 and 6 illustrate males versus females in time periods 0-10 and 11-41, respectively.

Figure 4. EPOC minutes 0-41, male vs. female response to MOD and HIAI conditions.

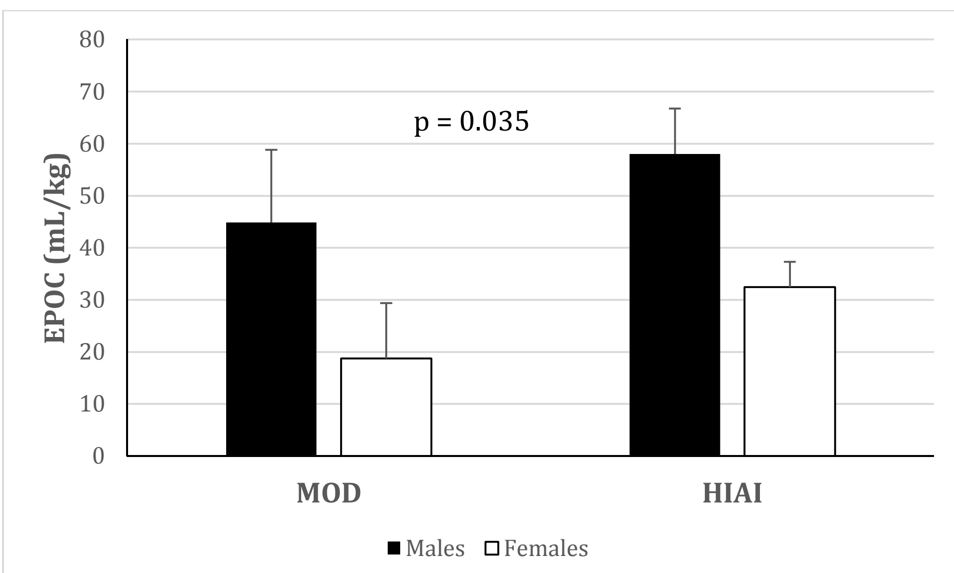


Figure 5. EPOC minutes 0-10, male vs. female response to MOD and HIAI conditions.

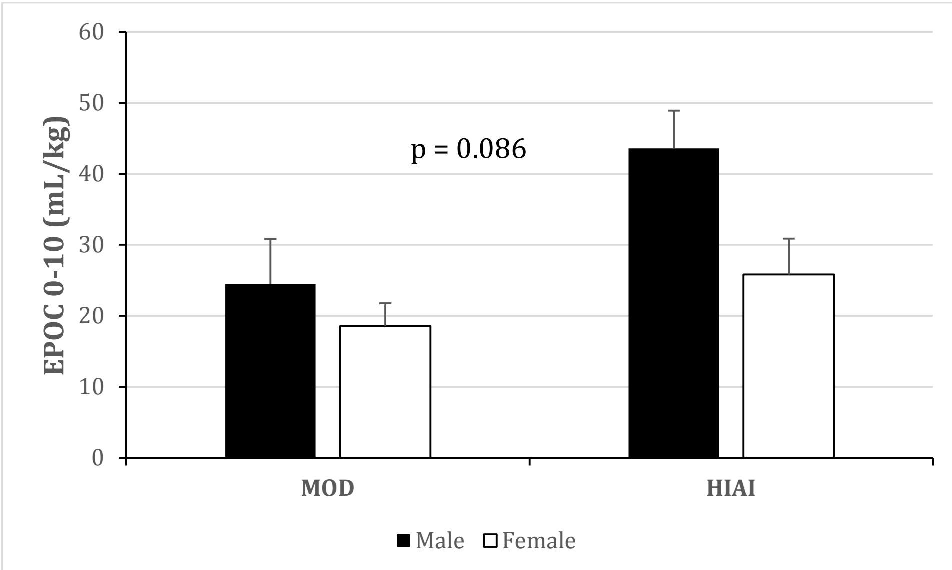
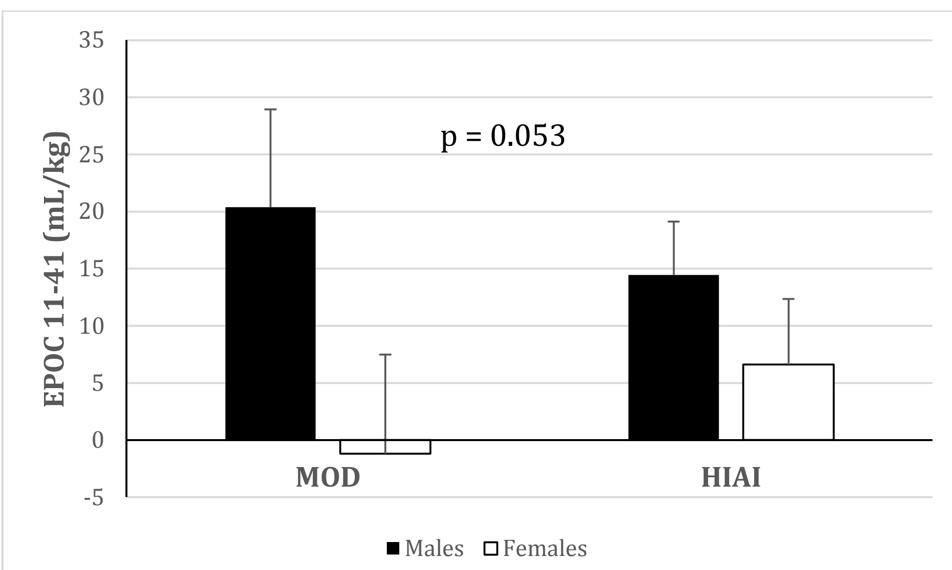


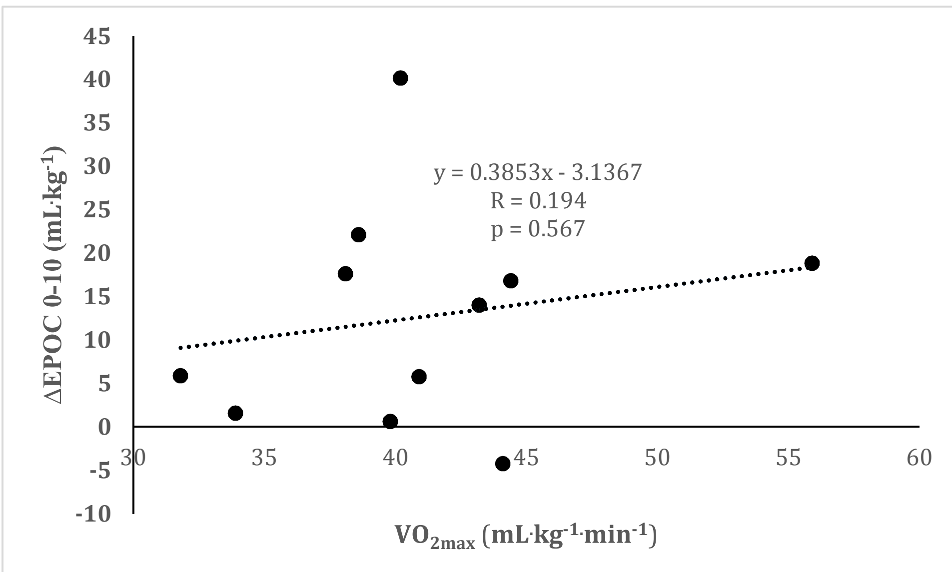
Figure 6. EPOC minutes 11-41, male vs. female response to MOD and HIAI conditions.



EPOC Relationship to VO_{2max}

There was no correlation between VO_{2max} and ΔEPOC ($mL \cdot kg^{-1}$), whether using the total time frame (0-41 min; $p = 0.574$), or the early (0-10 min; $p = 0.567$) or late (11-41 min; $p = 0.294$) phases of EPOC. Figure 7 illustrates the relationship for early EPOC. In addition, there was no significant correlation between VO_{2max} and relative EPOC during either the MOD ($p = 0.293$) or HIAI ($p = 0.457$) trials individually.

Figure 7. ΔEPOC minutes 1-10 vs. VO_{2max}



CHAPTER V

DISCUSSION

The purpose of this study was to investigate EPOC magnitude following HIAI exercise and MOD exercise, accounting for potential differences in fitness levels and sex. We hypothesized that EPOC following the more vigorous HIAI trial would be greater than that following the MOD trial, even though the total energy expenditure of both trials was equated. Over the entire EPOC period there was a weak trend for a difference between MOD and HIAI conditions, and minutes 0-10 revealed a significantly greater EPOC following HIAI than MOD exercise in absolute and relative terms; supporting the hypothesis. We further hypothesized that males and females would exhibit similar EPOCs. Males demonstrated a significantly higher EPOC following each condition in absolute and relative EPOC magnitudes. Finally, we hypothesized that individuals with a higher fitness level, as indicated by VO_{2max}, would have a faster recovery from intense exercise and thus demonstrate a lesser difference in EPOC between the MOD and HIAI trials. Contradicting this hypothesis, there was no correlation between fitness level and ΔEPOC.

EPOC Response: MOD vs. HIAI

The majority of EPOC occurred during minutes 0-10 and was significantly greater following HIAI than MOD, which supports similar findings of previous authors demonstrating an exponential increase in EPOC with increasing intensity (Gore & Withers, 1990; Knuttgen et al., 1970; LaForgia et al., 1997; Matsuo et al., 2012; McGarvey et al., 2005; Phelain et al., 1997). The magnitudes of EPOC in L following MOD (1.98 L) and HIAI (3.02 L), both with AEUs of 16.8, were comparable to that of some studies with similar AEUs (Knuttgen et al., 1970; Sedlock et al., 1991), however some authors found more or less EPOC. Gore & Withers (1990) found that 20 minutes of treadmill running at 70% (14 AEUs) elicited a magnitude of 5.7 L in six trained males. One possible reason for the greater EPOC in that study was the longer post-exercise measurement period (8 hours), which included a standardized meal feeding at 3 h post-exercise. For most of the exercise conditions in Gore & Withers (1990), as discussed in the literature review, post-exercise VO_2 returned to baseline within one hour: 80 minutes at 50% returned to baseline in 2 hours, 50 minutes at 70% in 4 hours, and 80 minutes at 70% in 8 hours. It is clear that the exercise stimuli needed to elicit a prolonged EPOC from previous literature is greater than 50 minutes at 70% when investigating submaximal aerobic exercise (Laforgia et al., 2006; Gore & Withers, 1990). With this information, it is not necessary to include standardized meals and VO_2 measurements beyond an hour or two post-exercise when comparing submaximal aerobic exercises of shorter duration, as it makes it more difficult to compare results.

In this study, an intensity increase from MOD to HIAI demonstrated a 1.5-fold increase in EPOC despite maintaining matching workloads, and a similar 2-fold increase was found when comparing 50% to 75% intensity at matched workloads by Phelain et al., 1997. Mann et al. (2012) found greater EPOC magnitudes following higher intensity levels (seven 30-s intervals at 120% with 15 s rest in-between) but lower overall AEUs, further supporting the exponential EPOC increase with intensity.

EPOC Response: Males vs. Females

This study demonstrated that males had a significantly higher total EPOC response following HIAI and MOD conditions, contradictory to the hypothesis. Of the 11 studies known to compare more than one intensity discussed in the literature review, only four included female subjects. Of these four, two had only female subjects, and two had both male and female subjects. Knuttgen et al. (1970) had 7 male and 5 female subjects, and no comparison was made between the sexes. In addition, this study did not control for the changes in metabolism with the menstrual phases. Short & Sedlock (1997) had 11 male and 11 female subjects but also did not assess sex differences in EPOC responses, or take the menstrual phase into account. Of the two studies with only females, Phelain et al. (1977) found that exercise of around 16 AEUs produced about 1.5 L of EPOC, and Sedlock et al. (1991) found that around 39 AEUs produced 9 L of EPOC when intensity was 75% and produced 4.8 L at an intensity of 50%. Table 3 below summarizes the male-only studies that had similar AEUs to those in the female-only studies. The intensity column indicates the intensity (%) of the work interval when the task consisted of HIIE.

Table 3. Summary of studies including only males, comparing AEUs to EPOC.

AEUs	EPOC (L)	Intensity (%)	Task	Mode	Author
8.1	4.5	~62	3 x 3min & 2 min @ 50%	Cycle	Matsuo et al., 2012
12	1.2	60	20	Treadmill	Mann et al., 2014
14	5.7	70	20	Treadmill	Gore & Withers, 1990
14	1.3	80	20	Treadmill	Mann et al., 2014
15	1.4	30	50	Treadmill	Gore & Withers, 1990
16	1.6	~85	20	Treadmill	Mann et al., 2014
18.9	7.6	70	7 x 2 min & 3 @ 30%	Cycle	McGarvey et al, 2005
19.5	7	90	30	Cycle	McGarvey et al, 2005
21	6.9	105	30	Treadmill	Laforgia et al., 1997
21	15	60	20 x 1-min & 2 min rest	treadmill	Laforgia et al., 1997
21	7.6		30	Treadmill	Sedlock et al., 2010
24	2.9	65	40	Cycle	Matsuo et al., 2012
35	10	70	50	Treadmill	Gore & Withers, 1990

When the intensity is $\leq$ 70%, and AEUs are equal to or less 16, male studies demonstrate a range of EPOCs between 1.2 and 4.5 L compared to the female study which resulted in 1.5 L at ~16 AEUs (Sedlock et al., 1991). When the intensity is $\geq$ 70% and AEUs are 16 or greater, male EPOC ranges from 1.6 L to 15 L compared to the females with 9 L who performed around 38 AEUs (Phelain et al., 1997). In addition, the range of fitness levels of the male studies is large: 49.7 $\pm$ 4.0 to 69.2 $\pm$ 4.0 $mL \cdot min^{-1} \cdot kg^{-1}$. The only female study to report VO_{2max} showed a lesser fitness level than any of the male-only studies: 47.4 $\pm$ 4.2 $mL \cdot min^{-1} \cdot kg^{-1}$. One possible reason for differences in male and female responses to EPOC is the amount of fat-free mass (FFM) in males versus females. Lamont et al. (2010) compared 1-h total EPOC responses following an exercise bout of 1 h at 50% of VO_{2max} in male and female subjects matched for age and fitness level. Results demonstrated a greater EPOC in males (Males: 45.3 $\pm$ 11.9, females 15.8 $\pm$ 5.3 $mL \cdot min^{-1}$). However, when EPOC was corrected for FFM, there were no significant differences between the sexes. A post-hoc test on our results revealed that when EPOC was corrected for FFM, there was no significant difference between men and women following MOD and HIAI exercise ($p = 0.162$). There is a difference in male and female response to exercise in our results and in Lamont et al. (2010) due to the difference in FFM. Due to the low number of subjects who completed this study, this should be confirmed in a study with a larger number of subjects that accounts for FFM and fitness level.

EPOC Relationship to VO_{2max}

In this study, there was no correlation between fitness level and ΔEPOC, despite the hypothesis that those with greater fitness levels would show a decrease in ΔEPOC. One limitation in the study was the narrow range of VO_{2max} values, with only one subject exceeding 45 $mL \cdot min^{-1} \cdot kg^{-1}$. Short and Sedlock (1997) demonstrated that trained subjects (males: 56.7, females 48.6 $mL \cdot min^{-1} \cdot kg^{-1}$) had a shorter EPOC duration than untrained (males: 39.8, females 35.9 $mL \cdot min^{-1} \cdot kg^{-1}$), and when exercising at an absolute intensity EPOC was significantly lower than untrained. When exercise was prescribed at the same intensity relative to each subject's VO_{2max}, EPOC responses were not significantly different. Sedlock et al. (2010) performed similar testing and found similar results in terms of relative and absolute intensity by training individuals and comparing them to a control. Our findings demonstrate that when the prescribed intensity is relative to each individuals' fitness level, no difference in EPOC is observed.

Conclusion

Though this study contained significant findings, more subjects are needed to increase the power of these results, particularly when examining the correlation between fitness level and ΔEPOC, and comparing males to females. We also assume that all of the subjects did not exercise, or consume alcohol or caffeine 24 hours prior to the testing time, and that diet remained the same for the 24 hours prior to each testing session. Because resting VO_2 was not significantly different prior to each condition, these factors most likely did not have a significant effect on resting metabolism. It is recommended that future studies contain larger numbers of males and females with a larger range of fitness levels to support the findings above. It was also difficult to compare absolute

EPOC values because many of the studies use various lengths of EPOC measurement, as well as including feeding times. From the literature reviewed, it might make comparisons easier if one general established duration and method of collection (e.g., not including a feeding time if collection is less than 2 hours) was used following submaximal exercise lasting less than 50 minutes. In addition, the lack of inclusion of female subjects makes it difficult to apply these results to the general population. There seems to be a clear difference between male and female subjects in what little literature exists. Although we compared separate studies with similar workloads and intensities, future studies should compare males and females controlling for menstrual phase, fitness level, and FFM. Finally, in this study, EPOC consisted of only 3.7% of the net oxygen consumption during the MOD exercise trial, and only 5.8% of the net oxygen consumption during the HIAI trial. While the HIAI EPOC was significantly greater than the MOD EPOC, neither EPOC contributes substantially to the overall energy cost of the exercise, and is thus unlikely to have significant effects on weight loss or other health outcomes. When prescribing exercise to those who are less fit or are new to exercising, it is important to consider the intensity and time that is most tolerable when the primary goal is weight loss.

Appendix A

Screening Questionnaire

Name________________________________ Sex_______ Age_____ Date:_______________________

Height _____________ Weight__________

Phone _____________________ Email ____

Please answer the questions below to the best of your ability:

1. Do you have any form of heart or vascular diseases, pulmonary disease, diabetes, known high cholesterol, or known hypertension (high blood pressure)? If yes, list below:

2. Do you use any form of tobacco product?

3. Have you exercised in the past three months? If so, how many days per week do you exercise? About how many minutes per week do you exercise?

4. Has anyone in your immediate family (father, brother, mother, sister) have cardiovascular disease or an event (heart attack)? If so, what was their age at the time of event or diagnosis?

5. Do you take any medication that may affect your heart rate such as beta-blockers or prednisone?

6. Have you had any recent changes in weight loss or gain in the past month (>5lbs)?

7. If you are female, do you think you may be pregnant?

8. If you are female, do you have regular periods?

Appendix B

INFORMED CONSENT DOCUMENT

OLD DOMINION UNIVERSITY

PROJECT TITLE: The effects of fitness level and sex on EPOC following high intensity interval and moderate intensity aerobic exercise.

INTRODUCTION

The purposes of this form are to give you information that may affect your decision whether to say YES or NO to participation in this research, and to record the consent of those who say YES. The research project will take place in the Human Performance Lab at Old Dominion University at the Student Recreation Center room 2003.

RESEARCHERS

David P. Swain, PhD, Responsible Project Investigator
Rachel Simmons
William Perez

DESCRIPTION OF RESEARCH STUDY

The elevated level of metabolism and caloric expenditure after cessation of exercise is known as excess post-exercise oxygen consumption (EPOC). Maximizing EPOC following various exercises can potentially be utilized for greater fat loss. In addition, EPOC magnitudes vary in the between sex and fitness level. Understanding these differences is important for exercise prescription, particularly if the goal is to maximize caloric expenditure for fat loss. The purpose of this study is to examine the caloric expenditure and oxygen consumption after a bout of moderate intensity cycling exercise, and after a bout if high intensity interval cycling exercise.

If you decide to participate, a 24-h dietary analysis form will be given to you to fill out for the 24 hours prior to each visit. All female subjects will also be given a menstrual cycle questionnaire to determine menstrual cycle phase; their start date will begin the day after their last period. In the following three visits, you will arrive at the same time of day for your first second and third visits.

Testing sessions: There are a total of three visits. The scheduled first visit will consist of collecting height, weight, and body composition using air displacement plethysmography. You will then rest seated for twenty minutes followed by a 10-minute resting metabolic rate test. You will then become familiarized with the Veletron cycle ergometer and the facemask used in VO2 assessment. You will then complete a maximal oxygen consumption test on the cycle ergometer increasing intensity in 3-minute intervals. Visit one should last no more than one hour. Visits two and three will be randomized and last approximately three hours each. You will be required to report to the lab at the same time on visits two and three: one visit will consist of a 7-minute warm up and a 40-minute moderate intensity cycle bout. The other visit will consist of the same warm-up followed by ten intervals of exercise: two minutes at maximal aerobic capacity followed by one minute of rest. After both exercise sessions, you will be required to spend thirty minutes with the facemask on immediately after exercise after being given water. Once finished with this initial measurement, you will be asked to re-don the facemask every thirty minutes for 10 minutes for a total of two hours. You will be asked to sit quietly and may bring a book or school work to do inbetween donning the facemask.

EXCLUSIONARY CRITERIA

You should have completed a health screening questionnaire to determine if you are eligible for the study. You must be between the ages of 18 and 45 years and you should not have cardiovascular disease, pulmonary disease, diabetes mellitus, or any symptoms of these diseases (to the best of your knowledge). If you are taking any medications or drugs that affect heart rate (beta-blocker, nicotine, prednisone), you may not participate in the study. If you think you may be pregnant or have irregular menstrual cycles, you may not participate in the study. If you have had a recent calorically restrictive diet and have cycled in weight loss or gain in the past month (> 5lbs) you may not participate in the study.

RISKS AND BENEFITS

RISKS: If you decide to participate in this study, then you may face risks associated high intensity exercise. These risks include but are not limited to physical discomfort, nausea, dizziness, fainting. Also, you will be carefully monitored during exercise testing and any and all preventative measures will be taken to avoid these risks. Should an emergency situation arise, EMS would be contacted and immediate and appropriate action will be taken.

BENEFITS: You may benefit by learning your body composition, aerobic fitness capacity, and your resting metabolic rate.

COSTS AND PAYMENTS

You will not receive any compensation for your time and participation in this study.

NEW INFORMATION

If the researchers find new information during this study that would reasonably change your decision about participating, then they will give it to you.

CONFIDENTIALITY

Information collected about you will be kept confidential by the researchers. The results of this study may be used in reports, presentations, and publications; but the researcher will not identify you.

WITHDRAWAL PRIVILEGE

It is OK for you to say NO. Even if you say YES now, you are free to say NO later, and walk away or withdraw from the study -- at any time. Your decision will not affect your relationship with Old Dominion University or otherwise cause a loss of benefits to which you might otherwise be entitled. The researchers reserve the right to withdraw your participation in this study, at any time, if they observe potential problems with your continued participation.

COMPENSATION FOR ILLNESS AND INJURY

If you say YES, then your consent in this document does not waive any of your legal rights. However, in the event of injury or illness arising from this study, neither Old Dominion University nor the researchers are able to give you any money, insurance coverage, free medical care, or any other compensation for such injury or illness. In the event that you suffer injury or illness as a result of participation in this research project, you may contact Dr. George Maihafer, the chair of the Institutional Review Board, at 757-683-4520, or the Office of Research, at 757-683-3460, who will be glad to review the matter with you.

VOLUNTARY CONSENT

By signing this form, you are saying several things. You are saying that you have read this form or have had it read to you, that you are satisfied that you understand this form, the research study, and its risks and benefits. The researchers should have answered any questions you may have had about the research. If you have any questions later on, then the researchers should be able to answer them:
Dr. David Swain, 757-683-6028
Ms. Rachel Simmons 757-651-1154

If at any time you feel pressured to participate, or if you have any questions about your rights or this form, then you should call Dr. George Maihafer at 757-683-4520 or the Old Dominion University Office of Research, at 757-683-3460.

And importantly, by signing below, you are telling the researcher YES, that you agree to participate in this study. The researcher should give you a copy of this form for your records.

Subject's Printed Name & Signature	**Date**

INVESTIGATOR'S STATEMENT

I certify that I have explained to this subject the nature and purpose of this research, including benefits, risks, costs, and any experimental procedures. I have described the rights and protections afforded to human subjects and have done nothing to pressure, coerce, or falsely entice this subject into participating. I am aware of my obligations under state and federal laws, and promise compliance. I have answered the subject's questions and have encouraged him/her to ask additional questions at any time during the course of this study. I have witnessed the above signature(s) on this consent form.

Investigator's Printed Name & Signature	**Date**

Appendix C

Menstrual Cycle Phase Questionnaire

You will be asked to report the last known date of your period, and begin the first testing session approximately one day after your next period. You will also be asked to complete both exercise testing sessions within two weeks (14 days) of the first testing session.

1. When was the last day of your last period?

2. Assuming regularity, will you be able to participate in the time frames mentioned above?

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