

THE EFFECT OF A CPET FAMILIARIZATION SESSION ON AEROBIC CAPACITY  
(VO<sub>2</sub>MAX) IN SEDENTARY MIDDLE-AGED FEMALES: PRELIMINARY RESULTS

Christopher Ryan Brooks

A thesis submitted to the faculty of the University of North Carolina at Chapel Hill in partial fulfillment of the requirement for the degree of Master of Arts in the Department of Exercise and Sport Science (Exercise Physiology).

Chapel Hill  
2017

Approved By:

Claudio L. Battaglini

Erik D. Hanson

Eric D. Ryan

© 2017  
Christopher Ryan Brooks  
ALL RIGHTS RESERVED

## **ABSTRACT**

Christopher Ryan Brooks: The Effect of a CPET Familiarization Session on Aerobic Capacity ( $VO_{2MAX}$ ) in Sedentary Middle-Aged Females  
(Under the direction of Claudio L. Battaglini)

Cardiorespiratory capacity ( $VO_{2Max}$ ) is assessed by performing a cardiorespiratory exercise test (CPET). Research postulates that a learning effect can compromise the accuracy of the CPET. Healthy populations produce high CPET reproducibility, whereas clinical populations are inconclusive. The purpose of this study was to examine the effect of a familiarization session on aerobic capacity in sedentary middle-aged females. This study recruited 6 female participants ages 35-65. Test protocol consisted of a familiarization session, a  $VO_{2Max}$  pre-test, and a  $VO_{2Max}$  post-test.  $VO_{2Max}$ , respiratory exchange ratio (RER), and maximal heart rate ( $HR_{Max}$ ) were analyzed using a paired samples t-tests. Relative and absolute  $VO_{2Max}$  significantly decreased by -9.68% ( $p=0.03$ ) and -9.33% ( $p=0.03$ ), from pre to post. No significant differences were observed for RER and  $HR_{Max}$  ( $p=0.23$ ;  $p=0.30$ ). This potentially indicates the familiarization effectively mitigated a learning effect; however, due to the small sample size a larger sample is necessary to re-evaluate these preliminary findings.

## **ACKNOWLEDGEMENTS**

I would like to extend my deepest gratitude and thanks to Dr. Claudio Battaglini, Dr. Erik Hanson, Dr. Eric Ryan, Jordan Lee, and Chad Wagoner for all of their help and support in the process of writing and conducting my thesis work. I would also like to thank the University of North Carolina for the use of their labs and facilities to conduct my research.

## TABLE OF CONTENTS

|   |      |
|---|------|
| LIST OF TABLES .....  | vii  |
| LIST OF FIGURES .....   | viii |
| LIST OF ABBREVIATIONS .....   | ix   |
| Chapter   |      |
| I. INTRODUCTION .....   | 1    |
| Statement of purpose .....  | 7    |
| Research questions .....  | 8    |
| Hypotheses .....  | 8    |
| Delimitations .....   | 8    |
| Limitations .....   | 9    |
| Significance .....  | 9    |
| II. REVIEW OF LITERATURE .....  | 11   |
| A Brief Overview of VO <sub>2</sub> Max and Cardiopulmonary Fitness ..... | 11   |
| Strength and Its Role in Aerobic Capacity .....                           | 14   |
| Cardiorespiratory Fitness in Clinical Populations .....                   | 17   |
| Cardiorespiratory Fitness in Healthy Populations .....                    | 22   |
| Study Rationale Based on Literature/Future Directions .....               | 24   |
| III. METHODOLOGY .....  | 26   |
| Subjects .....  | 26   |
| Overview .....  | 27   |

|   |    |
|---|----|
| Instrumentation .....   | 28 |
| Procedures .....  | 29 |
| Data Analysis .....   | 34 |
| IV. RESULTS .....   | 35 |
| Subjects .....  | 35 |
| Hypothesis Analysis.....  | 36 |
| Exploratory Analysis .....  | 37 |
| V. DISCUSSION .....   | 39 |
| Overview .....  | 39 |
| Aerobic Capacity .....  | 40 |
| Future Research .....   | 46 |
| VI. APPENDIX 1.1 STUDY BROCHURE:.....                               | 47 |
| VII. APPENDIX 1.2: PRE-ASSESSMENT GUIDELINES.....                   | 48 |
| VIII. APPENDIX 1.3: PHYSICAL ACTIVITY READINESS QUESTIONNAIRE ..... | 49 |
| IX. APPENDIX 1.4: MEDICAL HISTORY QUESTIONNAIRE .....               | 50 |
| X. APPENDIX 1.5: DATA COLLECTION SHEET .....                        | 54 |
| REFERENCES .....  | 56 |

## LIST OF TABLES

|   |    |
|---|----|
| Table 1 (Standard Balke Protocol For Normal Sedentary Subjects) .....   | 3  |
| Table 2 (Bruce Protocol For Young Active Subjects) .....                | 3  |
| Table 3 (Subject Characteristics; n = 6 (mean $\pm$ SD)).....           | 36 |
| Table 4 (CPET Variables by Subject (mean $\pm$ SD)).....                | 37 |
| Table 5 (Exploratory Strength Variable by Subject (mean $\pm$ SD))..... | 38 |

## LIST OF FIGURES

|                                |    |
|--------------------------------|----|
| Figure1 (Study Overview) ..... | 28 |
|--------------------------------|----|



## LIST OF ABBREVIATIONS

|                       |   |
|-----------------------|---|
| A-VO <sub>2diff</sub> | The Difference in Arterial Oxygen Content and Venous Oxygen Content |
| CPET                  | Cardiopulmonary Exercise Test                                       |
| CSA                   | Cross Sectional Area  |
| EMG                   | Electromyography  |
| HR                    | Heart Rate  |
| HR <sub>Max</sub>     | Maximal Heart Rate  |
| PA                    | Peak Amplitude  |
| PT                    | Peak Torque   |
| Q                     | Cardiac Output  |
| RER                   | Respiratory Exchange Ratio  |
| RPE                   | Rate of Perceived Exertion  |
| SV                    | Stroke Volume   |
| US                    | Ultrasound  |
| VCO <sub>2Max</sub>   | Maximal Volume CO <sub>2</sub> Produced                             |
| VE <sub>Max</sub>     | Maximal Pulmonary Minute Ventilation                                |
| VL                    | Vastus Lateralis  |
| VO <sub>2</sub>       | Volume of Oxygen Consumed   |
| VO <sub>2Max</sub>    | Maximal Oxygen Uptake   |
| VO <sub>2Peak</sub>   | Highest Volume of Oxygen Consumption Attained During CPET           |

## CHAPTER I

### INTRODUCTION

#### *Aerobic Capacity*

Maximal oxygen uptake was coined in the 1920's by A.V. Hill (Hill and Lupton, 1923) and defined as the maximal capacity ( $VO_{2Max}$ ) of the body to transport and utilize oxygen during exercise.  $VO_{2Max}$  can be assessed by performing a graded cardiopulmonary exercise test (CPET). It is considered by many exercise scientists to be the gold standard of cardiovascular fitness (Powers and Howley, 2012).  $VO_{2Max}$  testing has many implications within the field of Exercise Physiology and it is considered the best single measurement of overall fitness of an individual. In the scientific literature, an increase in  $VO_{2Max}$  is the most common method of demonstrating a training effect on improving overall fitness. In addition,  $VO_{2Max}$  is frequently used in the development of aerobic training exercise prescription. Furthermore, cardiorespiratory capacity has been associated with the development of chronic diseases (Morris et al., 1953), and all cause mortality in individuals with low  $VO_{2Max}$  (Myers et al., 2002; Gulati et al., 2005). Most recently,  $VO_{2Max}$  has been proposed to be an independent factor that predicts post-treatment complications and mortality in lung and bone marrow transplant cancer patients (Jones et al. 2008; Wood et al., 2013). Given these applications of CPET, there has been great interest in identifying the physiological factors that limit  $VO_{2Max}$  and determining the role of this variable in endurance performance (Howley et al., 1995) and many other applications associated with overall health.

### *Maximal Exercise Testing*

Oxygen uptake ( $\text{VO}_2$ ) is a function of two major components; the first is cardiac output (Q), which is the product of heart rate (HR) and stroke volume (SV). The second component of  $\text{VO}_2$  is arteriovenous difference ( $\text{A-VO}_{2\text{Diff}}$ ), which is the amount of oxygen taken up by the peripheral tissue expressed by difference between the arterial oxygen content to that of the venous oxygen content.

Performing a graded exercise test on a treadmill or on a cycle ergometer are two of the most common modes of assessing cardiopulmonary function. However the choice of any graded exercise test should always depend on the population of which is being tested (Powers and Howley, 2012). Graded protocols can either be maximal or submaximal. For maximal tests,  $\text{VO}_{2\text{Max}}$  is estimated with equations that allow for the calculation of  $\text{VO}_{2\text{Max}}$  from the last work rate achieved on the graded exercise test. During submaximal testing however,  $\text{VO}_{2\text{Max}}$  is calculated using heart rate that is plotted against work rate until the termination criteria of the age predicted maximal heart rate is reached (Powers and Howley, 2012). There are a vast array of testing protocols, maximal and submaximal, in which the progression of the test differs by manipulation of the intensity, grade, or speed at a given stage. Below in Table 1. and Table 2. are two examples of differing graded exercise testing protocols (Powers and Howley, 2012).

**Table 1: Standard Balke Protocol For Normal Sedentary Subjects (Balke, 1970)**

| Stage | METs | Speed (mph) | % Grade |
|-------|------|-------------|---------|
| 1     | 4.3  | 3           | 2.5     |
| 2     | 5.4  | 3           | 5.0     |
| 3     | 6.4  | 3           | 7.5     |
| 4     | 7.4  | 3           | 10.0    |
| 5     | 8.5  | 3           | 12.5    |
| 6     | 9.5  | 3           | 15.0    |
| 7     | 10.5 | 3           | 17.5    |
| 8     | 11.6 | 3           | 20.0    |
| 9     | 12.9 | 3           | 22.5    |

\*Stages last 2 minutes each.

**Table 2: Bruce Protocol For Young Active Subjects (Bruce, 1972)**

| Stage | METs | Speed (mph) | % Grade |
|-------|------|-------------|---------|
| 1     | 5.0  | 1.7         | 10      |
| 2     | 7.0  | 2.5         | 12      |
| 3     | 9.5  | 3.4         | 14      |
| 4     | 13.0 | 4.2         | 16      |
| 5     | 16.0 | 5.0         | 18      |

\*Stages last 3 minutes each.

There are five criteria used for determining whether or not an individual reached  $VO_{2Max}$ . These criteria include: 1) a plateau or decrease of  $VO_2$  with increase in work rate, 2) a blood lactate level greater than 8 mmols, 3) a respiratory exchange ratio (RER) of 1.1 or greater, 4) an RPE greater than or equal to 17, and lastly 5) a heart rate within 10 beats per minute of the predicted maximal heart rate. If three of these criteria are reached then it can be concluded that in fact the individual performing the test did achieved his/hers maximal oxygen uptake capacity known at  $VO_{2Max}$ . If these criteria are not reached instead of deeming it a maximal test, it is referred to as a  $VO_{2Max}$  test.

## *Reproducibility*

The criteria mentioned above are what exercise physiologists currently use to determine whether or not an individual has truly performed a maximal test. However, the accuracy and reproducibility of the results obtained during the tests can be affected by many factors and thus making it in some occasions difficult to interpret if the test in fact was a true maximal cardiopulmonary determination. Factors affecting a test's reproducibility can be broken down into two groups: 1) random error, which includes within-patient biological and temporal variations and device/operator/reporter variability, and 2) systematic error, which includes phenomena such as familiarization or peripheral fatigue (mainly during cycle ergometer testing protocols) which cause results to be very difficult to interpret and use (Shun-Shin, Francis, 2012; Barron et al., 2014). Examples of factors that may affect the overall outcome of a cardiopulmonary test include encouragement during the test, pre-test conditions in subjects, as well as the subject's familiarity with the equipment. In many cases of test-retest research, to see if an intervention has in fact worked with regards to improving aerobic capacity, the interpretation of the results are often questioned due to a potential learning effect of the initial test in relation to the post test. This issue is believed to be especially augmented in individuals who are not familiar with or have never performed this type of test before. Barron and colleagues (2014) calls into question this learning affect in his article "*Test-retest Repeatability of Cardiopulmonary Exercise Test Variables in Patients With Cardiac or Respiratory Disease*" which concluded that a familiarization effect was observed in a number of variables, with persistently better values on the second test. This effect however, was much smaller than in a study by Elborn et al. (1990), which showed a 17% increase in peak  $\text{VO}_2$  from test 1 to 2 in heart

failure patients, compared with only 2.5% in Barron et al. study (Barron et al., 2014; Elborn et al., 1990). What can be concluded from these two separate studies is that there is a difference between test-retest experiments, but to what degree and how large are these differences, is yet to be determined.

In 1989, Cox and colleagues (1989) demonstrated that in patients with obstructive lung disease, even though his subjects were not familiar with the cycling test, that there were no important learning effects of his test-retest procedure which produced relatively low duplicate error. There was no significant difference between the values of the first CPETs performed on a cycle ergometer. Reproducibility of the test was deemed good in that workload (4.5%),  $\text{VO}_{2\text{Max}}$  (3.5%), RER (3.4%), HR (3.7%),  $\text{VE}_{\text{Max}}$  (6.6%), and  $\text{VCO}_{2\text{Max}}$  (6.0%) were moderately reproducible. This indicates that there was no apparent learning effect observed in this patient population. In the same study, Cox and colleagues (1989), healthy control subject's variability from pre to post test was reported at approximately 3% and those with chronic airflow obstruction were reported at approximately 6%. In fact, when the subjects of this study were asked, 5 of the 11 subjects expected better performance on their second test because of the knowledge and experience gained during the first test. However, the study concluded that there was no impact on their maximal workload and maximal oxygen uptake, and thus, there seemed to be no indication of a learning affect (Cox et al., 1989). In another population with restrictive lung disease, Marciniuk and colleagues (1993) demonstrates some of the same concepts and concluded that practice exercise studies are not necessary in patients with restrictive lung diseases even if they have not previously exercised on a cycle ergometer (Marciniuk et al., 1993). Therefore, it appears that in patients with cardiopulmonary disease, a practice or familiarization session does not seem to affect subsequent tests due to a possible learning effect.

Conversely, even with numerous studies indicating no learning effect, the literature is unclear. In a similar population, those with chronic airflow limitations, subjects underwent a repeated walking test, Knox and coworker found a significant increase in walking distances with repeated testing (Knox et al., 1988). Swinburn also found in patients with chronic airflow limitations a 29% increase in performance between the first and fourth exercise test on a cycle ergometer (Swinburn et al., 1985). Obviously the literature seems to be inconclusive as to whether there is or is not a learning effect occurring during many various forms of CPET testing. To date, it appears that a learning effect is population specific and more research must be conducted to continue to shed light on this potential testing issue.

Although the evaluation of the learning affect and how it relates to exercise testing has been observed in many different populations in literature, mainly those with various lung diseases and those with various heart conditions, it has rarely been reproduced in overall healthy individuals. A familiarization session is believed to play a crucial role in limiting the learning effect that occurs across multiple testing sessions with the same equipment in different populations. Without a familiarization session, maximal oxygen uptake recordings could be negatively affected and possibly underestimates of the  $VO_{2Max}$  of individuals could occur. This could have a myriad of negative implications including underreporting of data values in literature (which could be the difference between significant and non-significance), low-recorded  $VO_{2Max}$  values in performance testing, and incorrect exercise prescriptions. A recent study by *Wagoner et al.* demonstrated that there was a significant increase in  $VO_{2Max}$  in sedentary middle-aged females after 2 weeks of lower body resistance training. The increase in  $VO_{2Max}$  was attributed to the ability of the subjects to pedal longer via reduced lower limb fatigue resulting from the 2 weeks of training (*Wagoner et al.* 2016). These findings show great applicability to clinical

populations in which certain fitness levels may be beneficial in the battling of various etiologies; however, a major limitation in Wagoner's study was the lack of a control group. Without a control group one is not able to confirm if the increase in  $\text{VO}_{2\text{Max}}$  was due to the strength training or due to the learning effect of the CPET.

### Purpose Statement

The purpose of this study was to evaluate the effects of familiarization on CPET performances in sedentary older women, to determine if there is a learning effect and how much different this possible learning effect can affect the assessment of cardiopulmonary capacity in this population. The reason sedentary older women were the target population in this study was because this study was designed to serve as a control group for the aforementioned *Wagoner et al.* (unpublished data). As a result of having no control group, the Wagoner study is subject to scrutiny due to a possible learning effect occurring. It could be argued that the subjects did not improve physiologically in performing these tests; rather they learned to perform the tests better and became more comfortable performing the tests. Another reason that sedentary older women were the target population in this current study is that it gives the best representation of how healthy individuals may be affected by the learning effect so that it can eventually be studied in clinical populations, such as cancer, which ultimately is the population of interest of our laboratory. It was hypothesized that there would not be a learning effect between the initial CPET and the final CPET. One would believe the values would differ slightly, however these differences would not be significant. It was also hypothesized that subjects would exhibit similar RER and HR at the same given intensity when comparing post-CPET with pre- CPET.



### Research Questions

RQ1. Will  $\text{VO}_{2\text{Max}}$  differ significantly from the pre-test and the post-test?

RQ2. Will RER differ significantly between pre-test and post-test?

RQ3. Will the  $\text{HR}_{\text{Max}}$  differ significantly between the pre-test and post-test?

### Hypotheses

H<sub>1</sub>: Subjects will not exhibit  $\text{VO}_{2\text{Max}}$  values that significantly differ between the first test and the last test.

H<sub>2</sub>: Subjects will not exhibit significantly different RER scores at any given intensity between the first and second CPETs.

H<sub>3</sub>: Subjects will not exhibit significant different  $\text{HR}_s$  at any given intensity between the first and second CPETs.

### Delimitations

- All subjects were female between 35-65 years of age.
- All subjects were cleared by a physician prior to participating in the study.

### Limitations

- The results of this study may only apply to those whom are women, sedentary, apparently healthy and between the ages of 35-65 years old. Results may not be applied to females of all ages and males.
- All subjects were recruited from Chapel Hill, NC and may not a true representation of the entire world population.
- Due to the selected age range and gender, menstrual cycle could affect study results in those whom were pre-menopausal.

### Significance

Test-retest reliability is an important factor to consider as it relates to selecting the most appropriate and precise testing protocols and instruments in research. The validity of cardiopulmonary exercise tests in exercise science studies is important, first and foremost, because it determines whether or not an exercise intervention actually had an effect on the subject's oxygen uptake ( $\text{VO}_2$ ); where maximum oxygen uptake ( $\text{VO}_{2\text{Max}}$ ) has been correlated with overall health, longevity, and even prescribed as main intervention in certain populations with chronic disease such as cardiovascular disease. Reliability reduces the effects of test-re-test limiting factors. Being able to minimize limitations in any testing protocol allows for more precise interpretation of the test results and ultimately the efficacy of an intervention. In populations where  $\text{VO}_{2\text{Max}}$  values may not be very high such as those with impaired heart and lung function as well as cancer populations, a precise evaluation on the “real” effect of an exercise intervention can be a difference between improving disease outcomes or even in the worst case scenario, death. Regardless of the population in question, the issue of being able to

confidently interpret the results of a test, in this particular study a CPET, and being able to confidently determine if a learning effect had or not an impact on the results of subsequent tests is necessary and paramount if one desires to confidently determine the accuracy of such tests as well as using this test as a mean to evaluate an intervention aimed to improve cardiopulmonary capacity.

## **CHAPTER II**

### **REVIEW OF LITERATURE**

For organizational purposes, Chapter II was organized into five sections: SECTION I. A brief overview of maximal oxygen uptake ( $\text{VO}_{2\text{max}}$ ) and cardiopulmonary fitness; SECTION II. Strength and its role in aerobic capacity; SECTION III. Cardiopulmonary fitness in clinical populations; SECTION IV. Cardiopulmonary fitness in healthy populations, and SECTION V. Study rationale based on literature/future directions. Results discussed from the articles within this section have been selected to provide a history and proper rationale for the study at hand.

#### **Overview of Maximal Oxygen Uptake ( $\text{VO}_{2\text{max}}$ ) and Cardiopulmonary Fitness**

Maximum oxygen uptake ( $\text{VO}_{2\text{Max}}$ ) is defined as the highest rate at which oxygen can be taken up and utilized by the body during severe exercise. It represents the integrated capacity of the pulmonary, cardiovascular and muscle systems to uptake, transport and utilize  $\text{O}_2$  (Poole et al., 2008). It is one of the main variables in the field of exercise physiology, and is frequently used to indicate the cardiorespiratory fitness of an individual. In the scientific literature, an increase in  $\text{VO}_{2\text{Max}}$  is the most common method of demonstrating a training effect. In addition,  $\text{VO}_{2\text{Max}}$  is frequently used in the development of an exercise prescription. These applications of  $\text{VO}_{2\text{Max}}$  are why there has been such great interest in identifying the physiological factors that

actually limit  $\text{VO}_{2\text{Max}}$  and in determining the role of this variable in endurance performance (Howley and Bassett, 1999). The term “maximal oxygen uptake was define in 1923 by Hill and Lupton. They postulated that 1) there is an upper limit to oxygen uptake, □ 2) there are inter-individual differences in  $\text{VO}_{2\text{Max}}$ , 3) a high  $\text{VO}_{2\text{Max}}$  is a prerequisite for success in middle- □and long-distance running, and 4) that  $\text{VO}_{2\text{Max}}$  is limited by ability of the cardiorespiratory □system to transport  $\text{O}_2$  to the muscles. □Hill and Lupton also stated that, “however much the work rate be increased beyond this limit, no further increase in oxygen intake can occur” in reference to this upper limit of oxygen uptake (Hill and Lupton, 1923). These postulations demonstrate that  $\text{VO}_{2\text{Max}}$  has a physiological limit which is measurable, it is individualized to each person, it can somewhat predict success in long distance aerobic evens, and it can be limited due to various factors within our body. These postulations beg numerous questions: What specifically causes the limit  $\text{VO}_{2\text{Max}}$ ? Can this value be reached through some type of intervention, and if so, how? Do various diseases or epidemiological problems affect maximal oxygen uptake? Many of the questions asked here frankly are the reason as to why  $\text{VO}_{2\text{Max}}$  has not only usefulness in strictly a research setting, but even more so in a clinical setting.

The measurement of maximal exercise does not necessitate an individual being taken to a maximal intensity. This value can be estimated by using a submaximal exercise test. Submaximal estimates of  $\text{VO}_{2\text{Max}}$  are based on the linear relationship among heart rate, workload, and  $\text{VO}_{2\text{Max}}$  (Morrow et al., 2000). The exercise test requires much less effort than compared to those of maximal exercise. One great example of a submaximal exercise protocol is the Astrand-Rhyming nomogram (Astrand and Rhyming, 1954). It was originally established as a cycle ergometer test that coordinates workload and heart rate responses into a prediction of  $\text{VO}_{2\text{Max}}$ . There are also other field methods used to predict  $\text{VO}_{2\text{Max}}$  and aerobic capacity such as distance

runs/walks, step tests, the Rockport 1-mile walking test, etc (Morrow et al.). The decision on which type of test to use (maximal, submaximal, or field) really depends on the population or individual being assessed and what is the safest and most effective way of assessing aerobic capacity.

Maximal testing criteria include: a plateau or decrease in  $\text{VO}_2$  with an increase in work rate, respiratory exchange ratio (RER) (1.00, 1.10 or 1.15); heart rate within 10 bpm or within 5% of the age-predicted maximum; and blood lactate level of greater than 8–10 mmols. (Poole et al., 2008). Poole et al. concludes that utilization of ramp or incremental exercise tests precludes achievement of a  $\text{VO}_{2\text{Max}}$  plateau prior to exhaustion in a substantial proportion of individuals. Investigators have therefore resorted to a selection of secondary criteria to provide confidence that  $\text{VO}_{2\text{Max}}$  has in fact been achieved. This investigation demonstrated that use of established secondary criteria of RER, greater than or equal to 1.10 or 1.15, a  $\text{HR}_{\text{max}}$  within 10 bpm and/or a blood lactate greater than or equal to 8 mmols can lead either to a significant under measurement of  $\text{VO}_{2\text{Max}}$  or rejection of a large proportion of participants who may actually have achieved  $\text{VO}_{2\text{Max}}$ . On the basis of this article it was concluded that these secondary measurements might not be as accurate as once believed.

A comprehensive overview of  $\text{VO}_{2\text{Max}}$  testing and various issues associate with the test was investigated by Bassett and Howley in an article titled *Limiting Factors for Maximum Oxygen Uptake and Determinants of Endurance Performance*. This article concluded that oxygen delivery, not skeletal muscle oxygen extraction, is the primary limiting factor for  $\text{VO}_{2\text{Max}}$  during exercise. This article also points out that metabolic adaptations in skeletal muscle are critical for improving endurance performance. Endurance training can actually cause an increase in mitochondrial enzyme, thus improving performance by enhancing fat oxidation. Another

observation of this article is that it is necessary to decrease lactic acid accumulation at a given  $\text{VO}_2$ , in order to achieve a higher  $\text{VO}_{2\text{Max}}$ . Running economy can also affect endurance performance (Bassett and Howley, 2000). All these factors play a role in the determination of  $\text{VO}_2$  and thus play a role in the determination of reproducing this maximal exercise test measurements. One factor neglected in a considerable amount of articles discussing  $\text{VO}_{2\text{Max}}$  is the role that the learning effect may play in these measurements.

### **Strength and Its Role in Aerobic Capacity**

The underestimation of  $\text{VO}_{2\text{Max}}$  discussed by Poole et al., brings about other interesting questions. One of which is whether or not other factors may lead to an underestimation of  $\text{VO}_{2\text{Max}}$ . It has been debated and shown within literature that sarcopenia may have a negative effect on  $\text{VO}_{2\text{Max}}$ . Sarcopenia is the natural loss of muscle mass and strength with age. All humans lose muscle mass and function as they age. This is true even of elite athletes who, although they continue to be physically active and perform at levels well above those of sedentary adults, demonstrate a decline in lean tissue with age. Aerobic capacity declines with age in congruence with this loss in muscle mass. This is reflected as both a decline in muscle and in cardiopulmonary reserves. Cross-sectional data also show that body cell mass is systematically lower in older adults than in middle-aged or young adults (5–7) and so is strength. The decline in cell mass with age is largely due to loss of muscle mass (Roubenoff and Hughes, 2000). Frontera et al., investigated 200 healthy subjects in a cross sectional fashion between the ages of 45 and 75, using the isokinetic strength testing of the elbow and knee extensors. The results show that a significant component of the age-related muscle weakness is a loss of muscle mass (Frontera et al., 1991). This decrease in muscle mass ultimately leads to a decrease in force production and can have evident implications when considering aerobic capacity testing

completed on a cycle ergometer. Another study by Hartman et al., aimed to determine the effect resistance training has on metabolic economy during typical activities of daily living in a geriatric population (Mean age = 66). 29 subjects participated in a 26-week heavy resistance-training program. Before and after the 26-week training intervention, heart rate and expiratory gases of each individual were measured while performing 3 tasks that would mimic common everyday activities encountered by this population. These tasks included: 1) walking at 3 miles per hour, 2) carrying a box to simulate holding a bag of groceries with 1 hand (30% of maximal isometric strength) while walking at 2 miles per hour and, 3) climbing stairs. Both strength and fat-free mass increased significantly after the training protocol. Oxygen cost decreased significantly by 6% for the carrying task whereas the RER decreased significantly for both walking and stair climbing. Heart rate decreased significantly for the carrying task. Subjects also reported a significant decrease perceived exertion during performance (RPE) of all functional task test conditions. This study suggests that a heavy-resistance training program might affect exercise economy during daily tasks and improve ease of physical activity. This could be a possible mechanism for increasing quality of life in an older/geriatric population (Hartman et al., 2007). As seen by Frontera and colleagues, the loss of muscle mass between the ages of 45-75 is a great contributor to sarcopenia and muscle weakness as one progresses in age. Hartman et al. then shows that heavy resistance training may improve some aspects of everyday activities in an older population. This leads to the rationalization that it may be possible that heavy resistance training can also attenuate some age related affects of sarcopenia as well in a middle-aged population.

These increases in muscle mass or attenuation of muscle mass loss may have direct implications to  $\text{VO}_{2\text{Max}}$  testing performance. If there is more muscle mass present for an



individual to recruit during exercise, then more force can be exerted by that individual during the exercise task at hand. This may especially true in an activity such as cycle ergometry, where the resistance within the bike pedals continually increases and necessitates more force to turn the pedals as stages progress. Being able to produce extra force may allow an individual to go to a higher stage during a maximal exercise protocol and thus achieve and higher  $VO_{2Max}$  values. Fleg and Lakatta investigate this very problem in, *The Role of Muscle Loss in the Age-Associated Reduction in  $VO_{2Max}$* . Specifically they investigated to what extent the decline in  $VO_{2Max}$  could be attributable to the age associated loss of metabolically active tissue, such as sarcopenia. In order to achieve this, creatinine excretion, an index of muscle mass, was measured in 184 non-obese individuals between the ages of 22 and 87 who performed a true  $VO_{2Max}$  during a graded exercise protocol. A positive correlation was found between  $VO_{2Max}$  and creatinine excretion in men as well as in women.  $VO_{2Max}$  also showed a strong negative linear relationship with age in men and women as well, which is expected.  $VO_{2Max}$  was normalized for creatinine excretion and expressed per milligram of creatinine excretion rather than in the customary fashion. When this was done only 14% of the decline in aerobic capacity was explicable by age compared to that of 60% when solely  $VO_{2Max}$  and age were compared in men. In women there were similar numbers, as the aerobic capacity explicable by age decreased from 50% to only 8%. In comparing the standard age regression of  $VO_{2Max}$  per kg body weight with that in which  $VO_{2Max}$  was normalized per mg creatinine excretion, the decline in  $VO_{2Max}$  between hypothetical 30 year old and 70 year old was reduce from 39% to 18% in men and from 30% to 14% in women. Essentially this means that a large portion of the age-associated decline in  $VO_{2Max}$  in non-endurance trained individuals can be attributable to loss in muscle mass (Fleg and Lakatta, 1988).

## **Cardiopulmonary Fitness in Clinical Populations**

Exercise testing is an important tool that can be used for early diagnosis of lung function decrements as well as determining treatment effectiveness in clinical populations where there may be impaired lung function. In 1989, Cox et al. designed a study to examine the reproducibility and the learning effect of an incremental exercise testing. Specifically, Cox and colleagues studied cycle ergometer testing in patients with obstructive lung disease who were untrained, and who also had never done such a test prior to this study. This study examined 11 total patients with obstructive lung disease and was designed in a test retest fashion, as the subjects performed the first test then the second test 24 hours after. This study found that there were no significant differences between values of the first and second testing procedure. This indicates that there was in fact no learning effect that occurred during this cycle ergometer test. Results indicated good reproducibility for workload (4.5%),  $\text{VO}_{2\text{max}}$  (3.5%), RER (3.4%), and HR (3.7%) (Cox et al., 1989). These results indicate that despite subject not being familiar with the test and testing protocol, there were not major learning effects between the two testing procedures in each subject.

In 1993, Marciniuk, Watts, and Gallagher also examined reproducibility of variables measured during exercise testing in subjects with stable restrictive lung disease. 6 subjects were used for this study, all of which had also never performed an exercise test of this capacity. These subjects underwent a total of 3 exercise testing protocols on an electrically braked bike, which spanned over 28 total days, with each test separated by at least 7 days. Variables of interest were measured at the beginning of exercise, 40% of the maximum work rate, 70% of the maximum work, and at the end of exercise. The results of this study show that there was no significant difference in exercise duration, work rate, or the peak  $\text{VO}_2$  at end of exercise among the three

exercise sessions.  $\text{VCO}_2$ ,  $\text{W}$ ,  $\text{VT}$ ,  $f$ ,  $\text{SaO}_2$ ,  $\text{HR}$ , and Borg scores also did not differ among the three studies at end of exercise. Interestingly, there was also no significant difference in these variables at 40%  $\text{W}_{\text{max}}$  and 70%  $\text{W}_{\text{max}}$ . Comparison of these results, measured at 40%  $\text{W}_{\text{max}}$  and 70%  $\text{W}_{\text{max}}$ , show that reproducibility at submaximal levels of exercise parallels the reproducibility of measurements made at end of exercise (Marciniuk, Watts, and Gallagher, 1993). This suggests that, in addition to generating reproducible end of exercise measurements, measurements made at submaximal levels of exercise may also be used in evaluating the results of clinical exercise testing in these types of patients with lung impairment.

However, not all of the literature supports the notion that there is no learning effect in this particular population. Knox et al. reports in his study the “Reproducibility of Walking Test Results in Chronic Obstructive Airways Disease”, that there is in fact a learning effect that occurs during cardiopulmonary fitness assessments. The study looked at 12 patients over 3 consecutive days with 4 separate 5 minute walking distances per day with visual cues. The other group used in the study consisted of 24 subjects who performed 3 walks per week (on the same day) over 4 consecutive weeks. This study concluded that a learning effect was in fact evident, especially when walking tests were carried out over short time intervals. This study also collected data that shows that this learning effect may persist for up to 9 walking tests. When the walking tests were carried out over consecutive weeks rather than carried out over a shorter time interval, the learning effects were found to be less pronounced. Knox et al. recommends that, specifically with walking tests, that a minimum of 5 practice walking sessions should be used in order to familiarize the individual with the procedure when walking tests are performed over consecutive days. As for tests performed over consecutive weeks, the study recommends a minimum of 4 walking familiarization sessions to allow for the most accurate measurement of

fitness level. Another study by Swinburn et al., *Performance, Ventilation, and Oxygen Consumption in Three Different Types of Exercise Test in Patients With Chronic Obstructive Lung Disease* show a similar learning effect when it comes to repeat testing of cardiopulmonary capacity. In this study subjects performed a 12-minute walking test, a paced step test, as well as a cycle ergometer test. This study found that there was a progressive and significant increase in performance in all three types of exercise test between the first and fourth measurements (analysis of variance  $p < 0.01$ ). The overall increases in performance were: steps climbed 96%, duration of cycle exercise 29%, and 12 MWD 16%. Swinburn concludes that an important observation is the continuing improvement in exercise performance clearly seen in all three types of test with repeated testing. This is unlikely to have been due to physical training over such a short period, but it was not surprising in symptom limited tests such as these because increasing familiarity with the tests may have led to an increase in confidence and therefore motivation (Swinburn, 1985). An important note from this study is that the walking test had the lowest percent increases and seems to be the most reproducible between these 3 tests. In most cases, some form of walking is completed each day by individuals and is a familiar task to most people. A step test or a cycle ergometer may however not be as familiar to the average individual.

These results and conclusions of Knox and colleagues and Swinburn and colleagues show that in clinically compromised populations there does seem to be a learning effect occurring. However, Cox and colleagues and Marciniuk, Watts, and Gallagher show that there is not a significantly large difference between specific variables associate with cardiopulmonary exercise testing. Given the nature of this clinical population and the fact that many individuals have never taken part in a graded cardiopulmonary exercise test, a learning effect could very well be occurring in this subset of the population. This data only supports the necessity for a

familiarization session when testing subjects with an exercise test that may not be familiar to them.

In 2014, Barron et al. investigated the different measures of test-retest reliability in patients with cardiac and respiratory disease. A familiarization effect was observed, however it was much smaller than had been previously reported. This study looked at a myriad of cardiorespiratory variables. Out of the myriad the variables observed, Peak  $\text{VO}_2$ , anaerobic threshold (AT), peak heart rate (HR), double product (DP), peak minute ventilation and respiratory frequency, and peak work rate showed significant positive differences between tests 1 and 2. Only a 2.5% increase was seen in this study between test 1 and test 2 compared to another study by Elborn et al. that saw as much as a 17% increase in  $\text{VO}_{2\text{Max}}$  in heart failure patients (Barron et al., 2014). One key difference to note here between the two tests however is the use of a treadmill in the Elborn study versus the use of a cycle ergometer in Barron's study. Barron concludes that the majority of variables calculated in this study from cardiopulmonary exercise testing shows excellent test-retest reliability. Peak  $\text{VO}_2$ , OUES,  $\text{O}_2$  pulse, peak circulatory power, and the  $\text{VE}/\text{VCO}_2$  ratio at nadir show excellent test-retest reliability. Measures of the anaerobic threshold and the  $\text{VE}/\text{VCO}_2$  slope show good test-retest reliability.

Elborn et al., an aforementioned study, also assessed whether or not there was a learning effect that occurred in cardiopulmonary exercise testing in cardiac compromised patients. The purpose of this study was to evaluate the reproducibility of peak  $\text{VO}_2$ , exercise time and other cardiopulmonary parameters during repeated treadmill exercise testing in patients with moderate to severe chronic cardiac failure patients (Elborn et al., 1990). 30 total subjects with cardiac function impairment performed 3 cardiopulmonary exercise tests with at least 2 weeks of separation between each test. Changes over the three tests at rest, at a submaximal intensity, and

at maximal intensity were analyzed. There were no significant changes in resting parameters over the three tests. Exercise time and the stage during which the test was terminated both significantly increased between tests 1 and 2. No difference was observed between tests 2 and 3, during which the values were again significantly greater than test 1. At maximal exercise, there was no significant difference in heart rate, systolic blood pressure or respiratory rate responses. There was a significant increase in  $\text{VO}_2$  as was peak VE. There was no significant change in perceived exertion score at maximum exercise. This study by Elborn shows that all the parameters measured during a cardiopulmonary exercise test at rest and during symptom-limited exercise were reproducible between the second and third test. However, the first exposure of patients with chronic cardiac failure to an exercise test such as this resulted in an underestimate of exercise time by about 20%. Patients also achieved a higher exercise stage in test 2, despite their perceived exertion being similar. Peak exercise  $\text{VO}_2$ ,  $\text{VCO}_2$  and VE were also significantly underestimated between the first and second test. Elborn postulates that there may be several factors that to the observed improvement in exercise performance between tests 1 and 2. The first explanation is that the patient becomes familiar with the technique of treadmill walking and adapts to the awkward breathing via a respiratory valve with the nose clipped. As this process is unnatural and somewhat uncomfortable, becoming more comfortable with this type of breathing after repeated tests seems logical. The initial fear is overcome, and confidence develops. Another explanation is a physical training effect, but because the tests were all at least 2 weeks apart, this seems unlikely. Since therapy and symptom status were unchanged, an improvement in cardiac function was also unlikely to have been a factor. Another important result of this study to observe is that an improvement between tests 1 and 2 was also observed at submaximal exercise levels, although the heart rate and blood pressure responses were similar. This indicates that

overall exercise performance was improved. These changes from the first test to that of the second test, not only in the respiratory/lung populations, but also here in populations with cardiac issues necessitates the need for that first exercise session to familiarize the patient with the testing procedures as well as the testing protocol to efficiently eliminate any discrepancies that may occur due to a learning effect.

### **Cardiopulmonary Fitness in Healthy Populations**

This learning effect phenomenon has also been studied to some degree in healthy subjects as well as elite athletes. One such example of this can be seen in *Reproducibility of Incremental Maximal Cycle Ergometer Tests in Healthy Recreationally Active Subjects* (Dideriksen et al., 2015). This study used a population of recreationally active triathletes who performed an average of 3 hours of endurance exercise per week for the last year. One particularly important note about this study is that none of the subjects had previously performed an incremental cycle ergometer test, however some were familiar with stationary bike riding. The 13 subjects in this study performed 3 identical cycle ergometer maximal exercise tests to determine  $\text{VO}_{2\text{Max}}$ . Because the study aimed to investigate whether a learning effect could be observed, no physiological familiarization to the experimental procedures or equipment were allowed. Instead, the subjects were simply carefully informed about the experimental procedures the day of the first test. The 3 tests were all separated by at least 48 hours as well. Coefficient of variation percentages and intraclass correlations were used to determine the reproducibility of  $\text{VO}_{2\text{Max}}$  and how time averaging intervals affected this value. This study concluded that there is high reproducibility of  $\text{VO}_{2\text{Max}}$  measures was obtained regardless of time-averaging interval (5–60 s), and no learning effect was observed. However, the absolute level of  $\text{VO}_{2\text{Max}}$  was higher using

shorter time-averaging intervals compared with longer.

Fielding et al. (1997) also investigated the reproducibility of  $\text{VO}_{2\text{Max}}$  exercises tests in healthy individuals, specifically in older women. 17 women were used for this study from 51 to 68 years old. Each subject performed 5 different exercise tests separated by at least 1 week's time. Subjects performed the Modified Bruce protocol on a motor driven treadmill. The protocol was as follows: with the following 3-min stages: Stage I (2 mph, 10% grade), Stage II (2.5 mph, 12% grade), Stage III (3.4 mph, 14% grade), and Stage IV (4.2 mph, 16%). Oxygen uptake was measured continuously and metabolic data was taken every 30 seconds. Differences between each test were determined using an ANOVA. Pearson's correlation coefficient was also used to determine agreement between tests. Reliability estimates for each variable were also calculated using intraclass correlation coefficients.  $\text{VO}_{2\text{Max}}$  values were found to be consistent between all tests, with the mean coefficient of variation being 6.5%. Mean maximal ventilation was also not significantly different between tests. RER and maximal HR reached between tests was also not significant. A Person product correlation was also used to determine the agreement between the tests. For each given measurement there were significantly high levels of agreement between tests with  $r$  values ranging from 0.70-0.89. In addition, intraclass correlation coefficients for a single test are high and suggest good agreement between tests. In general, the associations did improve between test 2, test 3 and test 4. This study concluded that the Bruce treadmill protocol for women who are older (51-68 years old) produces highly reproducible results with  $\text{VO}_{2\text{Max}}$ , maximum heart rate, as well as ventilation. It is suggested that performance of only a single  $\text{VO}_{2\text{Max}}$  test is sufficient to assess aerobic capacity in middle-aged and older women.



## **Study Rationale Based on Literature/Future Directions**

Given the above literature, it is logical as to why the testing of middle-aged sedentary older women is important. It is necessary to determine the reproducibility of  $\text{VO}_{2\text{Max}}$  exercise tests in this population with hopes that future studies that use this test measurement in an attempt to quantify aerobic capacity can be precisely interpreted. While testing of healthy subjects generally seems to be in agreement with each other, specific clinical populations such as lung and heart diseased patients is not so clear. Determining the reproducibility of this test has numerous ramifications in research and the clinical setting. First, it would decrease the number of tests necessary to be sure that a precise measurement of  $\text{VO}_2$  value was obtained. Many times a familiarization session (or multiple) is needed to mitigate learning affect that may be present in a test-retest situation. In clinical populations where such things as sitting up or walking are difficult, this does not seem feasible. Another issue associate with this reproducibility is the accuracy of values reported within literature. Underestimation of  $\text{VO}_2$  was clearly demonstrated in the aforementioned study by Poole et al. (2008) and could lead to misrepresentation of data within literature as well as incorrect aerobic capacity assessments for athletes and clinical populations. The choice of sedentary older women was purposeful in nature in that this group was selected due to the age range and activity style and most likely be experiencing sarcopenia, which somewhat is similar to women who have been diagnosed with breast cancer. It is our intent to reproduce this study in women diagnosed with breast cancer so to be able to better understand the potential importance to conduct or not a familiarization session with these women to maximize the accuracy of the test and therefore better interpret the effects of exercise intervention in this cancer population. To be clear, the evidence found in this study is not directly generalizable to the breast cancer population, but knowledge obtained about sedentary middle

aged women will hopefully provide the rationale to further explore the findings in a similar population of women with similar age who have been diagnosed with breast cancer. Few studies have investigated this phenomenon in middle-aged women of in any cancer population; therefore it would add to the wealth of knowledge in the field of exercise as it relates to assessment of cardiopulmonary capacity.

## **CHAPTER III**

### **METHODOLOGY**

#### **Subjects**

Recruitment for the study was completely voluntary, as subjects were made aware of the study via flyers, emails, phone calls, as well as face-to-face interaction with research team members. Recruitment sites included areas that fell within that of Chapel Hill, North Carolina. Approval from the Institutional Review Boards in Exercise and Sport Science and School of Medicine at UNC-Chapel Hill were obtained from all subjects before participating in any aspect of the study.

All subjects participating in the study were between 35 to 65 years of age and sedentary. The sedentary nature of the participants was defined by not having participated in regularly scheduled exercise more than once a week for at least 6 months prior to beginning the study. Subjects were enrolled in the study if they present no cardiopulmonary and musculoskeletal disease that would preclude their participation in any aspect of the study as determined by a physician physical evaluation prior to any testing. Subjects were screened for exclusion based upon the criteria presented by the American College of Sports Medicine (ACSM) as contraindications to exercise testing (Pescatello & American College of Sports, 2014).

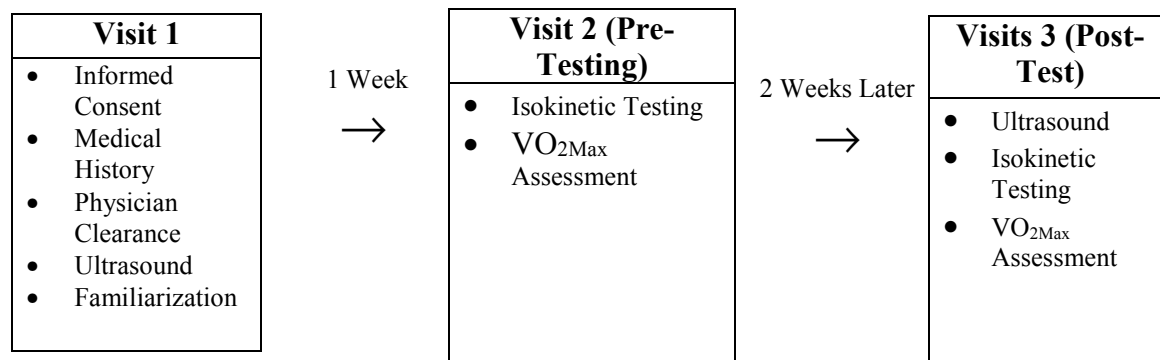
## Study Overview

This study was designed to collect data in a group of Sedentary Middle-Aged Females following a similar protocol of an unpublished Master's Thesis completed at UNC Chapel Hill titled, *The Effect of a Two-Week Lower Body Resistance Training Protocol on Aerobic Capacity ( $VO_{2Peak}$ ) in Sedentary Middle-Aged Females* (Wagoner et al. 2016). The difference between Wagoner et al. study and the current study was that the group of Sedentary Middle-Aged Females that were recruited to participate in the current study did not undergo an exercise intervention as subjects did in the Wagoner et al. 2016 study. In Wagoner et al. 2016 study, the researchers employed a two-week resistance training exercise intervention aimed to provide subjects a better muscular activation capacity believed to allow for the subjects improve their performance in a post CPET administered just two weeks after the pre CPET. Since Wagoner et al. study did not have a control group; the current study was designed to examine the issue associated with a potential learning effect that could have potentially confounded Wagoner et al. study where the improvements observed in  $VO_{2Max}$  may or may not have been attributed to the training and perhaps to the learning effects of testing.

The study procedures, parameters, and equipment used in Wagoner et al. study were exactly the same as the study design and procedures of this current study with the exclusion of a 1 rep maximum (1RM) tests, that were performed in Wagoner et al. with the goal of informing the training loads for the study intervention and the exercise intervention itself.

Figure 1 below is a graphic representation of the study events.

**Figure 1. Study Timeline**



## Instrumentation

### Anthropometric / Screening

Height was measured to the nearest 0.1 cm via a portable stadiometer (Perspective Enterprises, Portage, MI USA), and mass measured to the nearest 0.1 kg via a mechanical scale (Detecto, Webb City, MO USA).

A medical history questionnaire (Department of Exercise and Sports Science) was used to log the subjects' medical history, age, race, and relative physical activity level within the past year. This was utilized in conjunction with the physical examination and resting electrocardiogram (EKG) in determination of subject safety to participate in the study. The resting EKG was conducted using a GE CASE Cardiosoft V. 6.6 ECG diagnostic system (General Electric, Palatine, IL USA). Additionally, resting blood pressure was measured

manually via a Diagnostix 700 aneroid sphygmomanometer (American Diagnostics Corporation, Hauppauge, NY USA) and a Littmann stethoscope (3m, St. Paul, MN USA).

### Cardiopulmonary Test (CPET)

VO<sub>2Max</sub> was assessed using a Parvo Medics TrueMax 2400 Metabolic System (Parvo Medics, Salt Lake City, UT USA) on a Lode electronically braked cycle ergometer (Lode, Gronigen, The Netherlands). The subjects' respiratory responses were obtained by the use of a Hans Rudolph 7450 Series V2 Respiratory Valve (Hans Rudolph Inc., Shawnee, KS, USA). Rate of perceived exertion (RPE) was assessed using a Borg 6-20 Rate of Perceived Exertion (RPE) scale. Heart rate was monitored via a Pacer Polar heart rate monitor (Polar Electro Inc., Lake Success, NY USA). Post CEPT lactate measurement was obtained 3 minutes post completion of the CPET using a portable lactate analyzer (Lactate Plus, Sports Resource Group, Hawthorne, NY).

### **Procedures**

All subjects reported to the Exercise Oncology Research Laboratory (EORL) and the Neuromuscular Research Laboratory (NMRL) on a total of three separate occasions related to familiarization and testing purposes. The first visit included a familiarization and the initial ultrasound (US) measurements. The second visit included a pre-testing session. The last visit included the post-testing session and another US measurement. Before reporting for testing sessions, subjects were asked to follow a set of pre-assessment guidelines and were questioned as

to if they in fact followed the guidelines upon arriving to the lab. These guidelines included maintaining a proper hydration status as assessed by an American Optical, Hand Held TS Meter (Keene, New Hampshire, USA) refractometer, being at least two hours fasted, no caffeine consumption at least eight hours prior, and no alcohol consumption at least twenty-four hours prior. All subjects within the study were required to undergo a physical screening and approval by a physician in accordance with a 12-lead EKG, medical history questionnaire, and PAR-Q form. All visits took place in the EORL and NMRL.

#### Visit One: Physical Screening, Ultrasound, and Familiarization of Testing Procedures

The first visit to the laboratory included signing of the informed consent form, completion of the medical history questionnaire, PAR-Q, and a 12-lead resting EKG as part of a physical examination by a physician member of the research team. Height and weight were also obtained during visit one.

#### *Ultrasound*

For the US assessment, subjects laid supine for ten minutes on a table to allow for fluid shifts with their right lower limb in a relaxed position at full extension. Scanning sites for the CSA of the VL were determined by taking half the femur length as obtained by measuring the full length from the greater trochanter to the lateral condyle using a Gulick tape measure (AliMed, Dedham, MA, USA) (Mangine et al., 2014). US transmission gel was then applied to the subject's skin and probe in order to enhance signal with the US imaging device. A probe support was used to ensure that the probe scans were perpendicular to the VL along the transverse axis (lateral to medial). Three consecutive panoramic scans of the VL at a gain of

50dB and a depth of 5.0 cm (Mangine et al., 2014) were taken in order to assess CSA. The greatest CSA value obtained from the 3 separate scans was used for data analysis.

#### *Familiarization: Isokinetic EMG Strength Testing*

Subjects were familiarized with the testing protocols so they were accustomed and comfortable with the entirety of testing procedures. Initially, subjects were taken to the NMRL to be familiarized with the isokinetic EMG assessment. For the EMG familiarization, subjects were asked to sit in the dynamometer chair, and a member of the research team adjusted all harnesses to replicate the actual testing session. Subjects were then instructed on the isokinetic leg extensions simulating exactly the procedure that subject would then be undergoing during the day of testing. Once procedures have been discussed, subjects performed three consecutive isokinetic leg extensions at 50%, 60% and 75% of their perceived maximal effort. Velocity of the isokinetic leg extension was set at 60° per second.

#### *Familiarization: Cardiopulmonary Testing*

Subjects were then familiarized with the cardiopulmonary exercise test (CPET) on a cycle ergometer. Subjects were fitted for a respiratory mask and seat height on the cycle ergometer was adjusted and recorded for proper cycling pedaling mechanics. Subjects wore a HR monitor and were given time to become accustomed to the mask and cycle ergometer. Subjects then preceded with a simulation of the test protocol to be used during the test day up to the point of 75% their target heart rate (THR), which was determined by the heart rate reserve method.



### Visit Two (Pre) and Three (Post): Testing Sessions

The pre and post visits have been combined in this section since the procedures for both sessions with exception of an US measurement, that was also taken on visit 3, were identical. Testing procedures occurred in this order: (1) Isokinetic EMG strength testing and (2) the VO<sub>2Max</sub> assessment. A rest period of a minimum of 10 minutes and a maximum of 15 minutes was utilized between the two tests and rehydration allowed in order to ensure proper recovery.

#### *Isokinetic EMG Strength Testing*

Isokinetic EMG strength testing was used in order to determine if there were any strength associated changes between the pre-test and the post-test, and to follow exactly the protocol employed by Wagoner et al. 2016. Prior to beginning the strength test, subjects warmed up on the cycle ergometer for 5 minutes at 50 Watts. Subjects were then taken to the NMRL for the isokinetic leg extension EMG assessment. Procedures were based off those conducted by previous studies in similar populations (Bottaro, Russo, & de Oliveira, 2005; Theou, Gareth, & Brown, 2008). Subjects were then placed in the dynamometer chair with harnesses placed over the shoulders, waist, and right leg. The right knee was aligned with the dynamometer's center of axis of rotation at 90° as measured by a goniometer (Model G800, Whitehall Manufacturing, Industry, CA, USA). EMG surface electrodes were then placed on the muscle belly of the VL. The location of the muscle belly was determined as 66% of the femur length. EMG electrodes were placed parallel to the muscle fibers of the VL. Once electrodes were in position, subjects were instructed to complete three warm-up isokinetic leg extensions at 50%, 60%, as well as 75% of their perceived maximal effort at a velocity of 60° per second. Subjects proceed to

complete three maximal isokinetic leg extensions at a velocity of 60° per second with two minutes of rest between each contraction. The greatest peak torque value recorded between the 3 contractions was used for data analysis, as the subsequent EMG amplitude.

Subjects then completed one maximal isometric contraction at a set knee angle of 120° (60° below horizontal) for purposes of normalizing EMG amplitude. Members of the research team provided verbal encouragement during each contraction. The highest PT and peak amplitude (PA) values of the three contractions were used for data analysis.

Signal processing was accomplished via a Biopac MP150WSW data acquisition system and AcqKnowledge software (Biopac Systems, Inc., Santa Barbara, CA, USA) recorded EMG signal. Raw PA and PT signal was stored on a personal laptop computer (MacBook Pro, Apple Inc., Cupertino, CA, USA) and analyzed with Labview 2014 software (Version 14, National Instruments, Austin, TX, USA).

### *Cardiopulmonary Fitness Assessment*

The final assessment was the cardiopulmonary exercise test for the assessment of  $\text{VO}_{2\text{Max}}$ . The test was performed on an electronically braked cycle ergometer using the Astrand Cycle Ergometer Maximal Test Protocol. Subjects began the test by sitting quietly on the cycle ergometer for three minutes while the researchers collected resting metabolic data. The first stage of the test was set to begin at 0 watts and lasted for three minutes. The preceding stage also lasted three minutes with an increase to 50 watts. Each of the subsequent stages increased by 25 watts until subjects reached volitional exhaustion. HR, RER, and RPE (6-20) were continually monitored and recorded during the last 30 seconds of every stage. Termination of the test was determined by the subjects' reaching volitional exhaustion and signaling to stop the test,  $\text{VO}_2$

plateau or decrease with increase in exercise intensity, or if an abnormal subject response to the test was observed. Upon the test being terminated, subjects were escorted to sit comfortably in a chair and after 3 minutes of rest, a finger prick was used to collect a drop of blood for lactate evaluation. Criteria established for  $\text{VO}_{2\text{Max}}$  set forth by the American College of Sports Medicine (ACSM) was used to determine if the test of maximal or peak.

### **Data Analysis**

Collected data for this current study was analyzed with SPSS Statistics version 23.0. The alpha level was set *a priori* for all statistical analyses at 0.05. Descriptive statistics of the population characteristics such as age, height, body mass, was presented in the form of mean and standard deviation. Paired samples t-tests, used to compare the results of the CPETs, were conducted for a pre-test and post-test (period of 2-weeks with no exercise intervention) for the following variables:  $\text{VO}_{2\text{Max}}$ , RPE, HR. Exploratory evaluations on PT, peak EMG amplitude, and CSA were also conducted using paired samples t-tests.

## CHAPTER IV

### RESULTS

The purpose of this study was to evaluate the effects of familiarization on CPET performances in sedentary middle-aged women, to verify whether or not there is a learning effect and how much this possible learning effect can affect the assessment of cardiopulmonary capacity in this population. More specifically, the research questions being addressed were: Will  $\text{VO}_{2\text{Max}}$  differ significantly from the pre-test and the post-test (RQ1), will RER differ significantly between pre-test and post-test (RQ2), and will the  $\text{HR}_{\text{Max}}$  differ significantly between the pre-test and post-test (RQ3)?

#### Subjects

The study included a total of 6 sedentary middle aged females. Subject characteristics are presented as means  $\pm$  standard deviations in Table 3 below. Subject characteristics appear similar to those in the aforementioned Wagoner et al. study with neither group being significantly different from the other in each of the characteristics other than in weights (Wagoner's study: Pre weight:  $78.0 \pm 17.2$ ; Post Weight:  $77.7 \pm 17.0$ ). Only 5 subjects data were used to assess peak torque and EMG amplitude from pre to post intervention, as one subject's pre-test file was excluded due to a sampling error. The subjects' characteristics are presented in Table 3 below:

**Table 3: Subject Characteristics; n = 6 (mean  $\pm$  SD)**

|  |                  |
|--|------------------|
| <b>Age (years)</b>                         | 53 $\pm$ 7       |
| <b>Height (cm)</b>                         | 160.3 $\pm$ 6.0  |
| <b>Pre-Weight (kg)</b>                     | 73.4 $\pm$ 16.7  |
| <b>Post-Weight (kg)</b>                    | 73.9 $\pm$ 16.6  |
| <b>BMI (kg*m<sup>-2</sup>)</b>             | 28.90 $\pm$ 7.67 |
| <b>Post Menopausal (# of participants)</b> | 5                |

### **Hypothesis Analysis**

Out of the 6 subjects included in the analyses, 5/6 achieved VO<sub>2Max</sub> during the pre-test session by complying with the VO<sub>2Max</sub> criteria determination adopted by the current study as given forth by ACSM. For the post-test, the results are different; only 4/6 participants achieved VO<sub>2Max</sub> according to the criteria determination adopted here. Below in table 4, pre and post means, standard deviations, of all primary variables, including Respiratory Exchange Ratio and Lactate values collected 3 minutes post completion of the CPET are presented for each subject below. It should also be noted here that subject 5 was experiencing illness during the post-test session and this may have affected her testing values.

**Table 4: CPET Variables by Subject (Mean  $\pm$  Standard Deviation)**

| Subject  | Rel. $\text{VO}_{2\text{Max}}$<br>$\text{mL} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$<br>Pre | Rel. $\text{VO}_{2\text{Max}}$<br>$\text{mL} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$<br>Post | Abs. $\text{VO}_{2\text{Max}}$<br>$\text{L} \cdot \text{min}^{-1}$<br>Pre | Abs. $\text{VO}_{2\text{Max}}$<br>$\text{L} \cdot \text{min}^{-1}$<br>Post | RER Last<br>Stage<br>Pre | RER Last<br>Stage<br>Post | $\text{HR}_{\text{Max}}$<br>BPM<br>Pre | $\text{HR}_{\text{Max}}$<br>BPM<br>Post | $\text{RPE}_{\text{Max}}$<br>Score<br>Pre | $\text{RPE}_{\text{Max}}$<br>Score<br>Post | Lactate<br>mmol<br>Pre | Lactate<br>mmol<br>Post |
|----------|---|--|---|--|--------------------------|---------------------------|--|---|---|--|------------------------|-------------------------|
| 1        | 16.87   | 15.90  | 1.16  | 1.10   | 1.13                     | 1.09                      | 133                                    | 132                                     | 17  | 17   | 5.2                    | 4.8                     |
| 2        | 27.13   | 25.27  | 1.75  | 1.63   | 1.08                     | 1.14                      | 172                                    | 177                                     | 19  | 18   | 6.7                    | 7.9                     |
| 3        | 38.33   | 34.67  | 2.63  | 2.37   | 1.16                     | 1.15                      | 162                                    | 166                                     | 17  | 19   | 9.4                    | 8.6                     |
| 4        | 17.57   | 17.43  | 1.86  | 1.85   | 1.19                     | 1.16                      | 167                                    | 166                                     | 18  | 19   | 9.3                    | 10.2                    |
| 5        | 35.40   | 28.47  | 2.10  | 1.69   | 1.15                     | 1.08                      | 150                                    | 126                                     | 17  | 14   | 9.2                    | 5.0                     |
| 6        | 30.43   | 25.70  | 2.26  | 1.94   | 1.16                     | 1.08                      | 177                                    | 160                                     | 20  | 18   | 6.4                    | 5.5                     |
| Mean     |   | 24.57 $\pm$  |   | 1.76 $\pm$   | 1.15 $\pm$               | 1.12 $\pm$                | 160 $\pm$                              | 155 $\pm$                               | 18 $\pm$                                  | 17.50                                      | 7.7 $\pm$              |                         |
| $\pm$ SD | 27.62 $\pm$ 8.95  | 7.00*  | 1.96 $\pm$ 0.50   | 0.42*  | 0.04                     | 0.04                      | 16                                     | 21                                      | 1.26                                      | 1.87                                       | 1.8                    | 7.0 $\pm$ 2.2           |

\*  $p < 0.05$  from pre to post intervention

Hypothesis 1, that subjects would not exhibit  $\text{VO}_{2\text{Max}}$  values that significantly differ between the pre-test and post-test, was evaluated using paired samples T-tests. Significant decreases in both, relative  $\text{VO}_{2\text{Max}}$  [ $\text{mL}/\text{kg}/\text{min}$ ],  $p=0.03$  (-9.68%) and absolute  $\text{VO}_{2\text{Max}}$  [ $\text{L}/\text{min}$ ],  $p=0.03$  (-9.33 %) were observed. Hypotheses 2 and 3, subjects will not exhibit significantly different RER scores at the final stage of the CPET between the pre and post CPETs and subjects will not exhibit significant different  $\text{HR}_{\text{Max}}$  were analyzed using paired samples T-tests. No significant differences in RER,  $p=0.27$  (-2.61%) or  $\text{HR}_{\text{Max}}$ ,  $p=0.29$  (-3.13%) were observed.

### Exploratory Analyses

Paired T-tests were used to assess the strength variables peak torque, EMG amplitude, and CSA data is presented in table 5. In addition, the mean percent change is reported in the following sentence. None of these variables were found to be significantly different from pre to post test; peak torque,  $p=0.546$  (-7.15%), EMG amplitude,  $p=0.817$  (+1.98%), and CSA  $p=0.704$  (+1.56%).

**Table 5: Exploratory Strength Variables by Subject (Mean  $\pm$  Standard Deviation)**

|               | PT Pre            | PT Post           | EMG Amp. Pre      | EMG Amp. Post     | CSA Pre            | CSA Post           |
|---------------|-------------------|-------------------|-------------------|-------------------|--------------------|--------------------|
| Subject       | (N*m)             | (N*m)             | (mV)              | (mV)              | (cm <sup>2</sup> ) | (cm <sup>2</sup> ) |
| 1             | N/A               | N/A               | N/A               | N/A               | 14.26              | 14.67              |
| 2             | 63.86             | 71.93             | 121.27            | 121.59            | 10.46              | 12.48              |
| 3             | 95.03             | 113.74            | 111.26            | 138.96            | 13.12              | 13.25              |
| 4             | 49.84             | 33.81             | 43.43             | 39.09             | 14.28              | 15.41              |
| 5             | 85.46             | 79.60             | 99.92             | 81.98             | 15.23              | 13.27              |
| 6             | 125.57            | 89.11             | 88.31             | 91.76             | 17.31              | 16.92              |
| Mean $\pm$ SD | 83.95 $\pm$ 29.26 | 77.64 $\pm$ 29.12 | 92.84 $\pm$ 30.25 | 94.68 $\pm$ 38.55 | 14.11 $\pm$ 2.27   | 14.33 $\pm$ 1.66   |

## **CHAPTER V**

### **DISCUSSION**

#### **Overview**

The purpose of this study was to evaluate the effects of familiarization on CPET performances in sedentary middle-aged women, to verify whether or not there is a learning effect. Previous literature has been inconclusive as to whether or not a learning effect can influence during CPET performance. Cox et al. and Marciniuk et al. have shown in clinical populations that reproducibility of these cardiopulmonary exercise tests are adequate, even at submaximal levels (Cox et al., 1989; Marciniuk, Watts, and Gallagher, 1993). However, not all studies have shown high reproducibility of cardiopulmonary exercise testing, suggesting there may in fact be an observed learning effect. Knox et al. (1988) concluded that a learning effect was in fact evident in patients with chronic airway obstructions, during walking tests when they were carried out over short time intervals (4 walking tests for 3 consecutive days; 12 total tests). This study shows the importance of familiarizing individuals with the testing procedures in order to mitigate this learning effect and inaccurately reported fitness values (Knox et al., 1988). Swindburn et al. (1985) also reports a learning effect in multiple tests of aerobic capacity in patients with chronic airway obstruction disease. In this study, subjects performed a 12-minute walking test, a paced step test, as well as a cycle ergometer test. This study found that there was a progressive and significant increase in performance in all three types of exercise test



(Swindburn et al., 1985). The learning effect is an important factor to consider as it relates to selecting the most appropriate and precise testing protocols and instruments in research. The validity of cardiopulmonary exercise tests in exercise science studies is obviously important in that it determines whether or not an exercise intervention actually had an effect on the subject's oxygen uptake ( $\text{VO}_2$ ) such as in the Wagoner et al. study. Recall that this study determined whether or not a 2-week lower body resistance protocol was enough to increase  $\text{VO}_{2\text{Max}}$  values in sedentary middle-aged women. The issue lies in being able to confidently interpret the results of a CPET and being able to effectively determine if a learning effect had an impact on the results of subsequent tests is necessary. Especially in clinical populations, placing these populations through as little stress as possible while still obtaining an accurate interpretation of the testing results is paramount if one desires to confidently determine and report the accuracy of such tests.

### **Aerobic Capacity**

Significant decreases in maximum oxygen uptake values from pre to post-test were observed, which was contrary to our hypothesis. These findings may be attributed to a multiple factors including, but not limited to, a diminished knowledge of the test during the 2 weeks between pre and post intervention (which may have been masked in the Wagoner et al. study due to subsequent increases in strength) , a really well performed familiarization session between visit 1 and visit 2, or potentially a loss of motivation. These concepts will be discussed in further detail below.

To our knowledge, no other study observed showed a significant decrease in aerobic capacity from pre to post test. While some studies have showed decreased  $\text{VO}_2$  values, these are usually in clinical populations in which no exercises has been assigned or with long time spans

with regards to the testing windows. Previous studies were inconclusive when it comes to determining whether or not a learning effect occurred. Previous studies without any exercise intervention? either showed an increase in aerobic capacity values, necessitating a familiarization session (Swindburn et al., 1985), or they showed no significant difference in aerobic capacity values meaning that a familiarization session was not needed (Cox et al., 1989).

### *Significant Increase in Aerobic Capacity Variables*

The following discusses a few of the studies that found an increase in aerobic capacity, which necessitates a familiarization session. One study by Swindburn in a clinical population had subjects perform a 12-minute walking test, a paced step test, as well as a cycle ergometer test. This study found that there was a progressive and significant increase in performance in all three types of exercise test between the first and fourth measurements. The overall changes in performance were: a 96% increase in steps climbed, 29% longer cycling duration, and a 16% improvement in 12 MWD (Swindburn et al., 1985). This indicates that up to 4 familiarization sessions may be needed in order to mitigate increases not due to the intervention! Unless you think there may be a training effect going on, which seems like a possibility to me. How long was the gap between tests in the other studies? This would allow you to compare.

A study by Elborn et al. (1990) had subjects with cardiac function impairment performed 3 cardiopulmonary exercise tests with at least 2 weeks of separation between each test. Changes over the three tests at rest, at a submaximal intensity, and at maximal intensity were analyzed. This study shows that all the parameters measured during a cardiopulmonary exercise test at rest and during symptom-limited exercise were reproducible between a second and third test as compared with the first test. The first exposure of patients with chronic cardiac failure to an

exercise test such as this resulted in an underestimate of exercise time by about 20%, which argues strongly for the importance of familiarization as shown here that the values were not reliable until the second testing session. Patients also achieved a higher exercise stage in test 2 compared to the first, despite their perceived exertion being similar. Peak exercise  $\text{VO}_2$ ,  $\text{VCO}_2$  and VE were also significantly underestimated between the first and second test (Elborn et al., 1990).

#### *No Significant Increase in Aerobic Capacity Variables*

The following discusses a few of the studies that found no significant increase in aerobic capacity, which would mean that a familiarization session is not needed. Cox et al. designed a study to examine the reproducibility and the learning effect of an incremental exercise testing. Specifically, Cox and colleagues studied cycle ergometer testing in patients with obstructive lung disease who were untrained, and who also had never done such a test prior to this study. This study was designed in a test retest fashion, as the subjects performed the first test then the second test 24 hours after. This study found that there were no significant differences between values of the first and second testing procedure. This indicates that there was in fact no learning effect that occurred during this cycle ergometer test. Results indicated good reproducibility for workload (4.5%),  $\text{VO}_{2\text{Max}}$  (3.5%), RER (3.4%), and HR (3.7%) (Cox et al., 1989).

In a healthy population, Fielding et al. also investigated the reproducibility of maximal exercises tests, specifically in older women. Women from 51 to 68 years old were used for this study, which is similar to the population used in this study. Each subject performed 5 different exercise tests separated by at least 1 week's time. Subjects performed the Modified Bruce protocol on a motor driven treadmill.  $\text{VO}_{2\text{Max}}$  values were found to be consistent between all

tests, with the mean coefficient of variation being 6.5%. Mean maximal ventilation was also not significantly different between tests. RER and maximal HR reached between tests was also not significant. In addition, intraclass correlation coefficients for a single test are high and suggest good agreement between tests. In general, the associations did improve between test 2, test 3 and test 4. This study concluded that the Bruce treadmill protocol for women who are older (51-68 years old) produces highly reproducible results with  $\text{VO}_{2\text{Max}}$ , maximum heart rate, as well as ventilation. It is suggested that performance of only a single  $\text{VO}_{2\text{Max}}$  test is sufficient to assess aerobic capacity in middle-aged and older women (Fielding et al., 1997).

### *Summary*

What these studies conclude is that in clinical populations a familiarization session is needed as these values can increase up until the fourth test. This increase up until the fourth test means that incorrect values of aerobic capacity may be obtained if taken in the first three tests. This necessitates a thorough familiarization session in order to properly instruct subjects on what they are doing so that they feel comfortable with performing what is being asked of them. While this familiarization session is important for every population, it may be even more important for clinical populations in which the accuracy of these exercise values may be a matter of life and death. In healthy older women there may not be a learning effect as Fielding et al. showed high reproducibility. However no study has shown a decrease in  $\text{VO}_{2\text{Max}}$  values. This study did also show that RER and  $\text{HR}_{\text{Max}}$  had no significant difference between pre and post-test. Because the current study showed that  $\text{VO}_{2\text{Max}}$  both relative and absolute all showed significant decreases from visit 2 to visit 3, it can be speculated that visit 1's familiarization session was somewhat effective. However, it can be speculated, that in this population, during the 2-week period

between visit 2 and visit 3, there was some de-familiarization with the test procedure; that forgotten knowledge of the tasks to be performed explains the decrements. However an alternative hypothesis is that the participants simply lost motivation. A maximal effort exercise test is difficult for anyone, especially those who are considered sedentary.. It is possible that they completed the first test and found out how hard the test actually was. This may have led to some hesitation and holding back of the same effort during the subsequent testing session. It is also possible that during the last session the participants were ready to end their time commitment to the study as 3 weeks is long time to commit oneself to a study. Some of these tests also took part near the end of the school year, which may have also been a factor in subjects losing motivation. This would mean that a learning effect was in fact mitigated through visit 1, however the opposite effect may have occurred in the 2-week period between visit 2 and 3 since the first visit was so thorough in the sense that the participants were instructed on everything to do, free to ask questions regarding the tasks, and allowed to redo portions of the test if their first attempt was not adequate. Also the second test was a week after the familiarization, which lessens the possibility of forgotten information, compared to the post test that was 3 weeks after the familiarization session. What this would mean in the context of the Wagoner et al. study, since the same familiarization session was performed, is that the increases observed in  $VO_{2Max}$  was in fact related to the strength protocol and not a learning effect; meaning that there may have also been a decrease in the Wagoner study, but this decrease was counterbalanced by the resistance training. With such a small sample size, these are all speculative at this time and a larger sample is needed to confirm or refute these preliminary study results.

When discussing the exploratory variables, the changes in muscular strength, EMG amplitude, and muscle size (CSA) observed from this study, it is important to mention that because a resistance training protocol was not a direct part of this study, it is not one of the main variables in question for this current study. However because of the Wagoner et al. study's hypotheses focused on these variables, these variables were measured in order to provide means for direct comparison as this study was designed to serve as the control group for Wagoner's study. Specifically, Wagoner's hypotheses two, three, and four, that stated peak torque (PT) and EMG amplitude would significantly increase whereas CSA would not in response to the training intervention. Wagoner found that PT and EMG amplitude significantly increased from pre to post intervention whereas CSA of the VL did not. Wagoner concluded that observed increases in PT and EMG amplitude were a result of increased neural activation and recruitment of muscle fibers considering that VL CSA did not increase in a hypertrophic way. With no changes observed in this current study for PT, EMG amplitude and CSA, which was expected, the improvements in some of these variables presented by Wagoner et al study, may have been attributed to the study strength training intervention. Once again, due to the small sample size of the current study, a larger sample size is necessary to confirm or refute this possibility.

### *Conclusion*

This current study found that there was a significant decrease in  $VO_{2Max}$  from pre to post-test in middle age sedentary women. This is a surprising result that may be explained in part by a potential de-familiarization of the women between pre and post-test, a long window between pre and post test, as well as a possible loss of motivation. However, the results of this preliminary evaluation should be cautiously interpreted due to the small sample size. Nevertheless, these

preliminary results appears to support the fact that a familiarization session is indeed important in this population and that a 2-week period may be enough to diminish the familiarization effect thus potentially requiring familiarization procedure to be implemented prior to each test.

### **Future Research**

This study is one of the few of its kind in terms of the population chosen to observe as well as the type of results obtained in that  $\text{VO}_{2\text{Max}}$  actually decreased . Future investigations exploring the influence of performing an additional familiarization session when the time between pre and post test is long is warranted. Furthermore, specificity in terms of the mode these tests are performed (treadmill vs. cycle ergometer) would be insightful to see how the mode of the test can influence the learning effect especially since treadmill walking/running may be a more natural pattern for most. Lastly, given the results of this study, this learning effect should be explored in different populations such as breast cancer population to see how this etiology affects aerobic capacity and strength measurements, especially given the wasting nature of this disease.

## APPENDIX 1.1: STUDY BROCHURE

### ☐ VOLUNTEERS NEEDED FOR RESEARCH STUDY



- Searching for volunteers that are female adults between the ages of 35-65 years.
- Must not have participated in regularly scheduled exercise (< 2 times / week) within the past 6 months.
- Participation involves 3 total visits:
  - Visit 1 → approximately 2 hours (Familiarization)
  - Visits 2 & 3 → approximately 1.5 hour each (Pre and Post Testing)
- Subjects will undergo 2 cardiopulmonary exercise tests and 2 isokinetic/isometric tests.

IRB: 15-1129



## **APPENDIX 1.2: PRE-ASSESSMENT GUIDELINES**

UNIVERSITY OF NORTH CAROLINA AT CHAPEL HILL  
Claudio Battaglini, Ph.D. FACSM.  
Department of Exercise and Sport Sciences  
105 Fetzer Hall, CB # 8700  
(919) 843-6045 / Email: claudio@email.unc.edu

### **Pre-Test Guidelines**

1. Avoid eating 2 hours prior to testing.
2. Void completely before testing.
3. Maintain proper hydration prior to testing.
4. Please wear appropriate clothing/shoes for testing (running shorts/shirt/shoes)
5. No exercise 12 hours prior to testing.
6. No alcohol consumption 48 hours prior to testing.
7. No diuretic medications 7 days prior to testing.

Source: Advanced Fitness Assessment and Exercise Prescription – Third Edition – Vivian H. Heyward

## APPENDIX 1.3: Physical Activity Readiness Questionnaire

Physical Activity Readiness  
Questionnaire - PAR-Q  
(revised 2002)

# PAR-Q & YOU

(A Questionnaire for People Aged 15 to 69)

Regular physical activity is fun and healthy, and increasingly more people are starting to become more active every day. Being more active is very safe for most people. However, some people should check with their doctor before they start becoming much more physically active.

If you are planning to become much more physically active than you are now, start by answering the seven questions in the box below. If you are between the ages of 15 and 69, the PAR-Q will tell you if you should check with your doctor before you start. If you are over 69 years of age, and you are not used to being very active, check with your doctor.

Common sense is your best guide when you answer these questions. Please read the questions carefully and answer each one honestly: check YES or NO.

| YES                      | NO                       |  |
|--------------------------|--------------------------|--|
| <input type="checkbox"/> | <input type="checkbox"/> | 1. Has your doctor ever said that you have a heart condition <u>and</u> that you should only do physical activity recommended by a doctor? |
| <input type="checkbox"/> | <input type="checkbox"/> | 2. Do you feel pain in your chest when you do physical activity?   |
| <input type="checkbox"/> | <input type="checkbox"/> | 3. In the past month, have you had chest pain when you were not doing physical activity?   |
| <input type="checkbox"/> | <input type="checkbox"/> | 4. Do you lose your balance because of dizziness or do you ever lose consciousness?  |
| <input type="checkbox"/> | <input type="checkbox"/> | 5. Do you have a bone or joint problem (for example, back, knee or hip) that could be made worse by a change in your physical activity?    |
| <input type="checkbox"/> | <input type="checkbox"/> | 6. Is your doctor currently prescribing drugs (for example, water pills) for your blood pressure or heart condition?                       |
| <input type="checkbox"/> | <input type="checkbox"/> | 7. Do you know of <u>any other reason</u> why you should not do physical activity?   |

If  
you  
answered

### YES to one or more questions

Talk with your doctor by phone or in person BEFORE you start becoming much more physically active or BEFORE you have a fitness appraisal. Tell your doctor about the PAR-Q and which questions you answered YES.

- You may be able to do any activity you want — as long as you start slowly and build up gradually. Or, you may need to restrict your activities to those which are safe for you. Talk with your doctor about the kinds of activities you wish to participate in and follow his/her advice.
- Find out which community programs are safe and helpful for you.

### NO to all questions

If you answered NO honestly to all PAR-Q questions, you can be reasonably sure that you can:

- start becoming much more physically active — begin slowly and build up gradually. This is the safest and easiest way to go.
- take part in a fitness appraisal — this is an excellent way to determine your basic fitness so that you can plan the best way for you to live actively. It is also highly recommended that you have your blood pressure evaluated. If your reading is over 144/94, talk with your doctor before you start becoming much more physically active.

#### DELAY BECOMING MUCH MORE ACTIVE:

- if you are not feeling well because of a temporary illness such as a cold or a fever — wait until you feel better; or
- if you are or may be pregnant — talk to your doctor before you start becoming more active.

**PLEASE NOTE:** If your health changes so that you then answer YES to any of the above questions, tell your fitness or health professional. Ask whether you should change your physical activity plan.

**Informed Use of the PAR-Q:** The Canadian Society for Exercise Physiology, Health Canada, and their agents assume no liability for persons who undertake physical activity, and if in doubt after completing this questionnaire, consult your doctor prior to physical activity.

**No changes permitted. You are encouraged to photocopy the PAR-Q but only if you use the entire form.**

**NOTE:** If the PAR-Q is being given to a person before he or she participates in a physical activity program or a fitness appraisal, this section may be used for legal or administrative purposes.

"I have read, understood and completed this questionnaire. Any questions I had were answered to my full satisfaction."

NAME \_\_\_\_\_

SIGNATURE \_\_\_\_\_

DATE \_\_\_\_\_

SIGNATURE OF PARENT  
or GUARDIAN (for participants under the age of majority) \_\_\_\_\_

WITNESS \_\_\_\_\_

**Note: This physical activity clearance is valid for a maximum of 12 months from the date it is completed and becomes invalid if your condition changes so that you would answer YES to any of the seven questions.**



© Canadian Society for Exercise Physiology www.csep.ca/forms

## APPENDIX 1.4: MEDICAL HISTORY QUESTIONNAIRE

### Medical History

Subject: \_\_\_\_\_ ID: \_\_\_\_\_ Telephone: \_\_\_\_\_

Address: \_\_\_\_\_

Occupation: \_\_\_\_\_ Age: \_\_\_\_\_

YES NO

#### Patient History

1. How would you describe your general health at present?  
Excellent \_\_\_\_\_ Good \_\_\_\_\_ Fair \_\_\_\_\_ Poor \_\_\_\_\_
2. Do you have any health problems at the present time? \_\_\_\_\_
3. If yes, please describe: \_\_\_\_\_  
\_\_\_\_\_
4. Have you ever been told you have heart trouble? \_\_\_\_\_
5. If yes, please describe: \_\_\_\_\_  
\_\_\_\_\_
6. Is there any chance of you being pregnant at this time? Yes: \_\_\_\_\_ No: \_\_\_\_\_
7. Is there any chance that you may become pregnant during span of the study?  
Yes: \_\_\_\_\_ No: \_\_\_\_\_
8. Have you had consistent menstrual periods for the last 3 months? Yes: \_\_\_\_\_ No: \_\_\_\_\_  
If no, when was your last period \_\_\_\_\_
9. Do you ever get pain in your chest? \_\_\_\_\_
10. Do you ever feel light-headed or have you ever fainted? \_\_\_\_\_
11. If yes, please describe: \_\_\_\_\_  
\_\_\_\_\_
12. Have you ever been told that your blood pressure has been elevated? \_\_\_\_\_
13. If yes, please describe: \_\_\_\_\_  
\_\_\_\_\_
14. Have you ever had difficulty breathing either at rest or with exertion? \_\_\_\_\_
15. If yes, please describe: \_\_\_\_\_  
\_\_\_\_\_
16. Are you now, or have you been in the past 5 years, under a doctor's care for any reason? \_\_\_\_\_
17. If yes for what reason? \_\_\_\_\_  
\_\_\_\_\_
18. Have you been in the hospital in the past 5 years? \_\_\_\_\_

19. If yes, for what reason? \_\_\_\_\_
20. Have you ever experienced an epileptic seizure or been informed that you have epilepsy? \_\_\_\_\_
21. Have you ever been treated for infectious mononucleosis, hepatitis, pneumonia, or another infectious disease during the past year? \_\_\_\_\_
22. If yes, name the disease: \_\_\_\_\_
23. Have you ever been treated for or told you might have diabetes? \_\_\_\_\_ 24. Have you ever been treated for or told you might have low blood sugar? \_\_\_\_\_
25. Do you have any known allergies to drugs? \_\_\_\_\_
26. If so, what? \_\_\_\_\_
27. Have you ever been “knocked-out” or experienced a concussion? \_\_\_\_\_
28. If yes, have you been “knocked-out” more than once? \_\_\_\_\_
29. Have you ever experienced heat stroke or heat exhaustion? \_\_\_\_\_
30. If yes, when? \_\_\_\_\_
31. Have you ever had any additional illnesses or operations? (Other than childhood diseases) \_\_\_\_\_
32. If yes, please indicate specific illness or operations: \_\_\_\_\_
33. Are you now taking any pills or medications? \_\_\_\_\_
34. If yes, please list: \_\_\_\_\_
35. Have you had any recent (within 1 year) difficulties with your:
- a. Feet \_\_\_\_\_
  - b. Legs \_\_\_\_\_
  - c. Back \_\_\_\_\_

#### Family History

36. Has anyone in your family (grandparent, father, mother, and/or sibling) experienced any of the following?
- a. Sudden death \_\_\_\_\_
  - b. Cardiac disease \_\_\_\_\_
  - c. Marfan’s syndrome \_\_\_\_\_

#### Mental History

37. Have you ever experienced depression? \_\_\_\_\_
38. If yes, did you seek the advice of a doctor? \_\_\_\_\_
39. Have you ever been told you have or has a doctor diagnosed you with panic disorder, obsessive-compulsive disorder, clinical depression, bipolar disorder, or any other psychological disease? \_\_\_\_\_
40. If yes, please list condition and if you are currently taking any medication.
- | Condition | Medication |
|-----------|------------|
| _____     | _____      |
| _____     | _____      |

---

---

---

### Bone and Joint History

41. Have you ever been treated for Osgood-Schlatter's disease? \_\_\_\_\_
42. Have you ever had any injury to your neck involving nerves or vertebrae? \_\_\_\_\_
43. Have you ever had a shoulder dislocation, separation, or other injury of the shoulder that incapacitated you for a week or longer? \_\_\_\_\_
44. Have you ever been advised to or have you had surgery to correct a shoulder condition? \_\_\_\_\_
45. Have you ever experienced any injury to your arms, elbows, or wrists? \_\_\_\_\_
46. If yes, indicate location and type of injury: \_\_\_\_\_
47. Do you experience pain in your back? \_\_\_\_\_
48. Have you ever had an injury to your back? \_\_\_\_\_
49. If yes, did you seek the advice of a doctor? \_\_\_\_\_
50. Have you ever been told that you injured the ligaments or cartilage of either knee joint? \_\_\_\_\_
51. Do you think you have a trick knee? \_\_\_\_\_
52. Do you have a pin, screw, or plate somewhere in your body as the result of bone or joint surgery that presently limits your physical capacity? \_\_\_\_\_
53. If yes, indicate where: \_\_\_\_\_
54. Have you ever had a bone graft or spinal fusion? \_\_\_\_\_

### Activity History

55. During your early childhood (to age 12) would you say you were:  
Very active \_\_\_\_\_ Quite active \_\_\_\_\_ Moderately active \_\_\_\_\_ Seldom active \_\_\_\_\_
56. During your adolescent years (age 13-18) would you say you were:  
Very active \_\_\_\_\_ Quite active \_\_\_\_\_ Moderately active \_\_\_\_\_ Seldom active \_\_\_\_\_
57. Did you participate in:
- a. Intramural school sports? \_\_\_\_\_
  - b. Community sponsored sports? \_\_\_\_\_
  - c. Varsity school sports? \_\_\_\_\_
  - d. Active family recreation? \_\_\_\_\_
58. Since leaving high school, how active have you been?  
Very active \_\_\_\_\_ Quite active \_\_\_\_\_ Active \_\_\_\_\_ Inactive \_\_\_\_\_
59. Do you participate in any vigorous activity at present? \_\_\_\_\_
60. If yes, please list:

| Activity | Frequency | Duration | Intensity |
|----------|-----------|----------|-----------|
|          |           |          |           |
|          |           |          |           |
|          |           |          |           |

61. How would you describe your present state of fitness?

Excellent\_\_\_\_\_ Good\_\_\_\_\_ Fair\_\_\_\_\_ Poor\_\_\_\_\_

62. Please list the type(s) of work you have been doing for the previous ten years:

Year Work Indoor/Outdoor Location (city/state)

---

---

---

---

63. Whom shall we notify in case of emergency?

Name: \_\_\_\_\_

Phone: (Home)\_\_\_\_\_ (Work)\_\_\_\_\_

Address: \_\_\_\_\_

64. Name and address of personal physician: \_\_\_\_\_

---

---

All of the above questions have been answered completely and truthfully to the best of my knowledge.

Signature: \_\_\_\_\_ Date: \_\_\_\_\_

## APPENDIX 1.5: DATA COLLECTION SHEET

Subject ID: \_\_\_\_\_

### **Visit 1**

Height (in/cm): \_\_\_\_\_ / \_\_\_\_\_ Weight (lbs/kg): \_\_\_\_\_ / \_\_\_\_\_

RBP: \_\_\_\_\_ RHR: \_\_\_\_\_

### **Ultrasound Assessment**

Half Femur Length (cm): \_\_\_\_\_

Pre CSA (cm<sup>2</sup>): \_\_\_\_\_ Post CSA (cm<sup>2</sup>): \_\_\_\_\_

### **Cardiopulmonary Assessment (VO<sub>2</sub>peak)**

Mask Size: \_\_\_\_\_ Seat Height (in): \_\_\_\_\_

#### **Pre**

RHR(Before): \_\_\_\_\_

RBP(Before): \_\_\_\_\_

RHR(After): \_\_\_\_\_

RBP(After): \_\_\_\_\_

Height (cm): \_\_\_\_\_

Weight (kg): \_\_\_\_\_

VO<sub>2</sub>peak: \_\_\_\_\_  
(ml/kg/min)

#### **Post**

RHR(Before): \_\_\_\_\_

RBP(Before): \_\_\_\_\_

RHR(After): \_\_\_\_\_

RBP(After): \_\_\_\_\_

Height (cm): \_\_\_\_\_

Weight (kg): \_\_\_\_\_

VO<sub>2</sub>peak: \_\_\_\_\_  
(ml/kg/min)

|                         |                         |
|-------------------------|-------------------------|
| _____ (L/min)           | _____ (L/min)           |
| Lactate (mmol/L): _____ | Lactate (mmol/L): _____ |

**Isokinetic / Isometric Strength Testing**

*Electrode Placement [66% Femur Length] (cm):* \_\_\_\_\_

**Isokinetic Leg Extension (60°/sec) CW**

|   |  |
|---|--|
| <u>Pre:</u><br>_____ (Basic Noise)<br><br>_____<br><br>_____<br><br>_____ | <u>Post:</u><br>_____ (Basic Noise)<br><br>_____<br><br>_____<br><br>_____ |
|---|--|

**Isometric**

|                      |                       |
|----------------------|-----------------------|
| <u>Pre:</u><br>_____ | <u>Post:</u><br>_____ |
|----------------------|-----------------------|

**Gravity Correction**

|                      |                       |
|----------------------|-----------------------|
| <u>Pre:</u><br>_____ | <u>Post:</u><br>_____ |
|----------------------|-----------------------|



## REFERENCE

- Astrand, P., Rhyming I. (1954). A Nomogram for Calculation of Aerobic Capacity (Physical Fitness) for Pulse Rate During Submaximal Work. *Journal of Applied Physiology*. 7:218-221.
- Bassett, D., Howley, T. (2000). Limiting Factors for Maximum Oxygen Uptake and Determinants of Endurance Performance, *Medicine & Science in Sports & Exercise*. 70-84.
- Balke, B. Advanced Exercise Procedure for Evaluation of the Cardiovascular System. Monograph. Milton: The Burdick Corporation, 1970.
- Barron, A., Dhutia, N., Mayet, J., et al. (2014). Test–Retest Repeatability of Cardiopulmonary Exercise Test Variables in Patients With Cardiac or Respiratory Disease. *European Journal of Preventative Cardiology*. 21(4), 445-453.
- Bottaro, M., Russo, A. F., & de Oliveira, R. J. (2005). The Effects of Rest Interval on Quadriceps Torque During an Isokinetic Testing Protocol in Elderly. *Journal of Sports Science & Med*, 4(3), 285-290.
- Bruce, R., Multi-Stage Treadmill Tests of Maximal and Submaximal Exercise. Exercise Testing and Training of Apparently Healthy Individuals: A Handbook for Physicians. New York: American Heart Association, 1972, p. 32-34.
- Cox N., Hendriks J., Binkhorst R. et al. (1989). Reproducibility of Incremental Maximal Cycle Ergometer Tests in Patients With Mild to Moderate Obstructive Lung Diseases. *Lung*, 167: 129–133.
- Dideriksen, K., Mikkelsen, U. (2015), Reproducibility of Incremental Maximal Cycle Ergometer Tests in Healthy Recreationally Active Subjects. *Clinical Physiology and Functional Imaging*.
- Elborn J., Stanford C., Nicholls D. (1990). Reproducibility of Cardiopulmonary Parameters During Exercise in Patients With Chronic Cardiac Failure. The Need For a Preliminary Test. *European Heart Journal*, 11: 75–81.
- Evans, E. S. (2012). The Impact of Acute Aerobic Exercise on Natural Killer Cell, Catecholamine, and Cortisol Responses in Breast Cancer Survivors (Order No. 3549667). Available from ProQuest Dissertations & Theses Global.
- Fielding, R., Frontera, W., Hughes, V., Fisher, E., Evans, W. (1997). The Reproducibility of the Bruce Protocol Exercise Test for the Determination of Aerobic Capacity in Older Women. *Medicine & Science in Sports & Exercise*. 29: 8, 1109-1113.

- Fleg, J. L., & Lakatta, E. G. (1988). Role of Muscle Loss in the Age-Associated Reduction in  $\text{VO}_{2\text{Max}}$ . *Journal of Applied Physiology*, 65(3), 1147-1151.
- Frontera, W. R., Hughes, V. A., Lutz, K. J., & Evans, W. J. (1991). A Cross-Sectional Study of Muscle Strength and Mass in 45- to 78-yr-old Men and Women. *Journal of Applied Physiology*, 71(2), 644-650.
- Gulati M., Black H.R., Shaw L.J. et al. (2015) The Prognostic Value of a Nomogram for Exercise Capacity in Women. *N Engl J Med*;353:468 – 475. □
- Hartman, M., Fields, D., Byrne, N., & Hunter, G. (2007). Resistance Training Improves Metabolic Economy During Functional Tasks in Older Adults. *Journal of Strength and Conditioning Research*, 21(1), 91-95.
- Heyward, V. H. (2006). Advanced Fitness Assessment and Exercise Prescription (Vol. 5): Human kinetics Champaign, IL.
- Hill A., Lupton H. (1923), Muscular Exercise, Lactic Acid, and the Supply and Utilization of Oxygen. *Quarterly Journal of Medicine*, 16: 135-71. □
- Howley E., Bassett Jr D., Welch H. (1995). Criteria for Maximal Oxygen Uptake Review and Commentary. *Medicine & Science in Sports & Exercise*, 32: 70-84.
- Jones LW, Eves ND, Mackey JR, et al. (2008). Systemic Inflammation, Cardiorespiratory Fitness, and Quality of Life in Patients With Advanced Non-Small Cell Lung Cancer. *J Thorac Oncol*, 3:194–195. □
- Knox A., Morrison J., Muers M. (1988). Reproducibility of Walking Test Results in Chronic Obstructive Airways Disease. *Thorax*, 43:388-92.
- Mangine, G. T., Fukuda, D. H., LaMonica, M. B., Gonzalez, A. M., Wells, A. J., Townsend, J. R., . . . Hoffman, J. R. (2014). Influence of Gender and Muscle Architecture Asymmetry on Jump and Sprint Performance. *J Sports Sci Med*, 13(4), 904-911.
- Marciniuk, D., Watts, R., Gallagher, C. (1993). Reproducibility of Incremental Maximal Cycle Ergometer Testing in Patients With Restrictive Lung Disease. *Thorax*, 48:894-898
- Morris JN, Heady JA, Raffle PA, et al. (1953) Coronary Heart Disease and Physical Activity of Work. *Lancet* 1953, 265:1053–1057; contd.
- Morrow JR, Jackson AW, Disch JG, Mood DP. (2000), Measurement and Evaluation in Human Performance, 2nd edition. Champaign, IL: Human Kinetics.
- Myers J, Prakash M., Froelicher V. et al. (2002). Exercise Capacity and Mortality Among Men Referred for Exercise Testing. *N Engl J Med*;346:793– 801. □

- Pescatello, L. S., & American College of Sports, M. (2014). ACSM's Guidelines For Exercise Testing and Prescription (9th ed.). Philadelphia: Wolters Kluwer/Lippincott Williams & Wilkins Health.
- Poole, D., Wilkerson D., Jones, A., (2008). Validity of Criteria for Establishing Maximal O<sub>2</sub> Uptake During Ramp Exercise Tests, *European Journal of Applied Physiology*, 102:403-410.
- Powers, S., Howley, E. (2012). Exercise Physiology: Theory and Application to Fitness and Performance. New York: McGraw-Hill Humanities/Social Sciences/Languages Print, 76-79, 334-347.
- Roubenoff, R., & Hughes, V. A. (2000). Sarcopenia: Current Concepts. *The Journals of Gerontology Series A: Biological Sciences and Medical Sciences*, 55(12), M716-724.
- Shun-Shin M., Francis D. (2012). Why Are Some Studies of Cardiovascular Markers Unreliable? The Role of Measurement Variability and What an Aspiring Clinician Scientist Can Do Before It Is Too Late. *Progress in Cardiovascular Disease*, 55: 14–24.
- Swinburn, C., Wakefield, J., Jones P. (1985). Performance, Ventilation and Oxygen Consumption in Three Different Types of Exercise Test in Patients With Chronic Obstructive Lung Disease. *Thorax*, 40:581-6.
- Theou, O., Gareth, J. R., & Brown, L. E. (2008). Effect of Rest Interval on Strength Recovery in Young and Old Women. *Journal of Strength and Conditioning Research*, 22(6), 1876-1881.
- Wagoner, C.W., The Effect Of A Two-Week Lower Body Resistance Training Protocol On Aerobic Capacity (VO<sub>2peak</sub>) In Sedentary Middle-Aged Females. Master's Thesis, University of North Carolina at Chapel Hill, 2016, 10120148
- Wood, W., Deal, A., Abernethy, A., Basch, E., Hie, Y., Whitley, J., Shatten, C., Serody, J., Shea, T., Reeve, B., Battaglini, C.L. (2013). Cardiopulmonary Fitness in Patients Undergoing Hematopoietic Stem Cell Transplantation: A Pilot Study. *Biol Blood Marrow Transplant*. 48(10):1342-9.