ABSTRACT

ENEGRY COST OF HEAVY LOAD CARRIAGE AND PROLONGED WALKING IN ROTC COLLEGIATE PARTICIPANTS

By Sara Marie Krajewski

April, 2014

Director of Thesis: Dr. Laurel Wentz

Major Department: Kinesiology

The load military service members carry while marching has increased throughout history, and the military routinely uses rucksacks that range in weight from 22-68 kilograms, depending on the type of mission and weather conditions (Van Dijk, 2009). Previous studies have examined the energy cost of load carriage under relatively short time frames, such as 30-45 minutes, but typical marching time greatly exceeds these limits during military operations. Few studies have examined marching times upwards of two hours with a fixed load during flat, incline, and decline conditions with trained military personnel. **PURPOSE**: The purpose of this study was to measure the energy cost of prolonged marching under heavy load. **METHODS:** Eleven members of East Carolina University's ROTC completed four days of testing, including a control day when they walked without load and three load carriage days that were randomized to a flat stage, -4% decline, and 5% incline. Load mass was 30 kg, and subjects attempted to walk for two hours under each treatment condition. Metabolic data was collected during the last five minutes of every twenty minute stage. **RESULTS:** Of the nine participants who completed four days of testing, only two were physically able to complete all of the two hour marches. In statistical analysis, Kaplan-Meier survival curves were used to measure load carriage fail time between groups (those who finished and those who did not) and by testing condition.

Significance was found for VO_2 between groups of those who completed versus those who did not complete the two hour tests at p=0.0098. VCO_2 was found to be significant between groups at p=0.0084. Significance by test was found to be significant for VO_2 during the flat phase. VE and RER were not significant. Based on thresholds, the incline proved to be the most physically challenging test, followed by the flat then the decline phases. Threshold cutoffs were clinically determined to observe if the load carriage indicators (VO_2, VCO_2, VE, and RER) were associated with load carriage fail time. Those who reached the incline, decline, and flat thresholds in the first 60 minutes were more likely to fail the two hour time endpoint. The trajectory of the data was relatively stable throughout and did not exponentially increase as was hypothesized during the second hour. While there was some variation, it appeared that the metabolic measures at which participants started each test did not vary tremendously over time, suggesting that if participants started around the threshold cutoffs, they were more likely to fatigue quicker. **CONCLUSION:** Overall, these findings suggest that the measured variables do not increase over ruck marches lasting two hours in duration. Initial metabolic values may be indicative of how long an indidvidual may be able to exercise beyond 60 minutes until fatigue. This information can help military commanders identify soldiers at high risk of fatigue and monitor their load, pace, and fueling strategies to limit loss of manpower and injury.

Energy Cost of Heavy Load Carriage and Prolonged

Walking in ROTC Collegiate Participants

A Thesis

Presented To

The Faculty of the Department of Kinesiology

East Carolina University

In Partial Fulfillment

of the Requirements for the Degree

Master of Science in Exercise and Sport Science

By:

Sara Krajewski

April, 2014

ENERGY COST OF HEAVY LOAD CARRIAGE AND PROLONGED WALKING IN ROTC

COLLEGIATE PARTICPANTS

by

Sara Krajewski

APPROVED BY:

DIRECTOR OF THESIS: ____
(Laurel Wentz, PhD)

COMMITTEE MEMBER: ____
(Jason Brinkley, PhD)

COMMITTEE MEMBER: ____
(Paul DeVita, PhD)

COMMITTEE MEMBER: ____
(Zachary Domire, PhD)

COMMITTEE MEMBER: ____
(Robert Hickner, PhD)

CHAIR OF THE DEPARTMENT OF KINESIOLOGY:

____
(Stacey Altman, JD)

DEAN OF THE GRADUATE SCHOOL:

____
(Paul J. Gemperline, PhD)

ACKNOWLEDGEMENTS

Without the guidance and direction of the faculty on my thesis committee, this project would not have been possible. I would like to thank Dr. Laurel Wentz, Dr. Jason Brinkley, Dr. Paul DeVita, Dr. Robert Hickner, and Dr. Zachary Domire for their invaluable feedback and support. I would also like to thank Dr. Brinkley for helping me with my complicated statistics. He not only encouraged me to think critically about my data but he was able help me improve my computer skills by learning how to use a statistical program I will use in the future.

I would also like to thank my family for their love and support. My brother Kevin Krajewski inspired this project with his attendance at The United States Military Academy at West Point. As a physician assistant I would like to continue working with those in the military or be part of continuing military research. I would also like to thank my classmates and roommates, Miranda and Christina for their wonderful friendship during my two years here at East Carolina University. I would especially like to thank Miranda for not only being there for me for support but for her endless help with formatting, making tables, library visits, and venting sessions. It was helpful to have a friend going through the same process and I couldn’t ask for a better person to go through it with. Lastly, I would like to thank my boyfriend James Sasser for giving me never ending support during this time and for believing in me when I did not think I could do it. I am very thankful to have been given this opportunity and I feel very proud to call this work my own.

TABLE OF CONTENTS

List of Figures

List of Tables

CHAPTER 1: INTRODUCTION

In certain occupations, the human body carries heavy loads across vast and arduous distances. Firefighters, rescue workers, construction workers, and military personnel carry heavy loads on a regular basis, which restricts their movement and may cause musculoskeletal injuries and metabolic system problems. The load military soldiers carry while marching has increased throughout history, as the military routinely uses rucksacks ranging from 22-68 kg, depending on the type of mission and weather conditions (Van Dijk, 2009). During military engagement, soldiers carry these heavy rucksacks and trek long distances prior to completing critical military tasks in which speed and accuracy are of great importance. In order to ensure the safety of military personnel, load carriage studies have examined the energy cost of walking under load. Previous studies have shown that load weight, walking speed, and body mass, together with terrain factors such as gradient and surface, all have a direct effect on energy expenditure (Patton, Kaszuba, Mellow, Reynolds, 1991). Since energy expenditure is a limiting factor in performance, it is an important component of combat readiness.

Previous research has shown that carrying heavy loads during ruck marches expends around 300 kcals of energy more per hour than marching without a pack (Van Dijk, 2009). The same study suggested that march speeds should be reduced when loads are heavier (25-60 kg) to stay under this rate. Matching loads and speeds for a given distance and terrain can be helpful for dictating the soldier’s capacity to perform his or her job for extended periods of time. An increase in metabolic load affects how fast soldiers can move, inhibits movement over obstacles, increases fatigue upon arrival, increases calorie needs, and increases the risk for injuries (Beekley, Alt, Buckley, Duffey, & Crowder, 2007).

The majority of research conducted on military load carriage and ruck marches have lasted for short bouts of exercise of anywhere from 30-45 minutes. These shorter bouts of exercise have shown lower mean oxygen uptake (VO_2) measures compared to those who have had subjects exercise for longer periods of time (Quesada, Mengelkoch, Hale, & Simon, 2000, Beekley et al., 2007, & Pal et al., 2009). Previous studies on load carriage assumed a steady-state condition occurs for continued marching and did therefore not address the possibility of an increase in metabolic variables with prolonged heavy load carriage (Quesada et al., 2000 & Beekley et al., 2007). The few studies that have examined load carriage for two hours noted conflicting results, as some did find linear increases in metabolic variables and some did not (Epstein et al., 1988 & Patton et al., 1991). Some research suggests that prolonged load carriage should not exceed the range of 33% to 40% of VO_{2max} so as to not induce fatigue (Quesada et al., 2000). It's also been shown that heavier personnel were able to march for a longer duration than lighter individuals while carrying the same load (Bilzon, Allsopp, Tripton, 2001).

In addition to duration of marches, speed and terrain influence energy cost. Typically military leaders set a ruck marching pace for members of a company or battalion. Walking at a speed of 1.3 meters per second accurately represents normal adult locomotion and mean walking speeds during military missions while carrying loads up to 40 kg (Grenier, Peyrot, Castells, Oullion, Messonnier, & Morin, 2011; Pal, Majumdar, Bhattacharyya, Kumar, & Majumdar, 2009). Previous studies examining load carriage at faster speeds have lasted for a shorter period of time, which may have given an incomplete examination of energy cost. The type of terrain also influences energy cost and previously conducted studies have failed to examine energy cost of a decline phase in ruck marching and focused solely on walking on a flat surface or at an incline (Knapik, Reynolds, & Harman, 2004). The eccentric muscle contraction of downhill walking can be taxing on the body and is important to study due to the change in muscle action and potential change in energy cost from incline and flat phases (Minetti, Moia, Roi, Susta, & Ferretti, 2002). In the arduous terrain such as the mountains of Afghanistan, an individual walking uphill eventually will walk downhill, a condition this study is examining. This study aims to fill gaps in research by examining the energy cost of prolonged load carriage at various treadmill gradients, including a decline, to determine if a linear relationship exists between duration of exercise and metabolic variables or if a steady state condition does really exist.

STATEMENT OF THE PROBLEM

Soldiers carry heavy loads for prolonged time periods and on arduous terrain. However, past research has not fully addressed the energy expenditure initiated by such activity. Previous studies by Quesada et al., (2000), Beekley et al., (2007) & Pal et al., (2009) have asked subjects to march for under one hour under loads of 10-25 kg, which may not realistic in the field. Furthermore, data extrapolated from these studies may not apply to prolonged walking, in which subjects may see increases in energy expenditure. Little data has been collected regarding their energy expenditure during walking of up to two hours at various treadmill gradients, including a decline phase. Therefore, the objective of this study was to measure the energy cost of prolonged load carriage over a range of treadmill gradients by East Carolina University collegiate ROTC members. Previous research of shorter duration has assumed a steady state situation occurs across each metabolic condition, which is not hypothesized to be the case. This research is significant because the results can help officials create recommendations for the amount of load soldiers can safely and reasonably carry for up to two hours with minimal risk of injury or fatigue at different gradients. The results could also help officials recommend refueling strategies to prevent fatigue and enhance performance when soldiers are required to carry loads of 30 kg.

RESEARCH HYPOTHESIS

1) Energy cost, as measured by VO_2, VCO_2, VE, RER, with a 30 kg rucksack, will increase significantly between 60 and 120 minutes for each treadmill condition compared to the first 60 minutes of walking.
2) The incline will be the most physically challenging as the thresholds for each metabolic value are hypothesized to be the highest during this stage followed by the decline stage, followed by the flat stage.

Definitions

For the purpose of this study, the following terms were defined:

Load Carriage - The ability of an individual to carry a weight, in this instance in a ruck sack, while walking

Maximal Oxygen Consumption (VO_{2max}) - The maximum possible oxygen uptake that a person can achieve.

Oxygen Uptake (VO_2) – The uptake of oxygen and a measure of the volume of oxygen used by the body to convert the energy from food into energy molecules.

Rate of Perceived Exertion (RPE) - How hard an individual believes they are work on a scale such as the Borg Scale (6-20). A 6 being the individual is not exerting themselves much and a 20 being maximal effort.

Respiratory Exchange Ratio (RER) - The ratio of carbon dioxide to oxygen uptake.

Ventilation Exchange (VE) – The exchange of gases, mostly oxygen and carbon dioxide, during the passage of air into and out of respiratory passages.

VCO_2 - Rate of Elimination of Carbon Dioxide

Delimitations

Age **-** East Carolina University's ROTC members range in ages from 18-25 years old.

Duration **-** The testing will last for two hours in duration.

Gradient **-** The treadmill will be set to 0%, 5% incline, and a -4% decline.

Sample Size **-** Based on previous research a sample size of eleven subjects was chosen, comprised of male participants. The majority of military load carriage studies have used approximately 6-15 participants (Beekley et al., 2007, Epstein et al., 1988, Pal et al., 2009, & Quesada et al., 2000).

Speed **-** Walking speed will be set at 1.3 meters per second.

Weight of Load **-** A 30 kg load will be used, representing the mean military approach march load

Weight of Subjects **-** Participants will vary in weight and will not be excluded based on their weight.

Limitations

Age **-** The results of this study can only be applied to college aged male participants. With age, VO_2 decreases so future work could examine the energy cost of older military personnel.

Treadmill **-** This study will not be completed in the field so the results are only a representation of how energy expenditure differs while on a military mission of approximately the same duration and gradient.

Abbreviations

HR - Heart rate

RER – Respiratory exchange ratio

RPE – Rate of perceived exertion

ROTC – Reserve Officers' Training Corps

VE – Minute ventilation

VCO_2 – Carbon dioxide produced

VO_2 – Oxygen consumption

CHAPTER 2: LITERATURE REVIEW

The loads soldiers carry have increased over time throughout military history and around the world. Until the 18th century, troops marched with loads that rarely exceeded 15 kg (Knapik et al., 2004). Auxiliary transport carried extra equipment through the 18th century. In current operations, soldiers carry considerable amounts of weight, some of which is removed if they contact hostile forces. Soldier mobility has been studied throughout history with an emphasis on acceptable soldier load carriage. Modern day battlefields require heavy loads; and despite the availability of transport, the need to carry loads is expected to continue (Van Dijk, 2009). This literature review examined research conducted among military personnel as it pertains to load carriage and energy expenditure. This chapter is divided into four sections: 1.) need for guidelines; 2.) metabolic cost of load carriage; 3.) decreased performance with load carriage; and 4.) duration of marching.

Need for Guidelines

Army rucksack weights vary depending on the type of activity in which the soldier is engaged. A training load is normally lighter than the rucksack used in the field. Fighting loads typically weigh more than the recommended 21.7 kg and the approach march load and emergency approach march load normally exceeds the recommended 32.7 kg (Van Dijk, 2009). Approach march loads are rucksacks military personnel use as a basic ruck and on a daily basis, whereas emergency approach march loads are heavier due to necessary emergency equipment that may be necessary if personnel find themselves unable to return to base for certain lengths of time. There are currently no guidelines suggesting what optimal exercise intensity soldiers should be working at while carrying specific loads. Most of the research on load has focused on weights relative to the subject’s body mass. In one such study, subjects walked with loads starting at 0% of their body mass and up to 75% of their body mass until they reached a steady state period, at least three minutes (Bastien, Willems, Schepens,& Heglund, 2005). Based on their analysis, energy expenditure was measured at steady state with a RER ≤ 1, suggesting that the exercise could be maintained over time. However, the authors suggested that the energy cost may increase over extended walking due to muscular fatigue (Bastien et al., 2005). Given that each service member is generally expected to carry similar weights, regardless of gender or body mass, these data are not realistic to apply to military operations. For example, a soldier who is 5 feet tall will likely be required to carry a load of approximately the same weight as a soldier who is 6 feet tall, assigned to the same unit and mission. Therefore, assessment of energy expenditure at a constant load creates scenarios observed in the field.

Metabolic Cost of Load Carriage

Metabolic or energy cost can be measured by oxygen consumption (VO_2), elimination of carbon dioxide (VCO_2) ventilation exchange (VE) and respiratory exchange ratio (RER). VO_2 is defined as a measure of the volume of oxygen used by the body to convert the energy from food into energy molecules and is influenced by a number of factors including age, diet, hydration status, gender, fitness level and military training. VCO_2 is the rate of elimination of carbon dioxide.VE is the exchange of primarily oxygen and carbon dioxide during inhalation and exhalation. RER is the ratio of carbon dioxide exhaled and oxygen inhaled in one breath. This ratio can be used to determine if the subject is metabolizing carbohydrates, fats, or a mixture of both as fuel to provide the body with energy. Typically, an RER of 0.70-0.85 indicates that fats are being used as the primary energy source. An RER between 0.701 and 0.999 indicates a combination of carbohydrates and fats as the energy source. Once the RER is above 1.0, carbohydrates are metabolized as the primary energy source and correlate with higher intensity activity. These four metabolic parameters are used to measure energy cost for this study when military participants are asked to walk under load for up to two hours in duration at various treadmill conditions.

Soldiers carry variable loads based on the task they are expected to perform. According to load carriage guidelines, approach march loads are recommended not to exceed 32.7 kg but typically do (Van Dijk, 2009). Many factors influence the soldier's ability to march under load including load mass, marching speed, terrain, distribution of the load, volume of the load, and the physical condition of the soldier (Knapik et al., 2004). The type of terrain is hard to examine in the laboratory so most research has used different incline phases to simulate soldiers walking uphill in the field. Previous research has examined the metabolic cost of walking at an increasingly steep grade over time. However, each study differs in the load carriage conditions. One study found physiological responses to prolonged, heavy load carriage increased significantly over time even at exercise intensities below 30% VO_{2max} (Patton et al., 1991). This study used grades of 0% and 5%. The grade increased by 2.5% until a plateau in VO_2 was observed. It was also observed that VO_2, VE, and heart rate were not constant but increased significantly during prolonged walking as the incline increased. Another study found that increases in load caused fatigue, once the work intensity exceeded 50% of VO_{2max} (Epstein et al., 1988). These data conflict with the results shown by Patton et al. (1991), who suggested a work intensity of 30% VO_{2max} causes fatigue. This discrepancy could have been due to differences in load carriage conditions between studies, i.e. speeds, loads, length of march, or higher aerobic capacity in subjects of this study compared to those of Epstein et al., (1988). Another study noted that as the load and treadmill gradient increased, VO_2 drastically increased as well. When carrying increasingly heavier loads, humans become less efficient and the energy expenditure per kilogram of load increases sharply (Lyons et al., 2005).

These previous studies have shown the metabolic cost of different inclines compared to each other with differing loads, as well as how energy cost with loads based on percentage of body mass differ at a flat condition. However, the metabolic cost during downhill walking, to our knowledge, has not been examined in military personnel walking under load. In the arduous terrain such as the mountains of Afghanistan, an individual walking uphill will eventually walk downhill, a movement that recruits different muscle actions and eccentric muscle contraction, which may alter metabolic cost. Walking downhill caused muscles to have problems coping with the tendency of the body to accelerate, rather than maintain a constant speed, which increased the risks for iliotibial band friction syndrome and intravascular hemolysis (Minetti et al., 2002). Walking downhill in a controlled manner is more important than walking downhill with power, in order to prevent injury.

Decreased Performance with Load Carriage

Previous research on metabolic cost has shown that increased metabolic cost causes increased physical fatigue and reduced performance (Grenier et al., 2011, Pal et al., 2009, Lyons et al., 2005, & Quesada et al., 2000). One study found a linear increase in heart rate, VO_2, and energy expenditure with increasing load weight (Pal et al., 2009). It was also noted that an increase in speed was associated with a greater increase in these physiological responses. Two other studies examining the effects of load carriage on walking energetics found the metabolic cost required (i.e., used by the muscles) to carry one unit of equipment mass was higher than the metabolic cost required to carry one unit of body mass. This shows how important the muscular demands are to carry military loads (Grenier, Peyrot, Castells, Oullion, Messonnier, & Morin, 2011; Bastien et al., 2005). Therefore, adding external weight to an individual's body increases energy expenditure. Even with constant velocity, increases in VO_2, heart rate, and ventilatory exchange were found with an increase in load (Beekley et al., 2007). Beekley et al., (2007) also found that increases in metabolic cost due to carrying heavier loads affected soldiers mobility, specifically speed, agility over obstacles, and fatigue. Elevated metabolic cost increased calorie or energy expenditure and increased the risk for injuries

From a military perspective, it is essential to understand the metabolic demands of load carriage if soldiers are expected to perform successful missions on the battlefield. Systematic decrements in the performance of long distance runs, jumps, short sprints, and obstacle course traversals while carrying a load have been observed (Knapik et al., 2004). Furthermore, very strenuous marches with loads of 34-61 kg over 10-20 km distances lead to post-march decrements in marksmanship and grenade throwing distance (Knapik et al., 2004). Therefore, increasing soldier's loads may negatively affect road marching performance and ultimately their performance in battle.

Fatigue with Prolonged Walking

Military personnel are commonly afflicted with lower extremity overuse injuries associated with high volume and high intensity of physical training (Wang, Frame, Ozimek, Leib, & Dugan, 2012). Major overuse injuries such as stress fractures, knee pain, and back pain result in significant medical cost and loss of training days to the military organizations (Wang et al., 2012). These types of injuries result in extended periods of recovery as well, delaying return to duty. Especially during basic training, strenuous training and walking with heavy loads exposes military personnel to significant physical fatigue. Wang et al., (2012) suggest that prolonged walking with load carriage could lead to significant neuromuscular impairment. In addition, carrying a load may increase lower limb loading, placing high stresses on the soft tissues surrounding the lower limb joints (Simpson, Munro, & Steele 2012). Furthermore, kinematics are altered during prolonged walking in an attempt to reduce impact forces and therefore increase injury potential. Quesada et al., (2000) showed that significant increases in peak plantar flexion and peak knee flexion occur in male soldiers who marched for 40 minutes with loads of 15% and 30% of body weight, demonstrating that kinematics change over time. In longer marches reaching two hours, Epstein et al., (1988) suggested that an increase in VO_2 over time is due to the reduction in mechanical efficiency due to altered locomotion biomechanics as the participant adjusts to the weight of the pack. Participants may also alter their gate and center of mass to compensate for the heavy load and gradient. Thus, load carriage and fatigue are two major stressors experienced by military personnel.

While examining military metabolic cost, studies have varied in focus from studying walking speed, gradients, loads, and lastly durations. One study examined energy cost of increasing loads while running at 10, 14, and 18 kilometers per hour for a total of 30 minutes (Beekley et al., 2007). Another study focused on the duration of exercise had subjects perform one hour of treadmill walking at 4 kilometers per hour (Lyons et al., 2005). Further research had subjects walk at a slow and moderate pace for 30 minutes while wearing body armor that increased body weight by 15.7% (Ricciardi, Deuster, & Talbot, 2008). The researchers noted a 12% and 17% increase in VO_2 with the increased weight, a difference that caused subjects to experience muscle fatigue and reduced physical endurance. In a different protocol, participants ran and walked for 40 minutes at six kilometers per hour, and it was determined that an exercise intensity should not exceed approximately 33 to 40% VO_{2max} so as not to induce fatigue. Based on these results, it was recommended the intensity of activity should not exceed approximately

33 to 40% of VO_{2max} to avoid early onset of fatigue (Quesada et al., 2000). Greater increases in metabolic and cardiovascular demands were noted using heavier loads and left subjects feeling more fatigued according to their VO_2 (Lyons et al., 2005). One of the few studies that has examined energy cost over prolonged walking found physical fatigue sets in once the work intensity exceeds 50% of maximal work capacity when subjects marched for two hours (Epstein et al., 1988). Relatively shorter duration studies have assumed a steady state occurrence during exercise but did not consider that energy cost of exercise may increase over time. In military operations, soldiers are likely going to be walking for longer than 30-45 minutes so therefore it is important to identify the effect of prolonged duration on fatigue and metabolic cost while carrying a fixed yet heavy load.

Due to the fatiguing consequences of prolonged walking under load, increases in exercise intensity are thought to increase metabolic cost over time. For example, metabolic cost is higher while walking on an incline than walking on a level surface and even a decline surface (Minetti, Ardigo, & Saibene, 1994). A hypothesized increase in metabolic cost over time is thought to result in accordance to results from Patton et al., (1991) who examined prolonged load carriage in highly endurance trained non-military men. Increases in VO_2 and VE were observed during prolonged walking with external loads due to increased body temperature, increased blood lactate, reduced mechanical efficiency, and a shift in substrate utilization (Patton et al., 1991). In the previously mentioned study, blood lactate was ruled out as a contributing factor due to the fact that it did not change from baseline levels following any of the load carriage trials. It is thought that energy cost increment is related to altered biomechanics due to the increase in extra weight that might induce fatigue in the individual (Epstein et al., 1988). Furthermore, work intensities that may elicit a VO_2 of more than 50% of VO_{2max} are characterized by significant anaerobic metabolism and lactic acid production, which may be the cause of reduced performance and an increase in energy cost over time (Epstein et al., 1988). As skeletal muscle fatigue occurs, the individual may recruit additional muscle mass and alter gait to carry the load (Epstein et al., 1988). A gradual increase in fatigue is thought to cause a linear increase in metabolic cost over time.

Summary

In general, military load carriage studies examining energy expenditure have been done, but differ greatly on whether speed, load, gradient, or duration cause an increase or steady state in metabolic parameters. This is an important topic of study due to the practical military implications that would help keep personnel safe and effective while carrying out military tasks. Current military recommendations suggest loads not exceed 33 kg whereas research suggests VO_2 should not increase beyond 35-50% of VO_{2max} so as not to induce fatigue. However, modern warfare creates mission requirements that make load recommendations impractical, placing soldiers at risk for injury and fatigue upon entering combat. Military walking generally exceeds 30 to 60 minutes so prolonged walking studies lasting much longer than one hour are needed. Therefore, this study aims to fill gaps in the literature by examining ruck marching with a 30 kg load, at a fixed speed, for two hours in duration at various treadmill gradients, to include a decline. These results could help military commanders create guidelines for refueling soldiers properly when heavy load carriage is required for mission performance so they exercise maximal combat effectiveness. Since it is hypothesized that energy expenditure will increase sharply during the second hour of testing due to decreased efficiency with neuromuscular fatigue, commanders should be aware of how far their men and women can ruck efficiently and safely.

CHAPTER 3: METHODOLOGY

In this study, the metabolic cost of one to two hour walking trials with and without a 30 kg load was measured across different treadmill conditions. On the first day of testing participants walked without load at the three treadmill conditions, for 20 minutes, with a five minute break, in a randomized order. On days two, three, and four participants walked with a 30 kg load for two hours at a randomized treadmill condition. A load of 30 kg represents an appropriate load soldiers carry in the field during an approach march. Participants were college-aged males outfitted in full combat uniform. Prior to beginning the study, participants completed a survey that asked about weekly training and recreational exercising habits as well as demographic and background information. Participants were instructed to consume a standardized breakfast one hour before testing and ensure that they were properly hydrated to standardize their pre-testing calories and macronutrient content. Anthropometric data were collected at the beginning of the first visit to the laboratory.

Participant Selection

Participants for this study were recruited from East Carolina University's Reserve Officer's Training Corps (ROTC). They were current members of ECU's ROTC program or officers affiliated with ECU's ROTC program. Participants were college-aged males ranging in age from 18-25 years old. Eleven participants were recruited for this study. Research by Pal et al., 2009, Beekley et al., 2007, and Grenier et al., 2011, all completed their studies with 10 male subjects. Bilzon et al., 2001 and Quesada et al., 2000 used 12 male subjects and Patton et al., 2001 used 15 male participants. Since females were recently admitted to serve in combat positions, they were invited to participate in this study, although none did. This sample of participants is accustomed to carrying loads, in the form of a rucksack, as well as walking for prolonged periods of time due to their ROTC training. Participants were excluded from the study if they had any current musculoskeletal injuries that would prevent them from walking for up to two hours. This study did not limit the participant pool to a given height and weight so ROTC members of all heights and weights were included in the study. All procedures were approved by the University Internal Review Board (IRB).

Equipment

In order to examine the VO_2, VCO_2, VE, and RER of each participant, the Parvo Medics' TrueOne 2400 automated metabolic measuring system was used. This is a compact, integrated metabolic measuring system typically used for cardiopulmonary stress testing, indirect calorimetry, and maximal oxygen consumption measurement. The machine was properly calibrated before each participant. Participants were fitted with a 2-way valve mouthpiece, attached to a hose, which was connected to the a TrueOne 2400 metabolic measuring system, Parvo Medics, Sandy UT that was calibrated prior to testing according to the manufacturer's instructions. The gas tank consisted of 16% oxygen, 4% carbon dioxide, and a nitrogen balance. A 3-liter syringe was used to calibrate flow rate and flow meter prior to each test. Height (cm) and weight (kg) was taken prior to start of the treadmill test. Participants wore the mouth piece during the last five minutes for every twenty minutes of walking to obtain metabolic data. To monitor heart rate, the participant wore a heart rate monitor and watch provided by the FITT building. To determine hydration status of the participant, urine specific gravity was measured, using a refractometer, before and after each testing session. Participants walked on a treadmill provided by the East Carolina University's Biomechanics Department (see figures 1-3)

Figure 1

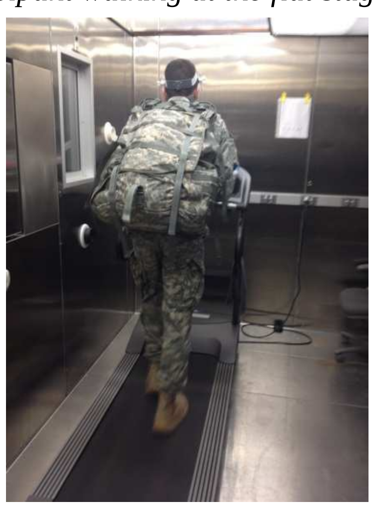
Treadmill used for testing

Figure 2

Environmental Chamber used for testing

Figure 3

Participant walking at the flat stage



Measurement Protocol

Each participant completed a survey about current and past ROTC training before testing began (Appendix D). The survey asked questions such as, "How many miles do you run each week?" or "How heavy is your average rucksack?" The purpose of the survey was to determine the amounts of physical activity participants engaged in each week as well as what types and how much military training they perform each week. After completing the survey, height and weight was recorded, using the Detecto scale attached to the wall. Height was recorded to the nearest 0.1 cm. Weight was measured using a calibrated medical scale and measured recorded to the nearest 0.10 kg. Body fat percentage was determined by Duel Energy X-Ray Absorptiometry (DXA) located in the FITT building. The measurements were taken with a Lunar Prodigy Advance Dual Energy X-ray Absorptiometry, General Electric, Madison WI. Prior to using the DXA machine, calibration was completed to ensure the machine would deliver a precise and accurate scan. Before the scan, the technician entered the subjects' weight, height, ethnicity, and gender into the software. The participant then lay supine with their body inside the rectangular reference box. Subject's ankles and leg were strapped together and participant's hands and arms were placed as close to their sides as possible making sure not to overlap onto their body. The subject was instructed to stay as still as possible during the scan that lasted between six to twelve minutes depending on body type.

Participants were asked to arrive hydrated before each testing day, which was confirmed with urine specific gravity measurements. Also, participants were given a list of seven breakfast meals, and asked to choose one and consume it an hour before each testing session. They were asked to consume the same meal before each testing session. The meals ranged in calorie content from 250-350 calories. Examples of meal options included 8 ounces of orange juice, one cup of frozen fruit, one half scoop of protein powder, and eight to twelve ounces of water. Another meal example was one granola bar, twelve ounces of a sport drink, one string cheese, and eight to twelve ounces of water. The walking portion of the study took place in the environmental chamber located in the Ward Sports Medicine Building with temperature set to 50% humidity and 20 degrees Celsius to eliminate temperature as a variable. When participants walked for two hours, they were given a five minute water and rest break 40 minutes and 80 minutes into the walking session. Metabolic data was collected during the last five minutes of each 20 minute stage.

There were a total of four testing days. The first day tested participants without load to establish baseline control measurements as well as to familiarize participants with the testing protocol. Participants were asked to walk for a total of one hour. They were randomly assigned to the flat, -4% decline, and 5% incline and completed one-twenty minute stage at each treadmill gradient. Metabolic data was collected with the VO_2 mask during the last five minutes of every twenty minutes. A five minute break was given between each twenty minute trial on the first day of testing without load. Participants walked at a speed of 1.3 meters per second, which translates to 3.0 miles per hour. Each day of testing was attempted to be separated by one week. However, due to outside commitments and circumstances, testing days were separated by anywhere from three days to one month. On the second day of testing, participants attempted to walk for two hours. Participants were randomized to one of the following conditions: 0% grade, -4% decline, or 5% incline while carrying 30 kg rucksack. Subjects performed the remaining walking phases on the third and fourth days of testing.

The treadmill gradients were chosen based on previous research methodology. The 5% incline was chosen based on research that examined moderate and higher inclines that used grades of 0, 3, 6, 9% as well as 5 and 10% (Lyons et al., 2005) and (Ricciardi et al., 2008). Both of these studies used speeds of 2.5-3.6 miles per hour. The decline was chosen to determine the energy cost while walking at a slight decline. Only water was permitted and no sports drinks were allowed until after the walking was complete. Snacks were also provided at the end to help with recovery from the exercise bout. Participants were permitted to quit exercise if they became too fatigued to complete two hour march. Since the goal was to have each participant reach the two hour end point, verbal encouragement was used to push the participants to walk until they could not go any longer. Once participants verbalized their fatigue and verbal encouragement no longer proved to be effective, the test was terminated. If participants were able to walk for two hours, the test was terminated after 120 minutes.

Statistical Analysis

For each participant, VO_2, VCO_2, VE, and RER were analyzed from the three days of walking for two hours at the various treadmill gradients with load. The JMP® and Microsoft Excel programs were used for survival analysis. Through the JMP software, the nonparametric method of using the Kaplan-Meier Estimator was used. The Kaplan-Meier method estimates survival probabilities computed using a product limit formula (Kleinbaum, 1996). Survival analysis is a collection of statistical procedures for the analysis of data in which the outcome variable of interest is time until an even occurs, in this instance, reaching the 120 minute time duration, or failing to meet that two hour time goal (Kleinbaum, 1996). Censoring is a key data analytical problem and occurs when some information about individual survival time is available but the exact survival time is not known (Kleinbaum, 1996). The data was right-censored because the true survival time interval, which is not really known, has been cut off, censored, at the right side of the time interval, giving the actual survival time observed. The resulting Kaplan-

Meier survival curves were compared to each other to determine significance using the Log-Rank test.

Survival graphs were created to determine significance for the log-rank test which was found at $p < 0.05$. The log-rank test of significance compared the three treadmill gradients to each of the four metabolic variables. The log rank test is a large sample chi square test that uses as its test criterion a statistic that provides an overall comparison of the Kaplan-Meier curves being compared (Kleinbaum, 1996). This determined whether or not significance was found between each the flat, decline, and incline stages compared to the threshold cutoffs of one variable. The survival graphs compared survival at specific time points, every twenty minutes of the two hour test. After the data was analyzed, spaghetti plot graphs were created to depict the metabolic variable among each of the individuals. The lines were connected between missing data points to further examine the slope of the lines over time. From there, a trend was observed that those who did not finish the two hour test had overall higher metabolic measures than those who were able to walk for the full two hours. Because of this, threshold cutoffs were created based on clinical predictions, to observe any potential increases in slope. It was hypothesized that the slope of the lines would increase over time during each treadmill condition. Also, if a participant had mean VO_2, VCO_2, VE, or RER values above the determined threshold during the first 60 minutes of the test, it was hypothesized they would be more likely to end the test early. Participants who did not reach the threshold cutoffs were believed to be able to walk further, possibly for the entire two hour test. The threshold cutoffs were made essentially to create groups so it was clear to differentiate between those who did not finish the two hour test and those who did.

CHAPTER 4: RESULTS

Descriptive Statistics

The participants were recruited from East Carolina University's ROTC program. Table 1 shows the subject demographics and training history. The participants were all male, therefore results cannot be applied to females, and they had a mean age of 21.3 ± 2.3 years, ranging from 18 to 25. The mean height and weight of participants was 177.8 ± 5.5 cm and 75.0 ± 18.5 kg. Participants weighed a mean of 77.5 ± 18.9 kg with their gear on, including their full combat uniform and boots. Participants had a mean body fat percentage of 17.8 ± 4.0%, considered healthy by the American College of Sports Medicine (ACSM, 2014). No relationships between body fat percentage and exercise duration were observed.

Table 1
Participant Demographics and Survey Results

Characteristic	Mean ± SD	Range
Age (years)	21.3 ± 2.3	18 – 25
Height (cm)	177.8 ± 5.5	112.3 – 183.3
Weight (kg)	75.0 ± 18.5	56.5 – 93.5
Gear Weight (kg)	77.5 ± 18.9	58.6 – 96.4
University Year (yr)	2.63 ± 1.36	1.3 – 3.99
Body Fat (%)	17.8 ± 4.0	13.8 – 21.8
Years in ROTC (yr)	1.81 ± 1.72	0.09 – 3.53
Days Ran (days/week)	4.5 ± 0.93	3.6 – 5.4
Miles Ran (miles/week)	12.7 ± 4.10	8.6 – 16.8
Resistance (days/week)	3 ± 1.26	1.7 – 4.3
Resistance (hours/week)	1.4 ± 0.60	0.8 – 2
P.A. per week (hours/week)	11.8 ± 3.37	8.4 – 15.2
Mean Ruck (kg)	23.9 ± 10.09	13.8– 34
Heaviest Ruck (kg)	37.9 ± 17.48	20.4 – 55.4
2 Mile Run time (mins)	13.2 ± 0.60	12.6 – 13.8

According to completed surveys, participants ran 4.45 ± 0.93 days per week and 12.72 ± 4.10 miles per week. Mean ruck weight was 23.9 ± 10.09 kg and the heaviest ruck participants had ever carried weighed 37.93 ± 17.48 kg. No data were collected as far as how long they have rucked with a rucksack or the typical weights of the ruck when they do go on ruck marches for ROTC.

Figure 4

VO_2 across time during flat, decline, and incline treadmill stages

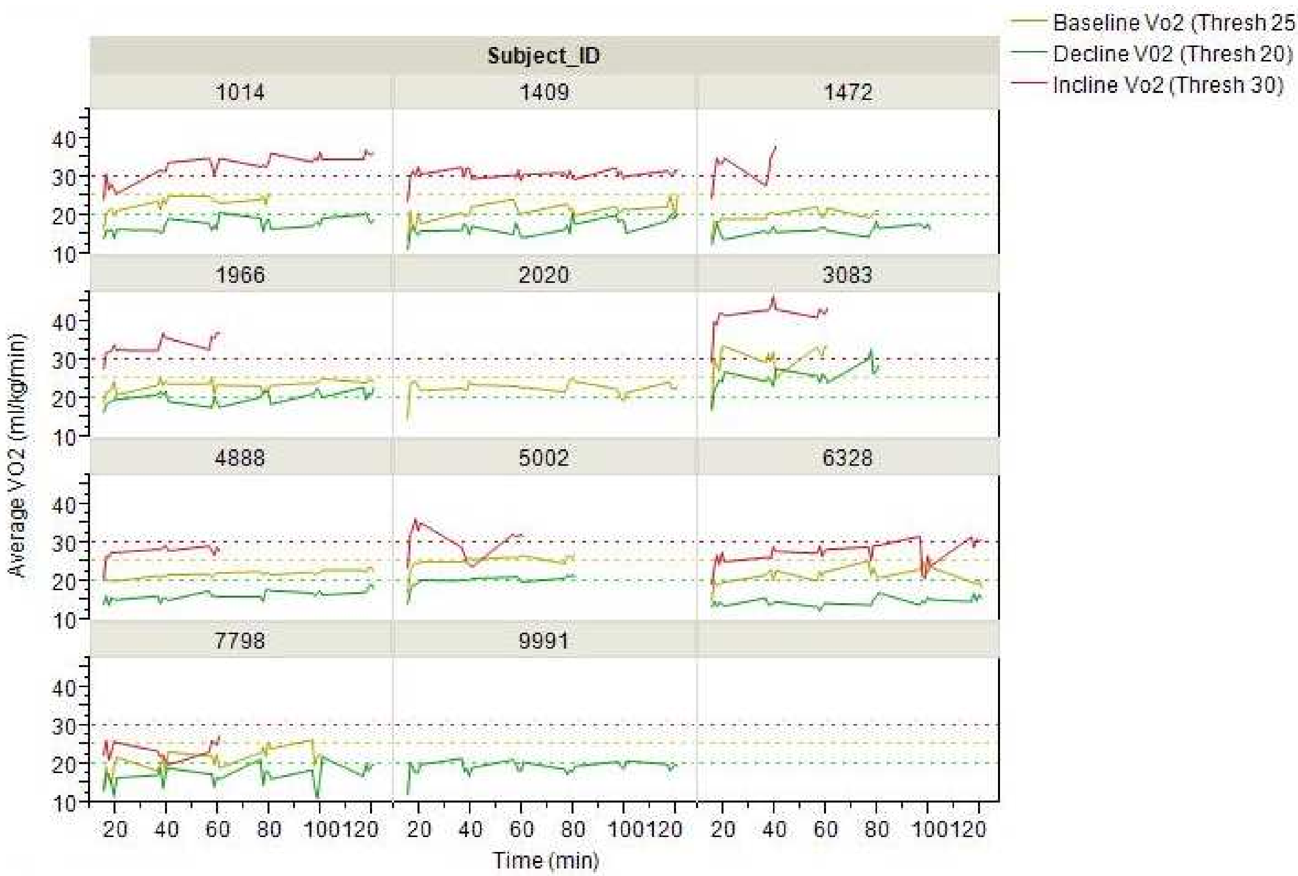
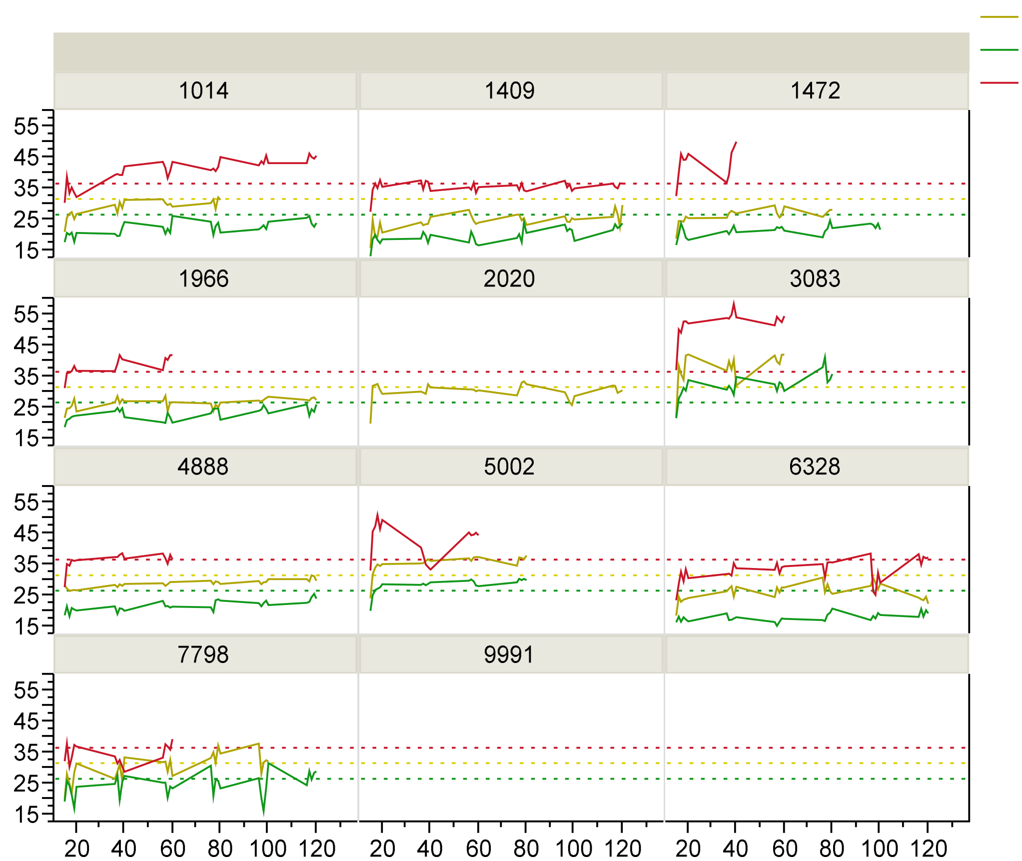


Figure 4 shows VO_2 levels for each treadmill condition. Of the three treadmill gradients, the incline condition had the highest VO_2 values with the threshold set at 30 ml/kg/min. Four subjects surpassed the incline threshold in the first 60 minutes and did not finish the full test. However, two of the three subjects who did finish the full test for the incline did rise above the threshold in the first 60 minutes. One subject who finished the test was able to stay under the threshold until around 80 minutes. The flat stage VO_2 threshold was established at 25 ml/kg/min. Four of the subjects who reached above threshold for the flat stage did not complete the two hour duration. Five subjects stayed under threshold and completed the full two hours. For the decline stage, the VO_2 threshold was set at 20 ml/kg/min. Two subjects who reached above threshold in the first 60 minutes did not complete the full two hours.

Figure 5

VO_2 across time during flat, decline, and incline conditions adjusted for fat free mass



After correcting for fat free mass in an attempt to observe differences in VO_2 related to body composition, no difference in VO_2 was observed. Threshold cutoffs for VO_2 were set at 36.5 kg/FFM/min for the incline, 31 kg/FFM/min for the flat, and 26.5 kg/FFM/min for the VO_2 decline. As in Figure 4, Figure 5 shows that those who finished the two hour test were generally under the clinically set thresholds and those who did not finish the two hour tests were generally above the VO_2 thresholds set for each condition.

Figure 6

VCO_2 across time during flat, decline, and incline treadmill stages

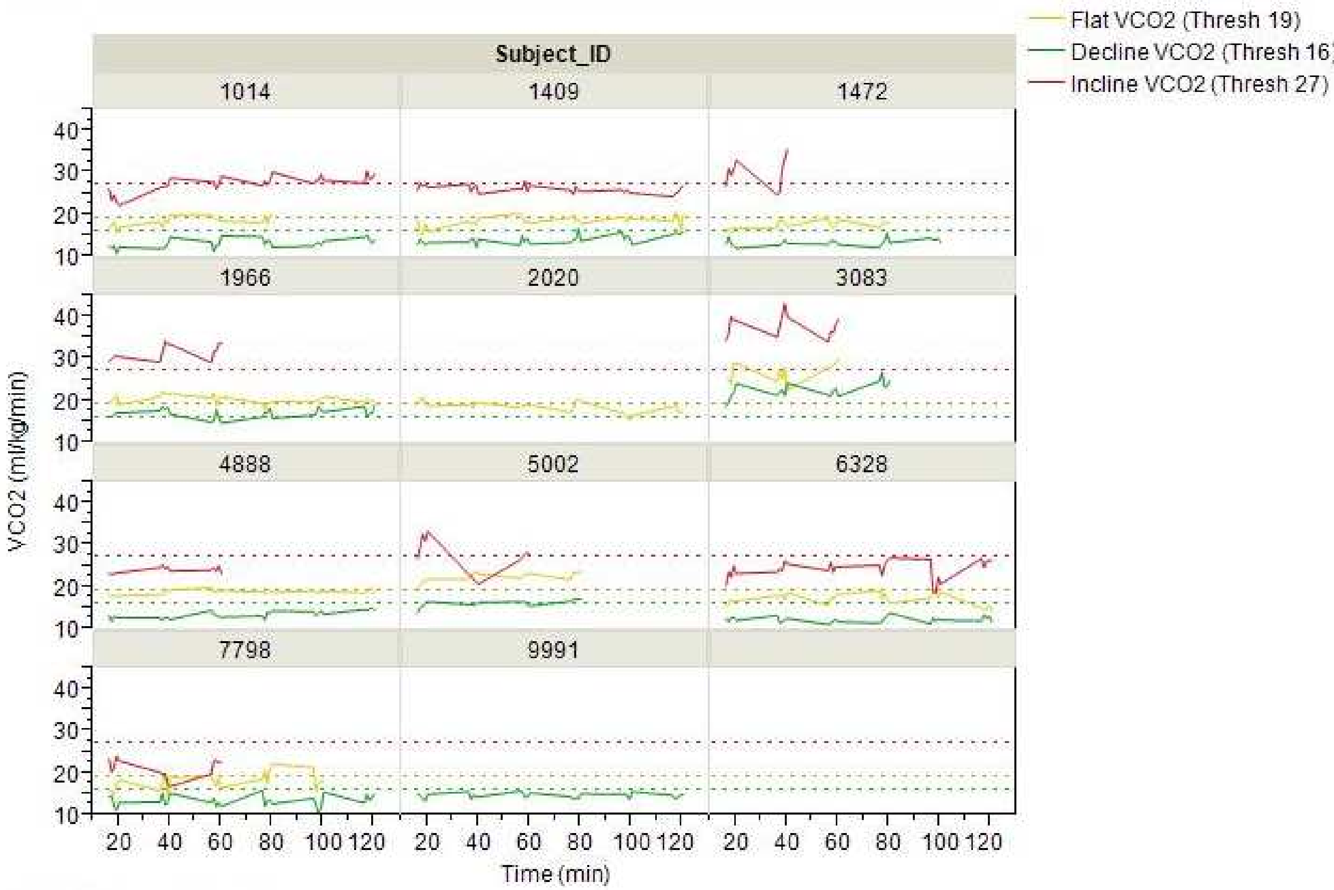


Figure 6 shows mean VCO_2 levels for each treadmill condition. Once again, the incline was the most strenuous of the three gradients, as mean VCO_2 measures were higher than the flat and decline stages. With the VCO_2 incline threshold set at 27 ml/kg/min, four participants passed threshold in the first 60 minutes and then stopped the test at 60 minutes. One subject who passed threshold in the first 60 minutes did finish the test but only slightly exceeded the threshold. The VCO_2 flat threshold was set at 19 ml/kg/min. Two participants surpassed threshold for the entire test and lasted 60 minutes. Those who did finish the full two hours were either below or right at the threshold and managed to walk the full two hours. The two subjects who ended the decline stage at 80 minutes were over the VCO_2 threshold at 16 ml/kg/min VCO_2 threshold most of the entire time at. Those who walked the full two hours were mostly below this threshold.

Figure 7

VE across time during flat, decline, and incline treadmill stages

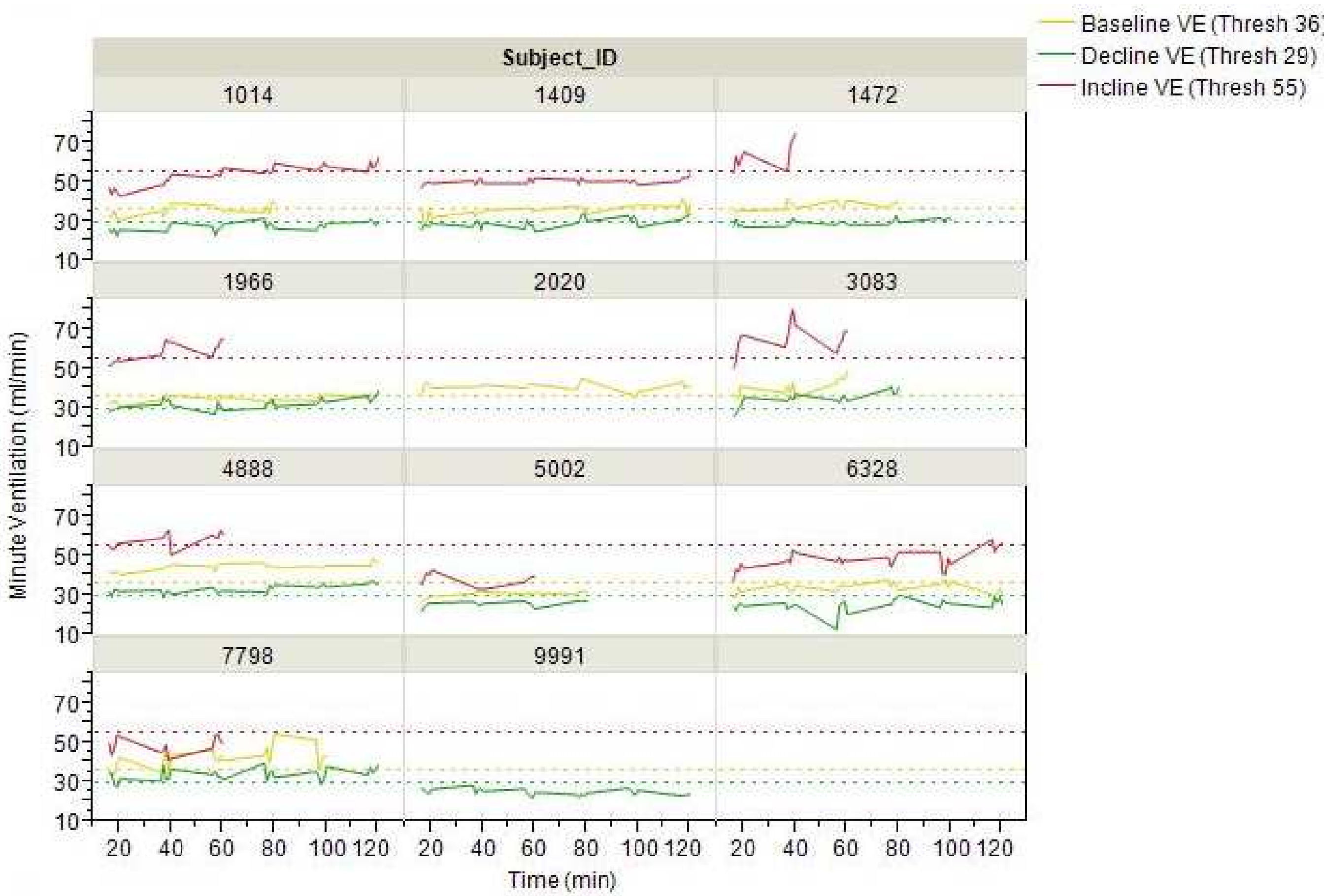
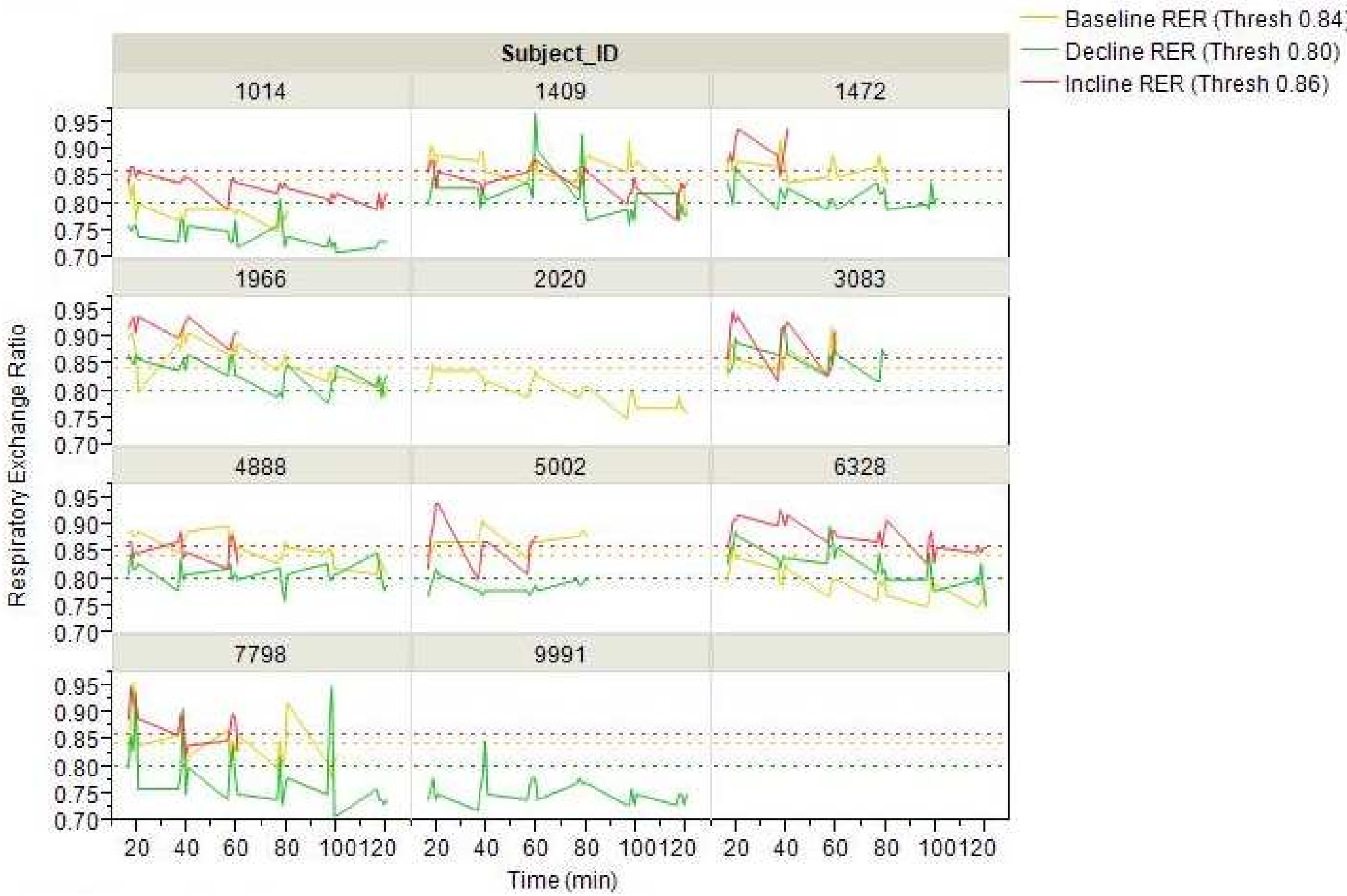


Figure 7 shows that VE for each condition. As in previous measurements, VE was highest during the incline stage. The VE incline threshold was set to 55 ml/min and four participants who crossed the threshold in the first 60 minutes did not finish the test. Those who did finish were right below or right at the threshold. Two participants walked only for 60 minutes and were under threshold, indicating they could have gone longer but ended early. Thresholds also could have been set at a level that may have been inaccurate. The flat VE threshold was set at 36 ml/min, and three subjects who did not walk for the full two hours were over the threshold. It is interesting to note that two subjects who finished the test were above threshold. However, their minute ventilation stayed constant throughout. The VE decline threshold was set to 29 ml/min. One participant was over threshold the entire time and did not finish the test. As in the flat stage, even some participants who crossed threshold managed to finish the test.

Figure 8

RER across time during flat, decline, and incline treadmill stages



RER produced different results than the other three variables. For RER, the incline threshold was set to 0.86, indicating a mixture of carbohydrates and fats being used as the fuel source. Those participants who did not finish the full test generally surpassed threshold. The RER flat threshold was set at 0.84, also indicating a mixture of carbohydrates and fats used as the fuel source. Some participants were able to stay below threshold and finish the test where as some were not. The RER decline threshold was set at 0.80 and there was variation between those who finished the test and crossed threshold and those who were below threshold and did not finish.

Kaplan-Meier Curves

Kaplan-Meier curves were used to determine the time to event, or failure, for participants who were not able to ruck march for the two hour duration that was asked of them for this study. While some participants had to leave early for class or another obligation, threshold cutoffs were made in attempt to identify participants who did not finish the two hour test due to fatigue versus an outside reason.

Figure 9

Kaplan-Meier Curve of time to failure by VO_2 threshold group between groups

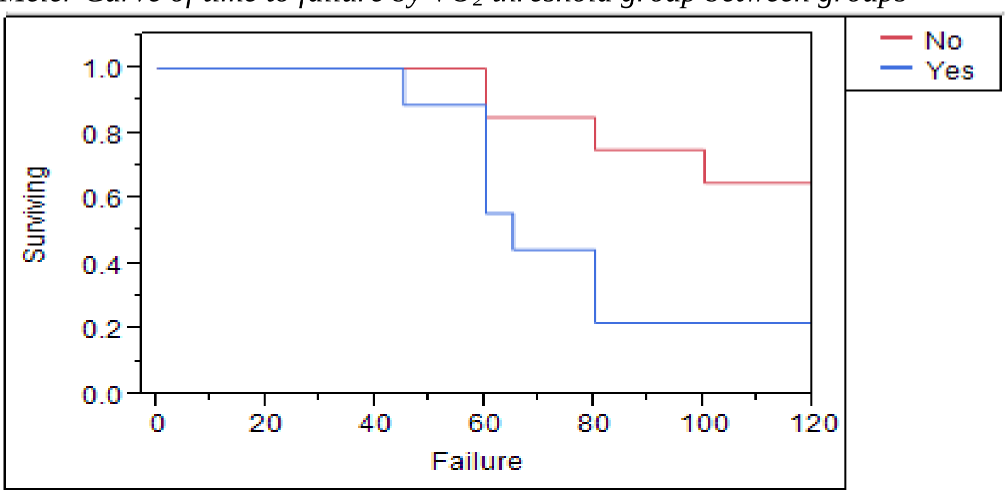
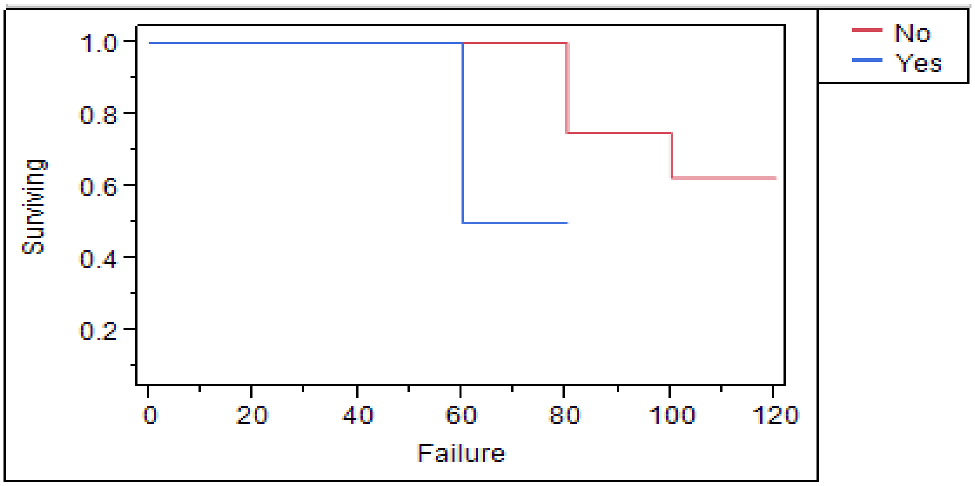


Figure 8 is the survival curve for VO_2 that demonstrats significance between groups with the Log-Rank test at 0.0098. VO_2 was found to be a significant measure for time to failure. 100% of particiapants made it to 40 minutes, but after that, those who failed the two hour test began to do so after 60 and 80 minutes respectively. Those who did not hit the VO_2 threshold in the first 60 minutes and walked the entire two hour test had a survival rate of around 0.70 (Figure 8). Those who did reach the threshold in the first 60 minutes had a survival rate of 0.20. Those who did not hit the VO_2 threshold had a higher survival rate than those who did suggesting that those who did not hit the VO_2 threshold were more likely to walk for a longer period of time. There was a significant difference in VO_2 between those who failed and those who survived the two hour test.

Figure 10

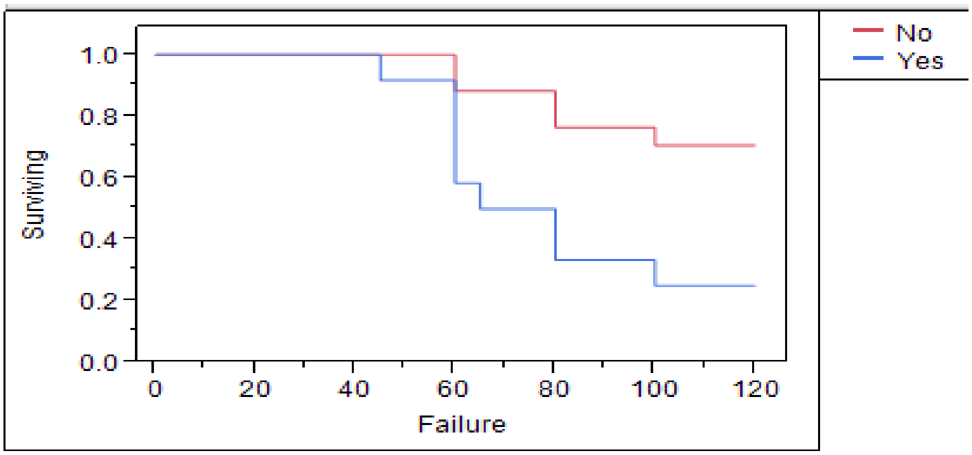
Kaplan-Meier Curve of time to failure by VO_2 threshold group at flat phase



When the data were computed to determine significance by the treadmill test, the flat phase was found to be significant for VO_2 with $p=0.0177$. Those who hit the VO_2 threshold during the flat phase had a survival rate of 0.50 at 60 and 80 minutes. Those who did not hit the VO_2 threshold during the flat phase had a survival rate of 0.60 at the two hour time endpoint. Similar to those who did not hit the VO_2 threshold during the flat phase had a higher survival rate than those who did hit the VO_2 threshold.

Figure 11

Kaplan-Meier Curve of time to failure by VCO_2 threshold group between groups



Significance for time to failure for VCO_2 thresholds between groups was found at $p=0.0084$. Those who did not hit the VCO_2 threshold had a survival rate of 0.70 and those who did hit the VCO_2 threshold had a survival rate of 0.20 indicating those who did not reach threshold had a higher survival rate.

No significance was found for VE and RER between groups as well as by test in the Kaplan-Meier curves. Significance was also not found for VO_2 by test for the decline and incline as well as VCO_2 by test for the flat, decline, and incline.

Table 2

Number of times participants reached thresholds

Subject ID	VO_2 Thresholds Yes	VCO_2 Thresholds Yes	VE Thresholds Yes	RER Thresholds Yes
1014	1	0	1	0
1409	0	0	0	2
1472	1	1	2	3
1966	2	3	2	3
2020	0	1	1	0
3083	3	3	3	3
4888	0	0	3	2
5002	2	3	1	2
6328	0	0	1	2
7798	0	1	2	3
9991	0	0	0	0

VO_2: Oxygen uptake

VCO_2: Rate of elimination of carbon dioxide

VE: Ventilation exchange. The exchange of gases, mostly oxygen and carbon dioxide, during the passage of air into and out of respiratory passages.

RER: Respiratory exchange ratio. The ratio of carbon dioxide to oxygen uptake.

Table 2 shows the number of times each participant reached the VO_2, VCO2, VE, and RER thresholds. Participants hit the RER threshold the most, followed by VE, VCO_2, and VO_2 thresholds. No significance was found for the VE and RER thresholds when those who went above it were compared to those who did not. One participant, ID 3083, hit the thresholds every time. He was not able to walk longer than 60 minutes for each of the three tests. Participant 9991 only completed the decline phase and did not reach threshold. This participant withdrew from the study for medical reasons. Participant 2020 only completed the flat stage and did not reach threshold.

Table 3

Percentage of participants who did or did not hit thresholds

	Decline	Flat	Incline
Thresholds	Percentage (%)	Percentage (%)	Percentage (%)
VO_2 No	72.73	81.82	63.64
VO_2 Yes	27.27	18.18	36.36
VCO_2 No	72.73	54.55	63.64
VCO_2 Yes	27.27	45.45	36.36
VE No	54.55	45.45	54.55
VE Yes	45.45	54.55	45.45
RER No	45.45	36.36	36.36
RER Yes	54.55	63.64	63.64

VO_2: Oxygen uptake

VCO_2: Rate of elimination of carbon dioxide

VE: Ventilation exchange. The exchange of gases, mostly oxygen and carbon dioxide, during the passage of air into and out of respiratory passages.

RER: Respiratory exchange ratio. The ratio of carbon dioxide to oxygen uptake.

In Table 3, thresholds were set in attempt to determine when participants were more likely to fail or not complete the two hour time duration. Table 4 shows the percentage of participants who did and did not reach threshold for VO_2, VCO_2, VE, and RER. For VO_2, 36.36% of the participants hit the threshold during the incline stage. Next, participants hit the VO_2 threshold 27.27% of the time during the decline stage. The flat stage had the lowest percentage of participants who did reach and did not reach threshold. For the decline, seven participants walked the entire time whereas three did not. For the flat stage, five participants completed the two hour duration and five did not. Even though more participants completed the two hour decline stage, more of them reached threshold than during the flat stage.

For the VCO_2 measure, the number of participants who reached the decline and incline thresholds was the same at 27.27% and 36.36% respectively. More participants reached the flat stage VCO_2 threshold than the VO_2 threshold at 45.45% compared to 18.18%. More participants reached threshold for the flat stage compared to the incline stage as well at 45.45% versus 36.36%. For VE, more participants reached the flat threshold as well. The percentage of those who reached the decline and incline thresholds was the same at 45.45%. Lastly, for RER, those who reached threshold were the same for the flat and incline stages at 63.64%.

In conclusion, spaghetti plot graphs were created to examine not only if a change in slope was observed but if participants who ended the tests early did so because they reached above the clinically predicted thresholds, suggesting participants may have been fatigued. There was no linear increased observed in the first or second hour of testing as was hypothesized and a steady state condition was in fact detected. The spaghetti plot graphs also were indicative of which of the tests was the most and least challenging due to having higher or lower threshold cutoffs. The incline proved to be the most physically challenging test as it had the highest threshold, followed by the flat and then the decline condition, proving to be the least physically challenging. The data was also treated with Kaplan-Meier survival curves to determine significance between those who were able and unable to finish the tests, as well as if the treadmill conditions were significantly different from each other. VO_2 and VCO_2 proved to be significant between groups of those who finished and did not finish for time to failure. VO_2 for the flat phase proved to be a significant measure of time to failure for the treadmill gradients. VE and RER were not significant. Therefore, the hypothesis that metabolic cost increases over time was not supported as steady state was observed. Also, the decline proved to be the least physically challenging test out of the incline, flat, and decline conditions.

CHAPTER 5: DISCUSSION

Military missions require personnel to walk for extended distances under heavy loads which exact a high metabolic cost. Shorter duration studies have assumed a steady state condition occurs for the metabolic cost at a given load or speed, meaning that service members' hourly energy expenditure will be the same throughout marching (Epstein et al., 1988). Few studies have examined energy cost during prolonged walking under load and those that have researched endurance load carriage have produced conflicting results. When carrying a 25 kg load, no linear increase in VO2 was observed but while carrying a 40 kg load, a significant linear increase was in fact observed (Epstein et al., 1988). In this study, it was first hypothesized that energy cost with a 30 kg rucksack would increase significantly between 60 and 120 minutes for each treadmill condition compared to the first 60 minutes of walking. Secondly, the incline was hypothesized to be the most challenging test as it was thought to result in the highest thresholds for VO_2, VCO_2, VE, and RER. The decline was hypothesized to have the next highest thresholds, followed by the flat condition.

Participant Demographics and Training History

The participants in the current study were not recruited based on various characteristics like height, weight, and body fat percentage as they were in the Patton et al., (1991) & Epstein et al., (1988) studies. Also, participants in the previous studies had relatively the same amount and level of physical activity training, allowing them to have similar aerobic capacity measures as well as the physical ability to carry out prolonged walking with load tests. Participants in this study varied in their training, physical activity, and deployment tours, as two of them had been deployed previously while some were in the middle of their first year of ROTC. The lightest participant weighed 61.05 kg and the heaviest participant weighed 89.13 kg, a difference of

28.08 kg. Participants also ranged in height from 170 to 183 cm. Body fat percentage ranged from 11.5%, considered healthy, to 23.8%, considered slightly overweight. Perhaps height, weight, and body fat percentage may have impacted why some participants could walk the full time and some fatigued faster. Also, since two of the participants had not carried weight over 27 kg, they were not trained to handle the level of weight that was asked of them in this study. Participants volunteered for this study and were accepted based on their desire to complete the study. The only exclusion criteria were if participants had a current or past musculoskeletal injury that would have prevented them for walking for two hours under load. Had stricter participant demographics been set in place, a more homogenous sample could have been recruited and the results may have shown different patterns.

Hypothesis 1: Energy cost, as measured by VO_2, VCO_2, VE, RER, with a 30 kg rucksack, will increase significantly between 60 and 120 minutes for each treadmill condition compared to the first 60 minutes of walking

Contrary to our hypothesis, there was no increase in the metabolic variables during the second hour of the two hour sessions. These data are consistent with previous research that assume a steady state condition occurs between 30 to 60 minutes into a walking with load bout and continues throughout two hours of walking under load (Pal et al., 2009 & Quesada et al., 2000). In a marching study lasting 40 minutes, no significant increase in metabolic cost was found for VO_{2max}, VE, and VCO_2 under load, which is consistent with results of this study (Quesada et al., 2000). Beekley et al., (2007) had participants walk for 30 minutes with heavy loads of up to 70% of an individual's body weight and systematic increases in VO_2 and VE were not observed, consistent with results from this study.

Two studies that examined prolonged load carriage reported mixed results. One study did not use military participants but used moderately active men and a distance of 12 km instead of a time endpoint. Their results showed that VO_2 and VE increased significantly during prolonged walking upwards of 200 minutes while carrying loads of 31.5 and 49.4 kg, which differs from the results of this study (Patton et al., 1991). Figures 4-7 from this study show instances in which metabolic variables increase very slightly but not in a linear fashion and were insubstantial. Another load carriage study lasted two hours in duration and used six highly endurance trained non-military men walking under two trials, with 25 kg and 40 kg of weight. Consistent with this study, no significant changes occurred in VO_2 and VCO_2 while carrying the 25 kg load, a mass similar to that used in this study. However, under the same conditions with 40 kg of weight, significantly higher energy costs were noted and the energy cost per kg gradually increased over time in a linear fashion (Epstein et al., 1988). The 30 kg of weight used in the present study may not have been enough to elicit such a linear increase as was hypothesized. Perhaps a heavier load was needed to observe an increase in energy cost linearly over time. Participants who are used to carrying heavier loads (30 kg and higher) may be more representative of military trained personnel and would be able to handle the load more efficiently than those in this study did.

VO_2

VO_2 was found to be a significant measure for time to failure based on the Kaplan-Meier Curve of time to failure by VO_2 threshold group was significant between groups at $p=0.0098$, showing that there was a significant difference in VO_2 between those who completed and did not complete the two hour testing sessions. When analyzing the VO_2 by the incline, decline, and flat conditions, the flat stage was found to be significant only for with $p=0.0177$, showing that there was a significant difference between those who were able and unable to reach the two hour time point during the flat stage. Six participants were unable to complete the two hour walking time and three were able to reach the two hour endpoint for the incline. For those who did not finish, an upwards trajectory was observed at the time of failure. If these participants had been able to walk longer, perhaps a linear increase in VO_2 would have been observed. In a previous study lasting two hours in duration, ruck marching with 25 kg over a level surface did not show an increase in VO_2 or VCO2. Only when rucking with a heavier weight of 40 kg did a linear increase become apparent (Epstein et al., 1988). One participant surpassed the threshold for the incline but was still able to walk for two hours. The same participant had a slight upwards trajectory over time and the other two participants did not. Out of the three who completed the incline stage, they ranged in height, weight, age, and body fat percentage, as well as their level of physical activity and ROTC training.

Although this study did not measure VO_{2max}, those who were not able to finish the two hour test due to fatigue could be in part due to the fact that they may have been exercising at an intensity equivalent to more than 50% of their VO_{2max} during the incline. Previous research has shown that well-trained men are unable to walk under load for prolonged periods at or above 50% of their VO_{2max}, and even working at an intensity that exceeds 35-40% of VO_{2max} may elicit fatigue in some individuals (Patton et al., 1991). Those who did not finish the incline reported this was a result of fatigue. A few instances exist when participants could have finished the study, specifically for the flat and decline stages, but were unable to do so because they had to leave early or were not feeling well, which may have altered the results. Participants could have been better informed as to how long the study would take from start to finish so they knew when to schedule the test during a time that worked best for them. This suggests thresholds could have been adjusted to differentiate between those who finished and did not finish the two hour test during the decline. However, no increases in VO_2 were observed during the flat condition. For the decline condition, one participant who did not finish the test appeared to have an upward trajectory for VO_2. For the other participants, whether they did finish or did not finish the decline stage, were just at or below threshold.

Since VO_2 thresholds for this study were set at 20, 25, and 30 ml/kg/min for the decline, flat, and incline stages respectively, participants who started at or exceeded these thresholds in the first 60 minutes may have reached anywhere from 35-50% of their VO_{2max}, which could explain why they were not able to finish the two hour study. Also, when VO_2 was corrected for fat free mass examine if body composition could have contributed to VO_2, no difference was found. This means that those who did not finish and above threshold when VO_2 was measured in ml/kg/min were no different than when VO_2 was measured in kg/FFM/min, suggesting body composition did not appear to have an impact on VO_2. Due to the small sample size and lack of power, body composition may not have been significant. Studies with a larger number of participants may observe differences in VO_2 relative to body mass and fat free mass. Those who were not able to finish the two hour tests were inefficient in their walking which could have contributed to the early finishing time. Those who were more efficient while walking may have been able to walk for longer.

A short duration study, lasting 50 minutes, found mean VO_2 measures of 18.8 ± 1.7 ml/kg/min while walking at a slow pace of 2.4 miles per hour while wearing an unknown weight of body armor (Ricciardi et al., 2008). This is comparable to the decline VO_2 threshold of this study set at 20 ml/kg/min while walking at 3.0 miles per hour. Also, with the 31.5 kg load and a speed of 1.35 meters per second, comparable to the 30 kg and 1.3 meters per second speed chosen for this study, mean VO_2 measures ranged from 17.1 ± 0.5 ml/kg/min to 18.9 ± 0.6

ml/kg/min (Patton et al., 1991). This is close to the 20 ml/kg/min VO_2 threshold that was observed for the decline condition. Lastly, when 40 kg of load was carried for two hours in a previous study, mean VO_2 started out at 17 ml/kg/min twenty minutes into the test and increased linearly until reaching about 18.7 ml/kg/min after 120 minutes while walking on a level surface (Epstein et al., 1988). This is comparable to the decline threshold set at 20 ml/kg/min but is lower than the 25 ml/kg/min threshold the flat stage was set at. Participants in the previous study were six healthy, highly endurance trained males who were in better physical condition than the majority of participants in the current study. Therefore, a major finding of this study was that many East Carolina University ROTC members in this sample could not ruck march for two hours with 30 kg of load at various gradients even though military missions may require this of them.

VCO_2

VCO_2 proved to be significant for time to failure thresholds between those who were able and unable to walk for two hours at p=0.0084 for all conditions. Five participants who did not finish the incline had upward trajectories at the time of failure. One participant who did not finish the incline was not only under threshold but appeared no show no increase in VCO_2, indicating he may have been physically able to walk longer. Similar to VO_2, three of the participants were able to walk the full time and only one out of those three had an upward trajectory over time. For the flat condition, two participants were under threshold but finished early. In this instance, had they allowed more time on the flat testing day, they may have been more likely to finish. Other participants remained at or below threshold and were able to finish the study. Only one participant was above threshold consistently during the flat stage, but was stable over time and did not finish the flat condition. During the decline, two participants were at or above threshold and did not finish this condition. In general, the majority of participants were under threshold and VCO_2 did not increase in a linear fashion. No overall increase in VCO_2 was observed as the majority of participants started out at the initial VCO_2 measure. Those who had higher initial VCO_2 means were more likely to fail the event than those who had lower VCO_2 means.

Perhaps 30 kg used in this study was not enough to elicit the linear increase observed in previous research (Epstein et al., 1988 & Patton et al., 1991). Although participants in this study could not efficiently ruck with loads heavier than 30 kg, another sample of participants who are used to carrying heavier loads on a regular basis may produce different results. However, none of the literature reviewed in this document has reported results on VCO2. Only one study examined the ratio of VCO2 to VO2 as a measure of obtaining RER (Quesada et al., 2000).

VE and RER

Due to the small number of participants who were able to walk for the full two hours during each treadmill condition, limited data and power could explain why VE and RER were not significant between those who made it to the two hour endpoint and those who did not. VE and RER were also not significant when examined by the incline, decline, and flat conditions. Quesada et al., (2000) reported no significance for RER when participants walked with load for 40 minutes finding that RER ranged from 0.80 to 0.90 without a linear pattern. This is similar to the current study where RER oscillated between the decline threshold of 0.80 and incline threshold of 0.86. Conversely, when walking at speeds of six km/hour, Beekley et al., (2007) found RER ranged from 0.85 to 0.95 with varying loads based on percentages of the individuals' lean body mass. This numbers are higher than was observed in the current study because participants were walking faster. When RER was examined, there was a high degree of variability among participants reaching and not reaching threshold. This variable is heavily influenced by meal composition prior to testing. Participants were instructed to eat the same meal an hour before each testing session, but this was not monitored. Even those who were able to finish the test had threshold spikes in the first 60 minutes for RER. For VE, participants who were able to walk for the full time also reached threshold or stayed consistently above threshold during the first 60 minutes or the entire test. In this instance, perhaps the thresholds could be adjusted to account for this discrepancy. Quesada et al., (2000) examined VE under various loads during 40 minutes of walking. At the heaviest load, 30% of participants' body weight, VE reached steady state after ten minutes around 50 L/min. The incline threshold was set to 55 L/min in the current study, comparable to the steady state VE found previously.

Thresholds were created in an attempt to determine when a participant would be more likely to fatigue. However, the thresholds established are unique to this sample and could be clarified further in future research. In general, those who were able to walk for two hours showed steady state among the majority of the metabolic parameters. Those who had elevated metabolic parameters did not finish the two hour sessions. Also, since seven participants were not able to walk for two hours during each condition, this study may have been too physically exhausting for the recruited sample. One study found similar results in that five out of fifteen participants could not complete the load carriage trials using 49.4 kg. In this study, participants were asked to walk upwards of 175-200 minutes on a level surface and results showed that VO_2 and VE are not constant but increase significantly during prolonged walking with external loads (Patton et al., 1991). This was a heavier load than was used in the current study. Also, another study had 34 participants walk with and without load walking at a slow pace, 2.4 miles per hour, and a moderate pace, 3.8 miles per hour (Ricciardi et al., 2008). Six participants were unable to walk at the moderate pace without load for 50 minutes and fourteen participants were unable to walk at the moderate place with load (Ricciardi et al., 2008). A different study examined varying loads based on the percentage of the individual's lean body mass and found that four participants out of ten quit of their own volition (Beekley et al., 2007). When the dropout was examined further, two participants were significantly shorter than the others and two had significantly higher percentages of body fat than the others (Beekley et al., 2007). This is an interesting finding that could be applied to the current study give more time to analyze participant demographics. Lastly this is the first time, to our knowledge, of survival curves and cutoff thresholds being used to differentiate between participants who were able and unable to reach the two hour duration of the different gradient conditions.

Despite differences in participant demographics; including height, weight, body fat percentage, physical activity level, and ROTC training, the results from this study show that VO_2, VCO_2, VE, and RER were relatively stable over time. Those who did not finish started out at or close to threshold, and those who did finish stayed under threshold for duration of test. There was no large linear increase over time in any of the metabolic measurements as hypothesized. Based on the two participants who walked for two hours on each testing day, the mean values collected during the first 60 minutes of walking may be predictive of the last 60 minutes of the walking time period although not certainty. It was hypothesized that VO_2, VCO_2, VE, and RER values would increase signifcantly after the first hour, but results indicated that this was not the case. For the most part, the variables were reasonably stable throughout the study. Those particpants with higher metabolic measuremnts, at or above threshold, were more likely to end early due to fatigue than those who remained under threshold.

No significant increase in metabolic parameters was observed for a variety of reasons. Many studies have examined energy cost and load carriage in relation to one's body mass, while few have examined energy cost of load carriage over time with a fixed load independent of one's body weight. Military load carriage practices are generally not based on soldier weight. This may account for why some participants were not able to ruck with the load. In a few instances, according to Figures 4-7, some participants who ended the test early did so because of fatigue and their metabolic parameters end with an upward trajectory. Perhaps if they had been able to walk longer a linear increase would have been observed. Also, some participants had more military training and were in better physical shape than others as shown in Table 1. Generally, those who were in better physical condition did not reach threshold and were more likely to finish the two hour tests. Since no linear increase was observed, the mean metabolic parameters during the first 60 minutes predicted the trajectories during the last 60 minutes, although not perfectly. To be consistent with previous findings, more power was needed to determine significance of a linear increase over time (Patton et al., 1991 & Epstein et al., 1988). Only two participants completed the two hour tests under each treadmill condition. If more had been physically able to complete each test, perhaps different trajectories would have been observed. Also, since participants walked in an environmental chamber with the temperature controlled at 40 degrees Celsius and humidity controlled to 50%, this may have eliminated an increase in metabolic cost over time. To our knowledge this is the first time an environmental chamber of this nature was used to examine the energy cost of military personnel walking for upwards of two hours under load.

Hypothesis 2: The incline will be the most physically challenging as the thresholds for each metabolic value are hypothesized to be the highest during this stage followed by the decline stage, followed by the flat stage.

As hypothesized, the incline was the most physically challenging test, producing the highest thresholds for VO_2, VCO_2, VE, and RER. However, the flat stage was the second most challenging and the decline the least challening. To our knowledge, this is the first study that examined these four metabolic variables during a prolonged walking under load bout using a load in military trained participants. Furthermore, reasearch has not examined threshold cutoffs of military personnel data similar to that collected in this study, nor has previous research compared incline, flat, and decline conditions to determine differences in metabolic cost. Incline and flat conditions have been examined, but not in relation to each other (Lyons et al., 2005 & Patton et al., 1991 & Epstein et al., 1988). Additionally, this is the first study that examined how a decline condition may differ from incline and flat conditions.

VO_2

According to results of this study, the incline had the highest threshold set at 30 ml/kg/min followed by the flat stage at 25 ml/kg/min followed by the decline condition at 20 ml/kg/min. The incline was the most physically challenging test as having the highest threshold as well as the highest mean VO_2 measures. This is consistent with results of another study that showed VO_2 increases as the gradient increases 0 to 9% (Lyons et al., 2005). Significant demands are placed on the cardiovascular system during inclined walking. As a result, metabolic demands also increase with inclined walking. An incline of 5% was used based off of research examining inclines from 3-9% (Lyons et al., 2005). A 5% incline was thought to be moderately high enough to show a metabolic difference between the flat and decline conditions yet a manageable incline for 2 hours of walking. For this sample of participants, it may have been too high and placed too much stress on their cardiovascular systems, inducing early fatigue. During the incline condition, load carriage may reduce work performance and increase fatigue. Since scheduling participants proved to be more difficult than expected, two out of the five participants who did not complete the flat stage reported they could have gone longer but had to leave early for class. Having more time to complete the study would have given participants the opportunity to reschedule during a time they could devote to the two hour walking duration. Since participants who were able to walk for the two hour duration at the flat stage were at or even above threshold, this suggests thresholds could have been adjusted to differentiate between those who finished and did not finish the two hour test. Lastly, the decline was the easiest condition resulting in the lowest threshold for VO_2.

VCO_2

As was observed for VO_2, the incline proved to the most physically challenging test, producing the highest values for VCO_2 set 27 ml/kg/min, followed by the flat condition set at 19 ml/kg/min then decline condition at 16 ml/kg/min. The decline stage appeared to be the least challenging test with thresholds set at the lowest level for VCO_2. The flat stage was hypothesized to be the least challening test, but results proved otherwise. Seven participants were able to walk for the full two hours at the decline stage whereas five participants walked for the full time at the flat stage. Downhill walking was hypothesized to be more difficult than level walking because of the muscle fatigue that comes with the eccentric musuclar contractions associated with walking downhill (Minetti et al., 2002). The decline could have been the least fatiguing condition because a greater recovery of elastic energy could be responsible for the increased economy (Minetti et al., 2002). To our knowledge, this is the only study that examined the energy cost of declined walking with a fixed load, lasting two hours in duration, using military trained participants.

VE and RER

Both VE and RER followed the pattern of VO_2 and VCO_2, as the incline had the highest thresholds, 55 L/min for VE and 0.86 for RER. VE thresholds were set at 36 L/min for the flat condition and 29 L/min for the decline condition. For VE, previous data observed that steady state began after approximately 10 minutes of walking under load (Quesada et al., 2000). With the exception of the incline, the majority of participants stayed under or just around threshold during the decline and flat conditions for VE. For the incline, more particiaptns were likely to fail the event as their VE means were generally above threshold. For RER, the flat condition had a threshold of 0.84 followed by 0.80 for the decline condition. One study examining RER determined that energy expenditure was measured at steady state with a RER less than or equal to 1, which supposes that the walking under load could be maintained over time (Bastien et al., 2005). However, it is possible that the energy cost may increase due to muscular fatigue (Epstein et al., 1988). RER has not been examined as thoroughly as VO_2, potentially due to the variability that was observed in this study. The thresholds for RER were relatively close together and therefore difficult to determine due to the various upward and downward trajectories of the data.

Limitations

This study used ROTC participants because they were thought to be more physically active and are trained to walk with loads. However, participants varied in their ROTC training, physical activity level, and military background. Data on heart rate was collected but due to timing it was not able to be analyzed. These data could have aided to our understanding of how physically exhausted participants were and at what percentage of the maximum heart rate they were at when they chose to end the test or when they reached the two hour endpoint.

This study also limitations due to outside factors that could not be controlled. First, since many subjects were unable to complete the protocol due to fatigue, the data analysis was complicated by failures. There was not enough power to determine significance between the two hour tests when only two participants completed the test. Also, some participants left early for reasons other than fatigue. Specifically, for the decline and flat testing sessions, it was not explained enough as to how long the entire tests take from start to finish. Participants were asked to walk on the treadmill for two hours but the entire time they came in for testing took anywhere from two and a half hours to three hours if they walked for the entire time. Therefore, some participants had to leave early for prior obligations. Therefore, some participants specifically for the decline and flat conditions, were unable to finish the two hour tests due to time constraints.

Despite ROTC training, some participants had never rucked with weight greater than 27 kg, and this study required them to carry 30 kg. Also, participants of any height and weight were permitted to participate in the study but were expected to carry the same weight. Two participants in particular had a difficult time on all three testing days with weight. The first one weighed the least out of all of the participants. He struggled each day of testing and never made it past one hour. The other participant was overweight and reported that his physical training was not as great as it has been in the past. It is also interesting to note that two participants have been deployed. One of them went the full time for every test, and the other one did not go the full time on two of his testing days. Testing days were aimed to be separated by one week to avoid delayed onset muscle soreness, but participants were unable to follow this regimen due to scheduling conflicts. The shortest time between tests was three days and the longest was almost two months.

Participants were instructed to choose from a standardized pre-testing meal and to consume the same meal one hour prior to arriving for each day of testing. Most likely this was not adhered to because it was not monitored. When asked before each testing session if participants had eaten the same meal an hour before coming in, some had complied and some had not. Since meals were not monitored, variable intake could partially explain why RER did not show significance over time and between testing conditions. Participants were also instructed to avoid exercise, especially lower body exercise, before each testing session under load. At least two participants came to their testing sessions having performed a lower body resistance training workout the day before, which they contributed to their failure to walk for the full two hours.

Possible improvements could be made to the study to increase the power and significance of data. The sample size was also small, with eleven who attempted the study and nine who completed each testing day. A larger sample size would be ideal. While the environmental chamber controlled for temperature and humidity, excessive noise distracted some subjects. Also, performing a VO_{2max} test would have been more appropriate than having participants complete one twenty minute session at each gradient without load. Having the participant's VO_{2max} would have allowed for analysis to examine what percentage of their maximal oxygen consumption they were working at during the load carriage days and it would be easier to determine those who failed to walk for two hours due to fatigue versus outside reasons. A lactate threshold test, using a finger prick to draw blood, could have been an even better measure of fatigue.

Future Research

This is one of the few studies that examined energy expenditure in military participants lasting two hours in duration while carrying a heavy load. It was hypothesized that the metabolic variables would increase over the two hour time period, especially after 60 minutes, which would be consistent with the findings of Epstein et al., (1988) and Patton et al., (1991). However, this was not observed and a steady state was observed, which is more consistent with findings from Quesada et al., (2000) that lasted for a shorter duration. Due to the varied characteristics of the participants in this particular study, future studies could look at a sample of participants who are more similar, with around the same body fat percentage, height, weight, and physical training level. Studies could also be done with better trained military participants, who have more strenuous physical training than the ROTC cadets at East Carolina University. Since many of the participants in this study were not used to rucking for upwards of two hours, those in the Army, Navy, or Air Force, who are used to long duration ruck marches, may be a more appropriate sample. Those who have more military training may be better able to safely and efficiently ruck with loads of this weight, but the results from using trained personnel still may not result in an observable increase in metabolic parameters. The combinations of load, speed, and gradient in this study may not be taxing enough to see increases in metabolic cost and therefore the duration of walking may need to be extended to see if increases are observed even after two hours. Also, since this was a small sample, future research could look at a larger number of individuals. The participants in this study were all male so since females were permitted to fight at the front of enemy lines over one year ago, future work could analyze if there are any differences for males and females. Also, this study only focused on a flat, -4% decline, and 5% incline. Future research could examine the energy cost of load carriage at different inclines and declines to determine if a truly steady state or linear increase exists between metabolic cost and prolonged walking. Also, in order to further examine if participants were truly fatigued, a lactate threshold test could have been performed to determine the exercise intensity at which lactic acid starts to accumulate in the blood stream. At higher exercise intensities, such as that of this study, a simple finger prick of blood could have determined when the lactate level in the blood reached anaerobic threshold, or the onset of blood lactate accumulation, suggesting fatigue was induced. Lastly, since the test was stopped after two hours of testing, future research could extend that time point to determine how long participants can walk for until they reach fatigue, whether it is in fact two hours, three hours, four hours, or longer. If the time component is removed, perhaps a more accurate time to event would have been observed. Data collected in this nature may be more representative of military ruck marching in the field and could help commanders set even better combinations based on load and speed given the duration of a ruck march to battle destinations.

Summary

Overall, the hypothesis that a linear increase would be over time and especially during the second hour of testing was not supported as a steady state condition was observed. Also, the incline did in fact prove to be the most challenging of the treadmill conditions, followed by the flat, then the decline condition. It was hypothesized that the 30 kg load in this study would be heavy enough to elicit an increase in VO_2, VCO_2, VE, and RER, which was consistent with results from a study lasting the same amount of time under a 40 kg load. Our results showed that the 30 kg load prompted a steady state like response in that no linear increase was seen over time and especially during the second hour of the test. Furthermore, the incline was hypothesized to be the most physically challenging test, whereas the decline appeared to be the least physically challenging, although the flat stage was hypothesized to be least physically exhausting test. The role of load carriage on energy expenditure is a topic of study especially important to the military so guidelines can be set as to the appropriate load and speed given a certain terrain or gradient. When soldiers perform strenuous ruck marches with a heavy load, it is essential that they are able to efficiently execute military tasks so as to avoid injury and fatigue. Military effectiveness may be compromised if soldiers are asked to carry a load and walk at a speed over various terrains that may induce fatigue. Estimating the metabolic cost of heavy load carriage during prolonged walking is essential to ensure military tasks are carried out as successfully as possible. Our results suggest that this sample of ROTC participants fatigued earlier than hypothesized and were unprepared to carry weight for prolonged periods of time lasting upwards of two hours. Significant results indicate that VO2 and VCO2 may be predictive of failure and are important metabolic measures to examine in military personnel. Metabolic cost over time is critical for military commanders to create optimal conditions of load and speed given terrain, gradient, and surface in order to ensure military combat effectiveness is not compromised due to fatigue.

REFERENCES

American College of Sports Medicine: Guidelines for Exercise Testing and Prescription (9th ed). (2014). Baltimore, MD: Wolters Kluwer & Lippincott Williams & Wilkins (pg. 73)

Bastien, G., Willems, P., Schepens, B., & Heglund, N. (2005). Effect of load and speed on the energetic cost of human walking. *European Journal of Applied Physiology, 94*, 76-83.

Beekley, M., Alt, J., Buckley, C., Duffey, M., & Crowder, T. (2007). Effects of heavy load carriage during constant-speed, simulated, road marching. *Military Medicine, 172*(6), 592-595.

Bilzon, J., Allsopp, A., & Tipton, M. (2001). Assessment of physical fitness for occupations encompassing load-carriage tasks. *Occupational Medicine, 51*(5), 357-361.

Epstein, Y., Rosenblum, J., Burstein, R., & Sawka,, M. (1988). External load can alter the energy cost of prolonged exercise. *European Journal of Applied Physiology, 57*, 243-247.

Grenier, J., Peyrot, N., Castells, J., Oullion, R., Messonnier, L., & Morin, J. (2011). Energy cost and mechanical work of walking during load carriage in soldiers. *Medicine & Science in Sports & Exercise, 12*, 1131-1140.

Kleinbaum, D. (1996). *Survival analysis: A self-learning text*. New York: Springer.

Knapik, J., Reynolds, K., & Harman, E. (2004). Soldier load carriage: historical, physiological, biomechanical, and medical aspects. *Military Medicine, 169*(1), 45-56.

Lyons, J., Allsopp, A., & Bilzon, J. (2005). Influences of body composition upon the relative metabolic and cardiovascular demands of load-carriage. *Occupational Medicine, 55*, 380-384.

Minetti, A., Ardigo, L., & Saibene, F. (1994). Mechanical determinants of the minimum energy cost of gradient running in humans. *Journal of Experimental Biology, 195*, 211-225.

Minetti, A., Moia, C, Roi, C., Susta, D., & Ferretti, G. (2002). Energy cost of walking and running at extreme uphill and downhill slopes. *Journal of Applied Physiology, 93*, 1039-1046.

Pal, M., Majumdar, D., Bhattacharyya, M., Kumar, R., & Majumdar, D. (2009). Optimum load carriage by soldiers at two walking speeds on level ground. *International Journal of Industrial Ergonomics, 39*, 68-72.

Patton, J., Kaszuba, J., Mello, R., & Reynolds. (1991). Physiological responses to prolonged treadmill walking with external loads. *European Journal of Applied Physiology, 63*, 89-93.

Quesada, P., Mengelkoch, L., Hale, R., Simon, S. (2000). Biomechanical and metabolic effects of varying backpack loading on simulated marching. *Ergonomics*, *43*(3), 293-309.

Ricciardi, R., Deuster, P., & Talbot, L. (2008). Metabolic demands of body armor on physical performance in simulated conditions. *Military Medicine*, *173*, 817-824.

Simpson, K., Munro, B., & Steele, J. (2012). Effects of prolonged load carriage on ground reaction forces, lower limb kinematics and spatio-temporal parameters in female recreational hikers. *Ergonomics*, *55(3)*, 316-326.

Van Dijk, J. (2009). Common military task: marching. *Human Performance Enhancement for NATO Military Operations (Science, Technology and Ethics), 3*, 1-46.

Wong, H., Frame, J., Ozimek, E., Leib, D., & Dugan, E. (2012) Influence of fatigue and load carriage on mechanical loading during walking. *Military Medicine*, *177(2)*, 152-156.

APPENDIX A: IRB APPROVAL

EAST CAROLINA UNIVERSITY
University & Medical Center Institutional Review Board Office
4N-70 Brody Medical Sciences Building· Mail Stop 682
600 Moye Boulevard · Greenville, NC 27834
Office **252-744-2914** · Fax **252-744-2284** · **www.ecu.edu/irb**

Notification of Amendment Approval

From: Social/Behavioral IRB
To: Sara Krajewski
CC:
Laurel Wentz
Date: 9/20/2013
Re:
Ame1 UMCIRB 13-000113
UMCIRB 13-000113
Energy Cost of Load Carriage on Marching in ROTC Subjects

Your Amendment has been reviewed and approved at the convened meeting on 9/18/2013 of the Social/Behavioral IRB.

Approval of the amended study and any consent form(s) is for the period of 9/18/2013 to 4/16/2014. The Social/Behavioral IRB determined that this revision does not change the overall risk/benefit ratio of the study.

Please note that any further changes to this approved research may not be initiated without UMCIRB review except when necessary to eliminate an apparent immediate hazard to the participant. All unanticipated problems involving risks to participants and others must be promptly reported to the UMCIRB. A continuing or final review must be submitted to the UMCIRB prior to the date of study expiration. The investigator must adhere to all reporting requirements for this study.

Approved consent documents with the IRB approval date stamped on the document should be used to consent participants (consent documents with the IRB approval date stamp are found under the Documents tab in the study workspace).

The approval includes the following items:

Informed Consent No More Than Minimal Risk.doc(0.01)	Consent Forms
Thesis proposal.docx(0.01)	Study Protocol or Grant Application

The following UMCIRB members were recused for reasons of potential for Conflict of Interest on this research study:

None

The following UMCIRB members with a potential Conflict of Interest did not attend this IRB meeting:

None

APPENDIX B: INFORMED CONSENT

East Carolina University

Informed Consent to Participate in Research

Information to consider before taking part in research that has no more than minimal risk.

Title of Research Study: Energy Cost of Load Carriage on Marching in ROTC Subjects
Principal Investigator: Sara M. Krajewski
Institution/Department or Division: Kinesiology
Address: 103 FITT Building, Greenville, NC 27858
Telephone #: (216) 387-0371

Researchers at East Carolina University (ECU) study problems in society, health problems, environmental problems, behavior problems and the human condition. Our goal is to try to find ways to improve the lives of you and others. To do this, we need the help of volunteers who are willing to take part in research.

Why is this research being done?

The purpose of this research is to determine the energy cost of walking with a heavy 30 kg load for long periods of time in ROTC military trained undergraduate male and female students. This research is significant because the results can help officials create recommendations for the amount of load soldiers are expected to carry for up to two hours with minimal risk of injury or fatigue at different gradients. The decision to take part in this research is yours to make. By doing this research, we hope to learn how energy costs change over long periods of time (up to two hours) while carrying a 30 kg rucksack across different treadmill grades during flat, incline, and decline phases. Your participation is voluntary.

Why am I being invited to take part in this research?

You are being invited to take part in this research because of your interest in response to the flyers posted around East Carolina University's campus. The amount of time it will take you to complete this study is four days. The first day you will be walking for one hour. One week later you will be coming in for the second day of testing and walking for two hours. One week later you will be coming in for the third and last day of testing which will last two hours. One week later you will come in for the final day of testing, lasting two hours. There is total of four days of testing for a total of seven hours and you will be compensated for your time. If you volunteer to take part in this research, you will be one of about 15 people to do so.

Are there reasons I should not take part in this research?

I understand I should not volunteer for this study if I am under 18 years of age or have any current or past bone or muscle injuries that would prevent me from walking for up to two hours per exercise session.

What other choices do I have if I do not take part in this research?

You can choose not to participate.

Where is the research going to take place and how long will it last?

The research procedures will be conducted at 245 Ward Sports Medicine Building, Greenville, NC 27858. You will need to come to the Human Performance Lab located on the third floor of Ward, in the Exercise Physiology department, for each day of testing during the study. The total amount of time you will be asked to volunteer for this study is 7 hours total over a total of 4 separate visits over the next 4 weeks.

What will I be asked to do?

You are being asked to do the following: You are being asked to first fill out a survey asking you a variety of questions based on your ROTC exercise training as well as some brief questions about your general health.

- After completing the survey, basic height, nude weight, and a dual x-ray absorptiometry scan will be completed. The DEXA scan will cause you no harm but you will be exposed to some radiation, although minimal, and comparable to a round-trip airplane flight from Chicago to Paris. A urine sample will be collected before you begin testing and after you complete testing on each day to check your hydration status.
- The first day of testing will require you to walk for one hour on the treadmill. There will be three stages lasting fifteen minutes each. You will walk on a flat surface, at a 0% incline, 5% incline, and decline phase. You will be wearing a mask during the last five minutes of each 15 minutes that fits inside your mouth that will measure gases you inhale and exhale
- On the second day of testing, you will walk on the same treadmill for a total of two hours at both at the 0% incline, 5% incline or at a decline phase.
- The third and last day of testing will require you to walk again for a total of two hours at the 0% incline, 5% incline, or decline phase.
- The last day of testing will be the time when you complete the treadmill gradient that you have left.
- You will be wearing the mask to measure gases for all four testing days. You will be given a 5 minute rest and water break after 40 and 80 minutes of walking, or at any time you request one.

What possible harms or discomforts might I experience if I take part in the research?
There are possible risks (the chance of harm) when taking part in this research. These risks include muscle soreness resulting from walking at the various inclines or declines. However, each testing day will be separated by one week so you will have time to recover from any unforeseen soreness. It will also be made sure that the testing days will not coincide with any ROTC training you will need to perform. Your body composition will be measured using dual energy x-ray absorptiometry (DEXA) technology, a state-of-the-art method that uses very low intensity x-rays. The x-ray exposure from DEXA is minimal and comparable to a round-trip airplane flight from Chicago to Paris. There is no pain associated with this test and it should not last longer than 10 minutes.

What are the possible benefits I may experience from taking part in this research?
Other people who have participated in this type of research have experienced learning about your own personal exercise test results including your heart rate during exercise, calories burned, oxygen consumption, bone density and body fat percentage. By participating in this research study, you may also experience these benefits.

Will I be paid for taking part in this research?
We will pay you for the time you volunteer while being in this study. You will receive payment at the end of testing. You will receive $20 for each day of testing and an extra $20 if you complete the study. If you complete the study, you will be paid $100. Parking will be taken care of for you.

What will it cost me to take part in this research?
It will not cost you any money to be part of the research

Who will know that I took part in this research and learn personal information about me?
To do this research, ECU and the people and organizations listed below may know that you took part in this research and may see information about you that is normally kept private. With your permission, these people may use your private information to do this research:

- The sponsor of this study, Dr. Laurel Wentz.

- Any agency of the federal, state, or local government that regulates human research. This includes the Department of Health and Human Services (DHHS), the North Carolina Department of Health, and the Office for Human Research Protections.
- The University & Medical Center Institutional Review Board (UMCIRB) and its staff, who have responsibility for overseeing your welfare during this research, and other ECU staff who oversee this research.

How will you keep the information you collect about me secure? How long will you keep it?
The research data will be stored on a secured web-based drive until graduation in May of 2014. The data will also be kept on a metabolic cart in the FITT building until graduation of May 2014. The data will be coded and secure so no one besides the PI will have access to the data. The data will not be released unless the subject would like to know their RER, VO2, heart rate, blood pressure, and VCO2. If the subject does want to know what their values are, they will ask the PI and the PI will write down the values and put it in an envelope and personally given to the participant.

What if I decide I do not want to continue in this research?
If you decide you no longer want to be in this research after it has already started, you may stop at any time. You will not be penalized or criticized for stopping. You will not lose any benefits that you should normally receive.

Who should I contact if I have questions?
The people conducting this study will be available to answer any questions concerning this research, now or in the future. You may contact the Principal Investigator at (216) 387-0371, Monday through Friday between the hours of 8:00am until 7:00pm.

If you have questions about your rights as someone taking part in research, you may call the Office for Human Research Integrity (OHRI) at phone number 252-744-2914 (days, 8:00 am-5:00 pm). If you would like to report a complaint or concern about this research study, you may call the Director of the OHRI, at 252-744-1971

I have decided I want to take part in this research. What should I do now?
The person obtaining informed consent will ask you to read the following and if you agree, you should sign this form:

- I have read (or had read to me) all of the above information.
- I have had an opportunity to ask questions about things in this research I did not understand and have received satisfactory answers.
- I know that I can stop taking part in this study at any time.
- By signing this informed consent form, I am not giving up any of my rights.
- I have been given a copy of this consent document, and it is mine to keep.

____

Participant's Name (PRINT) **Signature** **Date**

Person Obtaining Informed Consent: I have conducted the initial informed consent process. I have orally reviewed the contents of the consent document with the person who has signed above, and answered all of the person's questions about the research.

____

Person Obtaining Consent (PRINT) **Signature** **Date**

APPENDIX C: DXA QUESTIONNAIRE

DXA Risk Statement and Subject Questionnaire
Human Performance Laboratory
Body Composition / Bone Mineral Density Analysis

During this visit, my body composition will be measured using dual energy x-ray absorptiometry (DXA) technology, a state-of-the-art method that uses very low intensity x-rays. The x-ray exposure from DXA is minimal and comparable to a round-trip airplane flight from Chicago to Paris. I will need to remove metal clothing accessories, jewelry and my shoes as these can affect the scan results; however, I will otherwise remain fully clothed. The DXA will scan my entire body very slowly; so, I will need to lie on a table without moving for almost 5 minutes, while the DXA is passed over my entire body. I will feel no discomfort associated with this test. There are no restrictions to my normal activity following this procedure. This test will determine the amount of fat, muscle and bone that I have. I have been informed that females with ANY chance of being pregnant should NOT undergo DXA scanning.

If I have further questions about the risks of DXA I may contact David Rushing, Clinical Radiation Safety Officer, Brody School of Medicine at East Carolina University, 252-744-2236.

Please complete the following questions to the best of your ability. If you have any questions, please ask the DXA technician for assistance.

Have you had any X-ray procedures within the last 3 days which use:	Iodine	Yes ☐	No ☐
	Barium	Yes ☐	No ☐
	Nuclear Medicine Isotopes	Yes ☐	No ☐
Do you have any of the following Medical devices in your body:	Ostomy devices	Yes ☐	No ☐
	Prosthetic Devices	Yes ☐	No ☐
	Surgical Devices	Yes ☐	No ☐
	Pacemaker leads	Yes ☐	No ☐
	Radioactive seeds	Yes ☐	No ☐
	Radiopaque catheters or tubes	Yes ☐	No ☐
Are you wearing any of the following Jewelry or clothing	Metal buttons	Yes ☐	No ☐
	Zippers	Yes ☐	No ☐
	Snaps	Yes ☐	No ☐
	Jewelry	Yes ☐	No ☐
Do you have any of the following Foreign Objects in your body:	Shrapnel, Buckshot	Yes ☐	No ☐
	Metal of any sort	Yes ☐	No ☐
	Other: Specify	Yes ☐	No ☐

Females of child bearing age only…. *(Bolded answers contraindicate DEXA)*		
Are you currently	Pregnant or Lactating	**Yes** ☐ No ☐
	Using birth control	Yes ☐ No ☐
Circle type of birth control	Condoms or diaphragm; birth control pill; depo-provera; IUD; partner has vasectomy	
When did your last normal period start?	Date: _____/_____/__________	
Have you has unprotected intercourse since your last period?	Note: if you are using some form of birth control, this means that you did not use birth control or you experienced a problem with your birth control method?	**Yes** ☐ No ☐

I certify that the information given above is true and correct to the best of my knowledge and that I have read all of the above information, asked questions and have received satisfactory answers in areas I did not understand. (A copy of this signed and dated consent form will be given to the person signing this form as the subject or as the subject's authorized representative.)

_______________________________ _______________________________
Subject/Authorized Representative (Sign) Date

____
Subject/Authorized Representative (Print)

Witnessed:

_______________________________ _______________________________
Technician, Human Performance Laboratory Date

I hereby certify that I have reviewed the pregnancy information given by this subject and the indication was that:

☐ -there was a very low risk of pregnancy. DXA was performed.

☐ -there was an unacceptably high risk of pregnancy. DXA was not performed pending further tests.

_______________________________ _______________________________
Technician, Human Performance Laboratory Date

____
If this is for a research study, please print the name of the Primary Investigator above.

Revision 1.30
Submitted for Approval: 09/06/2011 Approved: 09/08/2011, David Collier, MD

APPENDIX D: ROTC SURVEY

ID____________________

Load Carriage

Date of Birth ____________________________

Please check your class:

___ Freshman
___ Sophomore
___ Junior
___ Senior

Please list your position within ROTC:

____

How many years have you been in ROTC?

___ less than 1
___ 1
___ 2
___ 3
___ 4
___ more than 4

How many **days** per week do you run?

___ 0-1
___ 2-3
___ 4-5
___ 6-7

How many miles do you run each **week**?

___ 0-4
___ 5-10
___ 11-15
___ 16-20
___ more than 20

How many **days** do you resistance train each week?

___ 0-1
___ 2-3
___ 4-5
___ 6-7

What is the length of your typical resistance training workout?

___ Not applicable
___ 0-30 minutes
___ 30 minutes—1 hour
___ 1 hour –1.5 hours
___ 1.5 hours—2 hours
___ more than 2 hours

In total, how many **hours per week** do you spend doing physical activity (*for ROTC and on your own*)?

___ 0-5
___ 6-10
___ 11-15
___ more than 15

ID________________

Load Carriage

What is the **average** weight of your ruck-sack?

___ 0-20lbs
___ 20-40lbs
___ 40-60lbs
___ 60-80lbs
___ more than 80lbs

What is the **heaviest** weight you have ever carried in your rucksack?

___ 0-20lbs
___ 20-40lbs
___ 40-60lbs
___ 60-80lbs
___ 80-100lbs
___ more than 100lbs

When (Month/Year)?

____

Do you have any **current** muscle injuries?

___ No
___ Yes

If yes, please list location:

____

Do you have any **previous** muscle inju-ries?

___ No
___ Yes

If yes, please list location and time of each injury:

What is your fastest 2-mile run time in the past 6 months?

___ 10-11 minutes
___ 12-13 minutes
___ 14-15 minutes
___ 16-17 minutes
___ 18-19 minutes
___ 19-20 minutes
___ more than 20 minutes

APPENDIX E: DAY 1 PROTOCOL

Ruck March Study

Subject ID:

Date:

Meal choice:

Height:

Body Weight:

Gear Weight:

Pre USG:

Post USG:

DXA:

Incline

HR	Time	RPE*
	5	-
	10	-
	15*	
	16	-
	17	-
	18	-
	19	-
	20	-

Flat

HR	Time	RPE*
	5	-
	10	-
	15*	
	16	-
	17	-
	18	-
	19	-
	20	-

Decline

HR	Time	RPE*
	5	-
	10	-
	15*	
	16	-
	17	-
	18	-
	19	-
	20	-

Incline Pre Grip Strength:

Incline Post Grip Strength:

Flat Pre Grip Strength:

Flat Post Grip Strength:

Decline Pre Grip Strength:

Decline Post Grip Strength:

APPENDIX F: DAY 2-4 PROTOCOL

Ruck March Study

Day 2, 3, 4

Subject ID:
Body Weight:
USG Pre:
USG Post:
Hand grip Pre:
Hand grip Post:
Steps at minute 10:
Steps at minute 60:
Steps at minute 110:

Time	HR	RPE*
5		
10		
15*		
16		
17		
18		
19		
20		
25		
30*		
35		
36		
37		
38		
39		
40		
45*		
50		
55		
56		
57		
58		
59		
60*		

Time	HR	RPE*
65		
70		
75*		
76		
77		
78		
79		
80		
85		
90*		
95		
96		
97		
98		
99		
100		
105*		
110		
115		
116		
117		
118		
119		
120*		