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The Effects of Exercise-Induced Muscle Damage on Aerobic Capacity Tested Through Repeated VO2max Tests

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THE EFFECTS OF EXERCISE-INDUCED MUSCLE DAMAGE ON AEROBIC CAPACITY
TESTED THROUGH REPEATED VO$_2$MAX TESTS

by
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ABSTRACT

LUKE BURNETT: The Effects of Exercise-Induced Muscle Damage on Aerobic
Capacity Tested Through Repeated VO_2max Tests

High-intensity resistance training alongside aerobic training has been seen to
improve aerobic performance due to the gains strength that accompany said
resistance training. However, it has been documented that the exercise-induced
muscle damage (EIMD) that results from the resistance training can impair aerobic
performance in the days following. Therefore, the purpose of this study was to
assess the time-course of recovery from EIMD for up to seven days following
muscle-damaging protocol. Eight recreationally trained participants (age 21.88 ±
0.99 yr., mass 70.02 ± 11.14 kg, height 171.61 ± 8.99 cm) volunteered to participate
in five testing sessions over the course of approximately two weeks. The first
testing session included a baseline measurement of treadmill VO_2max. During the
second testing session, muscle-damaging protocol consisting of eccentric split
squats was performed, followed by another VO_2max test. Testing sessions 3, 4, and
5 all consisted of VO_2max tests 48 hours, 96 hours, and 7 days following muscle-
damaging protocol. The participants' baseline VO_2max and time to exhaustion
scores served as their control group for the experiment. Significant ($p < 0.05$) losses
in mean VO_2max values were observed for the entire testing period. The only
significant decrease in mean time to volitional exhaustion was observed

immediately after muscle-damaging protocol, but time to exhaustion did not reach

that of baseline measures until 7 days post-EIMD, where they were observed to have

eclipsed that of baseline measures. Therefore, coaches/trainers are advised to allot

their athletes at least one week of active recovery following high-intensity

resistance training before return to competition.

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CHAPTER I

INTRODUCTION

Improved motor unit recruitment patterns and increased strength that results from chronic high-intensity resistance training has been shown to have a positive impact on endurance running performance (50, 30). However, the high eccentric forces associated with this resistance training can result in Exercise-Induced Muscle Damage (EIMD) (4, 19). The EIMD is a result of sarcomere disruption (36) that leads to reduced force production (40) and muscular endurance (5) capabilities. The sarcomere disruption initiates an inflammatory response from the body's immune system (37) that leads to soreness and decreased range of motion to the affected muscles (19). Cellular metabolites, including decreased levels of creatine kinase in the blood (47) and inorganic phosphate in skeletal muscle (14) can be used as markers do determine the severity of EIMD. EIMD has also been shown to increase Rating of Perceived Exertion (RPE) and Ventilation (V_E) demands during high-intensity exercise (15, 45).

Maximal Oxygen Uptake (VO_2max) has a high correlation with athletic performance and is the most commonly used test to denote cardiorespiratory fitness and prescribe aerobic fitness training (1). A criteria of cut-off values at time to volitional exhaustion for variables with a high correlation to maximum performance and effort has been established to ensure that a VO_2max test has elicited a true physiological maximum (25). These variables include: plateau in

oxygen consumption (23), blood lactate concentration (7), Respiratory Exchange Ratio (RER) (26, 27), percentage of maximal heart rate (25), and rating of perceived exertion (RPE) (1). Researchers often disagree on said criteria's ability to determine VO_2max (25), but it is generally accepted that the peak value obtained from a maximal-effort, full-body VO_2max test is a valid index of the physiological maximum (16). The terminology as it applies to physiological maximum of peak VO_2 when conducting tests using different exercise modalities has also been debated (25).

VO_2max is directly limited by the diffusing capacity of the lungs, maximal cardiac output, the oxygen carrying capacity of the blood, and several characteristics of skeletal muscles (9). While VO_2max is the best indicator of cardiorespiratory fitness, it is not the most reliable predictor endurance performance (32). A better running economy, or the ability to use less oxygen at a given speed, has been shown to be a more accurate predictor of performance for those with a similar VO_2max (41). Running economy is highly related to the mechanical characteristics of a person's running gait (3), which can be improved through endurance training (31). A combination of the percentage of VO_2max that can be maintained under the lactate threshold along with running economy is likely the best indicator of performance (9).

Reduced performance has been seen in the days following EIMD in endurance-type cycling exercises such as time-trial performance (45) and time to exhaustion at a constant workload (15). Reductions in running economy (12, 13) and VO_2max (10) give further insight as to how EIMD contributes to these

detriments in aerobic performance. However, the first study to conclude that EIMD reduces VO_2peak (10) only extended testing to 48 hours following muscle-damaging protocol. Thus, an understanding of the extended time-course recovery of VO_2max in the days following EIMD could aid athletes and coaches in prescribing resistance training protocol during their competition season to maximize the potential benefits while avoiding the potential acute reductions in performance. Therefore, the purpose of this study was to analyze the effects of EIMD on VO_2max and time to exhaustion during maximal treadmill exercise immediately after and for up to seven days following muscle-damaging protocol.

Purpose

The purpose of this study was to examine the effects of EIMD of the hip and knee extensor muscles from eccentric squats on VO_2max and time to volitional exhaustion through repeated maximal treadmill tests immediately after and two, four, and seven days following muscle-damaging protocol.

Hypothesis

VO_2max

H_{oA}: There will be no difference in VO_2max following EIMD.

H_{A1}: There will be a significant decrease in VO_2max following EIMD.

Previous research on the effects of EIMD on VO_2max showed a significant decrease for 48 hours post-EIMD but did not extend the testing protocol after this time period. It is expected that significant VO_2max decrements will be present

immediately after and at 48 hours post-EIMD similar to the previous literature, but for the significant decrease in VO_2max values to disappear after four days of recovery.

Time to Volitional Exhaustion

H_{oB}: There will be no difference in time to volitional exhaustion following EIMD.

H_{B1}: There will be a significant decrease in time to volitional exhaustion following EIMD.

Previous research on time to exhaustion indicates that exercise time for a VO_2max test following EIMD will significantly decrease. It is expected that this will be the case immediately after and 48 hours following damage protocol, but that this significance will fade after four days of recovery.

Definitions

Exercise-Induced Muscle Damage → structural damage to the muscle's sarcomere (36) that leads to decreased force production capabilities (47) and triggers an inflammatory response (37) which causes soreness and a decreased range of motion to the affected muscles (19)

Concentric muscle action → muscle action that results in muscular shortening with movement of a body part (34)

Eccentric muscle action → muscle action that results in muscular lengthening with movement of a body part (34)

Isometric muscle action → muscle action that results from increased muscle tension and no muscle shortening or lengthening (34)

Maximal Voluntary Contraction → measure of the voluntary force production capability of the quadriceps. With a subject seated upright and their leg strapped to an immovable force reader, the subject kicks maximally against the force reader as hard as possible when instructed (40). The torque from this contraction is directly proportional to the force produced and is measured via isometric contraction to ensure standardized muscle length and velocity (47).

Fatigue → failure to maintain force or power output during sustained or repeated muscle contractions (21)

Maximal Oxygen Uptake → the greatest rate of oxygen consumption by the body measured during severe dynamic exercise (34)

> **Max** → highest oxygen uptake achieved during a treadmill test of adequate intensity and duration that meets predetermined objective criteria (38)

> **Peak** → highest oxygen uptake obtained under a specific set of conditions (including cycle and arm ergometry) but is not necessarily considered a maximal test (38)

Running Economy → the energy demand for a given velocity of submaximal running, determined by measuring the steady-state consumption of oxygen (41)

Lactate Threshold → point during graded exercise when lactate production increases to a level that it cannot be removed and begins to accumulate (34, 1)

Respiratory Exchange Ratio → ratio of carbon dioxide output to the volume of oxygen consumed (VCO_2/VO_2). This ratio is used to determine the proportion of fats or carbohydrates are being burned during exercise. At higher exercise intensities,

the body burns more carbohydrates than fats because fat oxidation requires more oxygen than carbohydrate oxidation (34).

Rating of Perceived Exertion → subjective method of measuring a participant's physical strain. When using this method, the participant indicates a point on a 6-20 visual analog scale. The increasing numbers coincide with increasing perceived exercise intensity (11).

CHAPTER II

REVIEW OF LITERATURE

Exercise-Induced Muscle Damage (EIMD) is known to be caused by repeated

muscle action, and there is a large body of evidence supporting that eccentric

muscle action produces significantly higher levels of EIMD than comparable levels of

concentric muscle action (Armstrong, Warren and Warren, 1991). This

phenomenon occurs due to the fact that the highest levels of force can be produced

during eccentric action, while less voluntary muscle activation is needed to

accomplish said force. This leads to a higher mechanical stress on those muscle

fibers, causing more muscle damage (Enoka, 1996). The functional extent of this

damage can be measured by Maximal Voluntary Contraction (MVC) torque, which is

directly proportional to the force that can be produced from the damaged muscle.

The best method to assess this is through an isometric MVC, where joint angle and

velocity can be controlled. When MVC is measured directly after damage protocol,

the torque production may be more due to fatigue from the damage protocol rather

than from the onset of EIMD (Warren, Lowe and Armstrong, 1999).

EIMD is a direct result of structural damage at the cellular level, particularly

sarcomere disruption and overextension. Eccentric muscle damage causes a more

violent stretch of the crossbridges than that of concentric muscle action. This

results in a mechanical detachment of crossbridges rather than having ATP

chemically break their bond, leading to more pronounced damage. Within the

sarcomere, disruption of Z-line interaction appears to be the main damage site (Proske and Morgan, 2001). The thinner, weaker Z-lines of Type II muscle fibers are more susceptible to damage than those of Type I fibers (Friden and Lieber, 1991). The cellular disruption of muscle tissue triggers an inflammatory response by the immune system to the affected areas, causing an influx of fluid and cells to the damaged site in order to remove the faulty contractile proteins and associated byproducts so that repair can take place (Pyne 1994). This local inflammatory response leads to soreness and decreased range of motion of the affected muscles (Enoka, 1996). The degree of accumulation of certain metabolites associated with this inflammatory response, including blood levels of creatine kinase (CK), can be used to assess the level of EIMD in the muscle, but the degree of correlation between increased metabolite concentration and functional impairment has been called into question (Warren, Lowe and Armstrong 1999).

Sargeant and Dolan (1987) measured the effects prolonged uphill (concentric) and downhill (eccentric) walking on MVC and contractile properties of the thigh over an extended period of time. Levels of plasma CK were also used as a marker for EIMD. Following uphill walking, there were only significant changes to MVC and contractile properties immediately and at 24 hours following damage protocol, with no significant change in plasma CK levels. On the other hand, there were significant reductions in MVC and muscle contractile properties for 72 hours following the eccentric muscle damage, and CK levels had increased significantly 4 hours post-EIMD and rose as high as 2-7 times higher than baseline values after 24 hours before they began to decline. These changes in muscle damage markers were

seen despite there being a much greater oxidative metabolic cost (VO_2) for the uphill waking protocol. Walsh, Tonkonogi, Malm, Ekblom and Sahlin (2001) used a protocol of eccentric cycling exercise to induce EIMD and found that while eccentric cycling increased perceived soreness, elevated CK levels were not observed at two or four days post-EIMD. The authors suggested that the completely non-existent concentric activity of eccentric cycling as compared to the impact force associated with the more pronounced concentric component of downhill walking lead to decreased EIMD.

Davies, Eston, Fulford, Rowlands and Jones (2011) used ^{31}P-magnetic resonance imaging (^{31}P-MRS) to test if levels of phosphocreatine (PCr) and inorganic phosphate (P_i) had an effect on exercise tolerance following eccentric EIMD. Perceived soreness, CK activity, and isokinetic peak torque values were used as markers to assess levels of EIMD, and time to exhaustion was used as a marker for exercise tolerance. All markers of EIMD showed significant damage for up to 48 hours following eccentric EIMD, as well as a reduced time to exhaustion. PCr values at post-exercise time to exhaustion were not significantly different at comparable pre-EIMD times, so this was not associated with the reduced exercise tolerance. On the other hand, levels of P_i at comparable exercise times were found to have a significant increase. Asp, Daugaard, Kristiansen, Kiens and Richter (1998) analyzed muscle glycogen content and Maximal Work Capacity following eccentric EIMD and found that both variables were also significantly reduced for up to 48 hours following eccentric damage compared to a control group. This led to the damaged muscle working at a higher relative intensity for the same given workload, leading

to decreased endurance. Both studies support that EIMD leads to changes in muscle characteristics that can inhibit performance for up to 48 hours due to both force measures and perceived exertion.

Twist and Eston (2009), analyzed the effects EIMD on rating of perceived exertion (RPE) and ventilatory effort during two separate cycling tests. The first cycling test involved the participants working at a constant submaximal load for a period of 5 minutes. The results from this test showed significant increases in RPE at comparable times at 48 hours following EIMD as well as increased minute ventilation (V_E) and ventilatory equivalent for oxygen (V_E/VO_2), indicating an increased ventilatory effort and decreased ventilatory threshold. The second cycling test consisted of a 5-minute time trial in which the participant covered as much distance as possible. RPE values did not vary significantly from baseline at 48 hours or seven days post-EIMD for each minute of the time trial. However, power output and distance traveled were significantly lowered at 48 hours post-EIMD, indicating a higher level of effort for a decreased performance capability. There was also no significant change in V_E or V_E/VO_2 for the time trial. In a similar study, Davies, Rowlands and Eston (2009) investigated the effects of EIMD on the RPE and ventilatory responses during moderate- and severe-intensity cycling exercise. During the moderate-intensity exercise, there was a significant increase in V_E for 48 hours post-EIMD, but there was no significant difference in V_E/VO_2 or RPE. For the severe-intensity exercise, participants pedaled against a pre-determined work rate until volitional exhaustion. Time to exhaustion was significantly reduced for 48 hours post-EIMD, and V_E, V_E/VO_2, and RPE were all significantly increased during

the severe intensities, which supports the findings of Twist and Eston (2009) as it applies to a constant, submaximal load.

According to the American College of Sports Medicine (ACSM) (2014) maximal oxygen uptake (VO_2max) is accepted as the gold standard to measure cardiorespiratory fitness. This variable measures the maximal amount of oxygen that can be consumed by an individual, and this measurement is expressed in liters per minute (L/min). This absolute measurement is generally converted into a relative measurement to allow for comparisons among individuals with different body mass. This relative measurement is expressed in milliliters per kilogram per minute (mL/kg/min). VO_2max is derived from the product of maximal cardiac output (Q_{max}), expressed in liters of blood per minute (L/min), and the arterial-venous oxygen ($a\text{-}vO_2$) difference, expressed in milliliters of oxygen per liter of blood ($mL\ O_2/L$ blood). Because of the many different procedures that can be used to conduct a VO_2max test, a set of criteria has been put together to determine if a test has elicited a true VO_2max.

A plateau in oxygen uptake has become the primary criterion for determining the validity of a VO_2max test. This concept was first brought about by Hill and Lupton (1923). While measuring the oxygen consumption of a subject running around an outdoor track at increasing speeds, they noticed that there was a similar roof to the oxygen consumption that the subject could attain, regardless of the work intensity. Taylor, Buskirk and Henschel (1955) analyzed expired air from participants running on an inclined treadmill at a 10% grade. On subsequent days, the participants ran at the same speed at a 2.5% higher grade. The test was

repeated until the highest two consecutive VO_2 values measured on different days were separated by less than 2.1 ml/kg/min (approximately 150 ml/min). Although this measurement has generally been used as the precedent for plateau criteria, the actual cut-off value and rationale can very among studies. Howley, Bassett, and Welch (1995) give examples of some varying plateau criteria and the rationales for those sets of criteria that have been used for a variety of studies. The cut-off values for these studies generally give a stricter definition for the attainment of a plateau, with values ranging from 50-100 mL/min.

The necessity of a plateau in oxygen consumption to confirm the validity of a VO_2max test has been called into question. Day, Rossiter, Coats, Skasick and Whipp (2003) noted that when the theory of a plateau in oxygen consumption was first brought about tests were generally performed using a discontinuous protocol. The tests would consist of a bout of exercise being performed at a constant work rate followed by a period of rest. The same test would then be performed at a higher work rate until the plateau had been achieved, as opposed to the more recent, continuous test design in which work rate is increased every one to three minutes until the participant reaches exhaustion. This is also known as an incremental ramp test. Upon investigation, the authors found no significant difference in VO_2max values obtained from the constant-load protocol and the progressively increased workload protocol performed on the same subjects. Because the same values were obtained from the different test protocols, they concluded that a plateau in oxygen uptake is not necessary to elicit a valid VO_2max test. Poole, Wilkerson, and Jones (2008) found that only five of their eight participants exhibited a discernable

plateau in oxygen consumption during an incremental ramp test although their

VO_2max values were confirmed using a separate test protocol, which supports the

conclusion made by Day et al. (2003).

Astorino, Robergs, Ghiasvand, Marks and Burns (2000) suggest that sampling

intervals may have a dramatic effect on the ability of an exercise test to produce a

plateau in VO_2, thus potentially explaining the reason for much of the discrepancies

reported among researchers. After conducting three separate VO_2max test

protocols on 16 participants, they concluded that the low prevalence of VO_2 plateau

reported in scientific literature is due to sampling intervals being too widely

separated. Their data showed a 100% incidence of VO_2 plateau for sampling

intervals were separated by 15 seconds, whereas the incidence of plateau dropped

to 57% and 8% when VO_2 was measured every 30 seconds and 60 seconds,

respectively.

Blood lactate concentration has been used as a secondary criterion for the

attainment of a valid VO_2max test. Astrand (1952) decided to use post-exercise

levels of blood lactate concentration to supplement his oxygen plateau criterion

when only 50% of his subjects achieved his criterion for a plateau in VO_2. He found

the average blood lactate concentration of the subjects who had shown a plateau in

VO_2 to be 7.9 – 8.4 mMol/L. The exact cut-off values of blood lactate concentration

can also vary greatly. Howley et al. (1995) illustrate the theoretical rationale used

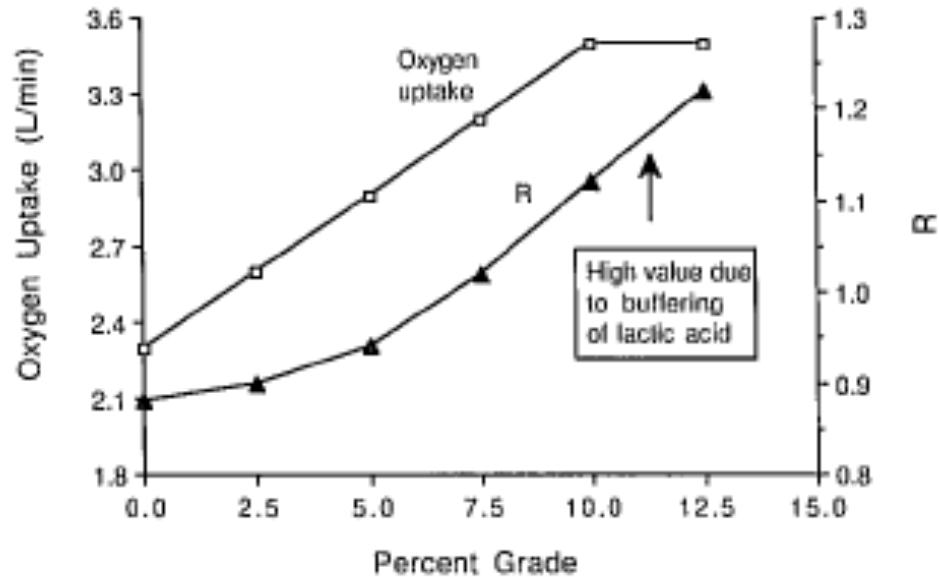by Astrand to establish his lactic acid criterion.
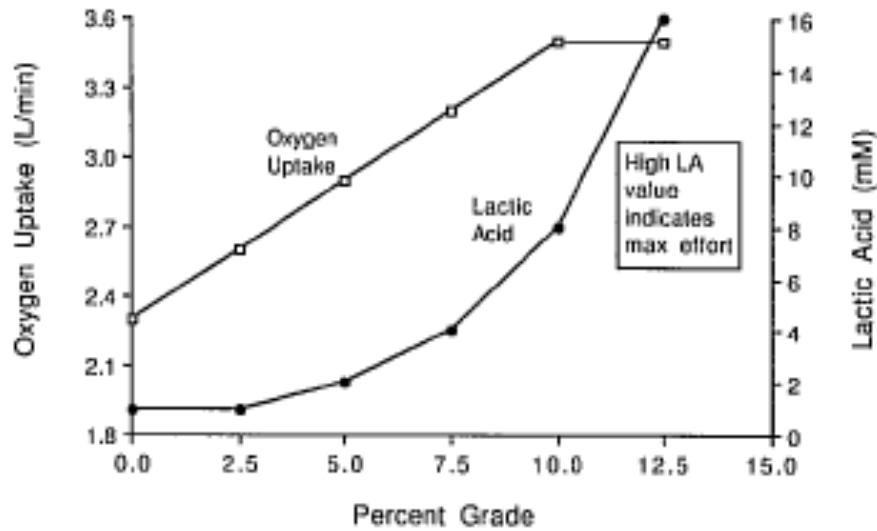
Figure 3—Theoretical plot of the change in blood lactic acid with increasing intensities of exercise, showing its relationship to a plateau in oxygen uptake.

Poole et al. (2008) concluded that the commonly used criterion value (≥ 8 mMol/L) for blood lactate concentration would have excluded six of their eight participants for a valid VO_2max test. On the other hand, they cited a study by Wasserman, Beaver, Davies, Pu, Heber and Whipp (1985) in which blood lactate concentrations as high as 15 mMol/L were recorded for participants exercising at a submaximal level, showing the high variability of lactate concentration for different populations.

Respiratory Exchange Ratio (RER) has also been used as a secondary criterion for attainment of a valid VO_2max test. The establishment of this criterion has its roots in the studies by Issekutz, Birkhead and Rodahl (1961) and Issekutz and Rodahl (1962), in which they used a cut-off value of RER ≥ 1.15. Howley et al. (1995) illustrate the theoretical rationale of changing RER values with increasing

work intensities as compared to a plateau in VO_2. RER cut-off values will also vary from study to study (Howley et al. 1995).

Figure 4—Theoretical plot of the change in the respiratory exchange ratio (R) with increasing intensities of exercise, showing its relationship to a plateau in oxygen uptake.

Poole et al. (2008) found that the RER values of 1.10 and 1.15, both of which are consistently used as criteria for attainment of a VO_2max, occurred at VO_2 values significantly lower than the maximal values attained at the end of testing protocol. While these findings suggest that a more strenuous cut-off value for RER should be used, the authors do recognize that it is possible for someone to attain VO_2max without reaching said cut-offs.

The achievement of some percentage of an age-adjusted estimate of maximal heart rate (HR_{max}) has also been used as a secondary criterion to determine the validity of a VO_2max test (Howley et al., 1995). The formula used to estimate this HR_{max} is 220 – age (in years). Londeree and Moeschberger (1984) concluded that

the standard deviation associated with this estimate is ±11 bpm, leading to possible significant over or under estimation in a VO_2max test.   In congruence with this finding, the ACSM (2014) states that this age-adjusted estimate should not be used as an absolute determination for the conclusion of a VO_2max test. The achievement of a pre-determined RPE level has also been used to ensure that the participant has exerted a maximal effort.

Scharhag-Rosenberger, Carlsohn, Cassel, Mayer, and Scharhag (2011) conducted a study to test the validity of an incremental ramp test protocol by use of a supramaximal verification test. Ten minutes after the completion of an incremental ramp test, participants completed a constant-load test at a treadmill speed of 110% of the maximal velocity attained during the first test. If the supramaximal test validated the incremental ramp test, participants returned the following day for an identical supramaximal test. If the incremental ramp test was not validated, a second supramaximal verification test was conducted after another ten-minute resting period at a treadmill speed of 115% of the maximal velocity attained from the incremental ramp test. The test showed that 85% (34 of 40 participants) of the incremental ramp tests were confirmed by the first supramaximal verification test, although time to exhaustion was significantly longer on the second day of testing. The second verification test validated an additional four subjects, with two reporting that they were physically unable to perform the final supramaximal test.

The terminology as it pertains to maximal oxygen uptake has also been scrutinized (Howley et al., 1995). Rowell (1974) used the term VO_2max to describe

the highest oxygen uptake achieved during a treadmill test of adequate intensity and duration that met his objective criteria. He used the term VO_2peak to describe the highest value obtained under a specific set of conditions but not necessarily considered a maximal test, including cycle or arm ergometry. Howley et al. (1995) point out that researchers and reviewers often use the term VO_2peak if the certain criteria are not met, although those same researchers and reviewers are unable to consistently agree on said criteria. Katch, Sady, and Freedson (1982) conducted a study to test biological variability as it pertains to VO_2max tests. Five participants completed a total of 80 incremental ramp tests on a treadmill, with each participant completing at least eight tests. After completion of the study, 49 of these tests (61%) met the criteria for a "true" VO_2max. Upon examining the remainder of the 31 tests, their data suggested that the peak VO_2 value obtained during an incremental ramp test was sufficient to justify it being termed a VO_2max. Likewise, Day et al. (2003) concluded that the peak VO_2 value obtained from a maximal-effort incremental ramp treadmill test for a participant exercising to volitional exhaustion is likely a valid index of VO_2max, although confirmation from an additional constant-load test is preferred.

There are four physiological factors that can potentially limit the body's ability to deliver oxygen to the muscles during maximal exercise. These factors include the oxygen diffusing capacity of the lungs, maximal cardiac output, the oxygen carrying capacity of the blood, and several characteristics of skeletal muscle (Bassett and Howley, 2000). The first three are known as central factors, and they pertain the aspects of oxygen delivery to the muscles. The characteristics of skeletal

muscle are known as peripheral factors and pertain to the utilization of oxygen once it is delivered (Bassett and Howley, 2000).

For an average person, the ability of the pulmonary system to fill the heart with oxygenated blood does not tend to inhibit VO_2max as much as it does a trained individual. Powers, Lawler, Dempsey, Dodd, and Landry (1989). In the study conducted by Powers et al. (1989), the authors used hyperoxic gas to test the effects of pulmonary diffusing capacity in trained versus untrained individuals. They found that there was no significant increase in a normal subject's average VO_2max with an increase of arterial oxygen saturation, whereas highly trained participants did show a significant increase in VO_2max when hyperoxic gas was substituted for normal air. This supported findings by Dempsey, Hanson, and Henderson (1984), who attributed cardiac output to the differences in VO_2max seen in trained versus untrained participants. They concluded that the much higher cardiac output seen in trained participants reduces transit time between red blood cells in the pulmonary capillaries, leading to the decreased arterial oxygen concentration with normal air. Because athletic events are not conducted in hyperoxic atmospheric conditions, the pulmonary diffusing capacity does not have as much of an impact on VO_2max as cardiac output (Bassett and Howley, 2000).

As it pertains to cardiac output, stroke volume has been shown to limit an individual's VO_2max more than their heart rate. This is due to there being less variation in a trained individual's heart rate as compared to an untrained individual's heart rate of the same age with a significantly lower stroke volume. Thus, training has a more pronounced effect on stroke volume than on heart rate

(Bassett and Howley, 2000). The oxygen carrying capacity, or more specifically the hemoglobin content, of the blood is the last of the central factors. The hemoglobin content of the blood can be artificially increased by removing blood from the body then reinfusing it once the body's red blood cell count has returned to normal, a process known as blood doping (Ekblom, Wilson, and Astrand, 1976). There are also artificial methods of increasing the hemoglobin content of the blood, but manipulation of blood and blood components is banned in virtually every organized sport (Barnes and Rainbow, 2013).

Peripheral limitations have to do with aspects of the skeletal muscle. These include mitochondrial enzyme levels and capillary density (Bassett and Howley, 2000). The mitochondria are the sites of oxygen consumption in working muscle, so in theory, increasing mitochondria levels should increase oxygen consumption. However, only minimal gains in VO_2max have been seen with as much as doubled mitochondrial enzyme density (Bassett and Howley, 2000). Capillary density has been observed to increase with training (Andersen and Henriksson, 1977) in order to elongate the mean transit time of red blood cells passing through the capillaries, allowing for more oxygen to be extracted during this time (Saltin, 1985). These peripheral limitations have been observed to contribute more directly to endurance performance than to a whole-body VO_2max test. Cardiac output is currently believed to be the greatest limiting factor for VO_2max (Bassett and Howley, 2000).

While VO_2max is used as the gold standard to measure cardiovascular fitness, it may not be the most reliable variable to examine as it pertains to endurance performance. Running economy, defined as the steady-state VO_2 for a given running

velocity (or workload), has been shown to account for a significant amount of

variation in distance running performance among runners with comparable VO_2max

values (Morgan, Martin, and KrahenBuhl, 1989). On a very basic level, runners with

good running economy use less oxygen than runners with poor running economy at

the same given speed, so for competitive runners with a similar VO_2max, running

economy is a better predictor of performance than VO_2max (Saunders, Pyne,

Telford, and Hawley, 2004).

Anderson (1996) provides a table that lists the biomechanical factors related

to better running economy.

Factor	Description for better running economy
Height	Average or slightly smaller than average for males and slightly greater than average for females
Ponderal index	High index and ectomorphic or mesomorphic physique
Body fat	Low percentage
Leg morphology	Mass distributed closer to the hip joint
Pelvis	Narrow
Feet	Smaller than average
Shoes	Lightweight but well cushioned shoes
Stride length	Freely chosen over considerable training time
Kinematics	Low vertical oscillation of body centre of mass
	More acute knee angles during swing
	Less range of motion but greater angular velocity of plantar flexion during toe-off
	Arm motion that is not excessive
	Faster rotation of shoulders in the transverse plane
	Greater angular excursion of the hips and shoulders about the polar axis in the transverse plane
Kinetics	Low peak ground reaction forces
Elastic energy	Effective exploitation of stored elastic energy
Training	Comprehensive training background
Running surface	Intermediate compliance

While this is noteworthy, it does not give insight as to the mechanisms that can be

improved by training. Moore, Jones, and Dixon (2012) designed a study to observe

what biomechanical factors can be improved from training. In this study, beginner

runners were subjected to a 10-week training program aimed at improving running economy. The results of their study showed a significant increase in running economy for the subjects, and of the 34 biomechanical factors that were analyzed, seven were identified to have significantly changed from prior to the training program. These factors include: decreased toe-off knee extension, decreased toe-off plantar flexion, increased touchdown plantar flexion velocity, slower touchdown eversion velocity, slower peak eversion velocity, timing of peak dorsiflexion occurring later in the stance, and increased peak propulsive force. Of these seven, it was determined that knee extension, timing of peak dorsiflexion, and eversion velocity at toe-off accounted for 94.3% of the improved running economy. These results support the findings of Williams and Cavanagh (1987). Likewise, the finding by Moore, Jones, and Dixon (2012) also support the notion of Williams and Cavanagh (1987) that suggests individuals self-adapt their running styles during training to produce a gait that is more economical than their original gait.

The lactate threshold, or particularly the level of oxygen consumption obtained at the lactate threshold (expressed as a percentage of VO_2max) also has a high impact on endurance performance (Bassett and Howley, 2000). While VO_2max is primarily limited by central factors of the cardiovascular system (Bassett and Howley, 2000), the percentage of VO_2max at the lactate threshold is limited more by peripheral factors of the skeletal muscles (Holloszy and Coyle, 1984). Endurance training elicits the peripheral adaptions that lead to a higher percentage of VO_2max at the lactate threshold, which include a higher concentration of mitochondria, a slower utilization of muscle glycogen and blood glucose, a higher rate of fat

oxidation, and decreased lactate production (Holloszy and Coyle, 1984). Together, the central and peripheral factors along with running economy contribute to overall running performance (Bassett and Howley, 2000).

Chen, Nosaka, and Tu (2007) conducted a study using downhill running to analyze the effects of EIMD on running economy. The results of their study showed that running economy was significantly reduced immediately after and for the following three days after EIMD. These results are supported by Chen, Nosaka, Lin, Chen, and Wu (2009), who used the same damage protocol and also found that EIMD has a greater effect on decreased running economy for increasing intensities. The results from Chen, Nosaka, and Tu (2007) also identified that stride frequency was significantly increased, while stride length and range of motion of the knee and ankle following EIMD were all significantly reduced, attributing to an overall reduction in running economy. Tsatalas, Giakas, Spyropoulos, Sideris, Kotzamanidis, and Koutedakis (2013) support all of these kinematic findings following EIMD except ankle range of motion. This discrepancy likely surfaced because the damage protocol used in their study did not put stress on the ankle due to the participants being seated, as opposed to downhill running creating an unnatural surface for the ankle to land on.

Black and Dobson (2012) conducted a study to test the effects of EIMD on VO_2peak during cycling exercise. Their results showed a significant decrease in VO_2peak 48 hours post-EIMD. This contrasted the only other published study (Gleeson, Blannin, Walsh, Field, and Pritchard, 1998) to examine the impact of EIMD on VO_2peak, although there was evidence by Gleeson et al. (1998) of an earlier onset

of lactate accumulation. Black and Dobson (2012) suggest that the reason for this difference was that damage protocol used in the study by Gleeson et al. (1998) did not elicit sufficient levels of EIMD.   The findings reduced VO_2peak by Black and Dobson (2012) were attributed to reductions in peak power output due to early exercise termination. Their findings of reduced V_E and HR at volitional exhaustion support the idea that the participants terminated exercise before their maximal physiological potential was reached, although they were giving a maximal effort. The authors state that possible explanations for the reductions in VO_2peak include reduced force production capability in the damaged muscles, an earlier onset of lactate accumulation as evidenced by a lower ventilatory threshold, and local muscle pain and soreness in the affected muscles.

The time period for the effects of EIMD are of interest because of the positive impacts that resistance training, especially high-intensity resistance training using explosive movements or heavy weight, has on endurance running performance (Yamamoto, Lopez, Klau, Casa, Kraemer, and Maresh, 2008). The improvements in running performance are attributed to neural adaptions leading to more efficient motor unit recruitment patterns and increased strength that improve running economy (Mikkola, Rusko, Nummela, Pollari, and Hakkinen, 2007). If VO_2max is reduced in the days following EIMD, it could contraindicate some forms of resistance training during the competition phase of training when an uninhibited VO_2max is desired. The present study looks to continue the work done by Black and Dobson (2012) by examining the time-course recovery from the effects of EIMD on aerobic performance by observing VO_2max test values for up to seven days

following EIMD. The extended time period should allow enough recovery to

examine the length of time over which VO_2max remains reduced.

CHAPTER III

METHODS

Experimental Approach to the Problem

The purpose of this study was to investigate the time-course of recovery from

Exercise-Induced Muscle Damage (EIMD) as it applies to oxygen consumption and

exercise time, via analysis of repeated Maximal Oxygen Consumption (VO_2max) test.

This study used a within participant, repeated measures design with each

participant serving as his/her own control.

Participants

N	Age	Weight (kg)	Height (cm)
8	21.88 ± 0.99	70.02 ± 11.14	171.61 ± 8.99

Eight participants (5 male and 3 female) completed the study. Recruitment took

place via flyers, emails, and class announcements in the Oxford, MS, and University,

MS, communities. The study was open to adults (18-35 years old) who did not have

a history of orthopedic injuries of the hip, knee, and/or leg. Also, participants who

had not participated in high intensity weight training with their legs in the past 6

months and were currently or had had a recent history of aerobic training

(running/cycling) at least three days per week were excluded as well. Additionally,

all participants were to be free from use of prescription pain or psychiatric

medication. Each subject read and verbally affirmed understanding before signing

an informed consent approved by the Institutional Review Board at the University of Mississippi. Participants also completed the physical activity readiness questionnaire (PAR-Q) and were excluded from participation if they answered "yes" to any of the seven questions on the questionnaire.

Procedures

Total time commitment for the study was approximately 6 hours over the course of 14 days. One familiarization session and five testing sessions were completed.

For the familiarization session, potential participants reported to the Kevser Ermin Applied Physiology Laboratory for a 45-minute session. During this initial session, written and verbal description of the experiment and all procedures were given, and any questions were answered. Informed consent was given. Next, the PAR-Q was completed, which identified any contraindications to exercise. Then, potential participants who were deemed eligible for the study were asked to orally state to the researchers what was expected from them in the study and explain the risks and benefits of the study to confirm that they understood the procedures, time commitment, freedom to withdraw, and risks and benefits of study participation. Next, participants were familiarized with the experimental procedure for assessing maximal oxygen uptake (VO_2max). A heart rate monitor was strapped around the chest of the participant. The researchers provided instructions about the use of the perceived exertion and leg muscle pain rating scales. The breathing mouthpiece involved in the VO_2max testing procedure was inserted into the participant's mouth, and a nose clip was placed on the participant's nose. This allows for expired gases to be collected during exercise. Researchers also instructed the participant on the

use of hand signals to indicate impending volitional exhaustion or the need to immediately stop the test. Next, participants ran on the treadmill at a "comfortable" pace, which the participants determined themselves, for ten minutes while breathing into the mouthpiece. The initial six minutes were at 0% grade of the treadmill incline, minutes 6-8 were performed at 2% grade, and minutes 8-10 were performed at 4% grade. At the end of each minute, each participant practiced providing ratings of perceived exertion (RPE) and muscle pain ratings.

For the first testing session, participants reported to the Kevser Ermin Applied Physiology Laboratory for a 45-minute session. During this session, participants completed a maximal exercise test to assess VO_2max during running. The researcher provided instructions about the use of the perceived exertion and leg muscle pain rating scales. The participant then strapped a heart rate monitor around his/her chest. A sterile mouthpiece and nose clip were used to collect expired gas. The participant walked at 3.5 mph on the treadmill for five minutes as a warm-up. Following this, the participant began running at a comfortable pace with the treadmill at 0% grade of incline. Every two minutes, the grade on the treadmill incline was increased by 2%. The participant ran until they reached volitional exhaustion. Once the participant had signaled that they had reached exhaustion, the treadmill belt was slowed back down to 3.0 mph to allow the participant to cool down. During this cool down period, the breathing mouthpiece and nose clip was removed.

For the second testing session, the participant reported to the Kevser Ermin Applied Physiology Laboratory approximately 48 hours following the first testing

session for a 90 minute session. First, the participant completed a 24-hour history of diet, exercise, and supplement/drug use. Next, muscle soreness in the quadriceps, gluteus muscles, and overall leg muscles was assessed. This was done by having the participant complete three separate body squats in a slow and controlled manner. Following the completion of each body squat, the participant was asked to rate the intensity of the pain/soreness in their quadriceps, gluteus muscles, and overall leg muscles, respectively, on a 10 cm visual analog scale. The participant was instructed to place a mark along the 10 cm line that corresponds to the intensity of pain experienced during the squat. Anchors of "no pain" and "worst pain imaginable" were placed on the left and right end of the 10 cm line, respectively. The participant then completed eccentric muscle actions (4 sets to failure with each leg) of the quadriceps and gluteus muscles by performing single legged split squats with a workload 60% of their body weight. The participant was instructed to perform each eccentric muscle action in a slow (approximately 3 seconds) and controlled fashion. Researchers verbally counted during each action to assist in controlling the speed of the movement. The researchers helped raise the weight prior to the start of each lift to ensure that the participant primarily performs eccentric muscle action. Upon completion of the eccentric exercise, participants were asked to refrain from the use of pain relieving medications and alcohol, until the experiment was complete. Participants were also reminded of the instructions for monitoring symptoms related to rhabdomyolysis and the instructions for proper hydration for the duration of the study. Next, the participant rested for 20 minutes before muscle soreness was again assessed in the same

manner as previously described. Finally, participants performed a VO_2max test on the treadmill as described previously.

For the third and fourth testing sessions, participants reported to the Kevser Ermin Applied Physiology Laboratory approximately 48 hours following the previous testing session for a 45-minute testing session involving the following. The participant completed a 24-hour history of diet, exercise, and supplement/drug use. Next, muscle soreness in the quadriceps, gluteus muscles, and overall leg muscles, respectively, was assessed as previously described. Finally, a VO_2max test during running was assessed as described previously.

Testing session five was conducted in the same manner as testing sessions three and four, except the participants reported to the Kevser Ermin Applied Physiology Laboratory approximately 72 hours following the previous testing session instead of 48 hours following the previous testing session.

Equipment

Indirect calorimetry was used to measure all metabolic data during treadmill walking. Before any metabolic testing was conducted, the system was calibrated against standard gases (O_2 = 16.0%, CO_2 = 4.0%). The treadmill used to conduct VO_2max tests was the Trackmaster by JAS (Newton, KS). During the tests, the TrueOne 2400 Metabolic Measurement System (Sandy, UT) was used to analyze the expired air of the participants. After each test was completed, a Nova Biomedical Lactate Plus (Waltham, MA) was used to analyze blood lactate levels. Standard weight and height scales were used to measure the participants' anthropometric information.

Statistical Analysis

A 1x5 Repeated Measures Analysis of Variance (ANOVA) was completed for two separate dependent variables using the SPSS 21 statistical software: oxygen consumption and time to exhaustion. Significance was determined by using Fisher's Least Significant Difference (LSD) test. If a probability value of less than 0.05 was scored on the LSD test, the value was determined as significant.

CHAPTER IV

RESULTS

VO$_2$max
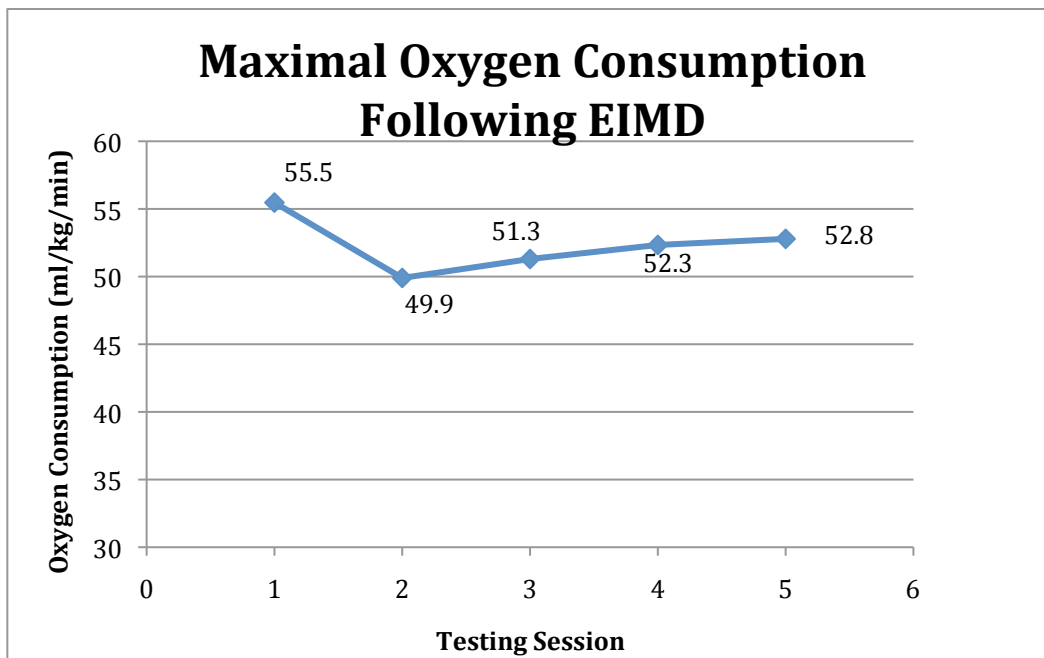
There was significant decrease (p < .025) in oxygen consumption following EIMD for up to seven days. This decrease was most evident immediately following damage protocol, with mean VO$_2$max values falling to 90%, 92%, 94%, and 95% of baseline measurements for immediately, 48 hours, 4 days, and 7 days post-EIMD, respectively. Although mean VO$_2$max levels did not return to significant values within baseline measurements, significant improvements were seen for 4 days (p = 0.027) and 7 days (p = 0.008) post-EIMD as compared to immediately after damage

protocol. Also, the improvement from 2 days post-EIMD to 7 days post-EIMD

showed a trend that approached significance ($p = 0.057$).

Time to Exhaustion

Time to Exhaustion Following EIMD
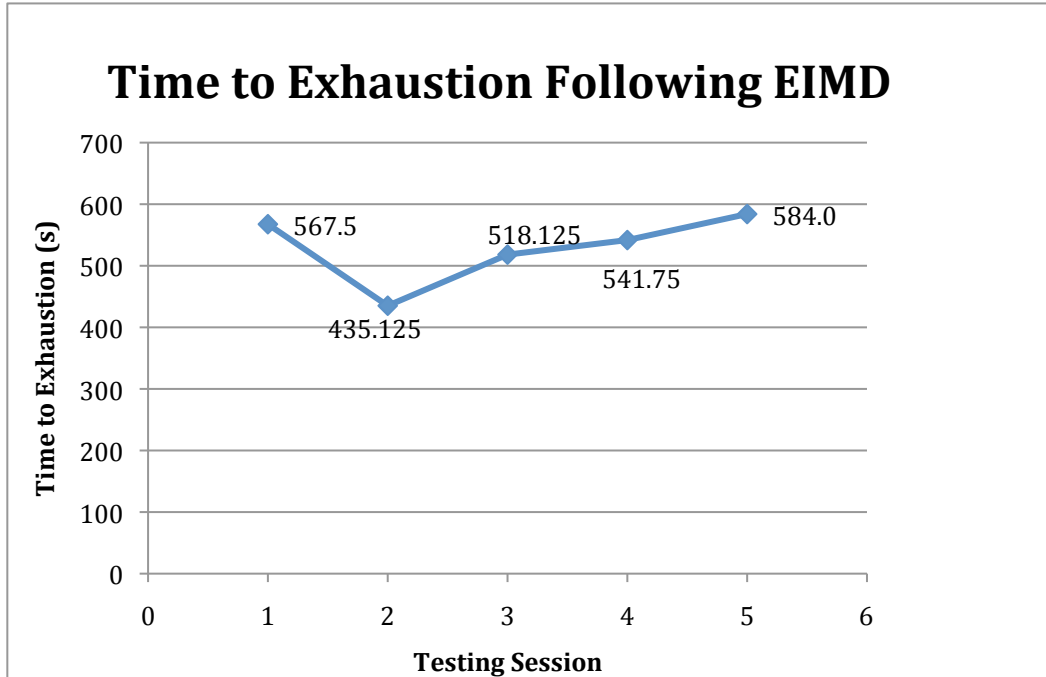
There was a significant ($p < 0.05$) mean reduction immediately following EIMD for

time to exhaustion, but this significance faded ($p > .050$) after 48 hours of recovery,

although the mean time to exhaustion did not fully recover to pre-EIMD values until

7 days post-EIMD, when mean times to exhaustion actually eclipsed those of

baseline values. Time to exhaustion was significantly ($p < 0.005$) higher at 48 hours,

4 days, and 7 days compared to immediately following damage protocol, and the

mean difference in time to exhaustion at 48 hours post-EIMD compared to 7 days

post-EIMD was found to be significant ($p = 0.001$). Percentages of baseline time to

exhaustion for post-EIMD values are as follows: 77%, 91%, 95%, and 103% for

immediately, 48 hours, 4 days, and 7 days post-EIMD, respectively.

CHAPTER V

DISCUSSION

The purpose of this study was to investigate the time-course recovery from EIMD of the hip and knee extensors on VO_2max and time to exhaustion for up to seven days by using repeated maximal treadmill tests. The results of the study found that VO_2max was significantly reduced for up to seven days following EIMD, which supports the finding by Black and Dobson (2012), who observed significant reductions in VO_2peak from maximal cycle tests for at least 48 hours. Time to exhaustion during the maximal treadmill test was found to be significantly reduced only immediately following muscle-damaging protocol. However, time to exhaustion did not reach levels of baseline measures until seven days post-EIMD. This indicates that EIMD could possibly be leading to reductions in time to exhaustion for as long as four days, supporting the finding by Davies, Rowlands and Eston (2009). This could in turn have an effect on the ability of the cardiovascular system to reach its maximum capacity, which was also in agreement with Black and Dobson (2012), but it does not explain why VO_2max was still reduced after a week of recovery. Also, the substantially pronounced decrements in time to exhaustion immediately following damage protocol could have been more strongly attributed to fatigue from the squats than the onset of EIMD, as suggested by Warren, Lowe, and Armstrong (1999).

Black and Dobson (2012) attributed their reductions in VO_2peak for a maximal cycling test two days following damage protocol to decreased force production capabilities of the muscles affected by EIMD and earlier exercise

termination due to an earlier onset of lactate accumulation and elevated muscle pain and soreness. Because there is evidence suggesting EIMD leads to a significantly decreased force production capability for the affected muscles for up to 72 hours (Sargeant and Dolan, 1987) and an earlier lactate accumulation during prolonged exercise for up to 48 hours (Gleeson, Blannin, Walsh, Field, and Pritchard, 1998), the conclusions by Black and Dobson (2012) are feasible. Although the results of this study do not lead to a rejection of these conclusions, Black and Dobson (2012) only continued testing for 48 hours post-EIMD. In the present study, VO_2max remained significantly lower than baseline for seven days post-EIMD, while time to exhaustion had recovered to values comparable to baseline after two days of recovery.

Because the protocol for the VO_2max tests in the present study had standardized time intervals for increasing intensity, it is possible to make an indirect, general analysis of running economy following EIMD by using the VO_2max and time to exhaustion data. Chen, Nosaka, and Tu (2007) found running economy to be significantly decreased immediately after and for up to three days following muscle-damaging protocol due to decreased force production capabilities, increased stride frequency, and reductions in stride length and range of motion in the knee and ankle joints. In the present study, mean VO_2 taken at four minutes into the post-EIMD VO_2max tests shows that the VO_2 measurement was highest immediately after damage protocol and declined progressively for two, four, and seven days post-EIMD. The higher VO_2 values at comparable exercise times indicate the lowest running economy immediately after damage protocol, with it improving gradually

over seven days. While statistical analysis was not run on these numbers, there is

still an indication of improved running economy with recovery time. Also, the lower

VO_2 levels accompanied by longer to time to volitional exhaustion after seven days

of recovery technically indicates a better running economy than that of baseline

times, which could mean that improvements in running economy can possibly be

seen from an intense bout of resistance training in seven days.

One of the limitations of this study was the sample size. Eight participants

may not be enough to draw concrete conclusions. Also, the damage protocol used in

this study likely caused more EIMD than a normal bout of resistance training would

elicit. While this leads to more pronounced symptoms from the EIMD, it may

overestimate the decrements to performance and elongated recovery time. Also,

some participants completed their baseline VO_2max test up to two weeks before

returning for the second testing session due to the university's spring break. This

could have led to a difference in the training status of the participants, leading to

varied results. Also, because the participants were all recreationally trained

volunteers, their inter-individual differences in level of training could have led to the

EIMD having varying effects. The participants of the study were also asked to

refrain from exercise between testing sessions. In a more applied setting, athletes

would have practice/training every day (or near every day), which would likely lead

to faster recover from EIMD due to more blood flow to the muscle during said

practice/training sessions.

In conclusion, EIMD can potentially lead to a significant decrease in VO_2max

for up to seven days and a decrease in time to exhaustion at a given workload for up

to four days. If this is known to be true, it should be recommended that coaches/trainers allow at least seven days of active recovery from resistance training before athletic competition. Future research on this subject should look to control some particular aspects of the experimental design when using a similar protocol. First, a larger sample size would be recommended in which training status can be distributed as evenly as possible. This would be best brought about if the researcher were able to conduct the study with an entire competitive team who had been exposed to the same training program. If this were possible, the most opportune timing for the start of the study would be immediately following the competitive season, allowing for as close to a realistic in-season training status as possible. Also, different protocols for the assessment of VO_2max should be taken into account depending on what sport/athletic event the participants would be competing in so as to elicit more generalizable results.

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