RELATIONSHIP BETWEEN

BLOOD LACTATE AND ELECTROMYOGRAPHY

DURING AEROBIC EXERCISE

by

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ABSTRACT

The assessment of lactate threshold (LT) is an important measurement to prescribe training intensities and monitor chronic adaptations in athletes. A non-invasive method, electromyography (EMG), has been suggested as an alternative approach to LT testing. Three experiments determined the ability of EMG incorporated into compression shorts to estimate LT, effect of exercise on LT and EMG threshold (EMG_T), and determined the most appropriate filtering method of the EMG signal to estimate LT. In the first investigation, participants performed an incremental exercise test while blood lactate and EMG were measured. EMG displayed no differences from blood lactate in the ability to predict LT ($p = 0.08$). EMG_T and LT showed a moderate correlation ($r = 0.68$, $p = 0.01$) between the determination of work rates. The EMG_T occurred at the same stage of the incremental test as LT in 11 out of the 13 participants (85%). No differences were seen between percentage of maximal oxygen consumption or percentage of maximal heart rate between LT and EMG_T. In the second study, the effect of exercise on LT and EMG_T measurement was evaluated. Participants completed two maximal exercise tests separated by 30-minutes of exercise. Individual agreement demonstrated that pre- and post-exercise LT occurred at the same work rate in 5 of 10 participants; while pre- and post-EMGT occurred at the same work rate in 6 of 10 participants. Results indicated no significant difference between the work rates of the pre-exercise LT and EMG_T (0.43), although post-trial LT was significantly lower than post-trial EMG_T ($p = 0.007$). No difference in test stage were seen between the pre- and post-exercise EMG_T; however, post-trial LT occurred at a lower work rate as compared to pre-trial LT ($p = 0.03$). In the final study, four popular methods of EMG

signal transformation were examined in order to determine their effectiveness in estimating LT. The methods used were root mean square (10- and 60-second epochs), 60- second Smoothing, and 60-secons peak-amplitude averaging. Results indicated no differences in the ability of any signal processing variations to predict LT or in relation to $\%VO_{2peak}$ at each threshold level. In conclusion, EMG has been demonstrated to be a viable tool to estimate LT and may provide a reliable low-cost, non-invasive method of prescribing training intensities based upon EMG_T testing.

DEDICATION

Dedicated to my Mom and the rest of my family and friends who have provided me with the much needed support throughout my academic career. Thank you for all you have done. I am truly grateful for you being there for me and I cannot express how appreciative I am of you.

LIST OF ABBREVIATIONS

AT	anaerobic threshold
ATHOS	wearable electromyography compression shorts
Dmax	maximal computational distance of two best fit lines
EMG	electromyography
EMG_T	electromyographical threshold
[La]	blood lactate concentration
LT	lactate threshold
MLSS	maximal lactate steady state
RMS	root mean square
VO_{2peak}	peak oxygen consumption
VL	vastus lateralis
VT	ventilatory threshold

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CHAPTER 1

INTRODUCTION

A long history of research in endurance performance exists, particularly in relation to the determination of lactate threshold (LT) and its' ability to predict sporting outcome [Cheng et al., 1992; Dumke et al., 2006; Candotti et al., 2008; Carey et al., 2008; Green et al., 2014]. Prior to research into lactate threshold training, it was believed that VO_{2max} was the best indicator for athletic performance in high-intensity long-duration exercise [Passfield et al., 2000]. However, the current model for performance prediction argues that among athletes with similar VO_{2max} levels, an increased capacity to maintain high-intense exercise at near maximal VO_2 level in combination with low blood lactate concentrations ([La]) may be a better determinant of success [Lucía et al., 2000].

Practitioners often use LT work rates in the field for the purpose of assigning training intensities for endurance athletes, due to the performance-limiting impact of increasing blood [La] during long-duration events [Rowlands et al., 2000]. Additionally, LT work rates can provide an insight into the intensity of the training program; whereas exercise performed at the level of LT is considered moderate, work rates below are low-intensity, and above are high-intensity. However, proper assessment of blood [La] and LT work rates requires trained personnel for handling blood samples, valid testing equipment, and can be costly; therefore testing is not always a feasible option in the field.

The importance of precise determination of lactate threshold power outputs was confirmed by Weekes et al (1996). It was determined that well-trained cyclists were unable to

maintain intensity levels even slightly above LT (i.e., 15 Watts) for a duration of 30 minutes; whereas intensities 15 W below could be maintained for periods lasting longer than 30 minutes [Weekes et al., 1996]. This provides an insight into the sensitivity of workload determined by LT analysis and necessity of prescribing exercise based upon LT values.

At rest, the process of [La] accumulation and clearance is highly related to an individuals' metabolic rate, but not a direct reflection of oxygen availability [Gladden 2004]. However, in working skeletal muscle tissue, blood [La] does not accurately estimate the metabolic state during exercise; yet an association can be assessed by the relationship between increases in exercise intensity and [La]. While the accrual of [La] during exercise is a normal function of cellular metabolism, aerobic exercise performance is typical unaffected as long as clearance exceeds production of blood lactate [Powers, 2012]. If the accumulation of [La] progresses, it can efflux from the cell and enter various areas of the body (e.g., Type I muscle fibers, cardiac tissue, liver, etc.) to be used as an energy source. However, if exercise intensity continues to increase and blood [La] production rises to a level that exceeds clearance capacity and [La] inhibitions in motor unit recruitment (i.e., muscular contraction) can occur and disrupt the ability to continue performance unless intensity is reduced.

Other factors that determine La production and clearance can be dependent upon fiber recruitment patterns (e.g., Type I versus Type II), mass of the primary musculature involved in the task, and secondary musculature recruitment [Beneke et al., 2001]. For example, during high-intensity exercise utilizing a small muscle group as the primary mover, larger muscle groups may be performing as secondary or assisting movers. In these cases, the lactate shuttle hypothesis states that the larger, secondary mover may utilize [La] as a fuel source, under the assumption La production has exceeded clearance in the prime mover, via oxidative metabolism, with the

conversion of lactate to pyruvate via lactate dehydrogenase. Observations of blood [La] during this type of exercise may result in consistent, steady-state values. However, during bouts of exercise where large muscle groups are the prime mover (e.g., incremental cycling), smaller muscle groups may not be able to meet the demands of clearance versus the rapid production of La during high-intensity work [Beneke et al., 2001; Gladden, 2004]. Furthermore, the type of muscle fiber recruited may have an effect on lactate production as isozymes of lactate dehydrogenase exist which vary amount aerobic and anaerobic fibers. Within fast-twitch fibers a form of LDH resides that favors the production of lactate from pyruvate; whereas slow-twitch fibers contain an isozyme that prefers the breakdown of La to be used as an energy substrate [Myers et al., 1997].

Additionally, during high-intensity exercise there is an elevation of circulating catecholamines (i.e., epinephrine and nor-epinephrine) which can indirectly stimulate phosphorylase, a key regulatory enzyme, responsible for the breakdown of glycogen to be used for energy production in glycolysis. The increase in glycolysis rate produces an increase in blood [La], while simultaneously inhibiting fat metabolism via the reduction in substrate availability [Powers, 2012]. Previous research has demonstrated increases in blood lactate production during infusion of circulating epinephrine [Stainsby et al., 1985]; while blockage of beta-adrenergic receptors caused decreases in blood [La] during exercise [Stainsby et al., 1990]. It can also be hypothesized that during high-intensity activity (e.g., during the latter stages on an incremental graded exercise test) intra-muscular pressure exceeds that of blood capillary pressure resulting in their occlusion [Petrofksy et al., 1981]. Blockage of capillaries during the intense activity could lead to an increase in anaerobic glycolysis and the product of lactic acid; thus promoting a sharp increase in blood [La] [Petrofsky et al., 1981].

Literature has provided many conflicting definitions of lactate threshold (LT). For example, the exponential method initially described by Lundberg (1986) describes LT as the point at which blood [La] begins to increase in an exponential manner [Lundberg et al., 1986]. Alternative methods include the rise of blood [La] by 1.0 $mmol \cdot L^{-1}$ during incremental exercise, a 4.0 $mmol \cdot L^{-1}$ critical threshold (i.e., onset of blood lactate accumulation), ventilatory threshold marker (i.e., rise in breathing frequency and the inability to hold conversation during exercise), or the log-log method by which lactate values begin to deviate upwards from a straight regression line during incremental work rates [Lundberg et al., 1986; de Sousa et al., 2011]. Additionally, LT is the rate in which cardiovascular exercise can be sustained for long durations without subsequent increases in VO_{2max}, as well as the marker to indicate the transition from moderate to heavy exercise intensity. This justifies the examination of the ability of field metrics, such as EMG, to predict LT and allowing practitioners to prescribe training and competition intensities based upon individual variations in blood La production.

The OBLA method, 4.0 $mmol \cdot L^{-1}$ threshold, has been recently criticized as inaccurate method to determine intensity thresholds as this does not reflect individual LT variations, particularly between trained and untrained individuals [McLellan, 1985; Zhou et al., 1997]. Currently there are several computational methods to determine LT (e.g., Dmax, log-log transformation, 3^{rd} order polynomials, etc.) which may provide a more objective measure of calculation. One of these methods, Dmax, has been suggested to allow for a more individualized approach to predicting LT via analysis of all points along the given curve, as opposed to observed [La] points during testing [Cheng et al., 1992]. This computational method also provides a reduction in observer error as no visual determination of LT is required. Zhou and Weston (1997) determined the reliability of the Dmax method during incremental cycling on two

separate occasions with an intra-class correlation coefficient ranging from 0.77 – 0.93.While multiple methods of LT prediction and testing exist, along with advantages and disadvantages of each, the Dmax method of calculating LT provides an approach, which can be applied to non-invasive metrics (i.e., EMG) and field applicable.

While blood [La] is often used to determined threshold workloads, other laboratory means have also been utilized in order to introduce a non-invasive approach. Such methods consist of ratings of perceived exertion (RPE) [Fabre et al., 2013], heart rate deflection points [Carey et al., 2008], expired gas [Cheng et al., 1992], near infrared spectroscopy [Borges et al., 2016], and electromyography [Candotti et al., 2008]. One such non-invasive method that has been suggested to determine LT power output levels during incremental exercise is electromyography. EMG is a method by which the electrical activity within a muscle is recorded to determine the relative amount of motor units activated in a particular area of the muscle. There are currently two forms of EMG which captures motor unit action potentials (i.e., intramuscular and surface). Intramuscular EMG involves the penetration of the skin and muscle to place a fine-wire electrode to directly capture a specific motor unit deep within the skeletal tissue. Unlike intramuscular EMG, surface EMG provides a non-invasive method of monitoring the muscle activation via surface electrodes placed over a general muscular area. Two electrodes are placed, typically 2 cm apart, over the muscle belly; while a third electrode serves as the ground and placed over a bony process.

Similar to lactate, EMG during incremental exercise typically contains a positive exponential break point, termed the EMG threshold (EMG_T). Since lactate accumulates during exercise with increasing intensity (e.g., VO_{2max} testing), the associated increased [H^+] may have an effect on cellular membrane potential [Moritani et al., 1982; Lucía et al., 1999]. This negative

effect results in a decreased ability of the muscle to provide the necessary strength of contraction for a given task. This effect also provides a coinciding increased signaling of additional motor units to supply adequate energy to maintain performance. Due to the size principle, the types of motor units to be recruited in times of increasing intensities are Type II (anaerobic) fibers which can provide a greater twitch rate compared to Type I. Although, exponential rises in lactate and other metabolic by-products can be expected with this recruitment due to metabolic properties of Type II muscle fibers [Lucía et al., 1999]. Furthermore, increasing usage of Type II fibers can be witnessed within the EMG recording with increased amplitudes and right mean frequency shifts of the signal. This shift from a primarily aerobic (i.e., Type I) to a focus on anaerobic (i.e., Type II) motor units represents the EMG_T. Only recently has the reliability of EMG_T been demonstrated using two incremental cycling bouts separated by 48 hour sessions [MahmutoviĆ et al., 2016]. Thus, the relationship of LT and EMG_T needs further exploration in order to provide practitioners and athletes a reliable, valid field tool to determine training intensities and reduce the need for expensive laboratory testing and personnel for lactate testing.

PURPOSES AND HYPOTHESES

The purpose of this dissertation was to examine the various relationships between surface EMG, blood [La], and LT during aerobic activity. These relationships provide an understanding of EMG frequency, amplitude, and signal processing in order to determine the acute metabolic and physiology strain of the lower limb musculature during cardiovascular training. Specific purposes for each individual study are described below.

Study 1. The purpose of the first study was to determine if surface EMG obtained from wearable compression shorts could be a valid non-invasive method of estimating lactate

threshold during a maximal aerobic-exercise bout. We hypothesized that the EMG would serve as a useful tool of estimating LT during incremental exercise.

Study 2. The purpose of the second study was to determine the agreement between LT and EMG_T after a bout of steady-state aerobic activity. We hypothesized that LT determined via blood lactate sampling would differ as blood lactate concentrations would be altered due to the aerobic trial; whereas EMG would recover immediately and produce a similar workload as established by the pre-trial EMG_T.

Study 3. The purpose of the third study was to examine the best method of LT estimation during incremental aerobic exercise using various filters and time averaging windows of the EMG signal. We hypothesized that root mean square (RMS) and smoothing filtering methods would show no significant differences; whereas average peak amplitude would yield a more precise estimation of blood [La] compared to RMS and Smoothing. Additionally, we hypothesized that no differences would exist between time segment epochs.

SIGNIFICANCE OF THE DISSERTATION

This dissertation is important in its determination of the capabilities of EMG to estimate LT during incremental aerobic exercise. This ability would be useful to athletes and practitioners as a non-invasive, cost-effective method to monitor the physiological and neuro-motor responses during cardiovascular training or competition. Study 1 provides an initial perspective on the ability of wearable EMG to estimate LT power output during incremental aerobic exercise. Study 2 is the first to examine the agreement between EMG and LT after a bout of steady-state aerobic exercise. Additionally, results provide an insight into motor unit recruitment and lactate production during a period of fatigue. Study 3 provides a viewpoint as to the best filtering and

time averaging method of the EMG signal to predict LT. This was the first investigation to combine the various means by which EMG is currently used to observe changes in blood [La].

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CHAPTER 2

VALIDITY OF WEARABLE ELECTROMYOGRAPHICAL COMPRESSION SHORTS TO PREDICT LACTATE THRESHOLD DURING INCREMENTAL EXERCISE IN HEALTHY PARTICIPANTS

ABSTRACT

The purpose of this investigation was to determine if wearable surface electromyographic (EMG) technology, acquired from compression shorts, can validly estimate the lactate threshold work rate during incremental cycling. Thirteen adult men (n = 8) and women (n = 5) volunteered to participate in this study. Participants completed an incremental, maximal test on a cycle ergometer. Blood lactate was measured every minute, while EMG was recorded throughout the test at the vastus lateralis. Results demonstrated no significant differences ($p = 0.08$) between lactate and electromyographical thresholds. Additionally, no differences existed between EMG and lactate thresholds for maximal heart rate ($p = 0.13$, Cohen's $d = 0.43$) and percent peak oxygen consumption ($p = 0.64$, Cohen's $d = 0.09$). Consistent with previous results, EMG provided a moderate correlation with the prediction of work rates associated with LT ($r = 0.68$, $p = 0.01$). The exponential rise in blood lactate concentration and EMG may be attributed to the increase in Type II muscle fiber recruitment. Thus, wearable EMG compression gear may provide a viable field tool for monitoring training intensity and predicting LT work rates.

KEY WORDS: Electromyography; Aerobic Training; Blood Lactate; Cycling

INTRODUCTION

Lactate threshold (LT) has been utilized as a predictive measure of performance, due to its representation of a balance between lactate (La) production and clearance [Yoshida, 1984]. For example, when comparing two athletes possessing the same VO_{2max}, the individual with the ability to maintain a higher lactate threshold has the greater chance of success during a long-duration competition [Lucía et al., 2000]. Thus, LT can be seen as an important predictor of endurance performance and a useful measure in training.

Training at levels at or above LT is useful in increasing cardiovascular endurance, as well as inducing reduced [La] levels for a given power output [Davis et al., 1979; Yoshida, 1984, Gollnick et al., 1986]. Therefore, the LT is a key determinant in programming aerobic exercise intensity. Weekes et al (1996) demonstrated that athletes were unable to maintain even slight intensities (i.e., 15 Watts) greater than the level of LT for a duration of 30 minutes [Weekes et al., 1996]. Additionally, training at oxygen uptake levels (i.e., $\%VO_{2peak}$) deemed below LT has been shown to acquire a typical steady-state response in 2-3 minutes; whereas $\%VO_{2peak}$ intensities above LT have shown a delayed steady-state response and led to early exhaustion of the individual [Barstow, 1994; Roston, 1987].

Unfortunately, monitoring [La] during exercise is a costly, invasive blood analysis, which requires either capillary blood samples or an indwelling venous catheter. However, electromyography is a potential new method of monitoring exercise intensity and may provide a novel, cost-effective field technique to monitor [La] during endurance training. Surface electromyography (EMG) is a non-invasive measure of the electrical activity, in microVolts (μV), of skeletal muscle tissue along with alterations in motor unit recruitment. Preliminary research indicates EMG can be used as a method of estimating the LT during incremental

exercise [Candotti et al., 2008]. As exercise intensity increases, the accumulation and production of by-products such as La exert a negative effect on skeletal muscle membrane potential leading to the need for additional motor unit activation [Vaz et al., 1996]. Increased high-threshold fiber recruitment thereby impacts the amplitude of the EMG signal (i.e., increased μV output) over time [Vaz et al., 1996; Candotti et al., 2008].

Hypothetically, a relationship exists between the EMG signal and La production. Increases in the amplitude of the EMG signal depend on changes in motor unit recruitment and the amount of skeletal tissue recruited [Beneke et al., 2001; Pires, 2006]. Type I muscle fibers record at low frequencies between 20-125 Hz; whereas type II muscle fibers produce signaling at 125-250 Hz. During incremental exercise, a shift in the recorded signal from low to high frequencies (i.e., type I, slow-twitch to type II, fast-twitch fibers) is stated to be the EMG threshold (EMG_T). Recent research by Candotti et al. (2008) demonstrated a positive association ($r = 0.87$) between EMG_T and LT; along with the ability of EMG to determine the work rate at which LT occurred in recreational cyclists [Candotti et al., 2008]. EMG was sampled over 1-second averages and EMG_T determined by the intersection of two linear regression lines. However, only one [La] sample was collected per stage during the exercise trials which can result in a decrease in the sensitivity of LT analysis, and those initial findings have not been confirmed.

Commercially-available devices are currently able monitor EMG to capture activity of lower-body musculature via wearable compression shorts with specialized electrodes. However, the ability of the EMG, via compression gear, to predict lactate threshold has yet to be investigated. As such, the primary aim of this investigation was to determine if EMG, acquired from compression shorts, can be a valid method of estimating the lactate threshold power output

during incremental exercise. Based upon previous findings [Petrofsky, 1979; Candotti et al., 2008], it was hypothesized that EMG_T and LT would display a strong positive correlation. The results of this investigation may provide practitioners and athletes with a non-invasive field method of identifying EMG_T and LT during training.

METHODS

Experimental Approach to the Problem

The current study was designed to determine if surface electromyography via a new commercially-available wearable product could be used as viable source to non-invasively predict lactate threshold during incremental aerobic exercise. Thirteen participants completed an incremental exercise test (VO_{2peak}) on a cycle ergometer. Blood lactate was analyzed via fingertip during each minute of the test, while EMG was recorded from the vastus lateralis continuously. This study was approved by the university ethics committee.

Participants

Thirteen male and female healthy individuals participated in this study. Descriptive statistics are displayed in Table 1 for all participants. The sample size was sufficient according to an a priori power analysis with G Power software (Heinrich-Heine University of Dusseldorf, Dusseldorf, Germany) which determined 11 participants would be needed to obtain statistical power at the recommended 0.80 level. Participants recruited met the following criteria: 1) Between the ages of 18-40; 2) Currently participating in regular moderate-to-vigorous aerobic training of a minimum of three days per week for at least 30 minutes per session; and 3) Free from cardiovascular, metabolic, or neurological disorders that would otherwise affect the results or negatively impact safety.

Table 2.1. *Descriptive statistics of the study participants*

	Men (n = 9)	Women (n = 4)	All (n = 13)
Age (yr)	23.67 ± 5.55	20.75 ± 1.50	22.77 ± 4.80
Height (cm)	175.71 ± 4.16	165.03 ± 8.41	172.42 ± 7.45
Body mass (kg)	85.61 ± 10.05	60.50 ± 6.39	77.88 ± 14.94
HR_{max}	181.33 ± 13.35	181.5 ± 5.51	181.38 ± 11.24
VO_{2peak}	35.24 ± 5.04	32.38 ± 7.28	34.36 ± 5.67

HR_{max} = heart rate maximum; VO_{2peak} = peak oxygen consumption

Procedures

Participants were asked to report to the Human Performance Laboratory for one visit. Upon arrival, participants reviewed and signed an informed consent and medical history questionnaire. After consent, participants had height and weight measured. Standing height was measured to the nearest 0.1cm using a stadiometer (SECA 67310, SECA©, Chino, CA); while weight was measured to the nearest 0.1 kg (Tanita BWB-800, Tanita©, Arlington Heights, IL). Participants then performed an incremental maximal cycling test while blood lactate, heart rate, and electromyographical signals were measured at the site of the vastus lateralis. Prior to testing participants were familiarized with all equipment and procedures used in the investigation. All testing was performed on a manually-braked cycle ergometer (Monark 484 E; Monark©; Dalarna, Sweden). Participants were provided an initial warm-up on the cycle ergometer at an output of 40 W for a period of three minutes. The testing phase began immediately following the warm-up at a work output of 80 W and increased in increments of 40 W every 3 minutes thereafter. Cadence was kept at 80 (±5) revolutions per minute. Test termination criteria were: 1) Subject was no longer able to maintain cadence; or 2) Volitional fatigue.

During each stage the following variables were measured: blood lactate, heart rate, VO_2, and EMG activity. Capillary blood samples (25 µL) were taken via finger prick and analyzed every minute throughout the testing procedures, including rest and warm-up. Lactate concentrations were analyzed via the Lactate Plus Meter (NOVA Biomedical, Waltham, MA). Lactate Plus monitors have been shown to have a standard error of the estimate of 0.6 $mmol \cdot L^{-1}$ and strong correlation of 0.94 when compared to laboratory measures [Tanner et al., 2010]. A heart rate monitor (Polar Electro Oy, Kemple, Finland) was placed on the participants' chest in order to accurately assess heart rate during the test. A metabolic cart (TrueOne® 2400, ParvoMedics Inc., Sandy, UT, USA) was used to determine the oxygen uptake at the mouth. Before each test, the metabolic cart was properly calibrated according to the manufacturer's instructions. VO_{2peak} was recorded as the average oxygen consumption, expressed as $ml.kg^{-1} \cdot min^{-1}$, among the last minute of the incremental exercise test.

Surface EMG signals were collected with compression shorts containing built-in surface electrodes (Athos©, Mad Apparel, Inc., Redwood City, CA). The compression shorts contain electrodes that were situated over the vastus lateralis for measurement of the EMG signal. Preparation for the skin sites included shaving, abrasion, and alcohol cleansing in order to reduce impedance of EMG signals. Recordings were deemed viable when impedance was below 5 kΩ. Raw myoelectrical signals were quantified in this product using root mean square (RMS) transformation along with a signal conversion from analog to digital. EMG signals from the compression gear were averaged at a 10-second window.

LT and EMG_T Determination (Dmax Method)

LT and EMG_T were determined in each participant via Dmax calculations. Dmax was defined as the point that yielded the maximal distance from the model fitting [La] and EMG curve, using a 3rd order polynomial equation, to the line formed by the lowest and highest [La] and EMG values taken during the incremental test [Zhou et al., 1997]. The Dmax method has been previously validated as computational method to provide a reliable training threshold during incremental exercise. The Dmax point was computed on a custom-written computer program. The percent of VO_{2peak} ($\%VO_{2peak}$) and percent maximal heart rate ($\%HR_{max}$) were recorded at the LT and EMG_T to allow for comparisons between each threshold.

Statistical Analysis

The data obtained were analyzed using a software package (SPSS version 22.0, Somers, NY). Due to the staged, ordinal nature of the incremental test, the level of LT and EMG_T were compared using the Wilcoxon Matched-Pairs Signed-Rank Test. A Spearman's rank order correlation was performed to determine the relationship between LT and EMG_T. Additionally, a paired sample T-test was applied to identify any statistically significant $\%VO_{2peak}$ and $\%HR_{max}$ at the EMG_T, provided by the compression garment for the vastus lateralis muscle, and LT. Individual agreement was examined by calculating the number of times EMG_T and LT occurred within the same stage of the maximal exercise test. Significance was set at $\alpha = 0.05$ for all statistical analyses.

A Cohen's *d* statistic [Cohen, 1992] was calculated as the effect size of the differences and Hopkin's scale of magnitude [Hopkins et al., 2009] was used where an effect size of 0-0.2 was considered trivial, 0.2-0.6 was small, 0.6-1.2 was moderate, 1.2-2.0 was large, >2.0 was very

large. The procedures of Bland and Altman [Bland et al., 2010] were used to evaluate the 95% limits of agreement of the EMG_T method compared to the LT.

RESULTS

Thresholds in both [La] and EMG were observed in all participants in the investigation. Individual data, along with means (± SD) for the $\%VO_{2peak}$ and $\%HR_{max}$ at the EMG_T and LT are shown in Table 2. The mean blood [La] at the level of LT was 3.6 ± 0.69 $mmol \cdot L^{-1}$. There were no significant differences observed between the work rates obtained through blood [La] or EMG compression gear ($Z = -1.732$, $p = 0.83$). The wearable technology EMG_T occurred at the same stage of the incremental test as the blood lactate threshold in 11 out of the 13 participants (84.6%). Out of the remaining participants (i.e., 2), EMG_T took place one stage higher than LT. Using the Spearman's rho correlation, there was a significant moderate correlation found between LT and EMG_T ($r_s = 0.677$, $p = 0.01$).

In terms of $\%VO_{2peak}$, there were no significant differences between LT and EMG_T. Non-significant ($p = 0.38$), small (Cohen's $d = 0.45$) differences were seen between males and females for $\%VO_{2peak}$ at the level of LT. A significant correlation existed between $\%VO_{2peak}$ from both measures (i.e., [La] and EMG) at LT ($r = 0.73$, $p = 0.003$). The Bland Altman procedure suggested that the EMG_T displayed a 95% limits of agreement that ranged ± 9.3% around a constant error of -0.97% of VO_{2peak} (Figure 1A), with the upper and lower limits at 10.3 and -8.4 %, respectively. There was no correlation between the x- and y-axes of the Bland Altman plot suggesting no proportional bias existed ($r = 0.48$, $p = 0.101$).

$\%HR_{max}$ did not differ between the LT (88.8 ± 2.95%) and EMG_T (90.4 ± 4.06%) ($p = 0.13$, Cohen's d = 0.43). Furthermore, the $\%HR_{max}$ values between LT and EMG_T showed a significant moderate correlation ($r = 0.58$, $p = 0.04$). The Bland Altman procedure showed that

the EMG_T displayed 95% limits of agreement that ranged ±6.6% around a constant error of -1.5% of HR_{max} (Figure 1B), with the upper and lower limits at 8.1 and -5.1 %, respectively. There was no correlation between the x- and y-axes of the Bland Altman plot suggesting no proportional bias existed (r = 0.37, p = 0.21).

Table 2.2. *Individual and group mean ± SD values among the participants for $\%VO_{2peak}$ and $\%HR_{max}$ values at EMG_T and LT (n = 13)*

Participant	**$EMG_{\%VO2peak}$**	**$LT_{\%VO2peak}$**	**DIFF**	**$EMG_{\%HRmax}$**	**$LT_{\%HRmax}$**	**DIFF**
1	71.67	66.17	5.50	89.93	85.88	3.95
2	72.56	72.56	0.00	88.16	89.74	-1.31
3	76.06	76.97	-0.91	85.25	85.79	-0.54
4	65.88	67.06	-1.18	95.74	92.02	3.72
5	76.04	69.36	6.68	90.34	89.20	1.14
6	74.24	74.10	0.14	91.35	92.43	-1.08
7	78.17	76.30	1.87	92.82	90.26	2.56
8	86.70	83.10	3.60	97.28	88.59	8.69
9	81.13	76.70	4.43	94.95	90.40	4.55
10	69.42	75.84	-6.42	83.24	84.37	-1.13
11	71.16	71.87	-0.62	88.64	86.36	2.28
12	83.37	75.60	7.77	89.33	93.82	-4.49
13	63.50	71.81	-8.31	87.70	86.10	1.60
Mean	75.26	74.65	-0.61	90.36	88.82	-1.53
SD	6.85	5.79	4.76	4.06	2.95	3.36
95% CI	13.5	11.4	9.3	7.9	5.8	6.6
p	0.64			0.13		
Cohen's d	0.09			0.43		

DIFF = difference in $\%VO_{2peak}$ between EMG_T and LT

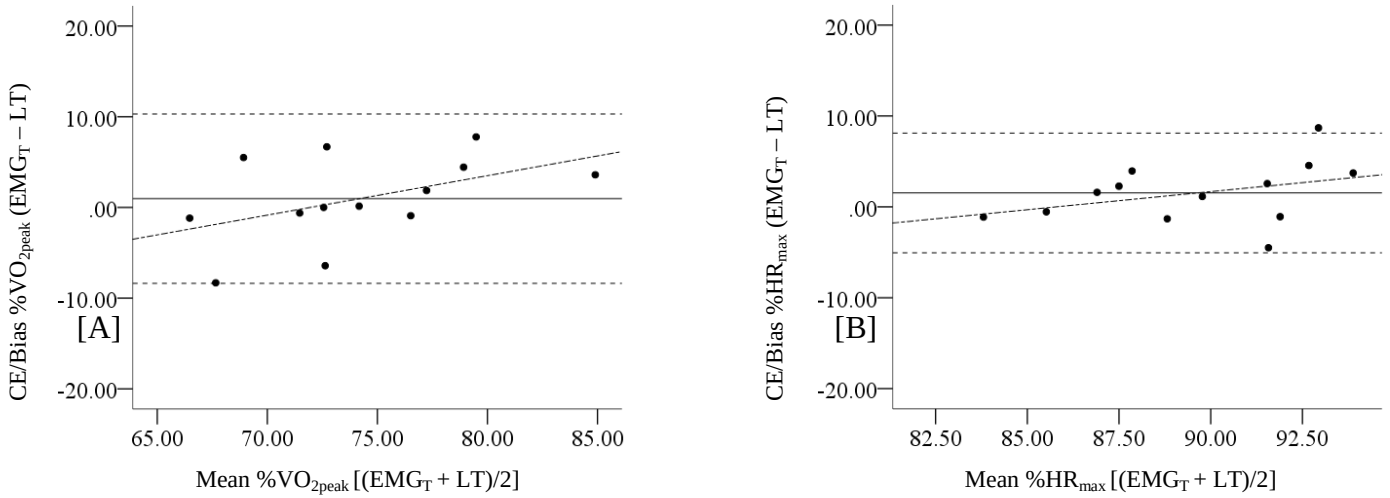

Figure 2.1. Bland and Altman plots comparing [A] $\%VO_{2peak}$ and [B] $\%HR_{max}$ (n = 13). The middle solid lines indicate the constant error or mean difference. The dashed lines represent the upper and lower limits of agreement (± 1.96 SD). The dashed-dotted regression lines represent the trend between the errors of each measurement. There was no significant correlation in the errors for either $\%VO_{2peak}$ ($p = 0.10$) or $\%HR_{max}$ ($p = 0.21$).

DISCUSSION

The purpose of this investigation was to determine if a field-wearable EMG device in compression shorts was a viable method of estimating LT via changes in the EMG signal. Consistent with previous results of EMG_T [Candotti, 2008], our findings indicate that EMG predicted the LT during incremental exercise. A key finding was the compression gear showed no significant differences in determining the LT when analyzed via $\%VO_{2peak}$ or $\%HR_{max}$. Therefore, wearable EMG technology such as the device analyzed in the study appears to provide a suitable method for estimating the LT within field settings by simply monitoring HR.

Whereas no other wearable EMG technology has been tested to predict LT, a compression sleeve, using near infrared LED technology, worn on the calf (i.e., BSX insight) has recently been shown to provide a reliable method of predicting LT [Borges, 2016]. The wearable sleeve provided oxygenation levels and measured threshold work rate at a different location (i.e., gastrocnemius) than the device of the present study. However, both that study and the present findings support the notion that non-invasive wearable technology may have the ability to be useful field measures of training intensities.

In contrast to the current study, Seburn et al. (1992) proposed that EMG was not a viable option to monitor the anaerobic threshold indicating that several criteria must be satisfied prior to establishing the relationship between blood [La] and EMG. First, non-linear associates must exist between work rate and muscle activation; however the data of the current study satisfies this notion as a curvilinear relationship existed in all participants examined. Secondly, EMG must respond immediately to the small changes in La production and decrements in pH that occur at the level of LT. The results of the current study suggest that this association holds true since both [La] and EMG showed inflection points at the same work rate in 11 of 13 participants tested.

These results also confirm the exponential rise in blood [La] during incremental-intensity exercise, thereby providing a LT level in all individuals examined. As previously stated, the rise in blood [La] occurs when production exceeds clearance; however, this cannot be simply explained, as this is a multi-factorial approach. One factor that accounts for this is the decrease in localized lactate-removal sites available in adjacent, non-working Type I muscle fibers. As the intensity of exercise increases, additional motor units are signaled for contraction leaving fewer lactate-removal sites available for the inter- and intra-cellular lactate shuttle process to occur leading to increased [La][Gladden, 2004; Candotti et al., 2008]. Another important factor in the production of La is the stimulation of phosphorylase, a key regulatory enzyme in carbohydrate metabolism. Throughout exercise, both elevations in epinephrine and Ca^{++}/Calmodulin from contracting musculature indirectly stimulate phosphorylase to break-down glycogen to be used during glycolysis to form energy, along with elevated [La] (in times of high-intensity). As the recruitment of type II fibers occur during high-intensity exercise, less reliance is placed upon oxidative metabolism, due to the low mitochondrial density of anaerobic muscle fibers leading to an increase in La production [Powers, 2012]. Furthermore, a redistribution of blood-flow from lactate clearance sites (e.g., kidneys, non-working skeletal tissue, liver, etc.) that would otherwise alleviate the accumulation of blood La during high-intensity activity impact the rapid rise in blood [La] [Candotti et al., 2008].

The EMG signaling also provided a positive breakpoint, EMG_T, which is primarily attributed to the rise in Type II muscle fiber motor unit recruitment during an incremental exercise bout. It was noted that independent of one's training status, thresholds can be observed within the EMG signal [Candotti et al., 2008]. Previous research into shorter cycling bouts (i.e., 3 minutes) while monitoring muscular activation revealed that there was a linear relationship

between EMG amplitude and the work load being performed [Petrofsky, 1979]. However, as the bouts were extended to fatigue, RMS signal amplitudes continually increased. These results support the current study demonstrating gradual increases in EMG amplitude throughout each workload of the incremental test. Additionally, with a change in the muscle fiber membrane potential during times of decreasing pH, the excitation-contraction coupling mechanism may be altered to reduce contraction ability or strength. Thus, a compensatory effect of increased recruitment of various motor units, particularly those with a greater twitch rate, is observed. Motor units with faster twitch rates (i.e., Type IIx) alter the EMG signal and result in recordings with a greater amplitude and mean frequency [Bergstrom et al., 2013].

The cause-effect relationship between the increase in [La] and EMG is not exclusive. Previous investigation into individuals with McArdle's syndrome (i.e., glycogen storage disease Type V wherein no lactate increase is seen) demonstrated that under fatigue, EMG amplitude changes can occur in the absence of blood [La] increases. This leads to a speculation that neural signaling in the central and peripheral nervous system may act to increase motor unit recruitment in times of increasing fatigue or work load [Mills et al., 1984]. Although, Moritani et al. (1982) suggested that decreases in the intracellular pH level, due to the accumulation of [La], increases the need for motor unit recruitment in an attempt to maintain proper cadence and force output during incremental or fatiguing bouts of exercise. While this relationship is not solely dependent upon each other, another factor (i.e., potassium concentration), not measured in the current study may have had an impact on the changes in EMG signal amplitude. Several studies have shown that increasing [K+] have a significant impact on motor unit conduction velocity and may affect EMG amplitude via inhibition of excitation-contraction coupling [Juel 1988; Camic et al., 2010]. In addition to the EMG recording, it has been previously shown that myosin heavy-chain (i.e.,

percentage of Type I fibers) distribution can be an important determinant of LT and VO_{2peak} [Farina et al., 2007].

Results of the current study demonstrated that the breakpoint in [La] occurred at a mean $\%VO_{2peak}$ of 74.7%. Previous research has also observed means similar to the current study, giving values ranging from 66.1% to 80% [Seburn et al., 1992; Farina et al., 2007; Green et al., 2014]. Whereas no endurance athletes participated in this investigation, Green et al. (2014) observed no significant differences in the $\%VO_{2peak}$ at the level of LT between aerobically and anaerobically trained individuals [Green et al., 2014]. However, as expected, they observed that untrained individuals showed significantly lower values of $\%VO_{2peak}$ compared to both groups of trained individuals.

Similar to the current results, Dumke et al. (2006) observed values of 91.0 $\%HR_{max}$ during an incremental cycling test at the level of LT, almost identical to 90.4% in this investigation [Dumke et al., 2006]. Additionally, Dumke et al. (2006) demonstrated that trained cyclists were able to maintain HR values of 90 % and 85 $\%HR_{max}$ during timed trials of 30 and 60 minutes, respectively, on separate occasions. These results reinforce the notion that a field metric, such as HR, may be used as training tool for intensity in lieu of blood [La] sampling.

Whereas these present results found no significant differences between EMG and LT in relation to work rate, EMG over predicted thresholds in two individuals. One possible cause of this occurrence may be the inability of the wearable technology electrodes to conform to the individual. For instance, differences in limb length of various individuals can alter the placement of the electrodes in or out of the innervation zone, be located atop a completely different muscle, or be positioned as to acquire signal acquisition (i.e., cross-talk) from neighboring muscle

groups. Therefore, misalignments or maladjustments may have an effect on the ability of the surface electrodes to provide valid information about the musculature being examined.

This study is not without limitations. First, while the Dmax method has determined to be reproducible in test-retest studies [Cheng et al., 1992] initial daily resting blood [La] concentrations may have an impact on the linear slope of the curve by which the polynomial curve is measured against; thereby giving a false sense of work output or LT [Zhou et al., 1997]. While blood [La] measured at the site of the fingertip does not directly reflect local muscular La values, it allows for an overall interpretation of the rate and type of cellular metabolism and accrual of metabolic by-products. Another limitation of monitoring thresholds with EMG is the training status of the individual. Whereas no trained cyclists were used in this investigation, previous research indicated that training may impact results. It was noted that trained cyclists have been shown to produce two significant breakpoints in EMG signaling during incremental exercise. This occurrence may be attributed to the ability of trained individuals to recruit higher threshold motor units when intensity approaches near-maximal levels, producing a second breakpoint in the EMG recording [Lucía et al., 1999].

Although most of the research in EMG_T has occurred in cycling, future research should examine the effectiveness of EMG to predict LT in various cardiovascular modalities (e.g., running, arm ergometry, etc.). Additionally, future research may be warranted to determine the chronic cardiovascular and physiological adaptations garnered from the prescription of training loads via EMG_T.

PRACTICAL APPLICATIONS

The results of this investigation determined that wearable EMG was a valid method of determining LT work rate. Additionally, no significant differences were seen between the

thresholds in EMG and LT expressed as $\%HR_{max}$ or $\%VO_{2peak}$. This finding indicates that wearable fitness technology may provide valid real-time measures of muscle activation in a practical and field setting. Practitioners may find this information useful, as monitoring blood [La] in the field can be impractical, time consuming, and may require trained personnel to handle blood sampling. Monitoring LT may be an important determinant of the ability of the athlete or individual to maintain pre-determined exercise intensities for extended durations (i.e., 30 minutes or more). Therefore, EMG, monitored via specialized compression gear, may provide a viable option in monitoring training intensity and predicting LT levels due to its ability to provide feedback in real-time.

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CHAPTER 3

AGREEMENT BETWEEN LACTATE AND ELECTROMYOGRAPHICAL THRESHOLDS FOLLOWING STEADY-STATE EXERCISE IN UNTRAINED HEALTHY PARTICIPANTS

ABSTRACT

The purpose of this investigation was to determine the agreement between electromyography (EMG) and lactate threshold (LT) after a 30-minute bout of exercise. Participants (n=11) completed two graded exercise tests (GXT) on a cycle ergometer separated by 30-minute steady-state exercise. Blood lactate was measured the last 30-seconds of each stage during each exercise test; while EMG of the vastus lateralis and oxygen consumption (VO_2) were monitored continuously. Individual agreement demonstrated that pre- and post-exercise LT occurred at the same work rate in 5 of 10 participants; and pre- and post- EMG_T occurred at the same work rate in 6 of 10 participants (1 participant elicited no EMG_T during the trials). Pre-exercise LT and EMG_T occurred at the same stage in 3 individuals; while post-exercise LT and EMG_T only occurred at the same work rate in 2 participants. Results showed no mean difference between work rates for LT or EMG threshold for the pre-GXT; however, LT during the post-GXT was significantly lower than EMG_T ($p = 0.007$). Post-GXT LT work rates were also determined to be significantly lower than pre-GXT LT ($p = 0.034$); whereas no differences existed in EMG thresholds. The utilization of EMG to estimate LT after a bout of exercise may not provide a valid metric due to poor individual agreement.

Key Words: Threshold Training; Electromyography; Cycling; Aerobic Training

INTRODUCTION

Lactate threshold (LT) training is a form of cardiovascular endurance training that consists of performing sustained aerobic activity at the speed, power output, or percentage of VO_{2max} at which blood lactate concentration ([La]) begins a rapid increase above baseline [Baechle et al., 2000; Yoshida, 1984]. Training at an intensity equal to or above the LT can result in favorable performance adaptations such as improved aerobic power and lactate clearance [Baechle et al., 2000; Davis et al., 1979;Gollnick et al., 1986]. In addition, steady-state training below LT for an extended period can be especially important for peripheral adaptations such as increased capillary and mitochondrial densities [Powers, 2012]. Though most investigations focus on the importance of threshold-based training in trained and elite endurance competitors, LT testing may also have important implications for baseline measures among recreationally trained healthy participants who are beginning an endurance training program.

Previous investigations using surface electromyography (EMG) to detect muscle fiber recruitment patterns during graded exercise testing (GXT) have reported a progression in recruitment of Type II muscle fibers with an increase in work rate [Guffey et al., 2012; Malek et al., 2006;Travis et al., 2011]. This has been identified as the electromyographical threshold (EMG_T) and defined as the greatest exercise intensity an individual can maintain without an exponential increase in the amplitude of the EMG signal [MahmutoviĆ et al., 2016]. Because an increase in EMG amplitude occurs near the LT, the EMG_T has been suggested as a non-invasive surrogate to blood lactate testing [Candotti et al. 2008; Dumke et al., 2006].

Previous investigation into the use of EMG_T to predict the LT has shown a significant positive association during incremental exercise [Candotti et al., 2008]. However, research with McArdle's syndrome patients has suggested that the EMG_T and LT display an independent

relationship and a more coincidental relationship than cause-effect [Mills et al., 1984]. The results of that research suggest that EMG amplitude and motor unit recruitment during exercise may be based upon fatigue in the muscle membrane and various ionic concentrations not limited to blood lactate production [Mills et al., 1984]. Furthermore, the relationship between EMG_T and LT following steady state exercise is unknown. This is an important consideration since the practical implications of both measures relate to the assumption that the work rate from each threshold should remain consistent across a steady state bout of exercise, hence their value for prescribing exercise intensity. However, the metabolic response of a decrease in glycolysis and increase fatty acid oxidation, and the neuromuscular response of a shift from Type II to Type I muscle fibers during steady state exercise may not occur at the same time-points [Beneke et al., 2001].

The purpose of this investigation was to evaluate the agreement between EMG_T and LT after a bout of moderate-intensity, steady-state aerobic exercise in recreationally active participants. Because of the previous suggestions of a disassociation between EMG_T and LT [Mills et al., 1984], and due to the possible different metabolic and neuromuscular responses to steady-state exercise [Beneke et al., 2001], it was hypothesized that the agreement between EMG_T and LT would decrease post-exercise.

METHODS

Experimental Approach to the Problem

A repeated measures design was utilized to evaluate if the agreement between surface electromyography and blood lactate concentrations after a 30-min bout of steady-state aerobic exercise. Eleven participants completed two graded exercise tests (GXT) on a cycle ergometer separated by 30-min of steady-state cycling 40 Watts below individual LT between the two tests.

Blood lactate was sampled during the last minute of each stage of the GXT's and every 5 minutes during the steady-state bout. Surface electromyography was recorded continuously throughout each test and the exercise bout at the vastus lateralis. The EMG signal and the blood lactate values pre- and post-exercise served as the dependent variables for this investigation.

Participants

Apparently healthy adult males (n = 5; ages = 26.4 ± 4.9 years; weight = 95.3 ± 15.5 kg; height = 181.7 ± 6.5 cm) and females (n = 6; ages = 23.8 ± 2.2; weight = 67.8 ± 6.9 kg; height = 162.3 ± 3.3 cm) volunteered for participation in this study. A power analysis determined that approximately 11 participants were needed to obtain statistical power at the 0.80 level. Participants were recruited with the following criteria: 1) Between the ages of 18-40; 2) Currently participating in regular moderate-to-vigorous aerobic training of a minimum of three days per week for at least 30 minutes per session; and 3) Free from cardiovascular, metabolic, or neurological disorders that would increase health risks. This study was approved by the local university research ethics committee.

Procedures

Participants reported to the Human Performance Laboratory for one visit. Upon arrival, participants reviewed and signed an informed consent and medical history questionnaire. After consent, participants had height, to the nearest 0.1 cm, and weight, to the nearest 0.1 kg, taken. Participants then performed an incremental maximal cycling test while blood lactate concentrations ([La]), heart rate (HR), and electromyographical (EMG) signals were measured. Prior to testing, participants were familiarized with all equipment and procedures used in the investigation. All testing was performed on a cycle ergometer (Monark 484 E; Monark©;Dalarna,

Sweden). Participants were provided an initial warm-up on the cycle ergometer at an output of 40 W for a period of three minutes. The testing phase began at a work output of 80 W and increased in increments of 40 W every 3 minutes thereafter. Cadence was kept at a pedaling rate of 80 (± 5) revolutions per minute. Test termination criteria consisted of: 1) Participant was no longer able to maintain cadence; or 2) Participant fatigue.

During each stage the following variables were measured: blood lactate, ratings of perceived exertion, heart rate, oxygen consumption (VO_2), and EMG activity. Capillary blood samples (25 µL) were taken via finger prick and analyzed the last 45 seconds of every stage throughout the testing procedures, including rest and warm-up. Lactate was analyzed using a single-blood-droplet device (Lactate Plus, NOVA Biomedical, Waltham, MA). This lactate monitor has been shown to have a standard error of the estimate of 0.6 $mmol \cdot L^{-1}$ and correlation of 0.94 when compared to criterion laboratory measures [Tanner et al., 2010].

A heart rate monitor (Polar Electro Oy, Kemple, Finland) was also placed on the participants' chest in order to accurately assess heart rate during the test. A metabolic cart (ParvoMedics Inc., Sandy, UT, USA) was used to determine peak oxygen uptake (VO_{2peak}). Before each test, calibration of the metabolic cart, according to the manufacturer's instructions, was performed. VO_{2peak} was recorded as the average oxygen consumption, expressed as $ml.kg^{-1.}min^{-1}$, the last minute of the GXT. Once the LT examination was completed, participants were given a 15-minute rest period before completing a 30-min cycling bout. This bout was performed on a cycle ergometer at the power output level one stage (i.e., 40 W) below the individual's LT at a sustained cadence of 80 revolutions per minute. This level of intensity was chosen to prevent any increase of blood [La] above LT during the exercise bout. During the cycling bout, blood [La] was assessed every 5 minutes; and EMG collected continuously.

EMG signals were collected with an electronic signal acquisition system, (MP-150 Physiograph, BIOPAC System, Inc., Goleta, CA). Disposable surface electrodes (Ag/AgCl) for the vastus lateralis were placed on the right side of the body 2/3 the distance between the anterior superior iliac spine and the lateral portion of the patella, spaced 2 cm apart and in the direction of the muscle fibers. A ground electrode was placed on the right anterior superior iliac spine. In order to reduce impedance of the EMG signal, skin preparation included shaving, abrasion, and alcohol cleansing.

Recordings below an impedance of 5 kΩ were deemed viable. Data were analyzed using software (Acqknowledge 4.2 BIOPAC System, Inc., Goleta, CA). All EMG signals were converted from analog to digital and raw myoelectrical signals were transformed using root mean square (RMS) calculations. All EMG signals were averaged with 10-second RMS windows consistent with previous research [Candotti et al., 2008, Green et al., 2014; Mahmutovic et al., 2016]. A bandpass filter of 20- to 400-Hz cutoff frequency, fourth-order Butterworth filter, and sampling rates of 2 kHz were used to filter the signal. The EMG signal was averaged at 10-second windows throughout the maximal exercise tests and plotted to determine the EMG_T using the procedures explained below. For the 30-minute exercise bout, the EMG signals acquired was averaged, using RMS transformation, at one-minute epochs surrounding the time point at which blood [La] is measured (i.e., 30 seconds pre- and 30 post-blood draws).

LT and EMG_T Determination (Dmax Method)

In order to determine LT and EMG_T, Dmax calculations were utilized. Researchers used a 3rd order polynomial curve, for both EMG and La, and determined Dmax to be the point that

provided the greatest distance to the binomial regression line formed by the lowest and highest [La] and EMG values taken during the incremental test. The method of Dmax was previously validated to provide reliable thresholds during incremental exercise [Zhou et al., 1997]. This method of assessment is able to provide a more objective measure of the LT and EMG_T as compared to more subjective, fixed [La] models (e.g., 4.0 $mmol \cdot L^{-1}$ threshold) and visual inspection. Additionally, Dmax allows for each individual lactate value for every subject to be equally considered for LT determination. The Dmax point was computed on a custom-written computer program. Additionally, percent of VO_{2peak} ($\%VO_{2peak}$) and percent maximal heart rate ($\%HR_{max}$) were recorded at both the point of LT and EMG_T to allow for comparisons. The LT and EMG_T at baseline and following the steady state exercise bout were labeled as LT-Pre and EMG_T-Pre, respectively, and LT-Post and EMG_T-Post, respectively

Statistical Analysis

The data were analyzed using SPSS version 22.0. Due to the staged, ordinal nature of the incremental test (i.e., each stage differing by 40 Watts), the power output at the threshold levels were compared using the Wilcoxon Matched-Pairs Signed-Rank Test. A repeated measures analysis of variance (ANOVA) was performed to determine if differences existed between the pre- and post-exercise thresholds when expressed as $\%VO_{2peak}$ and $\%HR_{max}$. Cohen's *d* statistic [Cohen, 1992] was calculated as the effect size of the differences in $\%VO_{2peak}$ and $\%HR_{max}$. Hopkin's scale of magnitude [Hopkins et al., 2009] was used where an effect size of 0-0.2 was considered trivial, 0.2-0.6 was small, 0.6-1.2 was moderate, 1.2-2.0 was large, >2.0 was very large. Pearson-product moment correlations were used to determine the relationships between the LT and EMG_T in $\%VO_{2peak}$ and $\%HR_{max}$. Intraclass correlation coefficients (ICC) were calculated to determine the relationship between pre- versus post-exercise LT and between pre-

versus post-exercise EMG_T when expressed as $\%VO_{2peak}$ and $\%HR_{max}$. ICC values less than 0.5 were determined to be poor, values between 0.5 and 0.75 were moderate, between 0.75 and 0.9 were good, and greater than 0.9 were deemed excellent [Hopkins, 2002]. The work stage at which LT and EMG_T occurred was noted and corresponding cases were counted to provide individual agreement for each measure comparison. Statistical significance was set at $\alpha = 0.05$.

RESULTS

Individual data, means (± SD), p-values, and effect sizes for all threshold values, expressed in $\%VO_{2peak}$ and $\%HR_{max}$ are shown in Table 3.1 and Table 3.2, respectively. The mean pre- and post- 30 minute exercise GXT VO_{2peak} in males were determined to be 34.9 ± 9.6 $ml \cdot kg^{-1} \cdot min^{-1}$ and 32.5 ± 10.3 $ml \cdot kg^{-1} \cdot min^{-1}$, respectively; while female values were 31.9 ± 2.4 $ml \cdot kg^{-1} \cdot min^{-1}$ (pre) and 30.8 ± 3.3 $ml \cdot kg^{-1} \cdot min^{-1}$ (post). Mean HR_{max} pre- and post-30 minute exercise GXT for males were 182 ± 12 beats per minute and 177 ± 15 beats per minute, respectively; while females were 178 ± 10 beats per minute (pre) and 175 ± 15 beats per minute (post). EMG thresholds were seen in 10 of 11 participants examined. Thus, the statistical comparisons represent a sample size of 10. For the 30-minute steady state bout, all means (± SD) for each 5 minute assessment ([La], EMG, $\%VO_{2peak}$, $\%HR_{max}$, and respiratory exchange ratio (RER)) are displayed in Table 3.3. The only significant mean difference was seen in the last 5-minute period of blood [La] sampling (i.e., minute 30) compared to all other previous time points. When comparing the pre- (33.3 ± 6.5 $ml \cdot kg^{-1} \cdot min^{-1}$) versus post- (31.6 ± 6.9 $ml \cdot kg^{-1} \cdot min^{-1}$) mean VO_{2peak} values, a significant difference was found ($p = 0.016$, Cohen's $d = 0.26$); whereas no significant differences existed in HR_{max} during the two incremental tests (HR_{max}-Pre = 179.9 ± 10.6 $beats \cdot min^{-1}$, HR_{max}-Post = 176.1 ± 14.1 $beats \cdot min^{-1}$, $p = 0.3$, Cohen's $d = 0.3$). Time to

fatigue was demonstrated to be significantly different between the two incremental cycling tests (GXT at Pre = 14.2 ± 4.15 min, GXT at Post = 9.7 ± 4.0 min, $p < 0.001$, Cohen's $d = 1.11$).

LT: Pre- Versus Post-Steady-State-Exercise

Resting [La] values prior to the pre- (1.55 ± 0.49 $mmol \cdot L^{-1}$) and post- (3.44 ± 2.04 $mmol \cdot L^{-1}$) GXTs were found to be significantly different ($p = 0.09$, Cohen's $d = 1.27$). No significant mean differences were seen in [La] values at the LT during the pre- (3.56 ± 0.84$mmol \cdot L^{-1}$) versus post- (3.50 ±1.99 $mmol \cdot L^{-1}$) GXTs.

When LT-Pre (128 ± 45 Watts) and LT-Post (104 ± 43 Watts) were expressed as Watts, there was a significant difference ($Z = -2.12$, $p = 0.03$). LT-Pre and LT-Post occurred during the same stage of the GXT in 5 out of the 10 participants (50%). LT-Post occurred one stage below LT-Pre in 4 participants, and two stages below LT-Pre in 1 individual. There were no significant mean differences between LT-Pre compared to LT-Post when represented as $\%VO_{2peak}$ ($p = 0.19$; Cohen's $d = 0.75$) (Table 3.1), with ICC procedures showing poor reliability (ICC = -0.36; 95% CI = -3.32 to 0.637; $p = 0.69$). When LT was expressed as $\%HR_{max}$, there were no significant mean differences seen between the pre- (81± 8%HR_{max}) and post-exercise (82 ± 4 %HR_{max}) values ($p = 0.74$, Cohen's $d = 0.17$) but the ICC procedures resulted poor reliability (ICC = -0.74; 95% CI = -13.64 to 0.62; $p = 0.76$).

EMG_T: Pre- Versus Post-Steady-State-Exercise

No significant mean differences existed in work rates, expressed in watts, pre- (140 ± 28 Watts) and post- (136 ± 47 Watts) exercise bout threshold level when determined by EMG ($p = 0.76$; Cohen's $d = 0.1$). EMG_T-Pre and EMG_T-Post occurred during the same stage of the GXT

in 6 out of 10 participants (60%). Out of the remaining participants (i.e., 4), EMG_T-Post took place at one stage higher in 2 individuals and one stage lower in the remaining 2 individuals. There was no significant mean difference in relation to for EMG_T-Pre when compared to EMG_T-Post when expressed as $\%VO_{2peak}$ ($p = 0.13$; Cohen's $d = 0.37$) with good reliability (ICC = 0.84, 95% CI = 0.38 to 0.96, $p = 0.004$). However, a significant difference existed between the pre- (74 ± 11 %) and post-EMG_T values when expressed as $\%HR_{max}$ (83 ± 7%) (p = 0.04, Cohen's d = 0.87), with good reliability (ICC was 0.85, 95% CI = 0.44 to 0.96, $p = 0.004$).

Comparison between LT and EMG_T: Pre-Steady-State Exercise

When LT-Pre and EMG_T-Pre were expressed as Watts, no significant mean difference existed (LT-Pre = 128 ± 45 Watts, EMG_T-Pre = 140 ± 28 Watts, Z = -0.791, $p = 0.429$). LT-Pre and EMG_T-Pre occurred at the same stage in 3 individuals (30%). EMG_T-Pre took place one stage higher than LT-Pre in 5 participants (50%) and one stage lower in 2 individuals (20%). When the threshold were expressed as $\%VO_{2peak}$, no mean differences were observed between LT-Pre and EMG_T-Pre ($p = 0.46$; Cohen's $d = 0.40$) (Table 3.1). However, Pearson product-moment correlations showed that the relationship between the pre-exercise threshold values to be non-significant ($r = -0.32$; $p = 0.36$). The Bland Altman procedures suggested that EMG_T-Pre compared to LT-Pre displayed 95% limits of agreement that ranged ± 26.05 $\%VO_{2peak}$ around a constant error of -3.28% (Figure 3.1A), with the upper and lower limits at 29.3 and -22.7 $\%VO_{2peak}$, respectively. Furthermore, there was no significant trend in the errors suggesting no proportional difference existed ($r = -0.31$, $p = 0.39$, Figure 3.1).

When the LT and EMG_T at pre-exercise were expressed as $\%HR_{max}$, no significant differences existed ($p = 0.25$; Cohen's $d = 0.48$) (Table 3.2), yet the Pearson-product moment

correlation between the two threshold values was non-significant (r = 0.23; p = 0.52). The Bland Altman procedures suggested that the $\%HR_{max}$ expressed EMG_T-Pre, compared to the LT-Pre, displayed 95% limits of agreement that ranged ± 20 $\%HR_{max}$ around a constant error of -4 $\%HR_{max}$ (Figure 3.2A), with the upper and lower limits at 24 and -16 $\%HR_{max}$, respectively. Furthermore, there was no significant trend in the errors suggesting no proportional difference existed (r = -0.04, p = 0.92).

Comparison between LT and EMG_T: Post-Steady-State Exercise

When LT-Post (104 ± 43 Watts) and EMG_T-Post (136 ± 47 Watts) were expressed in Watts, a significant difference was observed (Z = -2.71, p = 0.01). LT-Post and EMG_T-Post occurred at the same stage in 2 individuals (20%). EMG_T-Post took place one stage higher than LT-Post in the remaining 8 participants (80%). When thresholds were expressed as $\%VO_{2peak}$ a significant mean difference existed LT-Post and EMG_T-Post (p = 0.004; Cohen's d = 0.71) (Table 3.1); however, Pearson product-moment correlations showed the relationship between the post-exercise threshold values to be non-significant (r = 0.02; p = 0.95). The Bland Altman procedures suggested that EMG_T-Post displayed 95% limits of agreement that ranged ± 18 $\%VO_{2peak}$ around a constant error of -11 $\%VO_{2peak}$ (Figure 3.1B), with upper and lower limits of 29 and -7 $\%VO_{2peak}$, respectively. Additionally, a significant trend was present in the errors suggesting proportional bias (r = 0.79, p = 0.01). This indicated that EMG_T-Post had a tendency to overpredict LT, expressed as $\%VO_{2peak}$, after a bout of exercise. When the LT and EMG_T post-exercise were expressed as $\%HR_{max}$, no significant mean differences existed (p = 0.11; Cohen's d = 0.69) (Table 3.2), yet the Pearson-product moment correlation between the two threshold values was non-significant (r = 0.35; p = 0.32). The Bland Altman procedures

suggested that the $\%HR_{max}$ expressed EMG_T-Post, compared to the LT-Post, displayed 95% limits of agreement that ranged ± 17 $\%HR_{max}$ around a constant error of -5 $\%HR_{max}$ (Figure 3.2A), with the upper and lower limits at 22 and -12 $\%HR_{max}$, respectively. Furthermore, there was a significant trend in the errors suggesting proportional difference ($r = 0.72$, $p = 0.02$).

Table 3.1. *Individual and group mean ± SD values among the participants for $\%VO_{2peak}$ values at EMG_T and LT (n =10)*

Participant	**EMG_T_Pre** *($\%VO_{2peak}$)*	**LT-PRE** *($\%VO_{2peak}$)*	**DIFF** *($\%VO_{2peak}$)*	**EMG_T_Post** *($\%VO_{2peak}$)*	**LT-POST** *($\%VO_{2peak}$)*	**DIFF** *($\%VO_{2peak}$)*	**ΔEMG_T** *($\%VO_{2peak}$)*	**ΔLT** *($\%VO_{2peak}$)*	**DIFF** *($\%VO_{2peak}$)*
1	65	63	2	79	64	15	-14	-1	-13
2	81	73	8	86	68	18	-5	5	-10
3	64	74	-10	63	63	0	1	11	-10
4	67	62	5	64	68	-4	3	-6	9
5	78	51	27	85	65	20	-7	-14	7
6	84	62	22	82	65	17	2	-3	5
7	73	73	0	74	64	10	-1	9	-10
8	66	80	-14	65	64	0	1	16	-15
9	72	70	2	80	57	23	-8	13	-21
10	72	82	-10	76	62	14	-4	20	-24
Mean	72	69	3	75	64	11	-3	5	-8
SD	7	9	13	9	3	9	6	11	11
95% CI	14	18	26	18	6	18	12	22	22
p	0.46			0.004			0.06		
Cohen's *d*	0.40			0.71			0.90		

Abbreviation: EMG_T = electromyographical threshold; LT = lactate threshold; DIFF = difference in $\%VO_{2peak}$ between EMG_T and LT;
Δ = Change in $\%VO_{2peak}$ from Pre- to Post-30 minutes of aerobic exercise

Table 3.2. *Individual and group mean ± SD values among the participants for $\%HR_{max}$ at EMG_T and LT (n = 10)*

Participant	**EMG_T_Pre** *($\%HR_{max}$)*	**LT-PRE** *($\%HR_{max}$)*	**DIFF** *($\%HR_{max}$)*	**EMG_T_Post** *($\%HR_{max}$)*	**LT-POST** *($\%HR_{max}$)*	**DIFF** *($\%HR_{max}$)*	**ΔEMG_T** *($\%HR_{max}$)*	**ΔLT** *($\%HR_{max}$)*	**DIFF** *($\%HR_{max}$)*
1	69	70	-1	66	80	-14	3	-10	13
2	94	81	13	91	81	10	3	0	3
3	84	84	0	82	82	0	2	2	0
4	76	73	3	92	92	0	-16	-19	3
5	91	67	24	93	80	13	-2	-13	11
6	93	80	13	91	82	9	2	-2	4
7	90	90	0	93	82	11	-3	8	-11
8	81	87	-6	78	78	0	3	9	-6
9	88	84	4	95	82	13	-7	2	-9
10	82	92	-10	86	79	7	-4	13	-17
Mean	85	81	4	87	82	5	-2	-1	0
SD	8	8	10	9	4	8	6	10	10
95% CI	16	16	20	18	8	16	12	20	20
p	0.25			0.11			0.76		
Cohen's *d*	0.48			0.69			0.11		

Abbreviation: EMG_T = electromyographical threshold; LT = lactate threshold; DIFF = difference in $\%HR_{max}$ between EMG_T and LT;
Δ = Change in $\%HR_{max}$ from Pre- to Post-30 minutes of aerobic exercise

Table 3.3. *Changes across 30-minute Steady State Aerobic Exercise Bout (n=10)*

	Mean ± SD				
	[La]	**EMG**	**%VO_{2peak}**	**%HR_{max}**	**RER**
5 min	5.1 ± 2.2	0.1262 ± 0.31	61 ± 9	80 ± 4	0.87 ± 0.09
10 min	4.6 ± 2.3	0.1245 ± 0.30	58 ± 8	81 ± 6	0.89 ± 0.07
15 min	5.1 ± 2.9	0.1237 ± 0.31	61 ± 10	80 ± 7	0.89 ± 0.06
20 min	4.4 ± 2.6	0.1155 ± 0.29	56 ± 11	79 ± 7	0.87 ± 0.05
25 min	4.0 ± 2.5	0.1159 ± 0.29	59 ± 9	80 ± 9	0.87 ± 0.06
30 min	3.4 ± 2.3*	0.1115 ± 0.27	57 ± 10	81 ± 9	0.86 ± 0.04

Abbreviation: [La] = blood lactate concentration; EMG = electromyography of vastus lateralis;
$\%VO_{2peak}$ = percentage of maximal oxygen consumption; $\%HR_{max}$ = percentage of maximal heart rate;
RER = respiratory exchange ratio
*Significantly lower than all other time periods ($p < 0.05$)

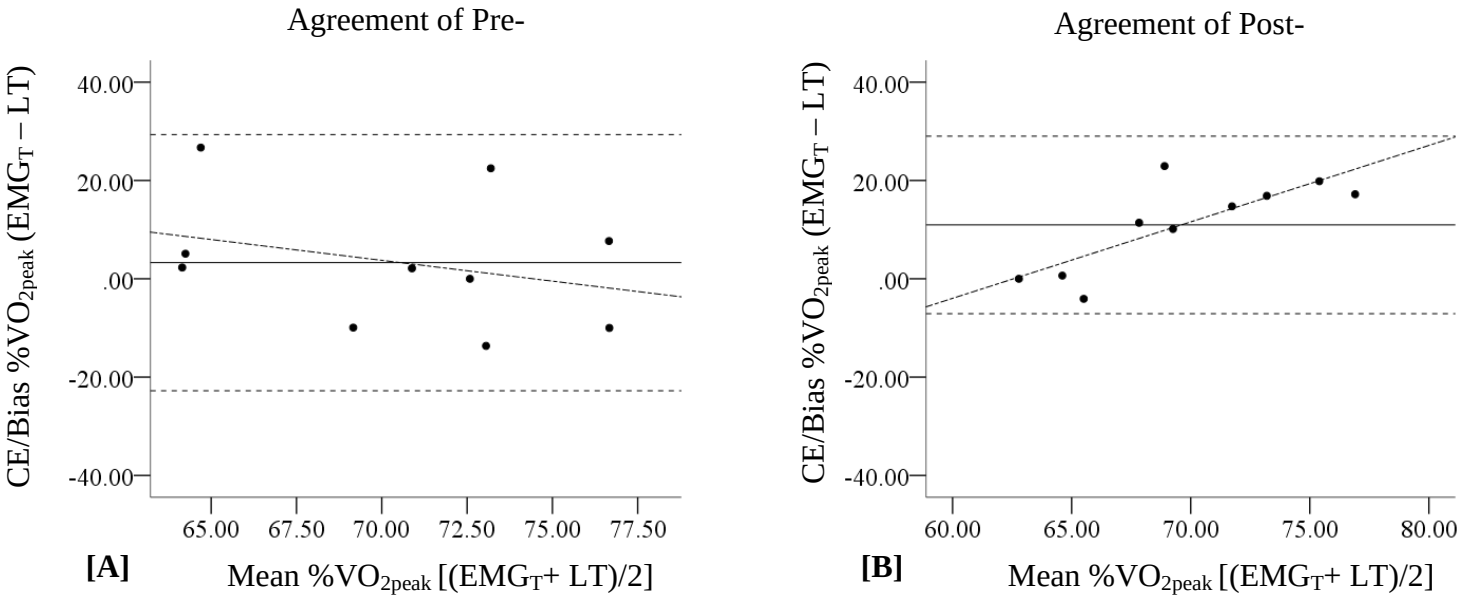

Figure 3.1. Bland and Altman plot comparing **[A]** EMG_T-Pre 30 min of aerobic exercise and LT-Pre $\%VO_{2peak}$; and **[B]** EMG_T-Post 30 min of aerobic exercise and LT-Post $\%VO_{2peak}$. The middle solid lines indicate the constant error or mean differences. The dashed lines represent the upper and lower limits of agreement (± 1.96 SD). The dashed-dotted regression lines represent the trend between the errors of each measurement when bias is present.

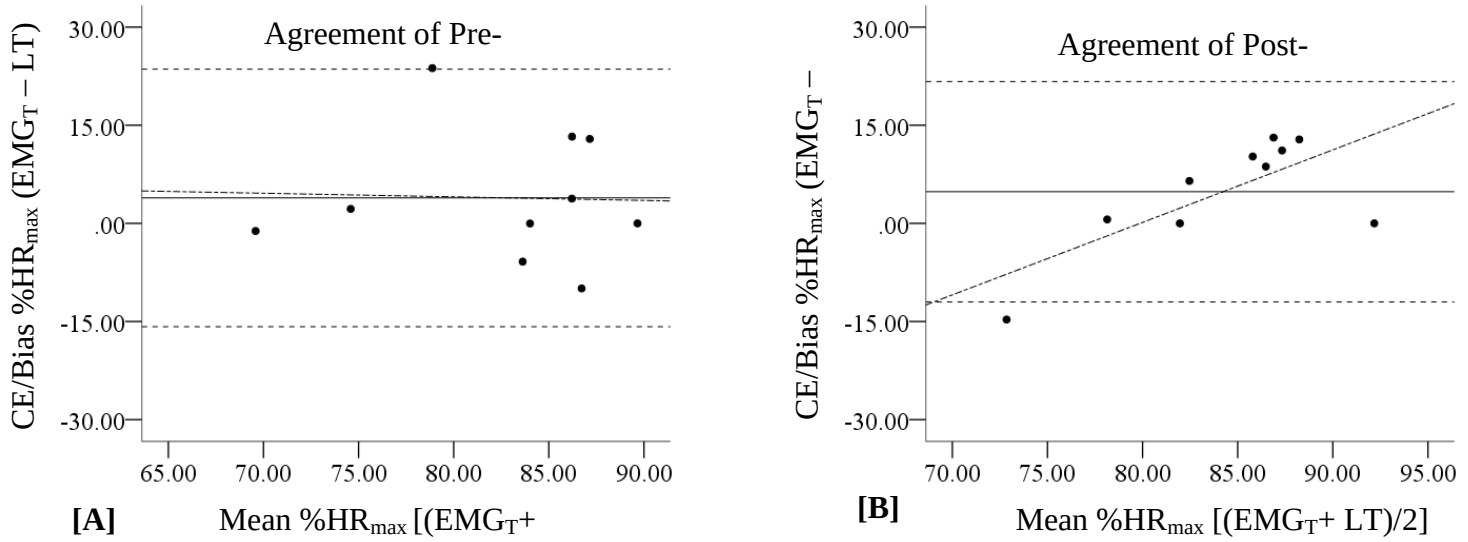

Figure 3.2. Bland and Altman plot comparing **[A]** EMG_T-Pre 30 min of aerobic exercise and LT-Pre $\%HR_{max}$; and **[B]** EMG_T-Post 30 min of aerobic exercise and LT-Post $\%HR_{max}$. The middle solid lines indicate the constant error or mean differences. The dashed lines represent the upper and lower limits of agreement (± 1.96 SD). The dashed-dotted regression lines represent the trend between the errors of each measurement when bias is present.

DISCUSSION

Because of the relationship between the recruitment of type II muscle fibers and blood lactate accumulation, the EMG_T has been suggested as a non-invasive estimate of the LT. Research is needed to examine the agreement between the EMG_T and the LT at baseline and after a steady state bout of aerobic exercise where a temporal disassociation between lactate concentration and muscle fiber type selection may occur, hence the purpose of the current investigation. The results showed that the LT and EMG_T occurred between the range of 64% to 80% of peak work rate, which is consistent with previous research [Demello et al., 1987; Farina, Ferguson et al., 2007; Green et al., 2014; Seburn et al., 1992]. However, the results indicated that LT-Pre and LT-Post were significantly different depending on the specific metric for work rate. For instance, the LT occurred at a lower power output during the post-GXT, yet there were no mean differences between the LT-Pre and LT-Post when expressed as $\%VO_{2peak}$ and $\%HR_{max}$. The ICC procedures suggested poor (ICC = -0.74 and -0.36, respectively) reliability, indicating differing patterns of individual variance between the LTs of the pre- versus post-GXT. The results between EMG_T-Pre and EMG_T-Post indicated a stronger ICC indicating a better degree of reliability when expressed as $\%VO_{2peak}$ and $\%HR_{max}$. Additionally, no significant mean differences were found when the pre- and post-EMG_T was represented as Watts or $\%VO_{2peak}$. When comparing the EMG_T and LT at the pre-steady-state-exercise time-points, no significant mean differences were found between the two thresholds when represented as any of the three work rate values. However, when comparing the post-steady-state-exercise time-points, the LT provided significantly lower work rate values expressed as Watts and $\%VO_{2peak}$. Furthermore, the Pearson-product correlation procedures showed no significant relationships between EMG_T and LT at either pre- or post-time points. The Bland-Altman procedures also displayed a wide

range of individual differences between the two threshold markers. In addition, a proportional difference was found in the post-steady-state values, indicating that the EMG_T provided a significant trend to over-estimate the LT with higher threshold values.

Furthermore, the individual agreement between pre- and post-GXT indicated that the LT occurred during the same stage in 5 out of the 10 participants; while EMG_T occurred in the same stage for 6 of the 10 individuals. Post-GXT LT took place one stage below pre-GXT in 4 of the remaining participants and two stages below in one participant. Whereas, post-GXT EMG_T occurred in one stage above pre-GXT EMG_T in 2 individuals and one stage below in the remaining two participants. Additionally, individual agreement demonstrated that pre-GXT LT and EMG_T occurred at the same stage in 3 individuals (30%). The pre-GXT EMG_T took place one stage higher than LT in 5 participants (50%) and one stage lower in 2 individuals (20%). For the post-GXT individual agreement, LT and EMG_T was observed at the same work rate for only 2 individuals (20%) with EMG_T occurring one stage higher than LT in the remaining 8 individuals. Collectively, the results of the study suggest that poor individual agreement existed between EMG_T and LT before and after 30 minutes of steady state aerobic exercise. Further, it appears that the EMG_T was a more reliable measure across the 30-minute steady state bout of exercise than the LT.

The study also demonstrated that observed VO_{2peak} in the post-GXT was significantly lower compared to pre-GXT. Because the sample examined consisted of untrained cyclists, the 30-minute steady state protocol may have induced volitional fatigue at a lower work rate in the post-GXT leading to a reduction in pedaling cadence (criteria for termination).The investigators observed consistent RPE values (data not provided) between the last stages of the GXTs despite VO_{2peak} being significantly lower during the 2nd test. Therefore, the decreased VO_{2peak} in the post-

GXT may have been due to the subjects perceiving a maximal-level of exertion at lower oxygen consumption. Furthermore, because LT occurred at a lower work rate (when evaluated as Watts) following 30 minutes of steady state exercise, anaerobic metabolism may have predominated earlier during the 2nd GXT. An earlier onset of predominating anaerobic metabolism may have also lead to a higher heart rate at a given oxygen cost [Powers, 2012], which may explain the lower VO_{2peak} in post-GXT compared to pre-GXT, despite no differences in HR_{max}.

EMG_T can be defined as the greatest exercise intensity an individual can maintain without an exponential increase in the amplitude of the EMG signal [MahmutoviĆ et al., 2016]. Previously, the EMG response to a GXT has been considered to be related to the response in blood lactate, hence the purported validity of EMG_T serving as a less invasive measure than LT. Whereas previous research in the relationship between [La] and EMG exhibited a relationship that may be construed as cause-effect [Candotti et al., 2008], an alternative approach may be indicative of a revolving cascade of events leading to both increases in both [La] and EMG signal amplitude during exercise. During incrementally increasing exercise a greater reliance on anaerobic metabolism occurs, initiating a rise in blood [La] due to the breakdown of glycogen in working skeletal muscle [Powers, 2012]. A rise in La production is accompanied by an increase in H^+ ions as a by-product of anaerobic glycolysis; thereby further decreasing blood pH [Powers, 2012]. The exponential rise in [La] is associated with an increase in motor unit recruitment, which is displayed as an increase in EMG amplitude. The concurrent increase in H^+ ion concentration activates group III and IV muscle afferents which inhibits α-motor neurons and decreases signal conduction to the working muscle [Amann, 2012; Amann et al., 2015].

While the physiological response to increasing exercise intensity between [La] and EMG signaling appear to be associated, their interaction may not be cause-effect. For instance, blood

[La] has been observed to gradually decrease over time during long-term, steady state training, mainly due to a decrease in glycogen content [Dumke et al., 2006]. However, Briscoe et al. (2014) demonstrated no change in EMG amplitude during 40 minutes of steady state exercise at or below the EMG_T. The current study also demonstrated a significant reduction in [La] during the 30-minute steady-state bout; yet no significant change in EMG signal amplitude occurred. Furthermore, the post-GXT LT was significantly lower than the pre-GXT LT, yet no difference in the EMG_T was found.

To extend the latter point in the previous paragraph, the current study indicated poor individual agreement between the two threshold markers, which supports the postulation that the EMG and [La] response are not a cause-effect relationship [Mills et al., 1984]. For instance, Mills and Edwards (1984) demonstrated that EMG and motor unit recruitment were not affected by the accumulation of blood [La] and did not play a role in muscular fatigue in patients with McArdle's Syndrome [Mills et al., 1984]. It was suggested that potential increases in potassium ion concentration induce greater fatigability of muscular membrane excitation, increasing motor unit recruitment. Similarly, additional research by Juel (1988) suggested that an increase in potassium concentration, which occurs independent of an increase in [La], decreased motor unit conduction velocity thereby increasing EMG amplitude [Juel, 1988]. Furthermore, Seburn et al. (1992) demonstrated no relationship between EMG and [La] during an incremental cycling test after a bout of exercise with the intent to increase blood [La] [Seburn et al., 1992], which was similar to the current findings since elevated resting [La] levels were seen before initiating the post-GXT. Further consistent with Seburn et al. (1992), the current results showed decreases in blood [La] during increasing post-GXT work rates following a pre-elevation in [La]. The decreases in blood [La] may be attributed to work rates below the level of LT in the early stages

of the incremental test. The clearance of [La] can be optimally observed at 30-40 $\%VO_{2peak}$ [Powers, 2012], which may have been the cause of early clearance from an elevated blood [La] at the onset of the post-GXT. Previous examination of [La] recovery suggests a longer duration to return to resting levels (i.e., minutes to hours) compared to EMG amplitude signal recovery, which has been suggested to recover immediately after an exercise bout [Komi 1984; Petrofsky, 1979]. Results of the current study also showed no significant differences in work rates between EMG_T-Pre and EMG_T-Post. This leads to the speculation that EMG maintains its relationship with changes in incremental exercise and associated increases in amplitude signaling independent of pH and [La].

The disconnection between LT and EMG_T does not implicitly imply that each of these markers is not regarded as equally important for the purposes of training considerations. The use of LT to prescribe training intensities may be useful and can based upon the metabolic demands of the given task (e.g., aerobic versus anaerobic); whereas EMG_T may have a greater application in the monitoring of motor unit recruitment and muscle fiber type utilization. Further research is needed to determine the specific importance of each threshold marker in regards to exercise prescription.

This study is not without limitations. One such limitation was that the population examined was limited to untrained cyclists between the ages of 18 and 40 years. Trained endurance athletes may have demonstrated a larger decrease in [La] removal during the 30-minute steady-state bout as compared to an untrained population [Barak et al., 2011; Martin et al., 1998]. This larger difference in clearance rate may have decreased [La] closer to resting values; thereby affecting the relationship between LT and EMG_T during the post-GXT. Nevertheless, the purpose of this study was to examine the agreement between the two

thresholds pre- and post-exercise in recreationally trained individuals. Second, the protocol used for the GXT used a 40 Watt increase between stages to determine the LT and EMG_T. Though the protocol has been used in previous investigations [Candotti et al., 2008], a smaller incremental increase in work rate (e.g., 20 Watts) may be a more practical method of LT and EMG_T determination in untrained participants as large incremental increases may have led to quicker fatigue in participants during the post-exercise GXT that are unaccustomed to higher intensity work rates. Additionally, the current study was performed upon a cycle ergometer, the results thus may not be extrapolated to different modalities of cardiovascular training as EMG patterns and [La] may differ during various GXT protocols. Therefore, future research should examine the agreement of EMG_T and LT within various modalities (e.g., running, arm ergometry, rowing).

PRACTICAL APPLICATIONS

Monitoring blood [La] levels has importance in endurance training and is typically performed in a laboratory setting. The capacity to accurately determine LT in the field is an important tool for research as well as prescribing training intensity, monitoring changes in performance, or examining physiological adaptations as a result of LT training. The use of EMG has been recently shown to provide a non-invasive way of capturing work rates and thresholds comparable to LT testing and may provide a reliable field metric for practitioners and athletes. The results of this investigation determined that EMG_T accurately assessed LT, when expressed as Watts, $\%VO_{2peak}$, and $\%HR_{max}$. However, after a bout of steady-state exercise, a disassociation was observed between LT and EMG_T, displaying significant differences in both Watts and $\%VO_{2peak}$. The results also indicated poor reliability in the ability to repeat LT after a bout of

cycling. These findings indicate that the recovery of the EMG signal may occur immediately and thus repeatable after training or day-to-day; while blood [La] may require additional recovery time to repeat LT testing. Practitioners should take note that both while LT and EMG_T testing may display similar properties during maximal exercise testing, they are not interchangeable. Therefore, further research is needed to determine the ability of these two metrics (LT and EMG_T) to prescribe training intensities and various applications in the field.

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CHAPTER 4

COMPARSION OF ELECTROMYOGRAPHICAL SIGNAL ANALYSES FOR ESTIMATING LACTATE THRESHOLD

ABSTRACT

Currently, no published research exists comparing filtering methods of electromyographical (EMG) analyses to estimate lactate threshold (LT). The purpose of this investigation was to evaluate and compare EMG transformations and time windows to predict LT. Participants (n=14) completed an incremental, maximal exercise test on a cycle ergometer until exhaustion. Blood lactate was measured every minute, while electromyography was recorded continuously at the vastus lateralis. EMG signaling was then transformed and filtered using two time-segment windows (i.e., 10 and 60 seconds), as well as three signal conversions (i.e., root mean square, smoothing, and peak amplitude averaging). Results indicated no mean differences between the EMG thresholds, for any of the filtering methods, when compared to the LT criterion. Moderate correlations were seen when comparing each of the EMG filters to LT ranging from 0.69 – 0.79. Root mean square and Smoothing filters accurately indicated LT in 10 out of 14 participants; whereas peak amplitude averaging indicated LT for 11 out of 14 participants. EMG may be a useful tool to estimate the work rate associated with LT. Averaging EMG over a minute of time and continual 10-second recordings demonstrate comparable readings and allow an easier application of EMG threshold in the field.

Key Words: Aerobic Training, Blood Lactate, Threshold Training

INTRODUCTION

Continual changes in blood lactate concentrations ([La]) during incremental exercise often make accurate analysis of lactate threshold (LT) challenging in laboratory and field settings. Most lactate analyses require laboratory equipment, trained personnel, as well as drawing and handling of blood. Additionally, various methods are utilized to determine a precise, individualized LT, particularly with the use of mathematical algorithms (e.g., D_{max}, log-log transformations, regression modeling, etc.) [de Sousa et al., 2011; Stanula et al., 2013]. As a result, simpler methods of prediction have been used to estimate [La] during incremental aerobic activity (e.g., electromyography (EMG)). By monitoring muscle activity via EMG, practitioners may be able to pinpoint the transition between slow-twitch (i.e., aerobic) and fast-twitch (i.e., anaerobic) muscle fiber recruitment. This transition is stated to be the EMG threshold (EMG_T). The relationship between LT and EMG_T has been previously investigated and has demonstrated no significant difference in work output [Candotti et al., 2008]. Reliability of the EMG signal to produce a similar threshold on two separate occasions has also been demonstrated [MahmutoviĆ, et al., 2016]; thereby increasing the value of a non-invasive field tool to provide thresholds which practitioners may use to prescribe training intensities.

The averaging of time windows (i.e., segments) has been used as a method of determining the EMG_T [Candotti et al., 2008; Guffey et al., 2012]. For instance, Candotti et al. (2008) used a sampling average of 1-second RMS values to determine the correlation coefficient between EMG and blood [La] during incremental cycling exercise. Results indicated a positive, significant association ($r = 0.83$) [Candotti et al., 2008]. Ten-second averages of the EMG during incremental testing have also been used to pinpoint the EMG_T with similar results ($r = 0.73$ –

0.83) [Jürimäe et al., 2007] further demonstrating a practical use of an EMG signal to provide indication of the shift of aerobic to anaerobic metabolism during physical activity.

Despite the non-invasive nature of EMG, understanding raw signaling transformations is an important step in monitoring muscular activation. Raw EMG signaling presents an unprocessed amplification of the summation of motor unit action potentials as they reach the skin. Additionally, raw signaling oscillates between the positive and negative poles of the electrodes, which prohibit segment averaging as the positive and negative amplitudes would cancel out and equal zero. Therefore, signal filtering and transformation of raw EMG is needed to provide a valid interpretation. Common methods of filtering are root mean square (RMS), integral averaging (Smoothing), and mean peak (maximal amplitude averaging).

Previous investigation into the prediction of the LT via the EMG_T is limited. However, several studies have examined the various methods by which to process the raw EMG signals (e.g., D_{max}, log-log, 3^{rd} order polynomials), but no study has yet to determine the most appropriate filtering method (i.e., root mean square, smoothing, mean peak) or compared time-window segments (i.e., 10- or 60-second averaging) of the EMG signals to estimate blood [La] activity during incremental exercise. Thus, the purpose of this investigation was to evaluate and compare various EMG transformations and time window segments to predict LT activity. It was hypothesized that maximal amplitude averages would have a stronger association with the estimation of blood [La] versus root mean square or smoothing signal filtering. Additionally it was hypothesized that no differences would exist between 10- or 60-second window averaging segments of the EMG recording.

METHODS

Experimental Approach to the Problem

A single-visit, multiple measurement was utilized to determine which filtering and averaging method of the raw surface electromyography signal would best predict lactate threshold during incremental exercise. Fourteen participants completed a maximal incremental exercise test during one visit to the lab. During the trial, blood lactate was sampled every minute via finger prick, while surface electromyography was continuously recorded from the vastus lateralis. The raw electromyography signal was transformed using three filtering and averaging methods (i.e., root mean square, smoothing, and mean peak). Each filtered signal was averaged over 10 and 60 seconds throughout the entire test and used to predict lactate threshold via inflection point calculations (i.e., Dmax).

Participants

Fourteen males (n = 9; ages = 23.67 ± 5.55 years; height = 175.71 ± 4.16 cm; weight = 85.61 ± 10.05 kg) and females (n = 5; ages = 20.80 ± 1.30; height = 163.12 ± 8.44; weight = 57.76 ± 8.25 kg) volunteered to participate in this study. A power analysis determined that approximately 11 participants would be needed to obtain statistical power at the Beta=0.80 level. Participants recruited met the following criteria: 1) Between the ages of 18-40; 2) Currently participating in regular moderate-to-vigorous aerobic training of a minimum of three days per week for at least 30 minutes per session; and 3) Free from cardiovascular, metabolic, or neurological disorders that would otherwise affect the results or negatively impact safety. This study was approved by the university ethics committee.

Procedures

Participants were asked to report to the Human Performance Laboratory for one visit. Upon arrival, participants completed and signed an informed consent and medical history questionnaire. After consent, participants had height and weight measurements taken. Standing height was measured to the nearest 0.1cm using a stadiometer (SECA 67310, SECA©, Chino, CA); while weight was measured to the nearest 0.1 kg (Tanita BWB-800, Tanita©, Arlington Heights, IL). All eligible participants performed an incremental maximal cycling test while blood lactate, heart rate, and electromyographical signals were measured. Prior to testing, participants were familiarized with all equipment and procedures used in the investigation. All testing was performed on a cycle ergometer (Monark 484 E; Monark©; Dalarna, Sweden). For the cycling protocol, participants began with an initial three-minute warm-up on the cycle ergometer at an output of 40 Watts (W). Following the warm-up the testing phase began at a work output of 80 W and increased in increments of 40 W every 3 minutes thereafter. Cadence was kept at 80 (±5) revolutions per minute. Test termination criteria were determined to be: 1) Participant was no longer able to maintain cadence; or 2) Volitional fatigue.

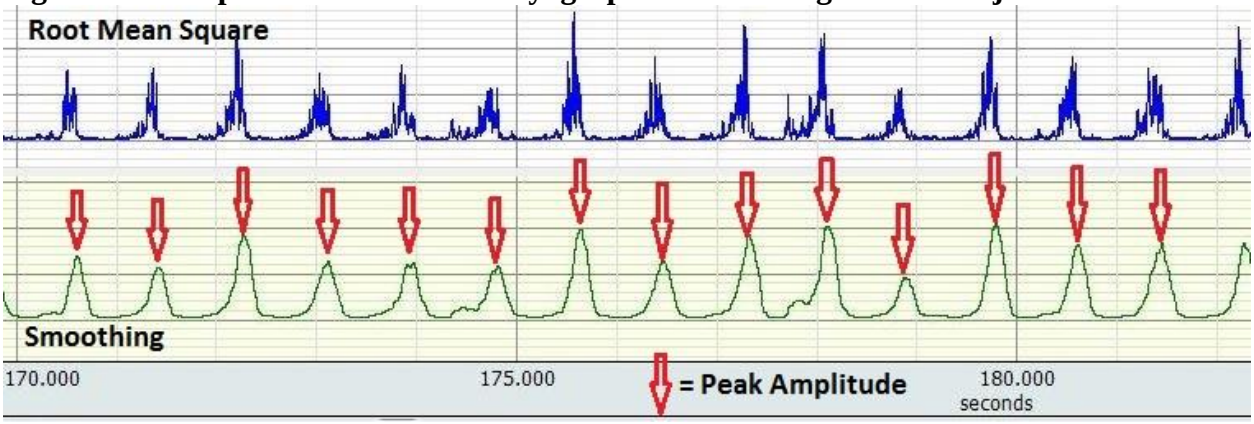
During each stage, the following variables were measured: blood lactate, heart rate, VO_2, and EMG activity. Capillary blood samples (25 µL) were taken via finger prick and analyzed the last 45 seconds of each stage throughout the testing procedures, including rest and warm-up. Lactate was analyzed via the Lactate Plus (NOVA Biomedical, Waltham, MA). The Lactate Plus monitor has been demonstrated to have a standard error of the estimate of 0.6 $mmol \cdot L^{-1}$with a 0.94 correlation when compared to laboratory measures [Tanner et al., 2010]. A heart rate monitor (Polar Electro Oy, Kemple, Finland) was also placed on the participant's chest to assess heart rate during the test. A metabolic cart (TrueOne® 2400, ParvoMedics Inc., Sandy, UT,

USA) was used to determine oxygen consumption. The metabolic cart was properly calibrated according to the manufacturer's instructions before each test. Peak oxygen consumption (VO_{2peak}) was recorded as the average oxygen consumption, expressed as ml.kg-1.min^{-1}, among the last minute of the incremental exercise test.

Surface EMG signals were collected with an electronic signal acquisition system, (MP-150 Physiograph, BIOPAC System, Inc., Goleta, CA). Disposable surface electrodes (Ag/AgCl) for were placed on the right vastus lateralis 2/3 the distance between the anterior superior iliac spine and the lateral portion of the patella, spaced 2 cm apart and in the direction of the muscle fibers. A ground electrode was placed on the right anterior superior iliac spine. Preparation for the skin sites included shaving, abrasion, and alcohol cleansing in order to reduce impedance of EMG signals. Recordings were deemed viable when impedance test measurement was below 5 kΩ.

Data were analyzed using software (Acknowledge 4.2 BIOPAC System, Inc., Goleta, CA). Raw myoelectrical signals were quantified by using the root mean square (RMS) as well as processed using a moving average of 300 samples per second (i.e., Smoothing) and maximal amplitude averaging. Prior to smoothing and PK averaging, the raw signal was rectified to provide the absolute values of the sample. Sampling rates of 2 kHz were used to record EMG along with a 20- to 400-Hz cutoff frequency bandpass filter and a fourth-order Butterworth filter. All signals were converted from analog to digital. All filtering methods of the EMG signal were average over 60-second epochs and compared to the criterion of 10-second RMS (RMS-10). A sample 10-second electromyographical recording during a portion of the maximal exercise test, for one subject, is provided in Figure 1.

Figure 4.1. Sample 10-second electromyographical recording for one subject



LT and EMG_T Determination (Dmax Method)

LT and EMG_T were determined in each participant via Dmax calculations. Dmax was defined as the point of maximal distance from a 3rd order polynomial equation, utilizing the [La] and EMG curves, to the line formed by the lowest and highest [La] and EMG values taken during the incremental test. The Dmax method has been validated as a reliable computational method to provide training thresholds during incremental exercise testing [Zhou et al., 1997]. The Dmax model of LT assessment is able to provide a more individualized approach and identification; whereas fixed concentration models (e.g., visual inspection, 4.0 $mmol \cdot L^{-1}$ threshold, etc.) may be subjective in nature and prone to bias. Additionally, Dmax allows for each individual lactate value for every participant to be equally considered for LT determination. The Dmax point was computed on a custom-written computer program. The percent of VO_{2peak} ($\%VO_{2peak}$) and percent maximal heart rate ($\%HR_{max}$) were recorded at all threshold levels to allow for comparisons.

Statistical Analysis

Statistical analysis was performed using a software package (SPSS Version 22.0; SPSS Inc., Chicago, IL). Due to the staged, ordinal nature of the incremental test, the various methods of EMG filtering (RMS-10, RMS-60, Smoothing, and PK) were compared to the criterion (i.e., LT) using the Wilcoxon Matched-Pairs Signed-Rank Test. Spearman's rank order correlation was performed to determine the relationship between LT and each of the various EMG filtering methods. The work stage at which LT and EMG_T occurred for each filtering and time epoch method was noted and corresponding cases were counted to provide individual agreement for each measure comparison.

RESULTS

LT and EMG thresholds were observed in all participants and used in data analysis. The Wilcoxon Signed-Rank test demonstrated no differences among RMS-10 (Z = -0.816, p = 0.41), RMS-60 (Z = -1.0, p = 0.32), Smoothing (Z = -1.0, p = 0.32), or PK (Z = -0.577, p = 0.56) when compared to the threshold criterion using blood [La]. Spearman's rho determined significant correlations between LT and RMS-10 (r_s = 0.69, p= 0.006), RMS-60 (r_s= 0.79, p = 0.001), Smoothing (r_s = 0.79, p = 0.001), and PK (r_s = 0.78, p = 0.001). Additionally, the results indicated that LT was accurately determined by RMS-10, RMS-60, and Smoothing for 10 out of 14 participants (i.e., 71 %); whereas PK predicted LT in 11 out of the 14 (i.e., 79%) participants. For the 4 participants that were not accurately assessed by RMS-10, RMS-60, and Smoothing, EMG underestimated LT in 3 individuals and over-estimated in 1 individual. In terms of PK, results indicated an underestimation in 2 individuals and 1 over-estimate in the ability of EMG to predict LT.

DISCUSSION

The purpose of this investigation was to evaluate practical filtering methods of the raw EMG signal in order to best estimate lactate threshold during an incremental exercise bout. The study also tested the difference between a 10-second average of the EMG signal versus 60 seconds. No significant differences were seen for any of the EMG filtering and time averaging methods when compared to LT. The results of this investigation supported previous work establishing EMG as a viable means of predicting LT during incremental exercise [Candotti et al., 2008; de Sousa et al., 2011; MahmutoviĆ et al., 2016]. However, differences existed between the EMG measures to estimate the stage at which LT occurred. PK predicted LT the greatest amount of times (79%) compared to RMS-10, RMS-60, and Smoothing, which predicted LT 10 out of 14 times (71%). Each of the EMG filtering methods over-estimated LT in the same participant by one stage; however, RMS-10, RMS-60, and Smoothing under-estimated LT by one stage in three individuals compared to two underestimations by PK.

Mahmutovic et al. (2016) demonstrated that the EMG_T was reproducible after a period of 48 hours using EMG averaged at 10-second RMS epochs during the end of each stage throughout a maximal aerobic test. Researchers found an excellent reliability with an ICC of 0.85 [MahmutoviĆ et al., 2016]. While Dmax was not used for signal processing, a common method of linear regression was performed, which found no difference in work rate during an incremental cycling test [de Sousa et al., 2011]. Additionally, Zuniga et al. (2013) demonstrated no significant differences, using the V-slope mathematical procedure, when examining anaerobic thresholds using 20-second epoch RMS EMG signaling compared to ventilatory and lactate thresholds. It was observed that EMG amplitude increased during times when the ability to produce force or power during an incremental treadmill test was reduced [Zuniga et al., 2013].

Whereas EMG was able to estimate the threshold level of lactate in the current study; previous results have seen inconsistent outcomes. The differences seen within studies demonstrating the ability or inability of EMG to estimate the aerobic/anaerobic transition may reside within the computational methods used to analyze the EMG signal. For instance, Seburn et al., (1992) observed a linear response to an incremental cycle exam while recording integrated EMG averaged for 60-seconds increments throughout the test [Seburn et al., 1992]. This may be possibly explained by the use of well-trained cyclists and smaller incremental increases (i.e., 23.5 watts) between stages as compared to the current study using untrained cyclists and a 40-watt increase for each stage. By utilizing a smaller increment between stages and well trained cyclists, the increase in motor unit recruitment to produce changes in the amplitude of the EMG signal may have not been sufficient to produce an exponential relationship. Taylor and Bronks (1994) also demonstrated significant differences between LT and EMG_T while using integrated EMG using an incremental treadmill test. However, the analysis used to determine EMG_T was a visual inspection of researcher-determined regression lines to determine a breakpoint in the signal [Taylor et al., 1994]. Visual inspection of the breakpoints in either LT or EMG_T would result in a more subjective determination, may exclude smaller breakpoints in the data for analysis, and be more prone to bias.

In terms of the filtering methods used within the current study, RMS provided an additional filtering option of the raw EMG signal through a process involving the initial squaring all values, summing those squares, dividing by the number of observations in the signal, followed by the square root to obtain the reported values [Criswell et al., 2011]. This is the most common method of EMG filtering, due to its transition of the signal from analog to digital and its propensity to provide less distortion of the signal [Criswell et al., 2011].

Integral averaging, or Smoothing, is often referred to as a running average of the EMG signal. For instance, if the smoothing filter is running at 50 samples per second, the first 50 points of raw data are averaged and then plotted. This filtering method reduces the variability of the signal by decreasing the probability of spontaneous artifact or variability in the recording. The current study utilized a running average of 300 samples per second. Using a longer averaged segment window diminishes the variance in amplitude estimation as compared to RMS, which is simply a digital conversion of the raw signal [Clancy et al., 2001]. However, the results showed no significant mean differences between RMS and Smoothing filters when estimating LT work rate. This signifies that with an overall estimation of a theoretical threshold level either filter may be used; however for more precise measurements of single dynamic movements or with the use of biofeedback a closer examination of the signal may be necessary.

Traditionally, there are challenges present while working with EMG recording: 1) the stochastic nature of the EMG amplitude; 2) the inability of EMG to directly represent muscular tension or force; and 3) "cross talk". First, during times of low intensity and rest, the variation in EMG signal amplitude and frequency is minimal; however during progressively increasing workloads and fatigue large variations in motor unit and muscle fiber type recruitments exist creating unpredictability in the EMG signal. This is often characterized by a decrease in frequency and subsequent increase in amplitude [Dimitrova et al., 2003]. While EMG is not a viable source to directly measure force of the muscle contraction, the rise in EMG is typically associated with times of progressively intensive exercise is the recruitment of Type II muscle fibers. This breakpoint in EMG is often stated to be the time at which a large increase in the recruitment of motor units occurs [Candotti et al., 2008; Zuniga et al., 2010]. This threshold can mistakenly be indicated as the initial recruitment in Type II fibers; however, Type II fibers may

contribute to early onset of muscular contraction, but not show signs of fatigue during this time. Therefore, no indication of a breakpoint may exist in the amplitude or frequency of the EMG recording. Third, "cross talk" between muscle groups within surface electromyography is inevitable, but may be reduced utilizing proper electrode placement and smaller electrode spacing (e.g., 2 cm). Since amplitude is a function of time and is defined as the standard deviation of the EMG signal between muscle fibers, any additional variation in the signal from another muscle group may alter the amplitude [Clancy et al., 2001]. While "cross talk" cannot be eliminated, any possible cross-over may affect EMG amplitude and in this case contribute to the exponential rise in the processed EMG.

Furthermore, Briscoe et al. (2014), using linear regression, determined that the amplitude of the EMG signal was more important than measuring frequency in order to determine EMG_T. Researchers also determined the importance of prescribing training intensities based upon muscle activation patterns as participants were asked to perform exercise bouts at 70%, 100%, and 130% of EMG_T. It was determined that individuals were able to pedal at both 70% and 100% for extended durations (i.e., >40 minutes); however, participants were unable to maintain the 130% intensity for a period greater than 12 minutes [Briscoe et al., 2014]. This represents the importance of amplitude based averaging when assessing EMG.

The use of surface EMG to estimate the work rate of LT has been shown to be a valid method; however, limitations may exist with the practically of this method in a laboratory or field-based setting. Surface EMG is affected by many factors that may distort or result in recordings that cannot be examined or interpreted properly. One such major limiting factor is weak conductors and insulators that may reduce the strength or block surface EMG signaling (i.e., skin and adipose tissue). Therefore, a key determinant of signal strength will vary

depending on the subcutaneous fat thickness of the individual being examined (i.e., a less subcutaneous fat will result in less signal impedance and vice versa).

The current study was performed on a cycle ergometer and may not extrapolate to incremental treadmill testing. Dynamic variations exist in the patterns recording via EMG through the vastus lateralis and may provide a difference when estimating LT. Lastly, the current study did not use trained cyclists for the study sample. Stanula et al. (2013) suggested that athletes who devoted a greater portion of their training time to aerobic endurance showed greater reliability between various mathematical methods of LT analysis (e.g., 4mmol·L^{-1}, log-log, D_{max}, etc.) compared to anaerobic athletes. It may be that trained cyclists' would yield more consistent results than our sample.

PRACTICAL APPLICATIONS

The use of EMG to predict [La] in the field is a convenient tool for practitioners due to its non-invasive nature as opposed to blood sampling. Additionally, when utilizing EMG to monitor intensity levels, signal transformation from raw capturing is a key component in understanding muscle activation and motor unit recruitment. All tested filtering methods, including RMS, smoothing, and peak amplitude averaging calculations provided valid filter options to monitor muscular activation to estimate blood [La] threshold during cycling. In particular, endurance and aerobic athletes may benefit from the ability to monitor threshold work rates established by EMG in order to properly prescribe training intensities.

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CHAPTER 5

CONCLUSION

Previous studies of the relationship between blood lactate and electromyography have demonstrated inconsistent results. Additionally, wearable EMG technology has yet to be examined to determine its' ability to provide a valid estimation of LT. This series of studies answered useful research questions by providing insight on the use of commercially-available EMG compression gear, the effect of aerobic exercise on LT and EMG_T testing, as well as, the various methods by which EMG is filtered and averaged to provide a more practical approach to monitoring EMG in the field.

The first study compared LT with EMG threshold of the vastus lateralis monitored with wearable compression shorts. No significant differences in work rate existed between LT and EMG_T during a bout of maximal incremental cycling. Additionally, no differences existed in $\%VO_{2max}$ or $\%HR_{max}$ at the work output level associated with LT and EMG_T. There were also a strong correlation between power output between [La] and EMG ($r_s = 0.68$, $p = 0.01$). EMG_T occurred at the same stage of the incremental test as LT in 11 of the 13 participants. These findings suggest that wearable EMG technology may be a viable field tool for the use of monitoring and predicting LT levels. Practitioners may find this information useful to prescribe training intensities for athletes to increase cardiovascular and muscular endurance.

The second study examined the agreement between LT and EMG_T after a bout of steady-state aerobic exercise. Consistent with previous results, the relationship between LT and EMG_T pre-exercise bout was found to have no significant difference in terms of power output. Post-

exercise there was a significant difference in work rate LT and EMG_T (Z = -2.71, p = 0.007). However, individual agreement demonstrated that pre- and post-exercise LT occurred at the same work rate in 5 of 10 participants; while pre- and post- EMG_T occurred at the same work rate in 6 of 10 participants. Additionally, LT and EMG_T demonstrated poor individual agreement (i.e., pre-exercise = 30% versus post-exercise = 20%); further demonstrating that EMG and [La] may not be a causal-effect relationship as previously suggested. Thus, future research is needed to determine the application of each metric for training and exercise prescription purposes.

The third study inspected various filtering methods of the EMG signal and determined their ability to monitor LT during an incremental exercise test. The results demonstrated no significant differences in the various methods used to transform the raw EMG signal (i.e., root mean square, smoothing, and peak amplitude averaging). Furthermore, no differences existed in the work rate or $\%VO_{2max}$ established at the threshold level for both lactate and EMG. Thus, the three popular transformation filtering methods of the raw EMG signal were each able to predict the work rate level associated with the laboratory method of lactate threshold testing.

The collective finds of this dissertation support the use of EMG as a potential field tool to monitor and predict LT training levels in a rested state. Furthermore, wearable EMG technology may be an inexpensive, non-invasive method for the testing of athletes and individuals as opposed to laboratory measures. However, future studies might seek to determine the validity and reliability of EMG fatigue thresholds for the use of setting training intensities.

APPENDIX

Office for Research

Institutional Review Board for the Protection of Human Subjects

THE UNIVERSITY OF ALABAMA RESEARCH

November 24, 2015

Ronald Snarr
Department of Kinesiology
College of Education
The University of Alabama
Box 870312

Re: IRB Protocol # 15-008-ME
"Estimation of the Lactate Threshold via Cycling While Wearing EMG Shorts"

Mr. Snarr:

The University of Alabama Medical Institutional Review Board has reviewed the revision to your previously approved full board protocol. The board has approved the change in your protocol.

Please remember that your approval period expires one year from the date of your original approval, May 14, 2015, not the date of this revision approval.

Should you need to submit any further correspondence regarding this proposal, please include the assigned IRB application number. Changes in this study cannot be initiated without IRB approval, except when necessary to eliminate apparent immediate hazards to participants.

Good luck with your research.

J. Grier Stewart, MD, FACP
Medical IRB Chair

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