

Neuromuscular Alterations in Middle-Distance Runners

**Prior Band-Resisted Squat Jumps Improved Running Time, Rating of Perceived Exertion,  
and Neuromuscular Performance in Middle-Distance Runners**

by

© Jonathan LeRon Low

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**ABSTRACT**

There is a need for more research that examines the time course of neuromuscular alterations that occur during middle-long distance running. A growing body of evidence suggests that post-activation potentiation (PAP) is a prominent neuromuscular alteration that aids in the enhancement and maintenance of force production. PAP conditioning contractions integrated into warm-up protocols have been shown to enhance subsequent performance, yet the role PAP plays in endurance performance remains under-studied. The aim of this study was to characterize the time course of the effects of a PAP conditioning stimulus (band-resisted jump squat protocol) on a subsequent 5 X 1 km running trial. This study examined neuromuscular properties (ITT, MVC, EMG, drop jump) and metabolic properties (RPE, HR). It was hypothesized that performing a 5RM band-resisted jump squat protocol as part of a standardized running-specific warm-up in a group of endurance runners would induce significant measurable PAP effects during the course of a subsequent 5 X 1 km time trial run and up to 10 minutes post-run protocol. The neuromuscular and performance changes resulted in decreased time to complete the running task (3.6%) in the intervention session, increased force generation (9.5%) throughout both trials, increased voluntary activation (10%) in the intervention session, and a lack of impaired evoked contractile properties. These results serve as evidence of measurable neuromuscular changes occurring during and after the subsequent running trial. It is plausible that the band-resisted jump squat protocol served to increase performance and physiological measures and is attributable to post-activation potentiation, heightened central-pacing strategies, and increased stretch-shortening cycle efficiency.

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**TABLE OF CONTENTS**

ABSTRACT..... ii

ACKNOWLEDGEMENTS..... iii

TABLE OF CONTENTS..... iv

LIST OF TABLES..... vi

LIST OF FIGURES..... vii

LIST OF ABBREVIATIONS..... viii

Chapter 1: Review of Literature..... 1

    1.1 Introduction..... 1

        1.1.2 Understanding Performance during Endurance Events..... 2

    1.2 Post-activation Potentiation (PAP)..... 4

        1.2.1 PAP Mechanisms..... 5

        1.2.2 Measuring Post-activation Potentiation..... 9

        1.2.3 Fatigue and Post-activation Potentiation..... 10

    1.3 PAP in Sport Performance..... 12

    1.4 PAP in Endurance Athletes..... 14

    1.5 PAP Conditioning Stimuli- Pre-Performance..... 16

    1.6 PAP During and After Endurance Performance..... 18

        1.6.1 Maximal vs. submaximal intensities..... 19

        1.6.2 Performance duration..... 20

        1.6.3 Field testing- jumps..... 21

        1.6.4 Neuromuscular adaptations during and after performance..... 22

    1.7 Justification of Testing Measures..... 24

    1.8 Conclusion..... 26

Chapter 2: Co-Authorship Statement..... 28

Chapter 3: Manuscript..... 29

    3.1 Abstract..... 30

    3.2 Introduction..... 32

    3.3 Methodology..... 35

    3.4 Results..... 46

    3.5 Discussion..... 58

    3.6 Perspectives..... 66

    3.7 Conclusion..... 67

3.8 References.....	69
3.9 Tables.....	80
3.10 Figures.....	82
3.11 Appendix.....	89

**LIST OF TABLES**

Table 3.1 Intra-session descriptive and reliability measures .....	80
Table 3.2 Evidences for PAP arising from within condition effects .....	81
Table 3.3 Evidences for PAP arising from between condition effects .....	81

**LIST OF FIGURES**

Figure 3.1 Estimated changes in mean triceps surae MVC force ..... 47

Figure 3.2 Comparison of triceps surae F100 between control and squat condition ..... 48

Figure 3.3 Graph of changes in resting time to peak twitch force ..... 49

Figure 3.4 Overall changes seen in percent voluntary activation ..... 51

Figure 3.5 Comparison of drop jump height over time and condition ..... 53

Figure 3.6 Comparison of changes in reactive strength index ..... 54

Figure 3.7 Total time to complete 5 kilometers compared between conditions ..... 55

Figure 3.8 Graph demonstrating uniformity in heart rate between conditions ..... 56

Figure 3.9 Graph demonstrating uniformity of rate of perceived exertion ..... 57

Figure 3.10 Experimental set-up: resistance bands ..... 82

Figure 3.11 Experimental set-up: 30 cm drop jump ..... 83

Figure 3.12 Experimental set-up: plantar flexors MVC ..... 84

Figure 3.13 Experimental set-up: electrode placement ..... 85

Figure 3.14 Experimental set-up: treadmill ..... 86

Figure 3.15 Experimental set-up:  $\dot{V}O_{2\max}$  ..... 87

Figure 3.16 Experimental set-up: running specific warm-up ..... 88

**LIST OF ABBREVIATIONS**

5RM	Five Repetition Maximum
ANOVA	Analysis of Variance
cm	Centimeters
EMG	Electromyography
ES	Effect Size
F100	Force Produced in the First 100 Milliseconds of an MVC
Hz	Hertz
HR	Heart Rate
ICC	Intraclass Correlation Coefficients
iEMG	Integrated EMG
ITT	Interpolated Twitch Technique
kg	Kilogram
km	Kilometer
μs	Microseconds
ms	Millisecond
mA	Milliamp
mV	Millivolt
MVC	Maximum Voluntary Contraction
MVIC	Maximum Voluntary Isometric Contraction
PAP	Post-activation Potentiation
PT	Twitch Torque Amplitude
R	Range
RLC	Regulatory Light Chain
RPE	Rate of Perceived Exertion
RSI	Reactive Strength Index
s	Seconds
SD	Standard Deviation
SSC	Stretch Shortening Cycle
TPT	Time to Peak Twitch Force
VA%	Percent Voluntary Activation
VO <sub>2</sub> max	Maximum Amount of Utilized Oxygen During Exercise



## Chapter 1: Review of Literature

### 1.1 Introduction

While the industrialized world has adopted a largely sedentary lifestyle, ‘fun runs’ and competitive road races have become increasingly popular in recent years (Running USA, 2017). With so many participating in the sport of middle-distance running, there is an increased need for research in this area. The current body of research has examined middle-distance running as a multifactorial concept that represents the influence of various metabolic, cardiorespiratory, biomechanical, and neuromuscular characteristics with varying opinions on their role in limiting performance (Barnes et al. 2015). The physiology of endurance running has, for some time, been studied with a focus on the metabolic system as the limiting factor for performance. Though it is well established that physiological properties such as maximal oxygen uptake, utilization of maximum aerobic power, running economy, and blood lactate levels are factors that impact endurance performance (Costill et al. 1973; Davies et al. 1979; Foster et al. 1978; Hill et al. 1925; Joyner et al. 1991), these properties cannot solely explain differences in endurance performance (Nummela et al. 2006).

Although our knowledge of the metabolic mechanisms governing endurance exercise has greatly increased, there is a need for further investigation into the limiting or enhancing effects that neuromuscular alterations have on endurance performance. Recent evidence suggests that factors relating to neuromuscular alterations in exercise (muscle recruitment and force production) play an important role in the performance of middle-distance runners (Jung et al. 2003; Paavolainen et al. 1999; Nummela et al. 2006; 2008). These studies examined how neuromuscular alterations can be enhanced with no significant changes in metabolic characteristics; demonstrating that neuromuscular alterations could be a factor for limiting or

enhancing endurance performance. Of the research that has examined neuromuscular alterations in endurance athletes (Finni et al. 2003; Girard et al. 2012; Millet et al. 2002; 2004; 2011; Nicol et al. 1991; Nummela et al. 2006; Nummela et al. 2008; Perrey et al. 2010; Ross et al. 2010; Skof et al. 2006), the majority of experiments have compared differences pre- and post-exercise with few focused on the progression (time course) of the alterations during exercise (Ross et al. 2010; Millet et al. 2004). Despite the body of past research, the specific limiting/enhancing effects of the time course of neuromuscular alterations in an endurance population have remained largely unknown. There is a need to examine the role and time course that neuromuscular alterations play in limiting or enhancing endurance running performance. Through an examination of the available literature, this review will serve to outline the gaps and future perspectives in relation to neuromuscular performance in endurance athletes. To do this, the review will first examine the basis of a phenomenon called post-activation potentiation (PAP) as a central factor in enhancing contractile performance. A discussion about the mechanisms and modes of measuring PAP will follow. Subsequently, a brief discussion of muscle fatigue and its relation to PAP will be presented before finally exploring the research themes and trends in PAP in the endurance athlete phenotype.

### **1.1.2 Understanding Performance during Endurance Events**

Performance during an endurance running event can be described by the physiological responses leading to the balance of muscle fatigue and force maintenance. Endurance running is characterized mainly by repeated stretch-shortening cycle properties and requirements for continuous effort.

Prolonged exercise (i.e. endurance running) is thought to reduce the capacity of the neuromuscular system to produce force via Renshaw cell inhibition, autogenic or recurrent

feedback (Golgi tendon organs), and type III and IV (i.e. nociceptor) afferent inhibition and decreased cortical excitability (Behm 2004). All of these factors can contribute to neuromuscular fatigue (Ross et al. 2010). Conversely, the mechanisms behind the maintenance and enhancement of force production, under fatiguing conditions, are thought to include increased motor unit recruitment, changes in rate coding, catch-like properties, alterations of motor-control, and post-activation potentiation (Behm, 2004). These mechanisms of force enhancement are especially applicable, as middle-distance running requires the maintenance of force for long durations, usually under fatiguing conditions. This is evident as endurance training has been demonstrated to promote faster recovery in neuromuscular performance (Hakkinen et al. 1990). Furthermore, enhanced running-specific neuromuscular performance has been found following fatiguing running exercises (Boullosa et al. 2011; Del Rosso et al. 2016; Rassier et al. 2000; Vuorimaa et al. 2006) with the suggested mechanism being post-activation potentiation.

A main aspect to consider in relation to neuromuscular performance during a prolonged running event is the ability to produce sufficient force in order to maintain velocity against the onset of muscle fatigue. Muscle fatigue can be defined differently depending on the task. Fatigue relating to high intensity or maximal contractions has been defined as a transient decrease in working capacity (Asmussen et al. 1978) or a decline in the force generating capacity of the muscle (Degens et al. 1994). In the case of endurance running, where the body is subject to prolonged contractions of submaximal intensity, fatigue can be defined as an increase in the perceived effort needed to exert a desired force and an eventual inability to produce this force (Enoka et al. 1992). The capacity to minimize muscle fatigue and maximize muscle force is critical to successful outcomes in middle-distance running performance. Contradicting results

have been reported on how the neuromuscular system achieves a balance of muscle fatigue against force production (Berg et al. 2003; Morio et al. 2016) with many attributing this balance to a phenomenon called post-activation potentiation (Boullosa et al. 2011; Doma et al. 2016; Latorre-Roman et al. 2014; Mettler et al. 2011; Sale 2004). It is the time course of this neuromuscular enhancement of force production (PAP) during running that is the focus of this study. The objective of this experiment was to examine the time course of neuromuscular alterations that could be elicited due to a post-activation potentiation (PAP) conditioning stimulus. It was hypothesized that a conditioning stimulus performed prior to a 5 x 1 kilometer running trial would elicit post-activation potentiation and will improve subsequent performance during a running trial.

### **1.2 Post-activation Potentiation (PAP)**

Post activation potentiation (PAP) is a phenomenon explained by an enhancement of muscle force output and rate of force development as a result of muscle contractile history (Robbins, 2005). This phenomenon is highly applicable to sports, and research has increasingly focused on methods for prompting a post-activation potentiation as a form of performance enhancement.

Over a hundred years ago, the idea of force potentiation was first described in the literature (Lee 1907). This idea of muscle force enhancement was furthered in the 1930's when Brown and Euler (1938) used the gastrocnemius, soleus, and tibialis anterior muscles from cats to demonstrate the enhancement of twitch tension from a prior tetanic contraction. Building on this idea, Perrie et al. (1973) used assays and electrophoresis with skeletal muscle from rabbits to examine the chemical changes leading to force enhancement. Perrie was one of the first to suggest that force enhancement could be attributed to the phosphorylation of the myosin

regulatory light chains (RLC). Since the publication of this fundamental research, many studies have reported the presence of PAP in skeletal muscle in both humans and mammals (Gossen et al. 2000; Gourgoulis et al. 2003; Grange et al. 1995; Grange et al. 1998; Gullich et al. 1996; Hamada et al. 2000; Hamada et al. 2003; Manning et al. 1982; Moore et al. 1984; Stuart et al. 1988; Vandenboom et al. 1993; Vandervoort et al. 1983; Szczesna et al. 2002).

### **1.2.1 PAP Mechanisms**

#### ***1.2.1a Phosphorylation of regulatory light chains.***

The current literature suggests that the most important mechanism for PAP is the phosphorylation of the myosin regulatory light chain, which leads to an increase in the  $\text{Ca}^{2+}$  sensitivity of the myofilaments (Baudry et al. 2007; Chui et al. 2003; Gossen et al. 2000; Hamada et al. 2000; Hodgson et al. 2005; MacIntosh, 2003; Sale, 2004). Subsequent research has supported the claim that RLC phosphorylation is associated with muscle force potentiation (Barany et al., 1979; Grange et al. 1993). The idea of RLC-induced muscle potentiation is currently believed to be the main mechanism in explaining PAP (Hodgson et al. 2005). Work over the past 20 years has greatly expanded our knowledge of RLC phosphorylation and is heavily based on the pioneering work done by Sweeney et al. (1986). In his research, Sweeney pioneered the idea of RLC phosphorylation by investigating the functional, mechanical, and biochemical features of this phenomenon in the ventricular and psoas tissue of rabbits.

On a molecular level, the adaptations of the regulatory light chain that occur at the myofilament of striated muscle consequently enhance the force production for a given  $\text{Ca}^{2+}$  concentration (MacIntosh, 2010). Attached to the end of the myosin molecule heavy chains within the myofilament are two myosin heads. Each myosin head contains a regulatory light chain that has a specific binding site for a phosphate molecule. When calcium molecules are

released from the sarcoplasmic reticulum during a muscle contraction, an enzyme known as myosin light chain kinase is released, which catalyzes the phosphorylation of the RLC's (Tillin et al. 2009). The phosphorylation of the RLC is believed to potentiate the following contractions by changing the structure of the myosin head, causing it to move away from its thick filament backbone (Hodgson et al. 2005; Szczesna et al. 2003). A study by Szczesna et al. (2002) demonstrated that RLC phosphorylation also makes the actin-myosin interaction more sensitive to myoplasmic calcium. A published review (Vandenboom et al. 2013) demonstrates this interaction and provides evidence of myosin phosphorylation and force potentiation in skeletal muscle of animal models. Though the RLC mechanism is the most widely accepted rationale for explaining PAP, in the past, its validity has been questioned with suggestions that other factors may provide a more comprehensive explanation of PAP (Stuart et al. 1988).

### ***1.2.1b Fiber types.***

The culmination of the previous animal-based evidence examining PAP mechanisms would suggest that the greatest enhancement of the potentiation effect could be seen in fast-twitch fibers, at certain physiological temperatures, during concentric contractions, and at shorter muscle lengths (Sweeney et al. 1986; Vandenboom et al. 2013). This is thought as mammals seem to potentiate after stimulation-induced elevations in RLC phosphate incorporation by skMLCK (enzyme controlling phosphorylation). These reviews suggest that mammalian fast twitch fibers have a greater content of skMLCK as compared to their slow twitch counterparts. In conjunction with this idea, it is thought that the levels of MLCP (enzyme regulating dephosphorylation) are highest in slow twitch muscle fibers as opposed to fast twitch fibers. This would suggest optimal potentiating effects in fast twitch fibers of mammals. Caution should be taken applying the conclusions of these specific findings to humans. Parallel to the evidence

regarding RLC phosphorylation in fast twitch fibers, there are two studies examining RLC phosphorylation using muscle biopsies in humans that suggest that similar levels of RLC phosphorylation exist in both fast and slow twitch muscle fibers (Houston et al., 1987; Stuart et al., 1988). This corresponds with the model that Boulosa et al. (2013) presents for endurance athletes which suggests that many physiological training adaptations (PAP in both fast and slow twitch fibers) have evolved as an outcome of the varying physical requirements of our *Homo* ancestors.

Differences in fiber type distribution in humans may be a viable explanation as to why there are inconsistencies in observing PAP after conditioning contractions (Tillins et al. 2009). Due to the greater capacity of the Type II fibers for RLC phosphorylation (Grange et al. 1993; Sweeney et al. 1993), it is assumed that muscles with increased Type II proportions will have greater PAP effects (Moore et al. 1984; O'Leary et al. 1997; Vandervoot et al. 1983). Interestingly, research has demonstrated that endurance athletes typically have a higher percentage of Type I fibers in their trained muscles (Bergh et al. 1978; Costill et al. 1976; Jansson et al. 1977) thus; these athletes might be expected to be less prone to PAP. However, it has been demonstrated that endurance training increases the maximum shortening velocity of Type I fibers (Fitts et al. 1977; Schluter et al. 1994). Widrick et al (1996) has further demonstrated this concept in elite masters distance runners. In their study, Schluter et al. (1994) also associated the increased maximum shortening velocity of Type I fibers with an increased content of 'fast' myosin light chains in Type I fibers. If this is the case, then the increased 'fast' MLC Type I fibers could also offer a greater capacity for MLC phosphorylation and well-trained endurance athletes might be expected to show higher PAP effects despite a lower percentage of Type II fibers (Hamada et al. 2000).

### ***1.2.1c Pennation angle.***

Other potential neuromuscular adaptations exist as proposed mechanisms for PAP. Much of the available literature examines the acute changes in muscle pennation angle (Reardon et al., 2014; Tillin et al., 2009) as an explanation for PAP. The pennation angle of a muscle is the angle formed by the fascicles and the inner aponeurosis (muscle fibers to connective tissue) and thus has an effect on force transmission from tendons to bones. Mahlfeld et al. (2004) suggested this mechanism after seeing a significant difference in pennation angle following three 3 second maximal voluntary contractions. Using ultrasonography, they measured a change in pennation angle from 15.7° pre-intervention to 16.2° post-intervention to 14.4° ( $p < 0.05$ ) 3-6 minutes post contractions. Though this change was equated to a 0.9% increase in force, it is plausible that this effect may aid in PAP.

### ***1.2.1d Recruitment of higher order motor units.***

Increased recruitment of higher order motor units (Gullich et al. 1996; Chui et al. 2003; Hodgson et al., 2005; Tillin et al., 2009), and a diminished motor unit discharge rate (Klein et al., 2001) have also been suggested as potential mechanisms. It is known that stimulating specific afferent fibers will activate  $\alpha$ -motoneurons (afferent neural volley) (Gullich et al. 1996). These stimulations are usually conducted in the form of a tetanic isometric contraction and are known to elevate the signaling of excitation potentials across spinal synaptic junctions for several minutes post-contraction (Tillin et al. 2009). Due to this change, during subsequent activity there is an increase in post-synaptic potentials for the same pre-synaptic potential (Luscher et al. 1983). This is explained through a mechanism proposed by Luscher et al. that suggests that an induced tetanic contraction (or conditioning contraction) will decrease transmitter failure during subsequent activity. Alpha-motoneurons are activated in an all-or-none fashion, yet due to an



‘autonomously protected activation reserve’, transmitter failure at synaptic junctions is common during a reflex or normal voluntary response. Certain contractions are believed to decrease transmitter failure via an increase in the quantity of neurotransmitters and an increase in efficacy of the neurotransmitters (Enoka, 2002). A decrease in transmitter failure prompts an increase in higher order motor units which consequently activates more muscle fibers producing an increased force output.

The H-reflex is a common way to further measure higher order motoneuron recruitment. This test is recorded from the muscle fibers using electromyography and is the result of an afferent neural volley (summation of H-reflex/M-wave action potentials) after a single-pulsed nerve stimulation. Thus, an increase in the H-reflex amplitude after a conditioning contraction could be a result of PAP and explained by a diminished transmitter failure and increased higher order motoneurons recruited at the spinal cord (Gullich et al. 1996; Maffiuletti et al. 2001).

Though the literature seems conclusive as to the explanation of PAP via neuromuscular mechanisms, some have minimized the contribution from spinal and supraspinal influences (Thomas et al., 2017; Xenofondos et al., 2014). It is notable here that all of these studies examined these changes post maximal or near-maximal conditioning activities and not in prolonged activities. Furthermore, what we know about PAP has been largely influenced by currently accepted methods of measuring such responses in different settings.

### **1.2.2 Measuring Post-activation Potentiation**

In the past, the reliability and validity of measurement tools for neuromuscular alterations (PAP) has been questioned (Buckthorpe et al. 2012); however, at present there are many widely accepted research methods for accurately measuring neuromuscular alterations. The studies discussed above primarily focused on using electrical stimulation in animal models as a way of

measuring neuromuscular function. Measurement techniques now include functional measures such as voluntary contractions in jumps, isometric contractions, and ground contact time in human participants.

Despite these techniques and given the relationship between PAP and its proposed neuromuscular mechanisms, it is important to note that many suggest the presence of PAP must be validated with a proper evaluation of the enhanced response using an evoked twitch (MacIntosh et al., 2010; Mitchel et al., 2011; O’Leary et al., 1997) or examination of the Hoffman (H)-reflex (Hodgson et al., 2005). Many studies have examined the twitch contractile properties of the muscle *in vivo* and have measured PAP as the difference between a pre- and post-electrically evoked twitch stimulus (Hamada et al., 2000a; Hamada et al., 2000b; Paasuke et al., 2000). In addition, Misiaszek (2003) furthered the idea of the use of the H-reflex as a measurement tool to study the effects of contractile history on neuromuscular response. In a review examining the factors modulating PAP in explosive maneuvers, Seitz et al. (2015) challenged the assumption of measuring PAP via pre/post stimulations by suggesting that PAP could be appropriately measured using certain standardized ballistic evaluations. A discussion of the most common methods for measuring PAP in endurance athletes will be explored further in this review.

### **1.2.3 Fatigue and Post-activation Potentiation**

Further to the modes of measurement and mechanisms of PAP is the application of PAP to sport performance. As examined in many of the aforementioned studies, one of the main inhibiting factors to the development of power and speed during a sport competition is the rate at which fatigue develops (Del Rosso et al. 2016). In the case of an endurance athlete, a high rate of fatigue development would decrease speed and negatively affect performance. Muscle fatigue

can originate at different levels of the motor pathway and can be divided into central and peripheral components. Peripheral fatigue is due to changes seen distally to the neuromuscular junction while central fatigue has its origin proximal to the neuromuscular junction representing activities within the central nervous system (Gandevia et al. 2001; Wan et al. 2017). Recent evidence has shown that in addition to peripheral fatigue; reduced neural drive (central fatigue) of the contracting muscles is likely to compromise motor performance during exercise (Girard et al. 2008). Although, Perrey et al (2010), concluded that acute muscle fatigue induced by intense running exercise is mainly peripheral. Conversely, it has been argued that the etiology of muscle fatigue is complex during prolonged running exercises and that impairment can come peripherally (neuromuscular propagation and excitation-contraction coupling) and centrally (central drive/activation) (Millet et al. 2003). Thus, the body's management of fatigue (central and peripheral) development, commonly known as pacing, becomes an important factor to consider when discussing PAP. Fatigue is present in both power and endurance activities and consequently, it has been hypothesized that the body employs PAP as an intervention to increase muscle output (Grange et al., 1993). Sale (2004) argues that PAP could counteract the onset of peripheral fatigue and further suggests that, in the case of endurance athletes, PAP could have its greatest effects in overcoming submaximal fatigue.

Submaximal fatigue can be defined as the balance between fatigue-induced impairments and neuromuscular strategies to sustain performance (Behm, 2004). Further to this, Behm (2004) suggests that ongoing contractions (as is the case with endurance activities) could also facilitate motor neuron excitation. With this in mind it is important to consider that the association between phosphorylation and the RLCs could be disguised by the balance of PAP and fatigue. Because contractile activity yields both fatigue and PAP, the balance between them will

determine whether the muscle response will be enhanced, diminished, or remain the same (MacIntosh et al., 2003). And in the case of this discussion, whether speed and jump height will increase, decrease, or remain constant. Several models have been proposed, and attempt to explain fatigue amongst endurance sports (Abbiss et al., 2005). Predominantly, the dialogue for explaining fatigue in endurance athletes has revolved around centrally mediated mechanisms but recent findings (Millet, 2011) suggest that when considering the muscle strength loss during and after prolonged running exercises, it is important to note both the peripheral and central mechanisms. It is on this basis that Boullosa et al. (2011) suggest that PAP responses post-running are specific for endurance-trained athletes and could be peripheral.

### **1.3 PAP in Sport Performance**

With increasing availability of performance enhancing technology and research, the pressure for athletes to perform and stand out during competition is increasing. Coaches and fitness professionals benefit from quality scientific research that can guide them in improving athletic performance. The aim of incorporating a PAP stimulus into an athlete's training or pre-competition program is to elicit an acute enhancement in muscle performance and consequently a higher jump, faster time, or further throw. A recent review of 141 PAP articles found that the highest effect sizes for conditioning stimuli designed for eliciting PAP were found with a moderate intensity, multiple-set conditioning activity performed with trained individuals (Wilson et al. 2013). Interestingly, this same review found that the effects of post-activation potentiation have been examined in different types of athletic populations in various conditioning settings.

PAP effects have been found in athletes from a variety of sports. From American football to hand ball (Okuno et al. 2013) to baseball (Naclerio et al. 2014) and soccer (Chatzopoulos et al. 2007; Chelly et al. 2009). For example, a higher mean propulsive velocity and lower blood

lactate levels were found in a group of judoists performing two sessions of resistance training equated by volume and total resting time (Iglesias-Soler et al. 2012), suggesting that resting times during training were more prone to eliciting post-activation potentiation. This contrasts with a study that found that volleyball players performing 5RM back squats induced a measurable PAP effect for up to ten minutes post-conditioning stimuli, irrelevant of resting time (Ah Sue et al. 2016). In direct opposition to this study was one that hypothesized PAP effects, but found no effects on countermovement jumps in NCAA division I volleyball players (Beaudette et al. 2015). A study by Arias et al. (2016) employed similar conditioning stimuli to the study conducted by Ah Sue, but recruited recreationally trained individuals and did not find any PAP effects, potentially due to the difference in training status. This is interesting as PAP effects have been shown in many studies recruiting recreationally-trained active individuals (Horan et al. 2015; Hughes et al. 2016; Lowery et al. 2012; Miyamoto et al. 2011; Turner et al. 2015; Ulrich et al. 2016; Wyland et al. 2015). For example, it was found that ballistic jumps caused a substantial PAP effect in the triceps surae muscle pair in recreationally trained participants and that drop jump performance increased (Bergmann et al. 2013). PAP has also been found in a sample of rugby players completing sprint trials after a bout of 3RM back squats (Bevan et al. 2010). Furthermore, in a study measuring PAP using twitch measures, national level cyclists, field hockey, and soccer players showed PAP effects after performing three different interventions of static squats and whole-body vibrations (Cochrane et al. 2009). Similarly, studies examining speed, jumping, and agility as PAP effects post-stretching have also found convincing results (Pojskic et al. 2015; Turki et al. 2011).

Despite this evidence, it is possible that not all performance improvements following a conditioning stimulus can be explained by PAP. The simple act of performing a warm-up can

yield positive acute adaptations such as muscle temperature elevation and increased metabolic activity (McGowan et al. 2015). Thus, suggesting the importance of differentiating these factors from potentiating contractile capacity. Further to the PAP-warm up relationship, it has been argued that inducing a PAP before a competition might be more effective than a conventional warm up at enhancing subsequent performance (Gullich et al. 1996; Saez Saez de Villarreal et al. 2007), though there is contradicting evidence to this (Docherty et al. 2007). In light of these different findings, future research is needed to compare the potentiating effects of a conditioning stimulus to conventional warm-up techniques (Tillin et al. 2009). In regards to middle-distance running, conventional warm ups have been shown to have a critical effect on performance and prevention of running-related injuries (van Mechelen et al. 1993; Zourdos et al. 2017). Comparatively, an increasing body of research has demonstrated that track and field athletes have also been shown to potentiate after certain conditioning stimuli incorporated into a warm-up (Bogdanis et al. 2014; Suchomel et al. 2016; Thomas et al. 2017; Tsoukos et al. 2016). And more recently, Pageaux et al. (2017) demonstrated a limited decrease in knee extensor force of 9 trained endurance athletes running 10 km after uphill walking and running.

### **1.4 PAP in Endurance Athletes**

In recent years, researchers in human performance have taken interest in the application of PAP to performance in the endurance athlete population. As is evident from the literature, traditionally PAP has been examined and investigated in maximal power activities with fewer studies examining its effects in endurance activities.

A widely cited article by Sale (2004) suggests that PAP should have its greatest effect during submaximal contractions in which the motor units are firing at relatively low frequencies, as is the case with endurance exercise. It follows then that if the levels of fatigue in a sport or

activity are relatively lower than the levels of PAP, not only athletes performing maximal activities will benefit, but any athlete could benefit from this mechanism. Furthermore, according to Sale (2004), PAP should be maximized at lower firing rates and these lower firing rates occur during endurance activities. This is explained as sensitivity to  $\text{Ca}^{2+}$  is maximized at low  $\text{Ca}^{2+}$  levels (lower firing rates at submaximal contraction intensities) and consequently limited at saturated  $\text{Ca}^{2+}$  levels (higher firing rates at maximal contraction intensities)(MacIntosh, 2003).

The interaction of pacing (balance of fatigue) with a prior conditioning stimulus, may provide a better PAP/fatigue balance and would be expected with the increased fatigue resistant slow-twitch fibers employed in endurance trained muscles. This balance in pacing between muscle fibers was examined in a study conducted by Hvid et al. (2013). They demonstrated a maintained  $\text{Ca}^{2+}$  sensitivity in only slow-twitch fibers after prolonged endurance activities (i.e. 4 hours), furthering the idea that an enhanced PAP/fatigue balance, and thus increased PAP effects, could exist with endurance trained muscles. Mettler and Griffin (2010) revealed a significant positive correlation between force potentiation and force-time integral produced by the stimulation train, suggesting a relationship between the volume of a conditioning activity and the degree of potentiation. These same authors furthered this idea in 2011 when they conducted a longitudinal study that demonstrated that the time to achieve maximal potentiation decreased with an increased intensity (Mettler et al. 2011). Interestingly enough, both of these studies were conducted on the *adductor pollicies* human muscle, which is a predominantly slow-twitch muscle, suggesting again that for the endurance phenotype, prolonged activities at submaximal intensities would induce a greater force production capacity and possibility greater potential for PAP.

In relation to the applicability of the PAP phenomenon to endurance athletes, there are two main themes that are present in the research. These are: the effect of PAP during warm-up activities as a means to improve subsequent performance (Feros et al., 2012; Silva et al., 2014) and the evaluation of PAP responses during training sessions, or after endurance tests and competitions (Del Rosso et al., 2016; Feros et al., 2012; Millet et al., 2003; Silva et al., 2014).

### **1.5 PAP Conditioning Stimuli- Pre-Performance**

With the existing evidence that PAP enhances subsequent performance, the most plausible application of PAP to sport is its use as a warm-up technique prior to training or competition in order to acutely enhance contractile potential. Conditioning stimuli yielding a PAP could have potential for enhancing subsequent performance in endurance events (Kilduff et al. 2011). Employing the proper conditioning stimulus to elicit a PAP could give athletes in high level competition the added advantage for success. Interestingly, few studies have examined the feasibility of this application in the endurance population (Burnley et al. 2005; Carter et al. 2005; Feros et al. 2007; Jones et al. 2003; Palmer et al. 2008; Silva et al. 2014; Skof and Strojnik et al. 2007).

In a study with trained cyclists, it was suggested that adding a 5RM strength exercise bout to a 5-minute warm-up improved performance in a subsequent 20-km cycling time trial (Silva et al. 2014). This study found no significant difference in blood lactate measures ( $p > 0.05$ ), but suggested that the improved performance (~6.1% reduction in time to complete the trial) after heavy resistance was explained by improved cycling economy. Though significant changes in the time to complete the trial were not correlated to changes in power output (PAP in pacing) or pedal cadence, the mean power output in the first 10% of the time trial tended to be greater (5.8%) after the heavy-intensity strength exercises. The authors also suggest the



possibility that, in addition to the neuromuscular effects observed, an increased capacity to access aerobic performance reserves could also aid in explaining increased performance during the trial.

Palmer et al. (2008) also examined the effects of prior heavy exercise (heavy cycling for 5 minutes) in well-trained cyclists but measured performance in a subsequent 4-km cycling time trial. Athletes were subjected to either no prior exercise, prior heavy exercise, or self-selected prior exercise, all conducted on a cyclist ergometer. Similar to Silva et al., they found faster times to complete the trial post heavy exercise, but different from the Silva study was the overall increase in the mean power output (5.4%) post-heavy exercise. These authors do not necessarily attribute the observed changes to neuromuscular mechanisms but instead suggest the main mechanism as a higher use of available energy.

Likewise, a study conducted by Feros et al. (2012) recruited national-level rowers and aimed to investigate the effects of PAP on rowing performance after a typical rowing warm-up, or a potentiated warm-up. The potentiated warm-up consisted of 5x5 seconds of ramp-up isometric contractions and was shown to result in a faster 1000-m rowing time (Effect Size (ES)= 0.21: small magnitude). More convincing results were reported from the first 500-m. Time to complete the first 500-m was faster (ES= 0.64: moderate magnitude,  $p = 0.009$ ) and the mean power significantly greater (ES= 0.64,  $p = 0.007$ ) after the potentiated warm-up, suggesting a time-limited PAP effect. The individual responses of the rowers were quite varied as the study lacked a control and standardization in the warm-up. Based on these results, it would be hard to assume that PAP was present in the musculature of all athletes or, if it was only specific to some, while being overcome by fatigue in others.

PAP effects were also observed in a study examining the effects of two different warm-up protocols on the neuromuscular systems of well-trained distance runners (Skof and Strojnik et al. 2007). This study found that adding sprinting and bounding exercises to an athletes' warm-up would improve neuromuscular efficiency via PAP, though the authors did not rule out increased efficiency of the central nervous system (improved central input or enhanced excitation of the  $\alpha$ -motor neuron) as a possible mechanism. A noticeable limitation to this study was the absence of a control condition and what more, though the study recruited well-trained runners, they did not perform a running task post warm-up to verify an enhanced neuromuscular function in a running-specific task.

As the evidence suggests, the application of PAP to warm-up protocols and its subsequent response in performance has been under-studied. Though it has been suggested that the effects of PAP conditioning stimuli during a warm-up last for less than 12 minutes (Seitz et al. 2016), more evidence is required to support or negate this claim. This assumption is in line with the original research conducted by Houston et al. (1990; 1991) that found a 10 minute limit to PAP after enhanced twitch force. Future studies should focus on combining continuous, below lactate threshold exercises, with high intensity strength bouts in order to better elucidate a specific PAP stimuli for endurance athletes (Boullosa et al. 2018).

### **1.6 PAP During and After Endurance Performance**

Further to examining PAP effects prior to performance, many studies have investigated the enhancement of contractile activity during and after endurance performance activities like testing, training, and competing. The available evidence seems to suggest that exercise load and training background are the two main modifiers of PAP effects in endurance athletes (Boullosa et al. 2018). Different styles of experiments have been performed to examine the effects of the

different levels of these modifiers in this population. The existing literature has also examined many forms of measuring enhanced contractile activity during performance.

### **1.6.1 Maximal vs. submaximal intensities.**

The current literature backs the idea that recording a PAP response using the interpolated twitch technique while performing a prolonged submaximal contraction (as opposed to a maximal contraction) seems to prompt the greatest response in endurance athletes. For example, the widely cited study by Hamada et al. (2000) demonstrated that 10 second maximal isometric contractions of the ankle plantar flexor muscles enhanced PAP in endurance athletes as compared to sedentary controls, suggesting that PAP may counteract fatigue during exercise. In contrast, another study concluded that 10 second conditioning maximal voluntary contractions of the knee extensor muscles did not enhance PAP in endurance trained females (Paasuke et al. 2007). In fact, the endurance trained females showed a faster decline in the magnitude of PAP when compared to a power trained group. When examining the response of endurance-trained athletes to 10 minute prolonged submaximal contractions of the knee extensors, Morana et al. (2009) found that peak twitch torque measurements increased during the first minute of the exercise and subsequently remained above baseline values in a group of endurance athletes. Contrasting these results with the Hamada (2000) and Paasuke (2007) studies, it would appear that when using the interpolated twitch technique to test for PAP, endurance athletes could benefit more from prolonged submaximal contractions as opposed to maximal isometric contractions. Considering the principle of training specificity, this idea runs parallel to the idea that power athletes benefit more from PAP effects after short, maximal to near maximal conditioning activities.

### 1.6.2 Performance duration.

An enhanced contractile capacity is also evident in endurance athletes after performing endurance exercises of different durations (Finni et al. 2003; Millet et al. 2002; Millet et al. 2003a; Millet et al. 2003b; Skof Strojnik et al. 2006; Simpson et al. 2018; Vuorima et al. 2006). This was initially demonstrated after a 65 km ultra-marathon running event in which a severely depressed maximal voluntary force capacity was reported concurrently with a surprising potentiated twitch mechanical response and shorter contraction/half-relaxation times (Millet et al. 2002). Another study by the same authors found similar results after a 140-km road cycling race (Millet et al. 2003a). The force produced by the cyclists' post-prolonged cycling race significantly decreased ( $p < 0.05$ ) while the average rate of twitch force development significantly increased ~12% though the authors suggest that caution should be taken in assuming enhanced neuromuscular function as EMG was not recorded. In a follow-up study of a group of endurance athletes completing a ski skating marathon, these authors suggested that potentiating effects were present (Millet et al. 2003b). The results were conclusive that maximal voluntary force decreased by 8.4% ( $p < 0.005$ ) yet enhanced neuromuscular function was observed as the peak mechanical responses were greater, rate of force development was faster, and contraction times were shorter. This contrasts with a study conducted by Finni et al. (2003) where reductions in EMG amplitude, superimposed doublet twitch, and rate of force development were found after participants completed a 10-km running trial.

More recently, it was discovered that 40 minutes of running at 80 and 100% of  $\dot{V}O_2$  produced jump potentiation and changes in coordination strategies for knee extension exercises (Vuorimaa et al. 2006). In this study, a continuous 40 minute run was compared to an intermittent 40 minute run (2 min run/2 min rest). It was found that an increased mechanical

power in the continuous run had a positive correlation with  $v\dot{V}O_{2\max}$ , while the correlation with the intermittent run was negative. In a similar study with endurance runners, participants ran 6km at anaerobic threshold ( $V_{OBLA}$ ) and the results showed no sign of contractile enhancement, suggesting a balance of fatigue and potentiation via an impairment of the excitation-contraction coupling (Skof et al. 2006a). The mean maximum twitch torque, EMG activation, and maximal voluntary force either dropped or showed no change at both stimulation frequencies though maximum twitch torque was enhanced 10 minutes post run. The results of this study directly contradict the results of the Vuorimaa study. Skof et al. (2006b) performed another study shortly after, that demonstrated potentiating effects in twitch torque 10 minutes after a 5X300 m interval workout despite evidences of low-frequency fatigue. Amid the conflicting results, a more recent study discovered that a 10 minute bout on a cycle ergometer performed at 60-70%  $HR_{\max}$  could elicit PAP (Simpson et al. 2018). Following the cycling bout, time to peak tension, half relaxation time, and contraction duration were reduced with no observed tendon alterations (as measured by ultrasonography). All of this evidence combines to suggest that an enhanced PAP/fatigue relationship is found in prolonged endurance activities of moderate duration as opposed to higher intensity exercises.

### **1.6.3 Field testing- jumps.**

A common method to examine neuromuscular performance in endurance athletes (power and rapid/explosive force development) outside of the laboratory is by measuring jump capacity. Recent studies have utilized jump test protocols in the field to demonstrate the acute effects of endurance running exercises on jump potentiation (Boulllosa et al. 2011; Boulllosa et al. 2009; Garcia-Pinillos et al. 2015;2016; Latorre-Roman et al. 2014). For example, an enhanced countermovement jump (CMJ) performance was observed after endurance athletes performed a

incremental running test and time limit running test at maximal aerobic speed (MAS) (Boullosa et al. 2009). The improved jump performance was observed in both conditions immediately following and in the incremental running condition up to seven minutes post-stimulus ( $p = 0.000$ ). To further this discovery, these authors had 22 endurance athletes perform an incremental running test (UMTT) and assessed power production via CMJ on a force plate and sprint performance (Boullosa et al. 2011). The results showed an increased CMJ height (3.6%) after the running test that correlated with an increment in peak power (3.4%). It was concluded that PAP could counteract the peak force loss after an exhaustive running test. Jump potentiation has also been demonstrated in other studies examining extended interval training effects on endurance athletes (Garcia-Pinillos et al. 2015; 2016). Thirty experienced sub-elite long-distance runners were recruited to perform 4 X 3 X 400 m intervals. Both metabolic (lactate, HR, RPE) and performance (CMJ, hand grip strength) measures were examined and significant ( $p < 0.001$ ) improvements were noted in CMJ height. Interestingly, it was also noted that the results suggested that improvement in performance may be due not only to metabolic adaptations but to specific neuromuscular adaptations (PAP). Concurrently, a study very similar to this one demonstrated that experienced long-distance runners can maintain their strength levels, and work capacity despite induced fatigue (Latorre-Roman et al. 2014). Based on these results it seems that potentiated responses in the field could be monitored via simple jump tests. These tests appear effective in evaluating the different effects of training exercises on acute neuromuscular adaptations.

### **1.6.4 Neuromuscular adaptations during and after performance.**

Many studies have focused on measuring neuromuscular adaptations during and after multi-stage trials created to simulate competitions (Del Rosso et al. 2016; McIntyre et al. 2012;

Girard et al. 2012; Pageaux et al. 2017; Perrey et al. 2010; Place et al. 2004; Ross et al. 2010; Rousanoglou et al. 2016). Most notably, a study (Del Rosso et al. 2016) demonstrated that fatigue and potentiation co-exist during long lasting endurance events and that a possible greater potential for PAP is evident at the end of an endurance race. They showed this by examining endurance athletes' responses to a 30 km running trial and found a significantly greater ( $p < 0.05$ ) CMJ height (as compared to baseline) throughout the trial despite significant decreases in speed and increases in RPE. This finding is also seen in a study by Place et al (2004). They were the first to suggest that knee extensor maximal voluntary force generating capability is significantly depressed after prolonged running (5 hours) at 55% of maximal aerobic velocity. Despite the suggested decrease in central activation, peak twitch potentiation was 18% greater ( $p = 0.01$ ) at the end of the exercise. In a study including a group of ten cyclists, the participants underwent repeated 20-minute stages of prolonged cycling at 70%  $\dot{V}O_{2peak}$  until volitional exhaustion (McIntyre et al. 2012). During the cycling, and at the end of the stages, some participants showed potentiating effects in peak power output. At the time of exhaustion, whole-body power muscle-function, and drop jump-performance had fallen (7-19%) in all participants. Ross et al. (2010) demonstrated impaired voluntary activation and neural drive but enhanced contractile processes in a study of eight experienced runners completing a 20-km running trial. Participant's RPE steadily increased throughout the trial while maximal voluntary contraction of the knee extensors decreased only during the final 5 km. The maintenance of the force was explained, in part, by the unchanged amplitude of the electrically evoked muscle action potential (M-wave) or potentiated twitch during and after the trial (Ross et al. 2010). This contrasts with a study (Girard et al. 2012) that found a reduction in plantar flexors strength after a 5 km running time trial, arguably explained by a failure of the neuromuscular transmission and excitation-

contraction coupling. More recently, a study conducted with nine endurance athletes performing a cycling or uphill walking protocol prior to a 10 km run found enhanced contractile properties explained by PAP. Though the cycling protocol was found to decrease knee extensor force, twitch torque, and rate of force development, uphill walking and running significantly limited the decrease in knee extensor force post exercise. Collectively, these studies (Del Rosso et al. 2016; McIntyre et al. 2012; Girard et al. 2012; Pageaux et al. 2017; Perrey et al. 2010; Place et al. 2004; Ross et al. 2010; Rousanoglou et al. 2016) would suggest that PAP could be observed in multi-stage trials designed to simulate competitive settings in endurance performance. Nonetheless, there is a need for more studies that examine PAP concurrently with different origins of fatigue in continuous prolonged endurance exercises (Millet et al. 2004).

The presented evidence substantiates the claim that endurance athletes demonstrate PAP responses during and after prolonged submaximal conditioning activities in various settings. In field settings, it appears that practical jump tests are valid for the observation of PAP responses and possibly origins of fatigue. Though the current evidence would suggest that an ideal PAP/fatigue balance, and consequently enhanced neuromuscular function, is best observed during submaximal contractions at moderate-high intensities and durations; more studies are needed to confirm or negate this current idea.

### **1.7 Justification of Testing Measures**

The following testing measures have been included in the methodology of the present study in order to quantify and elucidate the neuromuscular alterations:  $\dot{V}O_2$  maximum test, MVC, evoked contractile properties (ITT, %VA, TPT), drop jump, heart rate, and rate of perceived exertion. The incremental treadmill protocol for testing  $\dot{V}O_2$  maximum has been examined as a reliable and valid estimate (Weltman et al. 1990). The results from this test help to



provide an indication of the athleticism and training status of the participants and, if stratified into groups, could provide an opportunity for further analysis into neuromuscular alterations. In addition, the peak treadmill running velocity (termed the maximum aerobic speed) during this test has been demonstrated as a good indicator of running performance (Noakes et al. 1990). The maximum aerobic speed achieved during the  $\dot{V}O_2$  test will then be compared to the maximum speeds achieved during the running trials. This comparison will enable an estimate of what percentage of  $\dot{V}O_2$  maximum the participants were running at during their respective time trials thus providing a possible discussion of metabolic properties and differences. The heart rate and rating of perceived exertion will also be employed as comparison measures between the  $\dot{V}O_2$  maximum test and the running trials in order to ensure uniformity in the level of intensity and exertion between the sessions.

*Central Measures:* The MVC performed with a knee angle of  $90^\circ$  is a widely accepted variable for the measurement of plantar flexors muscle strength (Merlet et al. 2018; Meldrum et al. 2009). Specifically, the MVC is used as a measure of central activation (Kent-Braun et al. 1996) and thus, an increase or decrease in MVC force could help specify the nature of potential PAP effects (central or peripheral). As PAP aims to explain an increase in muscle function, an increase in MVC force could suggest the presence of a centrally mediated PAP mechanism. What more, as the plantar flexors play a central role in running performance (Bezodis et al. 2008), a maximum contraction of the plantar flexors will provide ample evidence as to the effects of the jump squats. In conjunction with the MVC, EMG will also be recorded as indicator of the electrical activity produced by actively contracting motor units as controlled by the central nervous system. Likewise, the resultant EMG activity will give an acceptable

indicator as to the increase or decrease of central activation and consequent implications for PAP (Chesler et al. 1997).

*Peripheral measures:* Evoked contractile properties (ITT, %VA, TPT) recorded from electrically evoked twitches in the plantar flexor muscles are good indicators for evaluating the neuromuscular system in exercise training (Pääsuke et al. 1999). These properties give a measure of the number of motor units activated as well as a measure of the excitation-contraction coupling function. This is relevant for the present study in that the main proposed mechanism for PAP is explained at the peripheral level and as such, a consequent increase (or decrease) in these evoked contractile properties would indicate altered function at the peripheral level.

*Performance measures:* The drop jump will be used in this study as a test of the lower limb muscle function under the stretch-shortening cycle. Drop jumps will be used as this gives a better indicator of the SSC during running than does a basic countermovement jump. The type of SSC that is tested in the drop jump is common in running and is known as the pre-stretch. An increased activation of the muscle fibers as a result of the pre-stretch could result in an increase in muscle force production (McClymont et al. 2005). Research suggests that differences in contact time, flight time, jump height, and reactive strength index could suggest improved running economy and consequent decrease in running time. The time to complete each kilometer repeat as well as the overall summed time will also be used as a measure of performance changes as the alteration seen at the neuromuscular level could affect pacing and would thus be made manifest in a faster running time to complete the trials.

## **1.8 Conclusion**

This review establishes the presence of neuromuscular enhancements (PAP) in endurance athletes and demonstrates the existence of PAP mechanisms during and after endurance

performance in endurance athletes. The exact role that PAP plays during endurance training, testing and competition is still debated and requires further research. What more, the evidence suggests that the examination of the time course of neuromuscular enhancements should be combined with PAP conditioning stimuli in order to better understand time-specific adaptations at the central and peripheral level. These adaptations can be measured most effectively with maximal voluntary contractions and the interpolated twitch technique and most simply via measured jump height. More studies are needed in order to examine the volume, intensity, and contraction type specifics regarding an ideal PAP eliciting conditioning stimulus in endurance athletes performing subsequent endurance activities. Studies involving endurance athletes should also include metabolic measures in order to control for metabolic changes evident from running. Future studies should also verify the effectiveness of different training interventions on PAP responses in endurance performance. Given all of the aforementioned evidence, it would follow then that the proposed thesis to examine the time course of the effects of conditioning stimuli on eliciting PAP in endurance athletes, has important implications. This thesis project fills the gaps in PAP endurance performance research as it focuses on examining the time course of how a PAP conditioning stimulus incorporated into a warm-up will affect a subsequent 5 km running trial. Further to the evidence above, it is hypothesized that performing a 5RM weighted squat protocol as part of a standardized running-specific warm-up will induce significant measurable PAP effects during the course of a subsequent 5 x 1 km time trial run and up to 7 minutes post-run protocol.

## **Chapter 2: Co-Authorship Statement**

The following details my role in preparation of this manuscript:

### **Research Design**

The methodology was developed based on previous research and manuscripts by Dr. David Behm in combination with other published evidence and work investigating post-activation potentiation, endurance athletes, and neuromuscular alterations. Discussions with Dr. David Behm and Dr. Duane Button helped to refine the details of the experiment. With assistance from Dr. David Behm I was able to obtain approval from the Health Research Ethics Authority (HREA) to conduct this research.

### **Data Collection**

All data was collected by me with assistance from Mr. Hamid Ahmadi.

### **Data Analysis**

I performed all data analysis procedures.

### **Manuscript Preparation**

I wrote the manuscript with assistance from Dr. David Behm.

**Chapter 3: Manuscript**

**Prior Band-Resisted Squat Jumps Improved Running Time, Rating of Perceived Exertion,  
and Neuromuscular Performance in Middle-Distance Runners**

**Authors:** Jonathan L. Low<sup>1</sup>, Hamid Ahmadi<sup>1</sup>, Liam P. Kelly<sup>2</sup>, and David G Behm<sup>1</sup>

**Institution:** <sup>1</sup> School of Human Kinetics and Recreation  
Memorial University of Newfoundland  
St. John's, Newfoundland, Canada, A1M 3L8

<sup>2</sup> Recovery and Performance Lab, Faculty of Medicine  
L.A. Miller Center  
Memorial University of Newfoundland  
St. John's, Newfoundland, Canada A1M 3L8

### 3.1 Abstract

**Introduction:** Post-activation potentiation is a prominent neuromuscular alteration that aids in the enhancement and maintenance of force production. PAP conditioning contractions (explosive-styled exercises) have been shown to increase subsequent performance. Though most research has examined PAP effects on strength or power activities, PAP and the role it could play in endurance sports has been widely understudied. The aim of this study was to characterize the time course of the effects of a PAP conditioning stimulus (band-resisted jump squat) on a subsequent 5 X 1 km running trial. **Methods:** A randomized within subjects, repeated measures experimental study design was used. Twelve healthy male, endurance-trained athletes completed 4 sessions: 1)  $\dot{V}O_2$  max test 2) a familiarization of testing equipment, estimation of the individual's 5RM, and testing of evoked contractile properties and 3/4) a running-specific warm up, the conditioning exercise intervention (4x5RM band-resisted jump squats or no squats) and a 5 x 1 kilometer time trial run. Tests were conducted immediately prior to the intervention, after each kilometer, immediately following the 5x1 km run, and at seven and ten minutes post 5 k run. These measures included the interpolated twitch technique (ITT), evoked contractile properties (peak twitch torque, rate of force development, M-wave), maximum voluntary isometric contractions (MVIC) to determine peak ankle plantar flexor force, force produced in the first 100 ms (F100), 30-cm drop jump (height, contact time and reactive strength index), rate of perceived exertion (RPE), and heart rate. **Results:** The PAP stimulus resulted in a 3.6% reduction in time to complete the run ( $p = 0.07$ ), and a significantly decreased time to complete kilometer one ( $p = 0.017$ ,  $d = 1.08$ ). Jump height increased by 9.2% while reactive strength index increased by 16% in the squat condition. F100 and MVC in the squat condition increased at kilometers 3 ( $p = 0.04$ ,  $d = 0.84$ ) and 4 ( $p = 0.034$ ,  $d = 0.29$ ), and 7 mins post run ( $p = 0.03$ ,  $d =$

0.60). %VA showed a significant increase 7 minutes post run ( $p = 0.04$ ,  $d = 0.59$ ) in the squat condition yet decreased 7 minutes post run in the control condition ( $p = 0.03$ ,  $d = 0.36$ ). No significant changes were noted in HR or RPE between conditions. **Conclusion:** This is the first study to demonstrate that a band-resisted squat protocol decreases running time and improves neuromuscular properties in endurance athletes running 5X1 km. It is hypothesized that heightened pacing abilities, improved stretch-shortening cycle efficiency (SSC), and PAP are the proposed explanations. We conclude that a 4X5RM band-resisted jump squat protocol improves running performance by increasing running economy and jump height while abating effects of fatigue.

**Key words:** post-activation potentiation; neuromuscular adaptations; running; endurance

### 3.2 Introduction

Success in endurance sports has been associated with an athlete's ability to transport and consume oxygen during intense, exhaustive exercise. Consequently, the physiology of endurance running has been studied with a focus on the metabolic system as the limiting factor for performance. Though it is well established that physiological properties such as maximal oxygen uptake, utilization of maximum aerobic power, running economy, and blood lactate levels are factors that impact endurance performance (Costill et al. 1973; Davies et al. 1979; Foster et al. 1978; Hill et al. 1925; Joyner et al. 1991), these properties cannot solely explain differences observed in endurance performance (Nummela et al. 2006). Recent research has suggested an alternative explanation for physiological limitations during performance. This theory proposes that, during endurance exercise, the central nervous system integrates feedback from multiple sources in the body and consequently regulates the recruitment of skeletal muscles such that the levels of intensity and duration do not exceed the threshold where potential damage could occur to the brain, heart, and other vital organs (Gibson et al. 2004; Kayser et al. 2003; Lambert et al. 2005; Noakes et al. 2000; Noakes et al. 2001). As the central nervous system coordinates the recruitment of skeletal muscle motor units (neural drive), the muscle's ability to produce power and speed is affected. A subsequent decrease in muscle performance is classified as muscle fatigue.

In the case of an endurance athlete, a high rate of fatigue development coupled with a heightened inability to tolerate fatigue, would decrease speed and negatively affect performance. Muscle fatigue can originate at different levels of the motor pathway and can be divided into central and peripheral components. Peripheral fatigue is due to changes seen distally to the neuromuscular junction while central fatigue has its origin proximal to the neuromuscular



junction representing activities within the central nervous system (Gandevia et al. 2001; Wan et al. 2017). Recent evidence has shown that in addition to peripheral fatigue; reduced neural drive (central fatigue) of the contracting muscles is likely to compromise motor performance during running (Girard et al. 2008). It has been argued that the etiology of muscle fatigue is complex during prolonged running exercises and that impairment can come peripherally (neuromuscular propagation and excitation-contraction coupling) and centrally (central drive/activation) (Millet et al. 2003). Thus, the body's management of fatigue (central and peripheral) development, commonly known as pacing, becomes an important factor to consider when discussing successful endurance performance. Further to the idea of the role the fatigue can play in limiting endurance performance, research also suggests that there are central and peripheral adjustments that take place at the skeletal muscle to enhance performance (Boullousa et al. 2009; 2011; Hamada et al. 2000; Seitz et al. 2016; Vuorimaa et al. 2006;). These peripheral adjustments are thought to enhance performance via a mechanism called post-activation potentiation (PAP).

Post activation potentiation (PAP) is a phenomenon explained by an enhancement of muscle force output and rate of force development as a result of muscle contractile history (Robbins, 2005). This phenomenon is highly applicable to sports, and research has increasingly focused on methods for prompting a post-activation potentiation as a form of performance enhancement. Unfortunately, due to the lack of applicable literature, scientists and performance professionals are often unaware of the potential PAP has to aid in both endurance training and endurance competition. The aim of incorporating a PAP stimulus into an athlete's training or pre-competition program is to elicit an acute enhancement in muscle performance and consequently a higher jump, faster time, or further throw.

In applying PAP to the endurance phenotype, a widely cited article by Sale (2004) suggests that PAP should have its greatest effect during submaximal contractions in which the motor units are firing at relatively low frequencies, as is the case with endurance exercise. It follows then that if the levels of fatigue in a sport or activity are relatively lower than the levels of PAP, not only athletes performing maximal activities will benefit, but any athlete could benefit from this mechanism. Furthermore, according to Sale (2004), these aforementioned lower firing rates occur during endurance activities and consequently help achieve the ideal balance between PAP and fatigue.

With the existing evidence that PAP enhances subsequent endurance performance, the most plausible application of PAP to sport is its use as a warm-up technique prior to training or competition in order to acutely enhance contractile potential. Interestingly, few studies have examined the feasibility of this application in the endurance running population (Burnley et al. 2005; Carter et al. 2005; Feros et al. 2007; Jones et al. 2003; Palmer et al. 2008; Silva et al. 2014; Skof and Strojnik et al. 2007). Further to examining PAP effects prior to performance, many studies have investigated the enhancement of contractile activity during and after endurance performance activities like testing, training, and competing. To our knowledge, there have been very few to no studies that have taken aim at combining a PAP conditioning stimulus into a warm up while subsequently monitoring the central and peripheral alterations during and after a training-styled run session.

Therefore, the aim of the present study was to evaluate the neuromuscular alterations of a PAP conditioning stimulus on subsequent performance during and after a running time trial. Specifically, we compared the neuromuscular effects of an applicable (Gooyers et al. 2012), in-field, 5RM band-resisted squat jump on a subsequent 5 X 1 kilometer time trial. Neuromuscular

measures (electromyography, maximal voluntary contractions, evoked contractile properties, and performance measures) provide substantially sensitive information to indicate the presence of central or peripheral changes. We hypothesized that performing a 5RM band-resisted jump squat protocol as part of a standardized running-specific warm-up would induce significant measurable PAP effects during the course of a subsequent 5 X 1 kilometer time trial run and up to 10 minutes post-run protocol.

### **3.3 Methodology**

#### **Experimental Design**

A within subject (repeated measures) experimental study design was employed in this study. Twelve healthy male endurance-trained athletes ( $28.33 \pm 1.37$  years,  $176.85 \pm 2.41$  cm,  $75.07 \pm 2.74$  kg,  $57.98 \pm 6.37$  mL\*kg<sup>-1</sup>\*min<sup>-1</sup> [ $\dot{V}O_2$  max] ) completed two familiarization, and two intervention sessions in a randomized order separated by a minimum of 72 hours. The familiarization sessions included: 1) an incremental running test to volitional exhaustion ( $\dot{V}O_{2max}$ ) and 2) a familiarization of testing equipment, estimation of the individual's 5RM, and testing of evoked contractile properties. The intervention sessions included a running-specific warm up, the intervention (4x5RM squats or no squats), and a 5 x 1 kilometer time trial run. Tests were conducted immediately prior to the intervention, during the 3 minutes after each kilometer, immediately following the 5 x 1 km run, and at minutes seven and ten post 5 k run. These measures included the interpolated twitch technique for contractile properties (peak twitch torque, rate of force development, percent voluntary activation, potentiated twitch force), maximum voluntary isometric contractions (MVIC) to determine peak ankle plantar flexor force, force produced in the first 100 ms (F100), 30-cm drop jump height (flight time, contact time and reactive strength index), rate of perceived exertion (RPE), and heart rate.

## **Participants**

Based on a statistical power of 0.8, an effect size of 0.5 (Cohen, 1969, p.348), and an alpha level of  $p < 0.05$ , an “F test, ‘A’ priori ANOVA: repeated measures” analysis was executed using *G\*Power* (software package 3.1.9.2) with a resultant minimum sample size calculation of 9. Consequently, twelve healthy male endurance-trained runners were recruited for this study. The participants were actively engaged in training (running an average 62 km/week, running 5 days/week, weight training 1 day/week), had average 5 km times ranging from 16:15 to 22:12, and had a minimum of two years’ experience competing in cross-country/road race events. Participants who had experienced any musculoskeletal injuries in the past six months or who had diagnosed neurological conditions were not eligible to participate in the study. Participants were willing and able to attend the laboratory on four occasions for familiarization and testing. Participants were required to refrain from consuming caffeine or alcohol 24 hours prior to testing and to abstain from food 4 hours previous to each testing session. The participants received instructions to avoid running and high intensity training of any kind 72 hours prior to testing sessions. To ensure healthy, ethically-informed participants, each participant read and signed a Physical Activity Participation Questionnaire (PAR-Q: Canadian Society for Exercise Physiology) and an approved informed consent form. The study adhered to the approval process of the institution’s Interdisciplinary Committee on Ethics in Human Research and was conducted according to the Declaration of Helsinki.

## **Procedure**

### ***Familiarization sessions.***

Participants attended the lab on two separate occasions to 1) perform the Université de Montréal Track Test ( $\dot{V}O_2$  consumption, ventilatory threshold, maximum aerobic speed, and

maximum heart rate) and 2) to estimate 5RM weighted squat, and establish evoked contractile properties measures. Due to the potentially exhausting effects of the familiarization sessions, they were separated by a 48-hour period.

***Université de Montréal Track Test (UMTT) & anthropometric measures.***

Upon arrival to the lab, the participant's height (cm) and weight (kg) was measured and age annotated. The participants began the session with a 5-10 minute warm-up/familiarization period on a treadmill (Cybex 751T, Medway, Massachusetts) at a self-selected pace. Following the warm-up, the participants were connected to an indirect calorimetric system (MOXUS CD 3-A/S-3A, AEI Technologies, Pittsburgh, Pennsylvania) via a mask secured to the oral/nasal regions of the face. The participants were outfitted with a heart rate monitor (chest strap; Polar H10, Polar, Kempele, Finland) and the test protocol was discussed and any questions answered. The test was conducted according to the protocol set out by Léger & Boucher (1980) in which the participant runs on a motor-driven treadmill at a constant slope of 1%. The test began at an initial speed of 7 km\*hr<sup>-1</sup> with a 1 km\*hr<sup>-1</sup> speed increase every two minutes until volitional exhaustion. After exhaustion was reached, a five minute rest/recovery period was allowed before commencing a verification phase (to ensure a true  $\dot{V}O_2$  maximum.) During the verification phase (Rossiter et al. 2006) the treadmill was set at 105% of the recently determined maximal aerobic speed (MAS) and the participant ran until their limit of tolerance was reached. A self-selected pace cool down period followed and continued until the recovery heart rate reached 120 b\*min<sup>-1</sup>. To further determine if the participants attained  $\dot{V}O_{2max}$  or true exhaustion, the respiratory exchange ratio (RER: >1.0), a plateau in  $\dot{V}O_2$  values at maximum intensity, Borg Scale perceived exertion scales (>17/20) and maximum heart rate (approximately 220 – participants' age) attained was also monitored (Aitken et al. 1988; Foster et al. 2007; Midgley et al. 2006;

Sánchez-Otero et al. 2014). Results from this session were used to provide a descriptive statistic into the training status of the participants. Maximum aerobic speed and maximum heart rate were used to compare and contrast the measured aerobic speed and heart rate reached during the running trials, enabling a better analysis of the metabolic properties during the trials.

***5RM estimation, evoked contractile properties & equipment familiarization.***

Based on the evidence demonstrating the potentiating effects of the squat, Rixon et al. (2007) suggest that potentiation effects can depend on an individual's training background. Squat (and other biomechanically similar) exercises are commonly performed in endurance athlete-focused training sessions (Chelly et al. 2009; Hickson, 1980 Storen et al. 2008; Willy et al. 2011). It was determined prudent to include squat exercises as the conditioning stimulus of interest as they are ubiquitous in studies examining the effects of post-activation potentiation (Bauer et al. 2017; Esformes et al. 2013; Healey et al. 2017; Tillin et al. 2009). As the in-field applicability (Behm et al. 1993; Newton et al. 2002; Wallace et al. 2006) of a variation on the squat (a band-resisted jump squat) is extremely high in endurance athletes, this exercise was implemented as the protocol. For every jump squat set, two looped resistance bands (Theraband CLX bands, Akron, Ohio) were attached with carabiners to heavy weights on either side of the participant. The resistance bands were then stretched over the head and placed cross-body on the contralateral clavicle/upper trapezius complex, such that the band secured to the weights on the left side of the participant was placed on the right shoulder of the participant and vice versa (see Figure 3.12). The jump squat began at the downward movement phase (thighs parallel with the floor) and proceeded with a maximally explosive jump (flight phase) against the resistance of the bands (Masamoto et al. 2003.) Every squat manoeuvre took place on a rubber mat where an exercise physiologist was present to monitor the participant. The same jump squat positioning

was ensured by the same test administrators in each instance. After a general low-intensity aerobic warm up on a cycle ergometer, a series of submaximal squats (five reps) with loads of 50-70% body mass were conducted to get an initial idea of the participant's 5RM. Band resistance (weight) was manipulated with four various coloured bands with standardized resistance (2.6 kg, 3.3 kg, 4.6 kg, and 6.4 kg). Following this, participants performed a series of sets with increasing band resistance (colour and number of band) until the 5RM was determined. Participants rested for 3 minutes between each 5RM trial. In each trial, if the squat jump was performed with the proper form (thighs parallel to the ground, feet left floor) for all 5 repetitions, the band resistance was incrementally increased by colour and number (dependent on the effort required for the previous lift) for each subsequent trial until the jump fell short of the full range of motion (thigh angle compromised, or evidence of improper gluteal and back form) on two separate trials (spaced by four minutes of rest between sets and 1-3 seconds rest inter-repetitions)(Mitchell et al. 2011; Jensen et al. 2003; Matthews et al. 2004). The 5RM was recorded as the maximum amount of resistance that could be lifted for five repetitions through the whole range of motion while maintaining proper form. Following the 5RM estimation trials, the participant was given a brief preview to the interpolated twitch procedure, the treadmill protocol and proper form for completing a 30-cm drop jump.

***Evoked contractile properties.***

Evoked contractile properties were recorded prior to MVIC assessment in the interventions in order to quantify the membrane excitability and excitation-contraction coupling function. Prior to the application of the stimulating and EMG electrodes, the underlying skin was prepared by shaving, lightly abrading, and cleansing (using isopropyl alcohol.) Peak twitch force of the triceps surae muscle group was evoked using bipolar surface electrodes placed on

the popliteal fossa overlying the medial popliteal nerve (cathode) and ~ 2 cm below the distal border of gastrocnemius (Hamada et al. 2000). The positioning of each electrode was recorded to allow between-session replication, though slight variation in positioning was expected as the electrode location at the beginning of each session was altered to ensure an optimal stimulus response. Both stimulating electrodes were connected to a high-voltage, constant current simulator (Digitimer Stimulator Model DS7AH, Hertfordshire, UK). The 200- $\mu$ s, 400-V square wave pulse with a current intensity (10 mA – 1 A) was progressively increased (by 10mA increments) until a maximum twitch force was achieved (no further increase in peak twitch torque.) Force-time data was recorded at 2000 Hz using a BioPac/AcqKnowledge data acquisition system (DA 150: analog-digital converter MP100WSW, Holliston, MA). The average of three trials was used to measure peak twitch force, time to peak twitch force, and rate of force development (Behm et al. 2001; 2013). This laboratory has previously reported excellent intersession reliability (ICCs of 0.98-0.99 for evoked twitch properties) using the above mentioned technique (Behm et al. 1996; 2002; 2013).

### **Testing Measures**

#### ***Ankle plantar flexors maximum isometric voluntary contraction (MVIC) force.***

The participant was seated in a standardized position with ankle, knee, and pelvis angles of 90°, as measured by a goniometer. The isometric ankle plantar-flexion of the dominant foot was measured using a custom built boot apparatus (Memorial University Technical Services) outfitted with a strain gauge deformed by a metal pedal. Participants were instructed to contract as quickly and powerfully as possible for the entire duration of the MVIC. The MVIC's were sustained for 4 seconds. All voluntary and evoked forces were detected by the built-in strain gauge (boot apparatus), amplified (x1000) (Biopac Systems Inc., DA 150; analog-digital



converter MP100WSW, Holliston, MA) and interfaced with a computer. The data was sampled at 2000 Hz and analyzed with a commercially designed software program (AcqKnowledge III, Biopac Systems Inc., Holliston, MA). Data analysis included triceps surae peak MVIC forces as well as the instantaneous strength defined as the force produced in the first 100 ms (F100).

***Interpolated twitch technique (ITT).***

The interpolated twitch technique (ITT) has been reported to be a valid and reliable measure of muscle voluntary activation (Behm et al. 1996; Behm 2009) and as such was used to achieve potentiated twitch measurements. The stimulation intensity was re-adjusted before each testing session with a simplified procedure based on the intensity used during the familiarization session (Rupp et al. 2010). The same electrode configuration and current intensity used for the maximal evoked twitches were used for the ITT. The ITT involved superimposing an electrically stimulated doublet with an inter-pulse interval of 10 ms (100 Hz) in the relaxed dominant triceps surae muscle pair, at 2.5 s after the onset of a 4-s MVIC of triceps surae, and 5 s after the MVIC (potentiated twitch) (Girard et al. 2012). An interpolation ratio was calculated by comparing the amplitude of the superimposed doublet with a post-contraction potentiated doublet (5 s following the MVIC) to estimate the extent of voluntary activation during the voluntary contraction, where  $\% \text{ of muscle voluntary activation} = (1 - (\text{interpolated doublet force} / \text{potentiated doublet force}) \times 100)$  (Behm et al. 1996).

***Electromyography (EMG).***

Muscle electromyography was recorded from triceps surae (soleus and medial gastrocnemius) during MVIC, evoked and ITT contractions. As mentioned previously, prior to application of the electrodes (1-cm 162 Ag/AgCl; MediTrace 133, Kendall, Technical products Toronto, Ontario, Canada), the designated area was prepared by shaving, lightly abrading, and

cleansing (using isopropyl alcohol.) The active recording electrodes were placed on the dominant medial gastrocnemius and dominant soleus (15-cm distal to the stimulating electrode) (Khosrawi et al. 2013), the reference electrodes were placed distally such that an inter-electrode distance of two centimeters was observed, and the ground electrode was secured on the lateral epicondyle of the femur (Behm et al. 2002). EMG activity was amplified (x1000), band-pass filtered (10-1000 Hz), rectified and directed to a computer. The integrated EMG (iEMG) activity was determined over a 1-s period following the peak force during the MVIC. The reliability of this EMG measurement technique has been demonstrated by this laboratory in previously published literature with reported ICC values ranging from 0.91 to 0.99 (Behm et al. 2001; 2002; 2004).

***30-cm drop jump (DJ).***

A series of two drop jumps were performed at 30 second intervals (French et al. 2003). During the jump, the participants were instructed to maintain their hands akimbo (hands placed on the iliac crest); this helped remove extraneous variance resulting from arm swing. The jump manoeuvre began with the participant standing on a 30-cm platform. When the participant felt inclined, they transferred from the platform, made a 2-footed landing on a jump mat (SmartJump, Fusion Sport, Chicago, Illinois), and jumped vertically for maximum height. Participants were instructed to maximize jump height by jumping as high as possible. Flight time, contact time, and jump height were measured by the SmartHub system (Fusion Sport, Chicago, Illinois) and reactive strength index (Jump height / Contact time) was computed accordingly (Young et al. 2003).

***Rating of perceived exertion (RPE).***

Participants were asked to measure their level of self-perceived effort in order to determine how hard they feel their body was working. They were told that this rating should reflect how heavy and strenuous the exercise feels to them; combining the feelings of physical stress, effort, and fatigue. They were instructed not to focus on any one factor (eg. shortness of breath) but to focus on the overall feeling of exertion. As the Borg scale ranges from 6-20, the participants were informed that a 6 represents ‘no exertion at all’ and a 20 means ‘total and maximal exertion.’ At each measurement interval, the participants were shown the Borg rating scale chart to assist in their response.

***Heart Rate (HR).***

Upon arrival to the laboratory, participants were outfitted with a heart rate monitor (Polar H10, Polar, Kempele, Finland) using a chest strap. Resting heart rate was recorded during a 5 minute period prior to commencing any activity (warm-up or intervention) and the monitor remained on and recorded the heart rate for the complete duration of the testing sessions. Following each testing session, the heart rate monitor was connected to a computer, interfaced with its accompanying software (Polar Weblink and Polar Flow), and the data uploaded for analysis and data extraction.

**Testing sessions**

***Running-specific warm-up.***

Prior to the first round of tests (pre-intervention), each experimental session began with a standard running warm-up (Bogdanis et al. 2014). The warm-up commenced with a five-minute running bout on a treadmill (Cybex 751T, Medway, Massachusetts) at a moderate self-selected pace. Following the treadmill warm-up, the participants performed a 4 x 50m dynamic warm up

(Skof et al. 2007; Bogdanis et al. 2014) including ‘A’s, B’s, and C’s (leg whips, knee kicks, heel-to-gluteal kicks) (see figures 3.18a/b). The warm-up component finished with the participants performing 4 x 50m accelerations (start 50 m repeat at a normal pace and accelerating through to a maximum sprint by the 50 m mark.) After each of the 50m repeats (dynamic and accelerations) the participants took active rest as they lightly jogged 50 meters back to the starting point (about 30-45 seconds between each 50m repeat.) All of the participants were accustomed to and had previous exposure to this running drill.

### ***Interventions.***

Using a random allocation selection on separate testing days, the participant performed one of two conditions (4 x 5RM squat conditioning stimulus or control). At the beginning of each testing session, proper EMG and stimulating electrode preparation and placement was ensured (as outlined above) and the heart rate monitor was situated on the participant with the resting heart rate observed. All testing measures were performed with uniformity (same tests in the same order) across all conditions.

### ***4 X 5RM squat conditioning stimulus intervention.***

The PAP stimulus intervention required the participants to perform the standardized running-specific warm-up and subsequently perform the pre-intervention tests (MVIC, ITT, DJ, RPE, HR). At the completion of the pre-intervention tests the participants moved directly to the mat to perform the conditioning stimulus (4 X 5RM band-resisted jump squats). The pre-determined 5RM resistance bands were anchored on either side of the participant and the resistance bands were placed cross-body on the clavicle/upper trapezius complex and the participant performed 4 sets of 5 repetitions (Ah Sue et al. 2016; Chatzopoulos et al. 2007; Linder et al. 2011; Silva et al. 2014). Each of the 4 sets of 5RM were monitored and

standardized such that the downward movement phase continued until the thighs are parallel with the floor. Each set was separated by a 2 minute rest. At the completion of the squatting protocol, the participant took an 8 minute rest according to ideal PAP stimulus protocol (Wilson et al. 2013; Silva et al. 2014) where sitting and walking were permitted. Subsequent to the 8 minute rest, the participants completed a 5 x 1 km running trial on a motor-driven treadmill (Paavolainen et al. 1999; Ross et al. 2010; Steele et al 2017). Participants were instructed to complete each kilometer repeat as fast as possible and verbal encouragement was given while the participant was running. All of the screens and controls on the treadmill were hidden and the participants were only given access to the control arrows that adjust the treadmill speed. Participants were encouraged to adjust the treadmill speed according to their physiological and pacing needs and were reminded to complete the trial as fast as possible. In an effort to mimic distance markers commonly found in running settings, verbal notice on distance was given when 500 and 250 meters remained in each repeat. The 5 km running trial was separated by test measures at each kilometer (participant dismounted treadmill at each kilometer, performed tests (3 minutes), and remounted the treadmill with 30 seconds allotted to return to speed). Following the completion of the 5 x 1km running bout, post-intervention tests (MVIC, ITT, DJ, RPE, HR) were conducted immediately and seven and ten minutes (Boullosa et al. 2009; Hamada et al. 2000; Millet et al. 2002) post-intervention.

***Control condition.***

The control condition required the participants to perform the running-specific warm-up and immediately following, the pre-intervention tests. Following the pre-intervention tests, the participants sat for a total of 13 minutes (5 minutes for control and 8 additional minutes as part of the PAP rest protocol). After the rest period, the participants ran the 5 x 1 kilometer trial

separated by test measures at each kilometer. Post-test measures were taken immediately following, and seven and ten minutes post 5 km run.

### **Statistical Analysis**

Statistical analyses were completed using the SPSS software (Version 23.0, SPSS, Inc. Chicago, IL). The assumption of sphericity and normality were tested for all dependent variables and if a violation was noted, the corrected values for non-sphericity with Greenhouse-Geisser are reported. A two-way repeated-measures ANOVA was used to compare triceps surae function (neuromuscular and metabolic) (2 conditions x 8 testing times) in each dependent variable. In the event of significant main effects or interactions, planned pairwise comparisons were made using the Bonferroni method to test for differences among mean value time points. Pearson correlation coefficients were calculated to determine relationships between selected parameters. The level of significance was set at  $P < 0.05$  and all results are expressed as mean  $\pm$  SD.

### **3.4 Results**

Other than the ITT measure (0.76), ICC reliability scores for each test measure exceeded 0.90 (Table 3.1). Resultant means, standard errors, confidence intervals, and test statistics can be found in Appendix I.

#### **Triceps Surae MVC**

There were no significant main effects of the conditioning squat stimulus or time period for triceps surae MVC force. There was a significant ( $F_{(7, 77)} = 2.3, p = 0.039$ ) interaction effect between the conditioning stimulus and the data time points for force produced between running trials. Between group post-hoc contrasts revealed significantly ( $F_{(1, 11)} = 4.679, p = 0.05, d = 0.55, 3\% \uparrow$ ) greater MVC force when comparing squat stimulus to control at 10 minutes post-run as compared to pre-test. Within group post-hoc analyses revealed significantly higher MVC

force for the squat condition at kilometer 4 ( $d = 0.29, p = 0.034, 8\% \uparrow$ ) and 10 minutes post-run ( $d = 0.36, p = 0.036, 9.5\% \uparrow$ ) compared to pre-test (Figure 3.1).

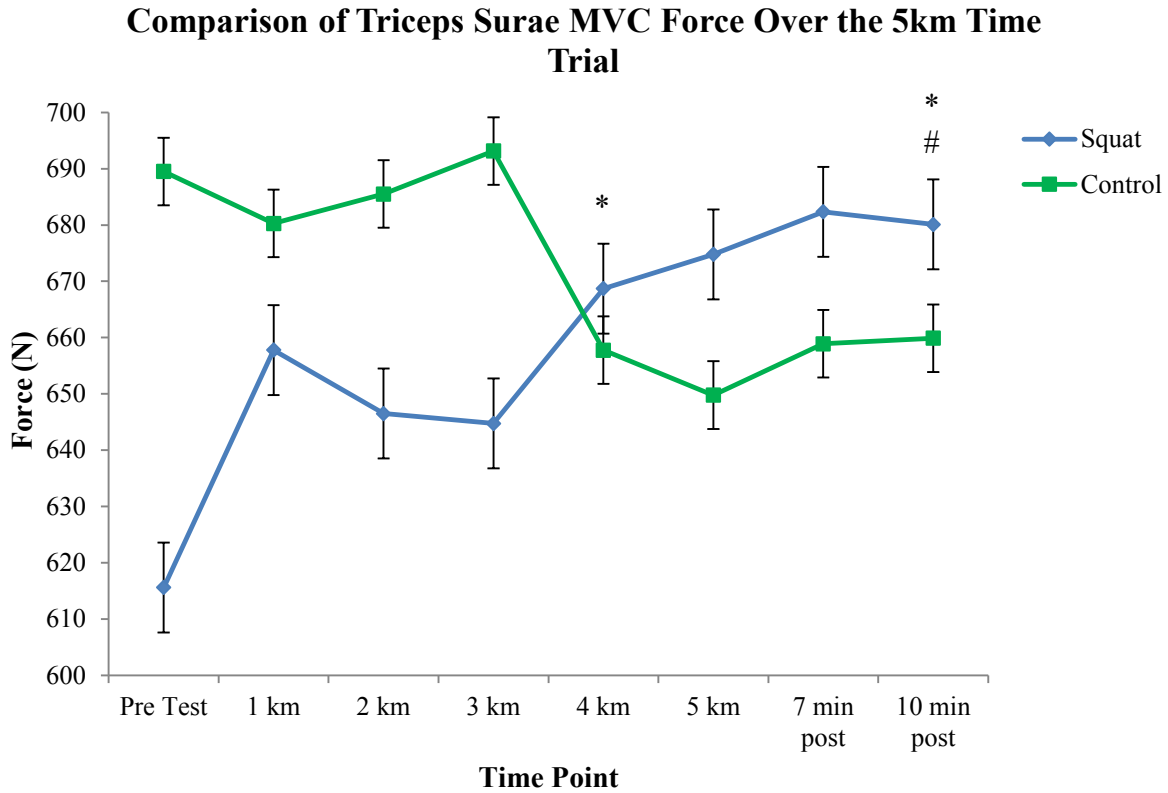


Figure 3.1. Estimated changes in triceps surae MVC force means between the control and squat condition. Overall increase noted in the squat condition. Asterisk (\*) indicates the squat condition value was significantly greater than pre-test. The hashtag (#) symbol indicates the squat condition value was significantly greater than the control condition.

**Gastrocnemius and Soleus EMG**

No significant overall main effects or interactions were observed.

**Triceps Surae F100**

There was a tendency for a main effect of time for F100 in the 5 km time trial ( $F_{(7, 77)} = 2.034, p = 0.061$ ). Contrasts revealed that F100 at kilometer 3 ( $F_{(1, 11)} = 9.605, p = 0.01, d = 0.7$ ,

24% ↑) and 7 minutes post run ( $F_{(1, 11)} = 6.042, p = 0.03, d = 0.6, 20\% \uparrow$ ) was significantly higher than pretest (see Figure 3.2).

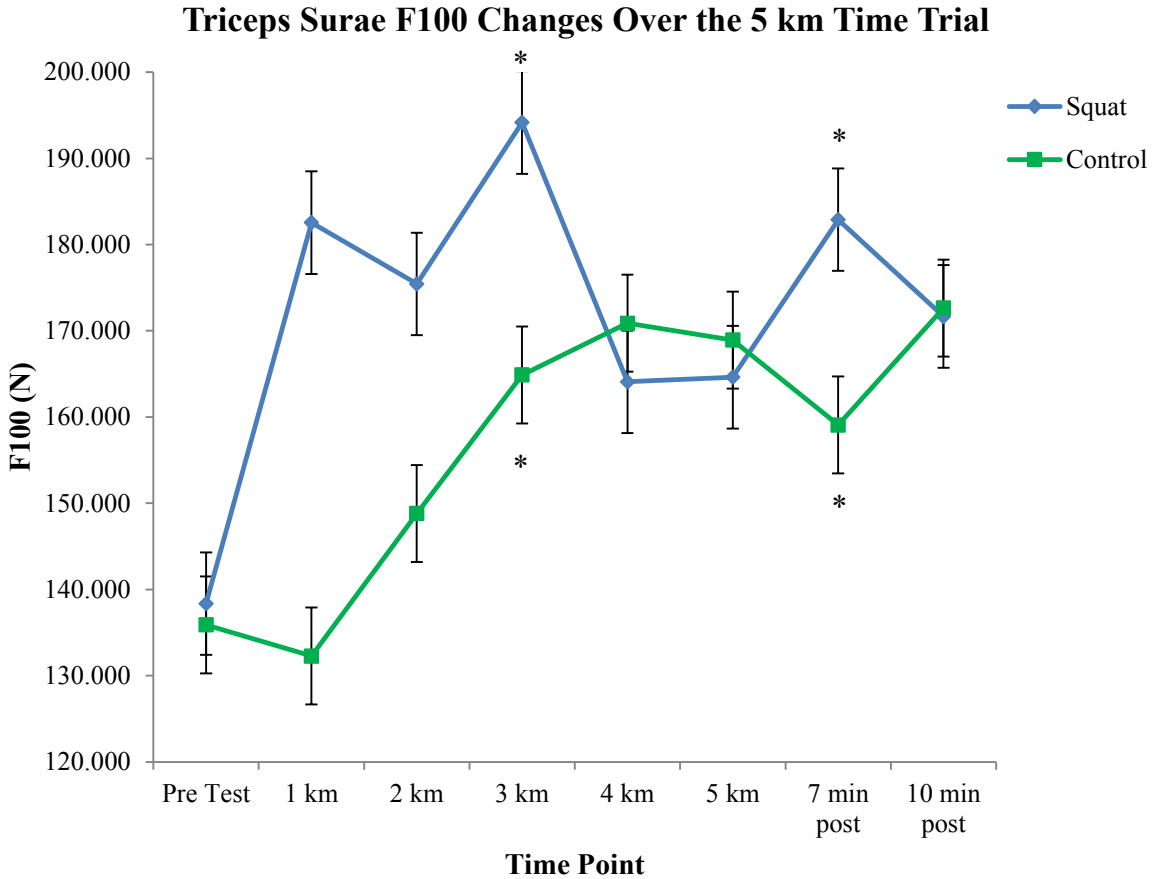


Figure 3.2. Representative graph of force developed in the first 100 milliseconds at each time point. Evident interaction of changes over time. Asterisk (\*) indicates the condition value was significantly greater than pre-test.

### Evoked Contractile Properties

#### *Resting Twitch Force.*

No significant trends, effects, or interactions were observed.

#### *Resting Time to Peak Twitch (TPT) Force.*

There was a main effect of condition for the resting TPT with the 5km time trial ( $F_{(1, 11)} = 3.684, p = 0.05$ ). Between group post-hoc contrasts revealed that resting TPT in the squat



condition ( $F_{(1, 11)} = 3.684, p = 0.05, d = 0.7$ ) was lower than control. The contrasts further revealed that resting TPT in the squat condition at kilometer 3 ( $F_{(1, 11)} = 4.365, p = 0.061, d = 0.5, 5\% \downarrow$ ) and 10 minute post run ( $F_{(1, 11)} = 6.057, p = 0.032, d = 0.6, 5\% \downarrow$ ) was significantly lower than control. The interaction graph (Figure 3.3) revealed that the squat stimulus enhanced resting TPT at kilometer 3 and 10 minutes post run.

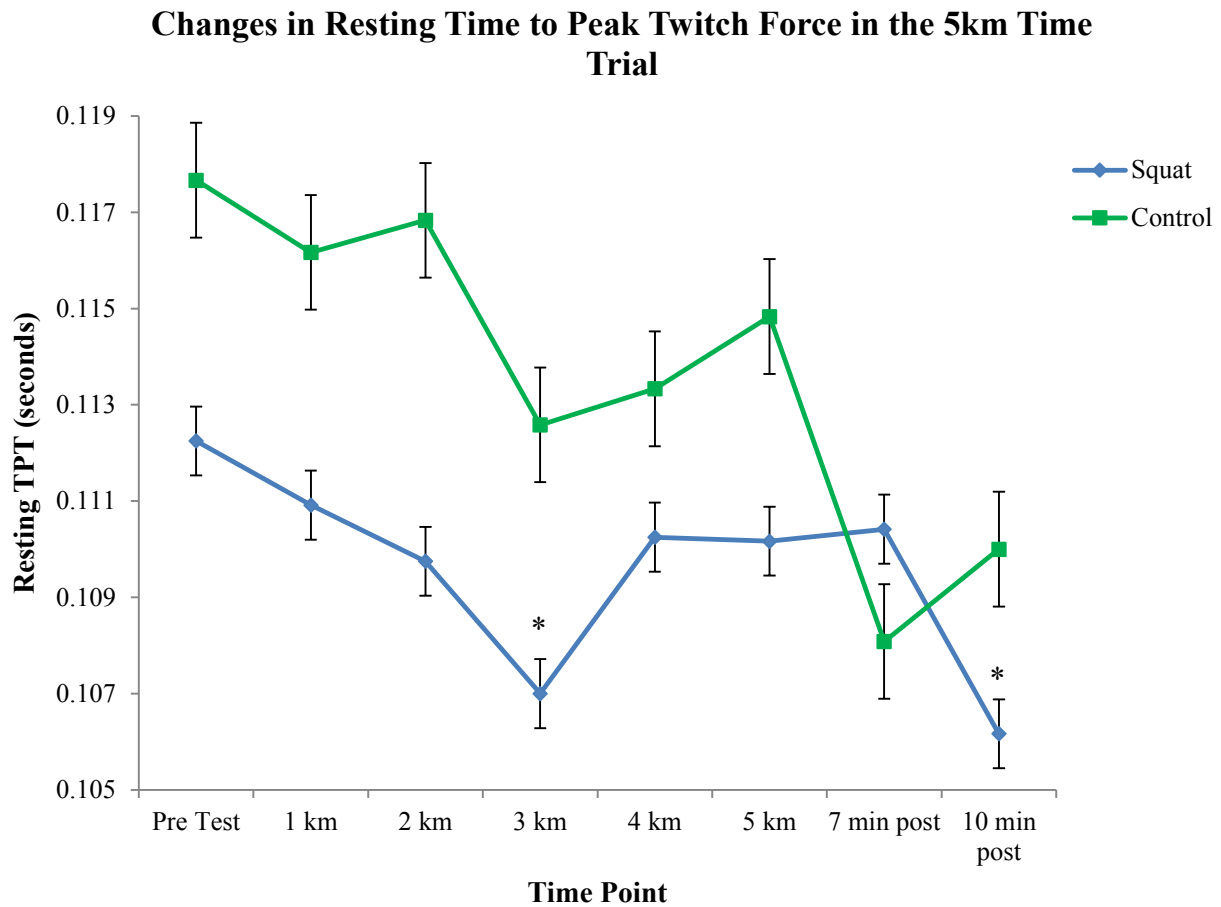


Figure 3.3. Estimated changes (as computed by means) noted between the two conditions. Overall decrease can be noted in the control condition. Asterisk (\*) indicates the squat condition value was significantly greater than pre-test.

**Potentiated Twitch Force.**

No significant trends, effects, or interactions were observed.

***Potentiated Time to Peak Twitch (TPT) Force.***

There was a significant main effect for potentiated TPT observed over time in the time trial ( $F_{(7, 77)} = 2.073, p = 0.05$ ). Contrasts revealed that potentiated TPT at kilometer 4 ( $F_{(1, 11)} = 4.447, p = 0.05, d = 0.54, 6\% \downarrow$ ) was significantly lower than pretest. A near-significant main effect for condition ( $F_{(1, 11)} = 3.908, p = 0.07, d = 0.51$ ) exhibited that potentiated TPT was lower in the squat condition.

***Voluntary Muscle Activation (%).***

There was a significant ( $F_{(7, 77)} = 2.699, p = 0.015$ ) interaction effect between the conditioning stimulus and the data time points for %VA between running trials. Between group post-hoc contrasts revealed significantly greater %VA when comparing squat stimulus to control at 7 minutes post-run ( $F_{(1, 11)} = 4.573, p = 0.05, d = 0.54, 10\% \uparrow$ ) and 10 minutes post-run ( $F_{(1, 11)} = 4.370, p = 0.05, d = 0.53, 11.5\% \uparrow$ ) as compared to pre-test. In the control condition, there was a significant decrease noted in %VA from pretest to 7 minutes post run ( $d = 0.36, p = 0.030, 19\% \downarrow$ ).

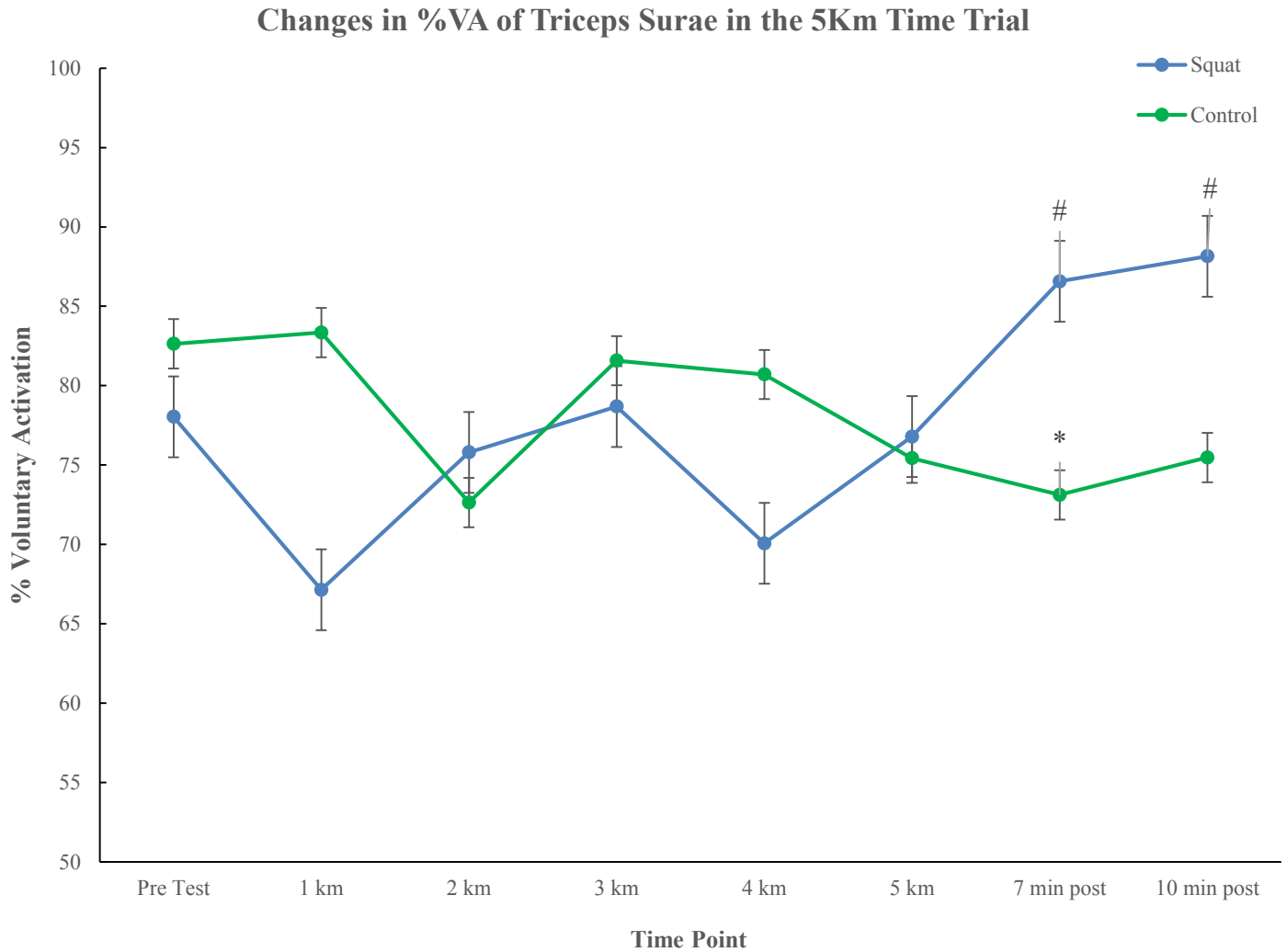


Figure 3.4. Representative graph demonstrating the change in %VA over the course of the interventions. Potentiated can be seen at seven and 10 minutes post run (squats). Asterisk (\*) indicates the control condition value was significantly lower than pre-test. The hashtag (#) symbol indicates the squat condition value was significantly greater than the control condition.

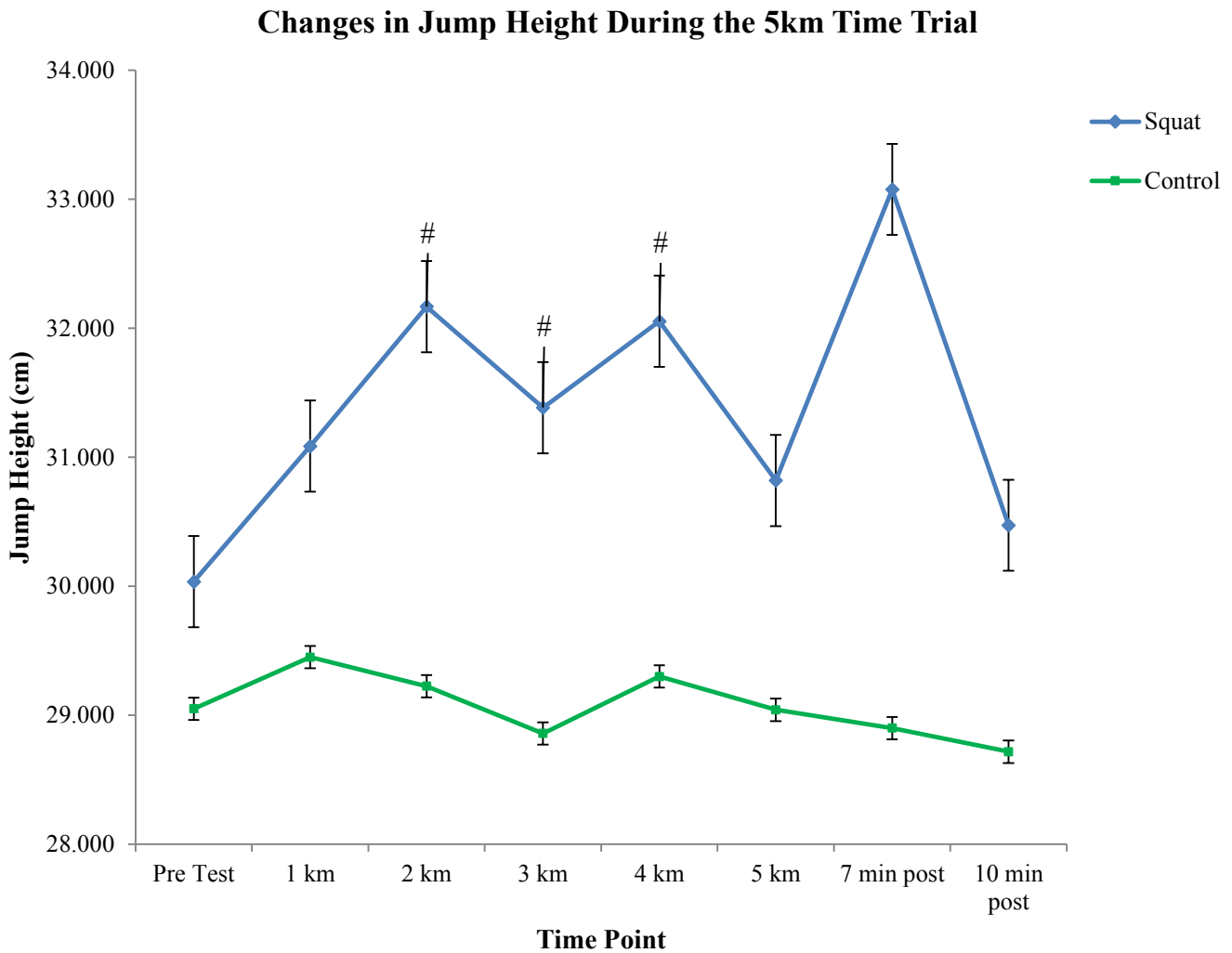
**Drop Jump Performance Measures**

**Contact Time.**

No significant trends, effects, or interactions were observed.

***Drop Jump Height.***

There was a main effect of jump height observed for condition in the time trial ( $F_{(1, 11)} = 6.815, p = 0.02$ ). Between group post-hoc contrasts revealed that jump height in the squat condition was significantly elevated ( $F_{(1, 11)} = 6.815, p = 0.02, d = 0.62$ ) as compared to the control condition. The interaction graph in figure 3.5 reveals that the squat stimulus increased jump height over the course of the entire time trial as compared to the control. When comparing the between group differences in the squat and control conditions, on average participants showed significantly increased drop jump height in the squat condition during kilometer 2 ( $d = 0.47, p = 0.015, 9\% \uparrow$ ), kilometer 3 ( $d = 0.42, p = 0.05, 8\% \uparrow$ ), and in kilometer 4 ( $d = 0.51, p = 0.011, 8.5\% \uparrow$ ) as compared to the control.



*Figure 3.5. Representative graph demonstrating the changes seen in jump height during the squat and control conditions. Significant changes observed in the squat condition. Hashtag (#) symbol indicates the squat condition value was significantly greater than the control condition*

***Reactive Strength Index.***

There was a main effect of time for the reactive strength index ( $F_{(7, 77)} = 2.305, p = 0.035$ ). Between group post-hoc contrasts revealed that reactive strength index was significantly higher at kilometer 2 ( $F_{(1, 11)} = 6.307, p = 0.03, d = 0.6$ ), kilometer 3 ( $F_{(1, 11)} = 13.132, p = 0.004, d = 0.74$ ), kilometer 4 ( $F_{(1, 11)} = 15.032, p = 0.003, r = 0.76$ ), and 7 minutes post run ( $F_{(1, 11)} =$

5.497,  $p = 0.039$ ,  $d = 0.58$ ). Within group contrasts in the squat condition revealed a significant increase in reactive strength index from pretest to kilometer 2 ( $d = 0.35$ ,  $p = 0.046$ , 11.1% ↑), from pretest to kilometer 3 ( $d = 0.35$ ,  $p = 0.019$ , 11.4% ↑), from pretest to kilometer 4 ( $d = 0.51$ ,  $p = 0.001$ , 15.7% ↑), from pretest to kilometer 5 ( $d = 0.38$ ,  $p = 0.008$ , 12.0% ↑), and from pretest to 7 minutes post run ( $d = 0.43$ ,  $p = 0.018$ , 13.5% ↑). Figure 3.6 represents the interaction graph and reveals that the squat stimulus increased reactive strength index throughout the course of the time trial, as compared to control.

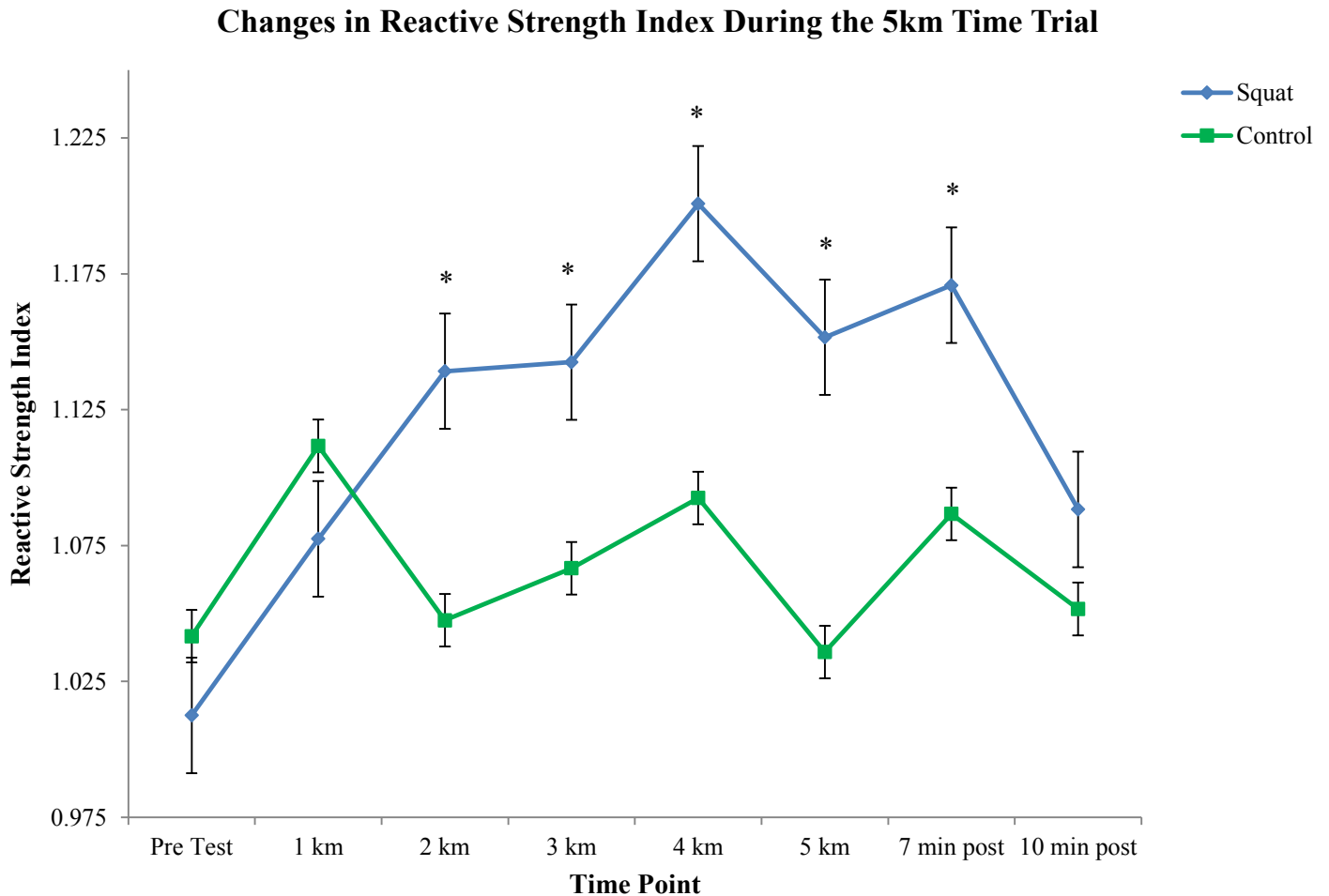
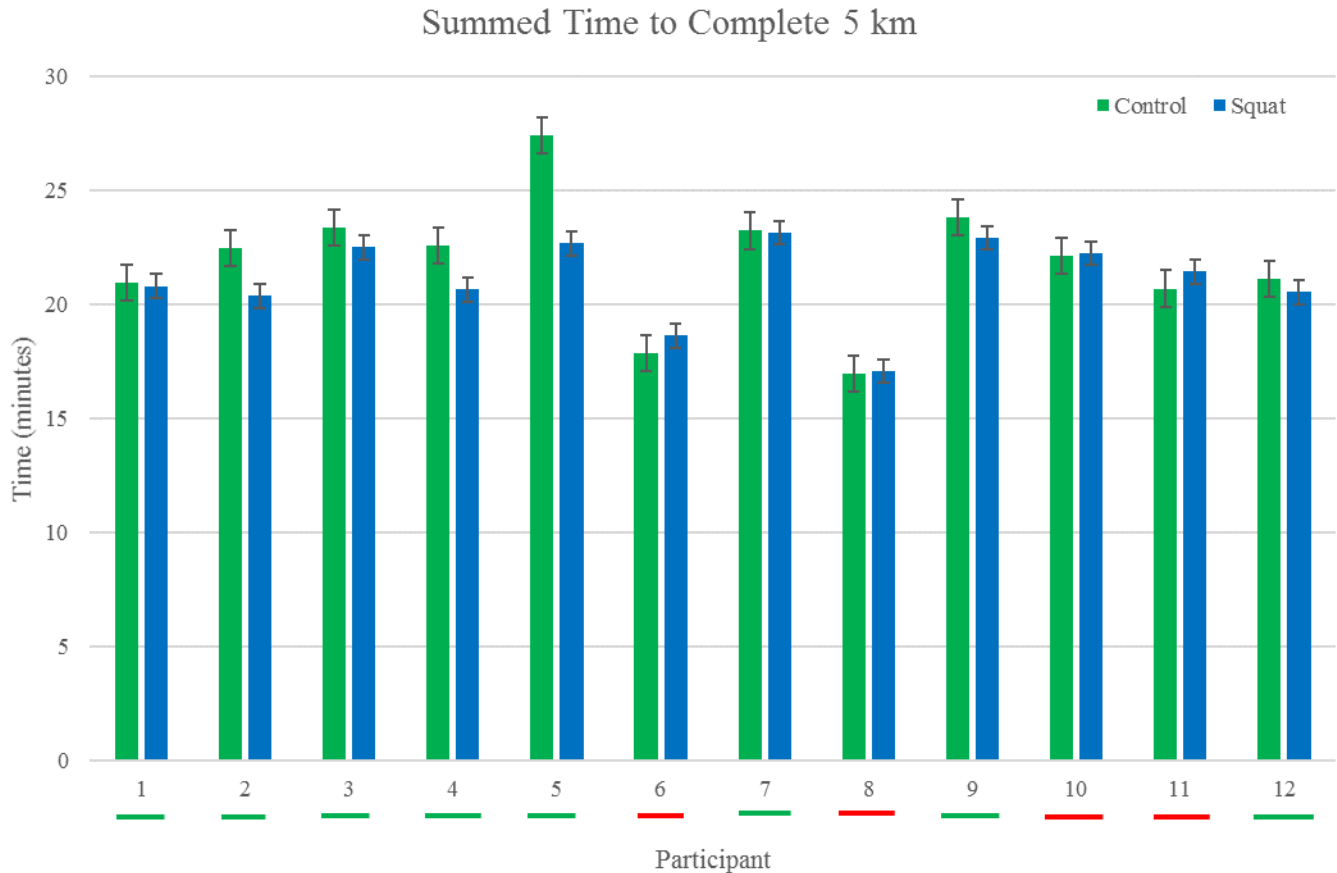


Figure 3.6. RSI means graphed to represent changes over time. Asterisk (\*) indicates the squat condition value was significantly higher than pre-test.

### Time to Complete the Time Trial

Eight out of the twelve participants ran the summed 5 kilometer time trial faster in the



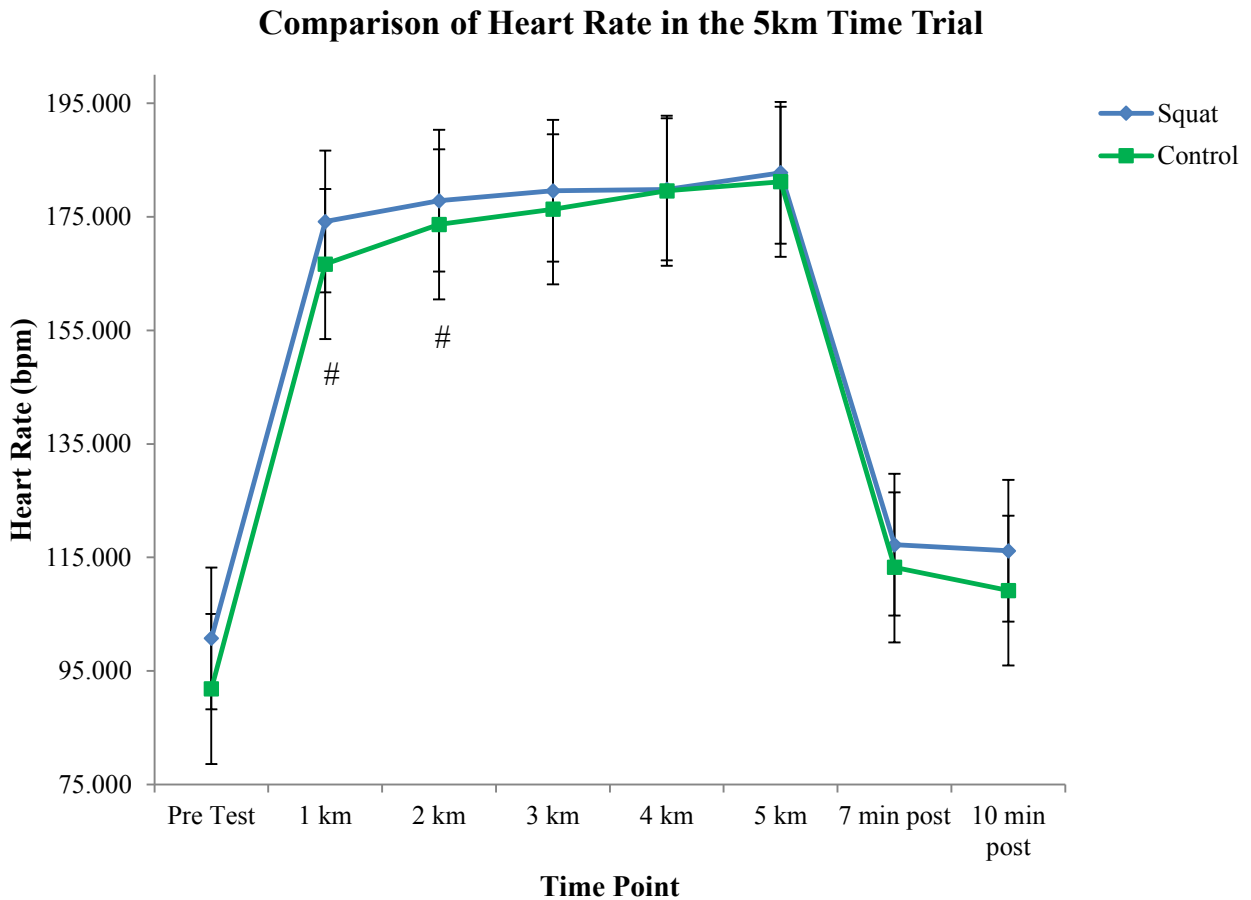
*Figure 3.7. Total summed times for each participant. Green underline indicates a decrease in time from control to squat and red underline indicates an increase in time.*

squat condition than in the control condition (see figure 3.7). The squat condition yielded a 3.6% reduction in the aggregate time to complete the 5 kilometers. A main condition effect ( $F_{(1, 11)} = 3.954, p = 0.05$ ) revealed that the average time to complete each kilometer was less in the squat condition than in the control condition. There was a significant decrease noted from time to complete kilometer 1 in the control condition to time to complete kilometer 1 in the squat condition ( $d = 1.08, p = 0.014, 8\% \downarrow$ ). Further contrasts revealed an interaction effect ( $F_{(4, 44)} = 2.694, p = 0.04$ ) between condition and time that demonstrated an increase in time to complete

the kilometer at kilometer 4 ( $F_{(1, 11)} = 7.974, p = 0.017, d = 0.65, 1.5\% \uparrow$ ) in the control condition.

**Heart Rate**

Figure 3.8 demonstrates the mean average heart rate in each condition throughout the experiment.



*Figure 3.8. Though significant changes were noted in the squat condition, heart rate in both sessions was uniform. Hashtag (#) symbol indicates the squat condition value was significantly greater than the control condition*

There was a main effect of condition for heart rate in the time trial ( $F_{(1, 11)} = 9.742, p = 0.01$ ).

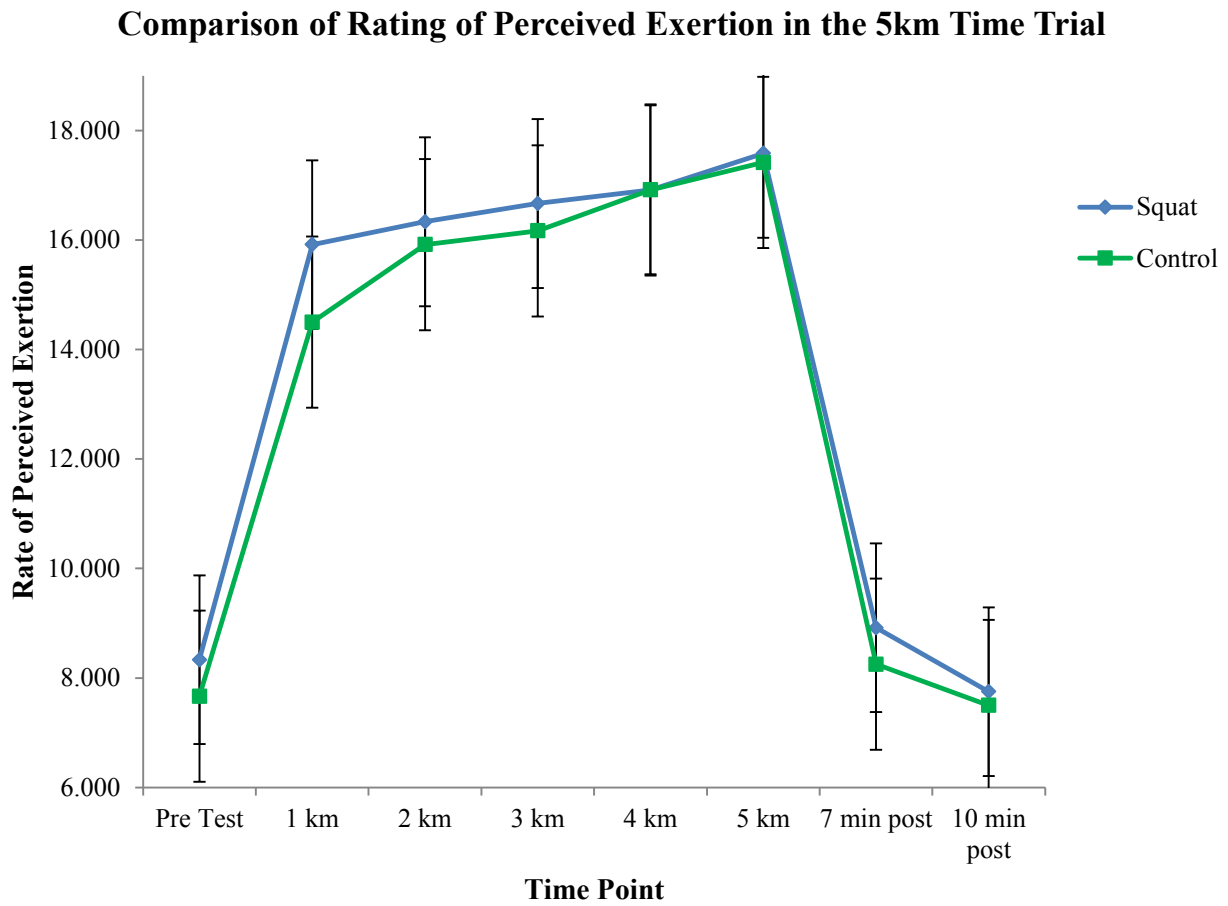
There was a significant increase noted in the heart rate at kilometer 1 in the control condition as compared to the squat condition ( $d = 0.84, p = 0.001$ ). There was another significant increase



noted in the kilometer 2 heart rate in the control condition as compared to the squat condition ( $d = 0.48, p = 0.009$ ).

**Rating of Perceived Exertion**

No significant trends, nor effects, or interactions were observed. Mean RPE was similar in both the squat; 13.552 (0.323) and control; 13.042 (0.387) condition respectively. The interaction graph revealed no significant correlations or interactions.



*Figure 3.9. Representative of the uniform perceived effort (Borg scale) between sessions.*

Tables 3.2 and 3.3 serve to give a comprehensive overview of the significant changes (along with effect sizes) that were noted in the experiment.

### 3.5 Discussion

The intention of the present study was to characterize the effects of a band-resisted jump squat protocol and subsequent five km running trial on the neuromuscular and metabolic properties of the plantar flexors. The main finding was that the PAP conditional stimulus incorporated into a warm-up protocol prior to the time trial run improved aggregate time to complete 5 km run trials better than a standard running-specific warm-up. Factors contributing to this improvement could be related to jump squat conditioning stimulus-induced increases in voluntary activation at seven and 10 minutes post-run, MVC force at four and ten minutes post-run, MVC F100 at three and seven minutes post-run, potentiated TPT at kilometers 4, drop jump height at kilometers 2, 3 and 4, as well as reactive strength index at kilometers 2-5, immediately and 7 min post-run. These performance improvements contrasted with control condition decrements in triceps surae MVC F100, potentiated time to peak twitch force, and time to complete the kilometer repeats.

Perhaps the most notable from these findings is the effect of the physiological and performance changes that combined to produce a faster time to complete kilometer one in the squat condition, as well as a 3.6% reduction in the aggregate 5 kilometer time after performing band-resisted squats as compared to the control. Principally, it is also important to note that the athletes uniformly performed the training protocol according to the criteria and intensity that was required. The similar changes in the descriptive metabolic measures illustrated the increased intensity during the intervention sessions, (RPE:  $17.5 \pm 0.31$ ; HR<sub>peak</sub>:  $181.96 \pm 1.85$ ; HR<sub>ave</sub>:  $151.25 \pm 2.07$ ) with no significant differences overall between squat and control condition. This fact eliminates and negates the possibility that these changes could be caused by varying levels of involvement between the two sessions.

To the author's knowledge, this study is the first to demonstrate such a result in an endurance-running population though similar studies conducted in endurance cyclist populations support this main finding. Comparatively, in a study with trained cyclists, it was found that adding a 5RM leg press strength exercise bout to a 5-minute warm-up improved performance in a subsequent 20-km cycling time trial (Silva et al. 2014). This study found no significant difference in blood lactate measures, but suggested that the improved performance (~6.1% reduction in time to complete the trial) after heavy resistance was explained by improved cycling economy. The authors also suggested the possibility that, in addition to the neuromuscular effects observed, an increased capacity to access aerobic performance reserves could also aid in explaining increased performance during the trial. In another related study, Palmer et al. (2008) examined the effects of prior heavy exercise (5 minutes of heavy cycling at 50% difference between lactate threshold and  $\dot{V}O_{2peak}$ ) in well-trained cyclists but measured performance in a subsequent 4-km cycling time trial. Athletes were subjected to either no prior exercise, prior heavy exercise, or self-selected prior exercise, all conducted on a cyclist ergometer. Similar to Silva et al., they found faster times to complete the trial post-heavy exercise although different from the Silva study was the overall increase in the mean power output (5.4%) post-heavy exercise.

Specific to the population of interest, these effects were also observed in a study examining the effects of two different warm-up protocols on the neuromuscular systems of well-trained distance runners (Skof and Strojnik et al. 2007). This study found that adding sprinting and bounding exercises to an athletes' warm-up would improve neuromuscular efficiency via PAP, though the authors did not rule out increased efficiency of the central nervous system (improved central input or enhanced excitation of the  $\alpha$ -motor neuron) as a possible mechanism.

Thus it is plausible that increased strength, improved voluntary activation, lack of impairment in contractile processes, equivalence in metabolic factors, and improved task-specific efficiency are responsible for the improved run times to completion seen in the squat condition. Further to performance adjustments, this multi-factorial explanation naturally includes peripheral and central adjustments and has been examined previously (Girard et al. 2012). Peripheral adjustments include changes in sarcolemmal excitability and excitation-contraction coupling, while central adjustments can be seen in the neuromuscular activity and voluntary activation.

Central adjustments were manifest in the present study as the results show that plantar flexor MVC in the squat condition increased by 8% at kilometer four and 9.5% 10 minutes post-run. The jump squat condition revealed increased F100 at various points throughout the subsequent running trial, EMG remained unchanged while voluntary activation (ITT) increased. As EMG and contractile force are key features of fatigue, this could suggest that a bout of band-resisted squat jumps could increase muscle performance by overcoming possible onset of fatigue. This idea of unchanged (or increased) central properties supporting endurance activities has been demonstrated in other running studies (Place et al. 2004; Ross et al. 2010; Skof and Strojnik et al. 2006), cycling studies (Booth et al. 1997; Sahlin et al. 1986), and intermittent isometric exercises (Vallestad et al. 1997). This unchanged, or increased neural drive, is in stark contrast to the results of previous studies that found a 10-30% decrease in contractile force (plantar flexors MVC) after completing running protocols lasting from one to several hours (Avela et al. 1999; Racinais et al. 2007; Saldanha et al. 2008). For example, Millet et al. (2011) found a 30-39% decrease in force production following an extreme running protocol lasting longer than 24 hours. These studies would suggest that central fatigue could be observed in endurance activities lasting longer than one hour, which was not the case in the present study.

Perhaps most relevant to this study are the contrasting results presented by Girard et al. (2012) and Nummela et al. (2006) where a decrease in plantar flexors (27%) and knee extensors (15%) MVC force was revealed after 5 km running time trials. A notable difference between these two studies and the present study is the 5 versus 3 (present study) minutes of neuromuscular testing that were conducted between each of the five-kilometer repeats. Though this testing included maximal contractions and plyometric testing, heart rate profiles notably dropped between each repeat. In a review, Behm (2004) further explains that a preceding contraction could provide improved central adjustments via motoneuron excitation from both supraspinal and afferent input. This review gives evidence that supraspinal motor-evoked potentials have been facilitated in studies of different durations and intensities (Balbi et al., 2002). Further evidence is presented in the review that suggests that at the spinal level, short term increases in explosive force following a conditioning contraction (MVC) are attributable to enhanced neuromuscular activation (Gullich and Schmidtbleicher et al. 1996). These changes are supported by observed increases in reflex amplitude 10 minutes post conditioning contraction (Trimble and Harp et al. 1998), and the possible 30% contribution of the motoneuron excitation to the discharge frequency from Ia afferents of the intrafusal stretch receptors (Gandevia, 1998).

Another possible explanation arising from the results of the well-trained runners in the present study is the presence of a centrally mediated pacing strategy that aims to inhibit substantial deterioration of muscle force, augmenting possible PAP effects. Noakes et al. (2004) suggests that at, or shortly after, the onset of exercise of different durations and intensities, well-trained endurance athletes adopt a centrally mediated pacing strategy in order to complete the activity as efficiently as possible, while maintaining physiological capacity. In the case of the current study, the results suggest that the participants effectively avoided significant strength loss

despite instructions to complete the running trial as fast as possible. This is not surprising as a review (Fuller et al. 1996) of pacing strategies demonstrates that well trained athletes effectively employ racing strategies that aid their ability to maintain a steady submaximal physiological effort. This idea of improved running economy during paced sub maximum activities is in line with the review by Behm (2004) wherein he indicates that PAP may be most evident with submaximal contractions. As an athlete employs an effective pacing strategy to maintain lower sub maximal frequency stimulation, suboptimal  $Ca^{2+}$  activation (less than full release of  $Ca^{2+}$  from the sarcoplasmic reticulum) may be present, enabling effects of PAP. This is explained by myosin regulatory light chain (RLC) phosphorylation (Grange et al., 1993; Houston et al. 1985; Houston and Grange, 1990; Sweeney et al., 1993) that takes place at the peripheral level under conditions of suboptimal  $Ca^{2+}$  activation and is thought to increase the number of force-producing cross-bridges (Sweeney et al., 1993), enabling an enhanced running economy. It follows then that any centrally mediated pacing strategy presented here could be developed from previous running experience and training. Thus, with the training background of the participants in this study, it is important to note that the results are specific to this type of athlete rather than less-trained individuals.

The lack of fatigue presented at the central level suggests and furthers the possibility of PAP performance improvements occurring at the peripheral level. The most notable finding at the peripheral level was a lack of impairment of the evoked contractile temporal properties, which is in line with other studies examining the peripheral effects of running (Millet et al. 2002; Pageaux et al. 2017; Ross et al. 2009; Vuorimaa et al. 2005). In fact, with the presented results it could be argued that evoked contractile temporal properties showed a tendency towards improving over the course of the running protocol. Potentiated TPT and resting TPT both

decreased by ~5% at kilometers 3-4 and 10 minutes post run, while percent voluntary activation rose in the squat condition by five and ten minutes post run. This enhanced contractile capacity has been demonstrated in endurance athletes after performing endurance exercises of different durations (Finni et al. 2003; Millet et al. 2002; Nicol et al. 1991). This was initially evident after a 65 km ultra-marathon running event in which a severely depressed maximal voluntary force capacity was reported concurrently with a surprising potentiated twitch mechanical response and shorter contraction/half-relaxation times (Millet et al. 2002).

These results are contradicted by a study examining the fatigue effects of endurance running (Nicol et al. 1991). Nine experience male runners were recruited to run a timed marathon with neuromuscular tests pre and post. In four out of the five parameters, the post-marathon performance values were lower. Sprint velocity, MVC force, ground reaction forces, and stride profiles were all negatively affected by the marathon intervention. This coincides with a study conducted by Finni et al. (2003) where reductions in EMG amplitude, superimposed doublet twitch, and rate of force development were found after participants completed a 10-km running trial. A notable difference between these studies and the present one is the timing of the neuromuscular tests, as the present study required testing at each kilometer interval instead of a basic pretest and posttest.

Furthermore, another obvious difference between these studies is the selected distance. Exertion during a marathon distance of 42 kilometers is more likely to cause low frequency fatigue and is presumably greater than that of a five kilometer time trial. Though these differences could explain the contradicting results, peripheral PAP effects have been demonstrated in other studies examining performance measures at intervals during running trials (Del Rosso et al. 2016). Additionally, in a similar study with endurance runners, participants ran

6km at anaerobic threshold ( $V_{OBLA}$ ) and the results showed no sign of contractile enhancement, suggesting a balance of fatigue and potentiation via an impairment of the excitation-contraction coupling (Skof et al. 2006a). The mean maximum twitch torque, EMG activation, and maximal voluntary force either dropped or showed no change at both stimulation frequencies though maximum twitch torque was enhanced 10 minutes post run. Skof et al. (2006b) performed another study shortly after, that demonstrated potentiating effects in twitch torque 10 minutes after a 5 X 300 m interval workout despite evidences of low-frequency fatigue.

With the nature of the shorter distance being run at a higher intensity in the present study, low frequency fatigue would have been the most likely to prevail. The lack of impairments and prevalence of improvements (increase interpolated and potentiated twitch force) would suggest that low frequency fatigue was not manifested to a great extent in the current study. An explanation for this finding could come from the proposed mechanism of PAP (membrane excitability and excitation-contraction coupling). Peripheral fatigue is often linked to excitation-contraction coupling failure and correlates with the reduced efficiency of the calcium cycle (Fitts et al. 1991). Sahlin et al. (1986; 1989) provided evidence that intracellular pH is directly connected to calcium cycle efficiency. Consequently, a drop in pH resulting from increased amounts of metabolites like  $H^+$ , ADP, Pi can inhibit the release of  $Ca^{2+}$  into the sarcoplasmic reticulum (Fitts et al. 1991; Metzger et al. 1987) and could reduce  $Ca^{2+}$  affinity for troponin (Hultman et al. 1986), reducing the muscle contractile force. The fact that the contractile properties remained the same, or increased demonstrates that peripheral fatigue was either not present or its effects were masked or balanced due to the effects of PAP. When calcium molecules are released during a muscle contraction, myosin light chain kinase is released, which catalyzes the phosphorylation of the RLC's at the end of the myosin head (Tillin et al. 2009).



The PAP-induced phosphorylation of the RLC is believed to potentiate the following contractions by causing it to move away from its thick filament backbone, more poised to efficiently interact with the actin (Hodgson et al. 2005; Szczesna et al. 2003). It is these proposed molecular-level changes that are thought to explain the noted enhancement in evoked contractile temporal properties and consequent decrease in time to complete the running trials.

Possibly, the most significant effects of PAP leading to faster run times were evident in the increased performance of the drop jump. This was manifest in the 15.7% increase in reactive strength index from pre-test to kilometer four in the squat condition. A 9% increase in jump height from pre-test to kilometer 1, 8% in kilometer two, and 8.5% in kilometer four in the jump squat condition also provide evidence for PAP. This is supported by two studies by Paavolainen et al. (1999; 2003) where positive correlations between jump performance and velocity in running a five kilometer time trial were observed. These results suggest that rapid force production can be beneficial not only for jumpers and sprinters but also for endurance runners. It is hypothesized that the protocol of band-resisted squat jumps performed prior to the running trial improved central and peripheral adjustments via PAP but that the performance adjustments can also be explained through a proposed leg stiffness-SSC model. It is well established that leg stiffness and SSC can contribute to more efficient running economy (Nicol et al. 2006). In this case, leg stiffness directly affects the SSC as increased leg stiffness gives rise to increased muscle force and power produced during SSC. This is evident as leg stiffness depends on the series elastic component (SEC) during the stretching phase (Rabita et al. 2008). Related studies have examined the relationship between the SSC (including leg stiffness) and continuous running performance. For example, Heise et al. (1998) observed an inverse relationship between leg stiffness and running economy and concluded that those with a less economical running style

were more compliant during ground contact. Likewise, Dalleau et al. (1998) discovered that energy cost during treadmill running was significantly correlated to the stiffness of the propulsive leg. The authors of these studies state that leg stiffness can be a good indicator of an individual's reactive power. These same authors further explain that increased leg stiffness would increase ability to absorb and reconstitute power during a forceful SSC (as seen in running). Furthermore, an increase in reactive power from increased leg stiffness would render the SSC more efficient and consequently decrease the energy cost of running, enabling a faster pace and time. Moreover, findings of other studies (Cleak et al. 1992; Foure et al. 2010; Spurrs et al. 2003; Torres et al. 2013) examining increased leg stiffness following intense exercise bouts could imply that the band-resisted jump squats served to increase leg stiffness and consequent running economy. It is on this basis that we propose that the increased jump height, reactive strength index, and subsequent time to complete the kilometer repeats can also be explained by an increased leg stiffness and reactive power, rendering the SSC more effective and consequently more economical, enabling the participant to run at a faster pace.

### **3.6 Perspectives**

The results of the present study provide strong evidence for physiological and running performance improvements following a band-resisted squat jump protocol. The metabolic measures employed herein were sufficient for comparing exercise intensities between sessions although future studies could include more in-depth measures (i.e. blood lactate) to verify metabolic responses. In regards to the testing measures, more familiarization could have been given to participants in regards to the maximal contraction test as there could have been a learning effect throughout the course of the study. The distribution of the number of participants familiar with performing MVC's seemed to be uniform, though more attention could have been

given in familiarizing those unfamiliar with the procedure. As with any study conducted on a treadmill in a laboratory setting, it is also important to note that different results could be obtained from conducting a similar study in a free-living environment. There is also potential for error within the results when participants are not familiar with treadmill running. We aimed to eradicate this error by conducting multiple familiarization sessions that included treadmill running and all participants appeared comfortable by the start of the first session. We also attempted to control for pre-conceived pacing notions by blinding the participants to speed, time, and pacing. It was the hope that participants would then run according to their internal physiological gauge instead of an external influence. The results from this experiment are of particular importance to coaches, athletes and practitioners, who must consider the most effective method of warming up to increase and maintain performance. Future research should continue to investigate various conditioning protocols that have the potential to illicit a PAP response in endurance athletes. With the proper technology, researchers could simultaneously investigate these same effects in participants while they perform the running trial without dismounting the treadmill for testing. Additionally, research comparing these effects to females, or populations with varying training status, merits inclusion in future studies in this field.

### **3.7 Conclusion**

This is the first study to attempt to examine the effects of a conditioning stimulus on a subsequent 5 x 1 kilometer time trial in well-trained endurance athletes. Neuromuscular and performance changes resulting in decreased time to complete the running task, increased force generation, voluntary activation, and evoked contractile temporal properties are likely due to changes in excitation-contraction coupling (PAP), central pacing strategies, and possible increased SSC efficiency. These changes appear to be dependent upon the conditioning

contraction as there were little to no significant changes noted in the control condition. It is plausible that the increase in performance following the conditioning contraction could be attributed to the expectations of the participants but the measurement observations demonstrated that the participants were not concerned with the outcome. The transient nature of the improvements observed post-conditioning contraction suggest that there are multiple factors responsible for mediating the changes in neuromuscular and performance measures during running. Based on the evidence presented, the researchers affirm the hypothesis that performing a 5RM weighted jump squat protocol as part of a standardized running-specific warm-up will induce significant measurable PAP effects during the course of a subsequent 5 km time trial run and up to 7 minutes post-run protocol.

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### **Conflicts of Interest**

The authors declare no conflicts of interest with the information contained within this manuscript.

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### 3.9 Tables

Table 3.1. Intra-session descriptive and reliability measures. R = range, ICC = intraclass correlation coefficient.

<b>Measure</b>	<b>Mean [R]</b>	<b>Intra-session ICC</b>
Gastrocnemius EMG	0.284 (0.066)	0.957
Soleus EMG	0.180 (0.040)	0.963
Triceps Surae MVC	665.331 (77.534)	0.984
Triceps Surae F100	164.200 (61.887)	0.976
Evoked Twitch Force	82.120 (23.917)	0.962
ITT Force	27.820 (36.218)	0.798
Potentiated Twitch Force	98.391 (30.511)	0.954
ITT % Voluntary Activation	66.661 (52.907)	0.763
Contact Time	300.083 (34.500)	0.994
Flight Time	490.677 (32.167)	0.991
Drop Jump Height	30.227 (4.358)	0.983
Reactive Strength Index	1.095 (0.188)	0.989
Time to Complete Kilometer	257.150 (22.750)	0.969
Heart Rate	151.25 (90.917)	0.943
Rate of Perceived Exertion	13.297 (10.083)	0.905



Neuromuscular Alterations in Middle-Distance Runners

Evidence for PAP Arising from Within Condition Effects (as compared to baseline)

	1 KM	2 KM	3 KM	4 KM	5 KM	7 MIN POST	10 MIN POST	Overall
MVC				↑ (0.29)			↑ (0.36)	
F100			↑ (0.67)			↑ (0.60)		
Potentiated TPT				↓ (0.54)				↓ (0.51)
%VA						↑ (0.36)		
Reactive Strength Index		↑ (0.35)	↑ (0.35)	↑ (0.51)	↑ (0.38)	↑ (0.43)		

Table 3.2 Effect sizes are illustrated for significant within condition interactions. White background/black text = squat condition, black background/white text = control condition, polka dot background = observed main time effect.

Evidence for PAP Arising from Between Condition Effects

	1 KM	2 KM	3 KM	4 KM	5 KM	7 MIN POST	10 MIN POST	Overall
MVC							↑ (0.55)	
F100								
Resting TPT			↑ (0.50)				↓ (0.60)	↓ (0.70)
Potentiated TPT								↓ (0.51)
%VA						↑ (0.54)	↑ (0.53)	
Jump Height		↑ (0.47)	↑ (0.42)	↑ (0.51)				↑ (0.62)
Reactive Strength Index		↑ (0.60)	↑ (0.74)	↑ (0.76)		↑ (0.58)		
Time to complete km	↓ (1.08)			↑ (0.36)				↓

Table 3.3 Effect sizes are illustrated for significant between condition interactions. White background/black text = squat condition, black background/white text = control condition, polka dot background = observed main time effect.

3.10 Figures



Figure 3.10. Experimental set-up and positioning of the resistance-bands for the squat jumps. The bands were attached with carabiners to heavy dumbbells and were then looped cross-body and positioned on the opposite shoulder of the side they were attached to.



Figure 3.11. Experimental set-up and method for conducting the 30-centimeter drop jump test.



Figure 3.12. Experimental set-up and method for measuring plantar flexors MVC using the boot apparatus.



Figure 3.13. Depiction of the placement of the stimulating (popliteal fossa) and recording electrodes.



Figure 3.14. Experimental set-up for the time trial kilometer repeats conducted on the treadmill.

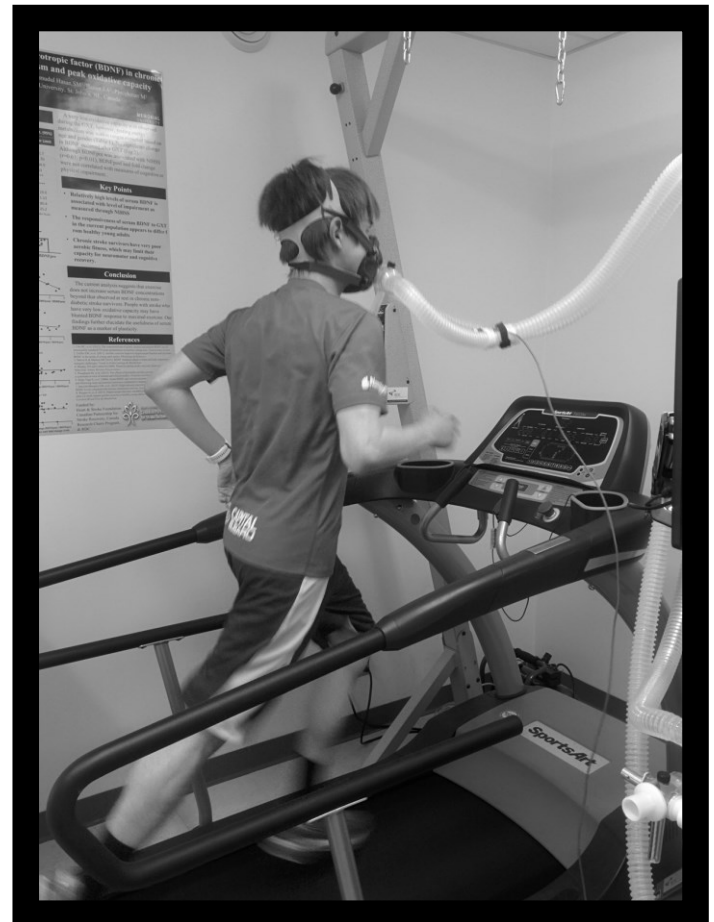
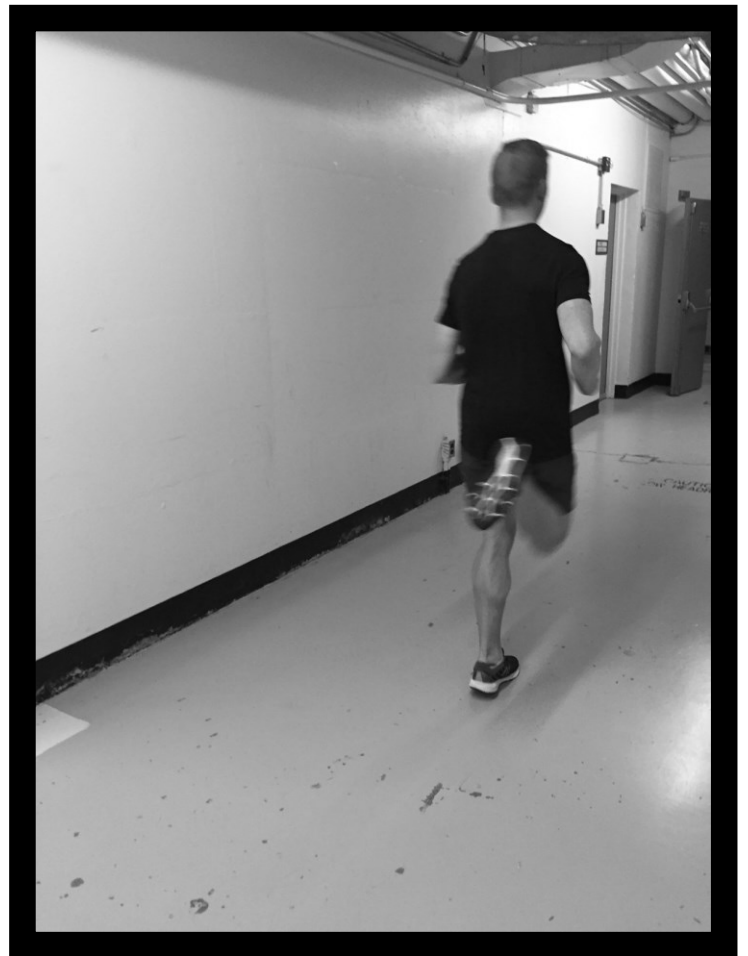


Figure 3.15. Photo of the set-up for conducting the  $\dot{V}O_2$  maximum test.



A



B

Figure 3.16. Photos of portions of the running-specific warm-up. *A*) shows the ‘A’s’ (high knees) while *B*) shows the ‘C’s’ (heel to glute kicks).



### 3.11 Appendix

Resultant contrast means, differences, and confidence intervals are displayed below as tables stratified into test measures (M = means, SE = standard error, BCa 95%/CI = confidence interval).

MVC					
Squat- 10 min post	M = 680.09, SE = 57.72	Squat- Pretest	M = 615.61, SE = 51.98	Squat- Pretest	M = 615.61, SE = 51.98
Control- 10 min post	M = 659.88, SE = 50.33	Squat- Km 4	M = 668.69, SE = 56.05	Squat- 10 min post	M = 615.61, SE = 51.98
Difference [CI]	-64.48, BCa 95% [-124.04, -4.93]	Difference [CI]	-53.09, BCa 95% [-101.29, -4.89]	Difference [CI]	-64.48, BCa 95% [-124.04, -4.93]
Significance	$t(11) = -2.38, p = 0.036$	Significance	$t(11) = -2.424, p = 0.034$	Significance	$t(11) = -2.383, p = 0.036$

Table 1.1. Triceps surae MVC 'between and within group' contrasts results with means, standard errors, test statistics, and confidence intervals included.

F100					
Squat- Pretest	M = 138.35, SE = 19.21	Control- Km 1	M = 132.28, SE = 21.63	Control- Pretest	M = 135.90, SE = 21.56
Squat- Km 3	M = 194.16, SE = 34.35	Squat- Km 1	M = 182.55, SE = 30.76	Control- Km 4	M = 170.88, SE = 23.37
Difference [CI]	-55.81, BCa 95% [-108.79, -2.83]	Difference [CI]	50.27, BCa 95% [0.144, 100.4]	Difference [CI]	-34.98, BCa 95% [-67.79, -2.17]
Significance	$t(11) = -2.319, p = 0.04$	Significance	$t(11) = 2.207, p = 0.049$	Significance	$t(11) = -2.347, p = 0.039$

Table 1.2. Triceps surae F100 'between and within group' contrasts results with means, standard errors, test statistics, and confidence intervals included.

Potentiated TPT Force					
Control- Pretest	M = 0.105, SE = 0.0031	Control- Pretest	M = 0.105, SE = 0.0031		
Control- Km 3	M = 0.0978, SE = 0.0014	Control- Km 4	M = 0.09783, SE = 0.00125		
Difference [CI]	0.0073, BCa 95% [-0.00003, 0.01453]	Difference [CI]	0.00717, BCa 95% [0.00009, 0.01425]		
Significance	$t(11) = 2.192, p = 0.05$	Significance	$t(11) = 2.228, p = 0.048$		

Table 1.3. Potentiated TPT 'between and within group' contrasts results with means, standard errors, test statistics, and confidence intervals included.

%VA					
Squat- 10 mins post	M = 88.15, SE = 3.59	Control- Pretest	M = 82.63, SE = 6.82		
Control- 10 mins post	M = 75.48, SE = 5.89	Control- 7 mins pot	M = 69.44, SE = 10.49		
Difference [CI]	12.67, BCa 95% [2.15, 23.20]	Difference [CI]	13.19, BCa 95% [1.54, 24.85]		
Significance	$t(11) = 2.650, p = 0.023$	Significance	$t(11) = 2.493, p = 0.030$		

Table 1.4. Triceps surae %VA 'between and within group' contrasts results with means, standard errors, test statistics, and confidence intervals included.

Drop Jump Height							
Squat- Km 2	$M = 32.17, SE = 2.07$	Squat- Km 3	$M = 31.38, SE = 2.17$	Squat- Km 4	$M = 32.05, SE = 1.97$	Squat- Pretest	$M = 30.04, SE = 2.14$
Control- Km 2	$M = 29.23, SE = 1.79$	Control- Km 3	$M = 28.86, SE = 1.73$	Control- Km 4	$M = 29.30, SE = 1.57$	Squat- Km 2	$M = 32.17, SE = 2.07$
Difference [CI]	2.94, BCa 95% [0.71, 5.18]	Difference [CI]	2.53, BCa 95% [-0.017, 5.07]	Difference [CI]	2.75, BCa 95% [0.76, 4.75]	Difference [CI]	-2.13, BCa 95% [-4.12, -0.15]
Significance	$t(11) = 2.895, p = 0.015$	Significance	$t(11) = 2.187, p = 0.05$	Significance	$t(11) = 3.040, p = 0.011$	Significance	$t(11) = -2.361, p = 0.038$

Table 1.5. Drop jump height ‘between and within group’ contrasts results with means, standard errors, test statistics, and confidence intervals included

RSI							
Squat- Km 4	$M = 1.20, SE = 0.104$	Squat- Km 5	$M = 1.15, SE = 0.107$	Squat- Pretest	$M = 1.01, SE = 0.106$	Squat- Pretest	$M = 1.01, SE = 0.106$
Control- Km 4	$M = 1.09, SE = 0.098$	Control- Km 5	$M = 1.04, SE = 0.095$	Squat- Km 2	$M = 1.140, SE = 0.113$	Squat- Km 3	$M = 1.142, SE = 0.114$
Difference [CI]	0.108, BCa 95% [0.009, 0.208]	Difference [CI]	0.116, BCa 95% [0.029, 0.202]	Difference [CI]	-0.127, BCa 95% [-0.251, -0.002]	Difference [CI]	-0.130, BCa 95% [-0.234, -0.026]
Significance	$t(11) = 2.398, p = 0.035$	Significance	$t(11) = 2.963, p = 0.013$	Significance	$t(11) = -2.245, p = 0.046$	Significance	$t(11) = -2.746, p = 0.019$
						Squat- Pretest	$M = 1.01, SE = 0.106$
						Squat- Km 5	$M = 1.152, SE = 0.107$
						Difference [CI]	-0.139, BCa 95% [-0.235, -0.044]
						Difference [CI]	-0.158, BCa 95% [-0.283, -0.033]
						Significance	$t(11) = -3.204, p = 0.008$
						Significance	$t(11) = -2.790, p = 0.018$

Table 1.6. Reactive strength index ‘between and within group’ contrasts results with means, standard errors, test statistics, and confidence intervals included

Time to Complete Km Trial	
Squat- Km 1	$M = 251.00, SE = 5.77$
Control- Km 1	$M = 272.50, SE = 10.92$
Difference [CI]	-21.50, BCa 95% [-37.62, -5.38]
Significance	$t(11) = -2.936, p = 0.014$

Table 1.7. Time to complete the kilometer trials ‘between and within group’ contrasts results with means, standard errors, test statistics, and confidence intervals included

Heart Rate					
Squat- Pretest	$M = 100.750, SE = 3.27785$	Squat- Km 1	$M = 174.167, SE = 2.581$	Squat- Km 2	$M = 177.833, SE = 2.262$
Control- Pretest	$M = 91.833, SE = 3.266$	Control- Km 1	$M = 166.667, SE = 2.568$	Control- Km 2	$M = 173.667, SE = 2.518$
Difference [CI]	8.91667, BCa 95% [1.599, 16.234]	Difference [CI]	7.500, BCa 95% [3.962, 11.038]	Difference [CI]	4.167, BCa 95% [1.276, 7.057]
Significance	$t(11) = 2.682, p = 0.021$	Significance	$t(11) = 4.666, p = 0.001$	Significance	$t(11) = 3.173, p = 0.009$

Table 1.8. Heart rate ‘between and within group’ contrasts results with means, standard errors, test statistics, and confidence intervals included