



**UNIVERSITY OF  
STIRLING**

**NEUROMUSCULAR MARKERS OF HIGH PERFORMANCE SPORT  
PREPARATION: MUSCLE CONTRACTILE MECHANICS**

BY

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*“By failing to prepare, you prepare to fail”.*

Benjamin Franklin

politician and scientist

(1706 – 1790)

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## List of Abstracts

### Radial Displacement Assessment of Skeletal Muscle Contractile Properties Following Electrically Elicited Fatigue

**Background:** Assessments of skeletal muscle functional capacity often necessitate maximal contractile effort, which exacerbates muscle fatigue or injury. Tensiomyography (TMG) has been investigated as a means to assess muscle contractile function following fatigue; however observations have not been contextualised by concurrent physiological measures. The aim of Chapter 2 was to measure peripheral fatigue-induced alterations in mechanical and contractile properties of the plantar flexor muscles through non-invasive TMG concurrently with maximal voluntary contraction (MVC) and resting muscle tension (RMT) in order to validate TMG as a gauge of peripheral fatigue. **Methods:** The plantar flexors of twenty-one healthy male volunteers were tested for TMG parameters (radial displacement and contraction velocity), along with MVC and RMT, before and after either a 5 minute rest period (control) or a 5 minute electrical stimulation intervention (fatigue). **Results:** Radial displacement demonstrated a fatigue-associated reduction ( $3.3 \pm 1.2$  vs.  $4.0 \pm 1.4$  mm,  $p=0.031$ ), while contraction velocity remained unaltered. Additionally, MVC significantly declined by  $122.6 \pm 104$  N ( $p<0.001$ ) following stimulation (fatigue). RMT was significantly increased following fatigue ( $139.8 \pm 54.3$  vs.  $111.3 \pm 44.6$  N,  $p=0.007$ ). **Conclusion:** TMG successfully detected fatigue, evident from reduced MVC, by displaying impaired muscle displacement, accompanied by elevated RMT. TMG could be useful in establishing fatigue status of skeletal muscle without exacerbating the functional decrement of the muscle.

## Radial Displacement Assessment of Bilateral Skeletal Muscle Asymmetry

**Background:** Bilateral muscular asymmetry can be quantified through differences in strength between contralateral limbs. Bilateral strength asymmetry of the knee extensors has been used in sports medicine to assess knee injury and inform rehabilitation programmes. Asymmetry is commonly assessed by MVC or countermovement jump (CMJ), providing information on isolated or multiple muscle groups, but not individual muscles. Assessment of muscle belly radial displacement (TMG) can provide information regarding asymmetry between individual muscles. The aim of Chapter 3 was to assess the relationship between bilateral asymmetry at the level of individual muscle twitch (radial displacement:  $D_m$  and contraction velocity:  $V_c$ ) and contractile function of the muscle group/ entire limb.

**Methods:** Twenty-five volunteers were assessed for VL, RF and VM  $D_m$ , knee extensor MVC, and CMJ. Measurements were performed on each limb, unilaterally, and symmetry index (S.I.%) was calculated. **Results:** Participants were categorised as asymmetric (S.I. > 10%) or symmetrical (S.I.  $\leq$  10%) based on MVC. S.I. of RF  $D_m$  ( $p = 0.003$ ) and  $V_c$  ( $p = 0.008$ ) were greater in the asymmetric group compared to symmetrical. There was no difference in CMJ between groups ( $p = 0.862$ ). **Conclusion:** TMG detected greater asymmetry in RF  $D_m$  and  $V_c$  of individuals with asymmetric MVC; however, symmetry was masked during a more complex task (CMJ). TMG could provide a useful screening measure for detecting asymmetry which may not be apparent during functional actions.

## Impaired Firing Rates of High-Threshold Motor Units Following Eccentric Exercise-Induced Muscle Damage of the Vastus Lateralis

**Background:** Exercise-induced muscle damage (EIMD) results in impaired muscle function and reduced neuromuscular recruitment. However, the specific nature of alterations in motor unit firing behaviour is unclear. The aim of Chapter 4 was to determine the acute effects of eccentric EIMD on alterations in motor unit firing rate (MUFR) and common drive (synchronisation of MURF) in the knee extensor muscles. **Methods:** Fourteen healthy active males completed a bout of eccentric exercise, with measurements of maximal isometric torque (MVC) and rate of torque development (RTD) and surface electromyography (sEMG) performed pre-exercise and 2, 3, 7 and 14 days post-exercise. **Results:** EIMD decreased MVC ( $235.2 \pm 49.3$  Nm vs.  $161.3 \pm 52.5$  Nm;  $p < 0.001$ ) and RTD ( $495.7 \pm 136.9$  Nm.s<sup>-1</sup> vs.  $163.4 \pm 163.7$  Nm.s<sup>-1</sup>;  $p < 0.001$ ) 48h post-exercise. Mean MUFR was reduced ( $16.4 \pm 2.2$  Hz vs.  $12.6 \pm 1.7$  Hz;  $p < 0.01$ ) in high-threshold motor units only, 48h post-exercise, and common drive was elevated (0.36 vs. 0.56;  $p < 0.001$ ) 48h post-exercise. **Conclusion:** EIMD was successfully induced, evident from reduced MVC and RTD. The firing rate of high-threshold motor units was reduced alongside impaired muscle function whilst, those of early recruited motor units remained unaltered. To maintain force output common drive of motor units increased to compensate for the firing rate impairment. This study provides fresh insight into the central mechanisms associated with EIMD.

## Self-Myofascial Release Increases Skeletal Muscular Efficiency without Affecting Range of Motion

**Background:** Self-myofascial release has been shown to alleviate some symptoms of exercise-induced muscle damage, and has been suggested to increase range of movement (ROM) without negatively impacting strength. However, the exact mechanisms of action are unclear. The aim of Chapter 5 was to investigate mechanisms associated with a single acute bout of self-myofascial release (MFR) on healthy rested muscle. **Methods:** VL and RF of sixteen healthy active male volunteers were measured using TMG; radial displacement (Dm) and contraction velocity (Vc) were assessed. Maximal voluntary contraction (MVC) and knee flexion ROM were also assessed, and surface electromyography (sEMG) was recorded during a submaximal isometric contraction (50% of MVC). Measures were performed before and after (0, 15 and 30 minutes) either a 2 minute rest period (control) or a 2 minute bout of MFR, and on 3 consecutive days. **Results:** MVC was reduced in control compared to with MFR ( $p < 0.001$ ), ROM was not altered ( $p = 0.125$ ). Dm of the VL ( $p = 0.001$ ) was elevated on the third day of MFR. sEMG was significantly reduced ( $p < 0.01$ ) 0, 15 and 30 minutes after MFR compared to control. **Conclusion:** Following MFR MVC was elevated compared to control and sEMG was transiently reduced during a submaximal task. This suggests that the activation efficiency of the involved muscles has been increased by MFR, which has spared the decline in MVC, observed in control.

## Skeletal Muscle Contractile Mechanics of High Performance Athletes

**Background:** Screening of muscles contractile mechanics, through TMG, may provide insight into functional and/ or technical imbalances among individual muscles. TMG has been established in laboratory settings, but has more sparingly been applied in the field, with elite athletes. The aim of Chapter 6 was to explore, through a series of case-studies, the practical application of muscle contractile assessment within high performance sports programmes. **Methods:** Three case-studies were performed, involving one female international hockey player and athletes within a group of 30 elite swimmers. TMG measurements were performed in each case-study, with radial displacement (Dm) and contraction velocity (Vc) parameters analysed. Measurements were performed pre- and post-anterior cruciate ligament (ACL) reconstruction surgery; on consecutive days, to assess variability; and during pre-competition taper. **Results:** Balance of contractile mechanics across knee extensor muscles, and between knee extensors:flexor was improved 20 weeks following ALC surgery. VL and GM reliability were greatest in rested state (9.1% and 7.2%, and 13.15% and 4.3%, for Dm and Vc respectively). Reliability of BF was greater in exercised state (14.5% and 11.7%, for Dm and Vc respectively). LD Dm was more reliable in exercised state (4.5%), but Vc was more reliable rested (8.2%). Dm was reduced in BF ( $5.1 \pm 1.3$  vs  $3.1 \pm 1.4$ mm) and GM ( $2.1 \pm 0.3$  vs  $1.6 \pm 0.5$ mm), following reduced training load; LD was unchanged. **Conclusion:** Imbalances were identified among agonist muscles, which may result in abnormal recruitment patterns, heightening the risk of injury. Increased day-to-day variability was detected among highly trained athletes compared to recreationally active individuals. Acute training adaptations were detected, in contractile mechanics, in individual muscles.



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## Publications

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The Measurement of Radial Displacement of the Skeletal Muscle to assess its Contractile Properties: The case of Tensiomyography. *Sports Medicine* [In preparation].

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Kennedy, P., **Macgregor, L.J.**, Barnhill, E., Johnson, C.L., Hunter, A.M., Brown, C., van Beek, E.J.R. & Roberts, N.

Application of Magnetic Resonance Elastography (MRE) to Measure the Effect of Warm-up Using Deep Heat Rub on Muscle Stiffness Following Exercise-Induced Muscle Damage (EIMD). *Radiology* [In review].

Dugdale, J.H., Hunter, A.M., Di Virgilio, T.G., **Macgregor, L.J.** & Hamilton, D.L.

The Influence of the “Slingshot” Bench Press Training Aid on Bench Press Kinematics and Neuromuscular Activity in Competitive Power Lifters. *Journal of Strength & Conditioning Research [In review]*.

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Substrate Metabolism at Rest and During Steady-State Cycling Following Four Weeks of n-3 PUFA Supplementation. *Journal of Sports Nutrition and Exercise Metabolism [In review]*.

### **Conference Presentations**

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**Macgregor, L.J.** & Hunter, A.M. Self-Myofascial Release Increases Skeletal Muscular Efficiency without Affecting Range of Motion. *International Sports Science & Sports Medicine Conference, Newcastle, UK, August 2016. [Abstract submitted].*



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## Glossary of Abbreviations

ACL	Anterior cruciate ligament
AIS	Anterior inferior iliac spine
BF	Bicep femoris
CK	Creatine kinase
CMJ	Counter movement jump
CNS	Central nervous system
CON	Concentric
CoV	Coefficient of variation
CSA	Cross-sectional area
dEMG	Decomposition electromyography
Dm	Radial displacement
DOMS	Delayed onset muscle soreness
ECC	Eccentric
EIMD	Exercise-induced muscle damage
EMD	Electromechanical delay
EMG	Electromyography
FR	Foam rolling
GM	Gastrocnemius medialis
HD-EMG	High density electromyography
ICC	Inter-class correlation coefficient

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LD	Latissimus dorsi
MFR	Myofascial release
MHC	Myosin heavy chain
MU	Motor unit
MUAP	Motor unit action potential
MVC	Maximal voluntary contraction
PMT	Pre-motor time
PNF	Proprioceptive neuromuscular facilitation
RF	Rectus femoris
RFD/ RTD	Rate of force development/ Rate of torque development
RMS	Root mean squared
RMT	Resting muscle tension
ROM	Range of movement
RPE	Rate of perceived exertion
sEMG	Surface electromyography
S.I.	Symmetry index
SS	Static stretching
Tc	Contraction time
Td	Delay time
Tr	½ relaxation time
Ts	Sustain time
TMG	Tensiomyography

VAS	Visual analogue scale
Vc	Contraction velocity
VL	Vastus lateralis
VM	Vastus medialis

# Chapter 1

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## Introduction & Aims

## 1.1 Introduction

Success within high performance sport is often the culmination of many years' worth of preparation. Throughout a typical week, hours of dedication from athletes coaches and associated practitioners are required. For an athlete to achieve optimal performance, it is important that best practice is observed during their preparation; ensuring that objective training and recovery strategies are evolved from informed practice is essential. Performance sport demands sensitive, applicable tools, capable of informing predictions regarding athletic preparation (Kellmann & Günther, 2000) and performance (García-García, Serrano-Gómez, Hernández-Mendo, & Tapia-Flores, 2015; Loturco et al., 2015). In this chapter we will review existing procedures that are commonly applied within sports settings. However, as will be discussed, many of these feature invasive techniques or carry intrinsic confounding variables (Bosco, Colli, Bonomi, von Duvillard, & Viru, 2000; Breil, Weber, Koller, Hoppeler, & Vogt, 2010; Fry et al., 1994). As such, the focus of this thesis is to establish the potential merit of objective, non-invasive assessments of skeletal muscle contractile mechanics within performance sport programmes.

The focus of this chapter is to explore the existing knowledge base surrounding skeletal muscle contractile mechanics, to examine established assessment techniques, and to relate the basic science of muscle contraction to sports preparation and ultimately performance; whilst identifying shortfalls in the existing evidence base. Accordingly, this literature review is not intended to critique sport-specific training concepts, nor is it within the scope of the review to explore in depth biochemical and metabolic processes. Instead, the objectives of the chapter are threefold. Firstly, to review seminal literature which has formed the foundation of our understanding of skeletal muscle contractile mechanics, and

to explore recent advancements in neuromuscular research. Secondly, to detail the relationship between basic science and both functional outcome and performance measures. Thirdly, to examine the role of skeletal muscle contractile mechanics in providing performance and preparation support within sport.

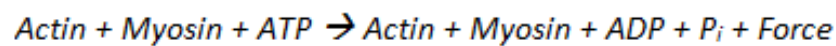
## 1.2 Physiology of Skeletal Muscle Contraction

Before exploring current developments in neuromuscular assessment, it is important to summarize the fundamental processes involved in skeletal muscle contraction. Although the word ‘contraction’ specifically implies a shortening action, throughout this thesis the term ‘muscle contraction’ will be used to describe the active generation of tension within a muscle, regardless of alterations in length (Kent, 1998). As such, the mode of contraction will always be stated (i.e. shortening contraction – concentric, lengthening contraction – eccentric, static contraction – isometric).

The principal function of skeletal muscle is to convert chemical energy into mechanical energy, to produce force and generate movement (Frontera & Ochala, 2015). However, the body of evidence which has developed our existing understanding of muscle contraction stems in the main from speculation, based on *in vitro* experimental model systems. It remains challenging to study muscle contraction at a basic level *in vivo* under physiologically relevant conditions. However, thanks primarily to the development of methods to isolate specific muscle proteins and to measure their biochemical properties (Webb & Trentham, 1981) we have long been able to identify myofibrillar proteins involved in cross-bridge cycling, and elucidate their role within skeletal muscle contractions. In simple terms cross-bridge cycling can be elegantly summarised by Formula 1.



The process of muscle contraction initiates with efferent signal transmission descending motor neurons. Each motor neuron terminates at multiple myofibres, forming motor units (MU). Simultaneous activity of several MUs excites thousands of fibres, producing palpable contractile tension within muscle. Each myofibre consists of thousands of myofibrils and contains billions of myofilaments (Frontera & Ochala, 2015). Myofilaments are arranged to form sarcomeres, the basic contractile units of skeletal muscle. Actin and myosin are the two most abundant myofilaments, comprising ~70–80% of the total protein content of a single myofibre.



**Formula 1 Representation of the cross-bridge cycle, generating contractile force. ATP = Adenosine Triphosphate; ADP = Adenosine Diphosphate; P<sub>i</sub> = Inorganic Phosphate. Adapted from Lieber & Bodine-Fowler (1993).**

The cross-bridge cycle is one of the final links in the chain of events that results in contractile force generation. At the site of the muscle, the process of excitation-contraction coupling (EC coupling) is paramount to muscle contraction (Rebbeck, Karunasekara, Board, Beard, Casarotto & Dulhunty, 2014). EC coupling begins with neurotransmitter acetylcholine (ACh) diffusing across the synaptic cleft, following propagation of action potentials downwards along motor neurons to the axon terminal. T-tubule depolarization follows, resulting in calcium (Ca<sup>2+</sup>) release from the terminal cisternae of sarcoplasmic reticulum. Ca<sup>2+</sup> binds to troponin, leading to deformation of tropomyosin. Inhibition of actin-myosin bonding is released, allowing formation of actin-myosin complex. Myosin ATPase splits ATP, releasing energy which is used to move actin-myosin cross-bridges, creating tension. ATP binds to myosin at the cross-bridge, breaking the actin-myosin complex. This dissociation

allows the myofilaments to slide past each other leading to muscle contraction. These steps are summarized in Figure 1.1. Although, it falls outwith the remit of this review, it seems pertinent to mention that the sequence of events involved in EC coupling is mediated by a wide number of genes, including: MTMR14, MG29 and KLF15 (Manring, Abreu, Brotto, Weisleder & Brotto, 2014).

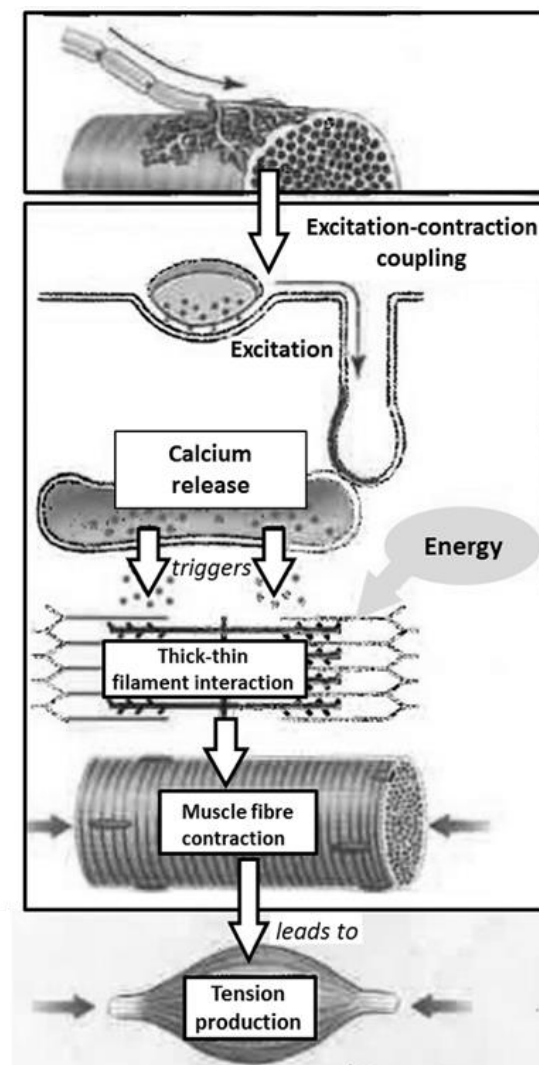


Figure 1.1 Schematic overview of the process of skeletal muscle contraction. Adapted from Martini (2004)

The tension generated through skeletal muscle contraction, producing force and generating movement, can be transmitted either parallel along, or perpendicular to, the longitudinal axis of a sarcomere (Bloch & Gonzalez-Serratos, 2003). Longitudinally, tension is

transduced from sarcomere to sarcomere until the end of the myofibril is reached. Perpendicular tension transmission, allows lateral transduction from one myofibril to another within a myofibre. This laterally transmitted tension is channelled across the sarcolemma to the extracellular matrix, by costameres. Indeed, longitudinal tension transmission only represents ~20-30% of the force generated by sarcomeres (Bolch & Gonzalez-Serratos, 2003). Structurally, costameres are associated with Z-lines (the border between neighbouring half-sarcomeres) (Peter, Cheng, Ross, Knowlton & Chen, 2011), a simplified overview of key costameric protein complexes is outlined in Figure 1.2.

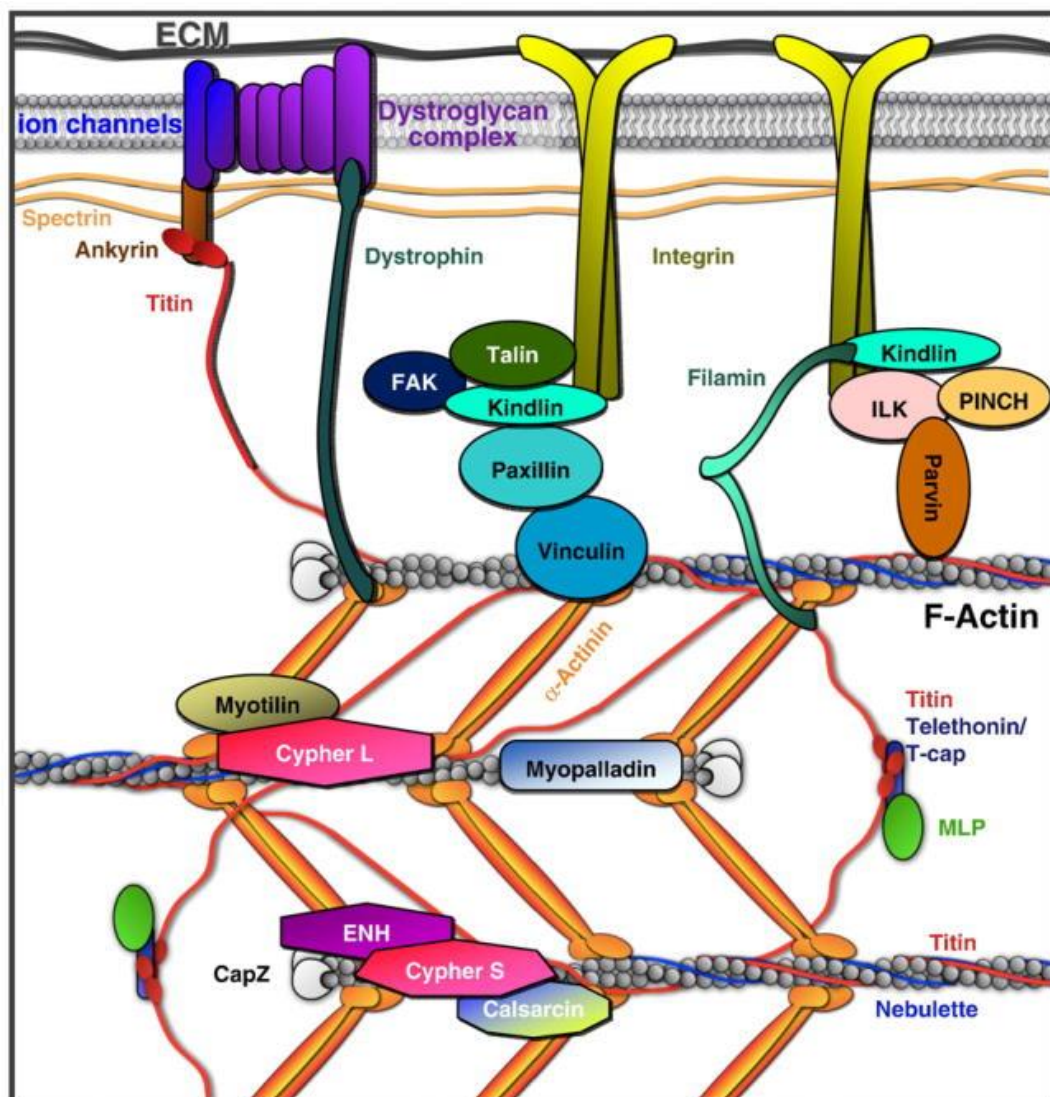


Figure 1.2 Schematic representation of key costameric proteins, revealing the bridges between sarcomere and costamere. Adapted from Peter et al (2011)

As well as lateral transmission of contractile force to the extracellular matrix, costameres also function to align the sarcolemma with nearby contractile elements, and to limit contraction-induced damage to the sarcolemma (Bloch et al, 2002). While a detailed review of costameric proteins is not within the scope of this thesis, a brief summary of the key components will follow. Two of the major costameric protein complexes are the dystrophin-glycoprotein complex and the integrin-vinculin-talin complex. Elements of the dystrophin-glycoprotein complex, and integrin (which interacts with the integrin-linked kinase-pinch-parvin complex and focal adhesion kinase), directly interact with Z-line components (such as filamin-C, which links costamere to sarcomere). Vinculin is a foundational protein within the costameric complex, interacting with a range of proteins, including: talin, paxillin, and  $\alpha$ -actinin. A full review of costameric proteins is provided by Bloch et al (2002).

### **1.3 Muscle Function**

#### **1.3.1 Strength and Power**

It has long been established that contraction force and contraction velocity are interdependent (Hill, 1938; Katz, 1939; Edman, 1979). Our current understanding of the force-velocity relationship can be accredited to Hill (1970) (Figure 1.3). When a muscle is required to move a load that is less than its maximum tetanic tension, the muscle will contract concentrically. Force generated by the muscle concentrically will always be less than the muscle's maximum isometric force capacity. As force requirement decreases, contraction velocity will increase until maximal velocity is reached, and force generation is zero (Figure 1.3). Maximum contraction velocity ( $V_c$ ) can be used to characterise muscle fibre type distribution and architecture (Lieber & Bodine-Fowler, 1993). If the external load

imposed on muscle outweighs the maximum force that the muscle can generate, forced lengthening of active muscle occurs which is an eccentric contraction. Absolute muscle tension during eccentric contractions is relatively larger than during concentric movements. Additionally, the absolute tension is less interdependent on  $V_c$  (Katz, 1939), therefore, ratio of concentric : eccentric force is likely to vary at different velocities (Hartsell & Spaulding, 1999). Although eccentric actions are physiologically common for many daily tasks, muscle injury and soreness are thought to be most strongly associated with eccentric contractions (Byrne et al., 2004; Clarkson, 1997; Proske & Morgan 2001).

Whole muscle contractile properties are fundamentally affected by arrangement of sarcomeres within the muscle. As such,  $V_c$  can be interpreted as being proportional to the number of sarcomeres in series, whilst force generation is proportional to the cross-sectional area (CSA) of sarcomeres (Powell, Roy, Kanim, Bello, & Edgerton, 1984; Bodine et al., 1982); these factors can be referred to as 'muscle architecture'. Since it is complex to determine sarcomere number or size precisely, muscle architecture is often assessed through whole muscle fibre characteristics. In this regard, muscle  $V_c$  can be described as proportional to muscle fibre length, with muscle force being proportional to total fibre CSA. Muscle power is primarily determined by muscle fibre type composition and muscle mass/architecture, as well as neuromuscular activation (Moritani, Kimura, Hamada, & Nagai, 2005). However, there is still uncertainty surrounding the extent to which fibre type composition predicts athletic performance (Terzis, Spengos, Kavouras, Manta, & Georgiadis, 2010). Research throughout the last few decades has allowed classification of muscle fibres according to criteria including: colour, in relation to myoglobin content; contractile properties of MUs in response to electrical stimulation;  $V_c$  during single twitch; fatigability

during sustained activation; predominance of metabolic pathway (oxidative or glycolytic);  $\text{Ca}^{2+}$  handling by sarcoplasmic reticulum and protein isoform expression (Lamboley, Murphy, McKenna, & Lamb, 2014). Accordingly, myofibres can be classified into one of three fibre types (Frontera & Ochala, 2015): type I (slow, oxidative, fatigue-resistant), IIA (fast, oxidative, intermediate metabolic properties), and IIX (fastest, glycolytic, fatigable). It is well established that fast type II muscle fibres correspond with higher power-generating capacity than slow type I fibres (Gilliver, Jones, Rittweger, & Degens, 2011), although type II fibres are also more susceptible to fatigue (Petrofsky & Lind, 1979; Greising, Gransee, Mantilla, & Sieck, 2013). Within high performance sport, a thorough understanding of strength and power producing capacity, as well as architecture and composition of muscle, are fundamental to effective preparation.

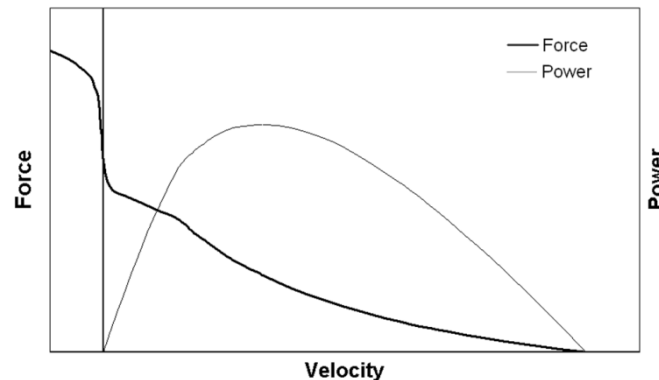


Figure 1.3 Typical Force – Velocity relationship and associated Power curve. Adapted from Edman (1979).

### 1.3.2 Muscle Length

It has been reported that around 50-70% of external work performed, during tasks such as running (Cavagna, 1970) or jumping (Thys, Cavagna, & Margaria, 1975), originates from stored mechanical energy in elastic structures of muscle, known as elastic potential energy (Gleim & Mchugh, 1997). Elastic potential energy is a function of musculotendinous

compliance or stiffness. With greater compliance, further contraction must occur in order for tension to be produced (Wilson, Wood, & Elliott, 1991). Elevated muscular stiffness is associated with functional strength output, suggesting that stiffer muscle allows for transmission of contractile force production in a more efficient manner (Gleim, 1984; Gleim & Mchugh, 1997). Gender differences in muscle stiffness have been reported (Wang, De Vito, Ditroilo, Fong, & Delahunt, 2015), with females displaying lower levels of stiffness; differences in neuromuscular control and/or muscle mass may be key mediators. Early work by Blix (1894) first demonstrated the principle which has since been described as the length-tension relationship (Figure 1.4A). Fundamentally, muscle produces lower tension at very long or very short lengths, while greatest tension is produced at an optimal length. The length-tension relationship describes that tension generated is directly a function of the extent to which actin and myosin myofilaments overlap one another (Figure 1.4B). The precise association between muscle tension and myofilament overlap was elegantly demonstrated through a series of studies, first by Gordon, Huxley, & Jitliant (1966) and then by Edman (1979). As illustrated in Figure 1.4A, passive tension (i.e. tension that is present in the absence of muscle stimulation) is near zero at the muscle length optimal for active tension. As muscle length is increased, so too will passive tension increase (Joumaa, Rassier, Leonard, & Herzog, 2008). Passive tension arises from myofibrillar structural protein, namely titin, which spans the half-sarcomere, connecting A-bands (the region containing the entire length of myosin filaments) to Z-lines (Magid & Law, 1985; Funatsu & Higuchi, 1990; Cornachione & Rassier, 2012; Rassier, Lee, & Herzog, 2005). Lengthening of muscle will result in greater series compliance, ergo greater active tension required to produce force or movement. This phenomenon has been most elegantly demonstrated by research examining muscle stretching protocols (Power, Behm, Cahill, Carroll, & Young, 2004; Behm

et al., 2011; Kay & Blazevich, 2012). Understanding the role that muscle length and stiffness play in tension production can allow athletes to optimize muscle functional output.

## 1.4 Assessing Muscle Function

### 1.4.1 Strength and Power

Muscular strength is a function of interactions between neural, mechanical and structural factors (Enoka, 1988; Rutherford & Jones, 1986). As such, there exists dissociation between muscle size and strength. The maximal force output of a muscle (i.e. strength) is measured as the force exerted by musculoskeletal elements against a transducer (Enoka, 1988). Although force is proportional to CSA, there is not a direct relationship between changes in muscle size and strength (Howald, 1985; Ikai & Fukunaga, 1970; Jones & Rutherford, 1987). For over fifty years, isometric maximal voluntary contraction (MVC) has been assessed, to quantify skeletal muscle strength *in vivo* (Wilson & Murphy, 1996). Isometric MVC has proven relatively uncomplicated and incorporates a high level of assessment control in laboratory settings utilising dynamometry (Figure 1.5). MVC has also displayed strong test-retest reliability ( $r = 0.998$ ) and sensitivity to both acute and chronic neuromuscular adaptations (Abernethy, Wilson, & Logan, 1995; Taylor, Sanders, Howick, & Stanley, 1991; Wilson & Murphy, 1996).



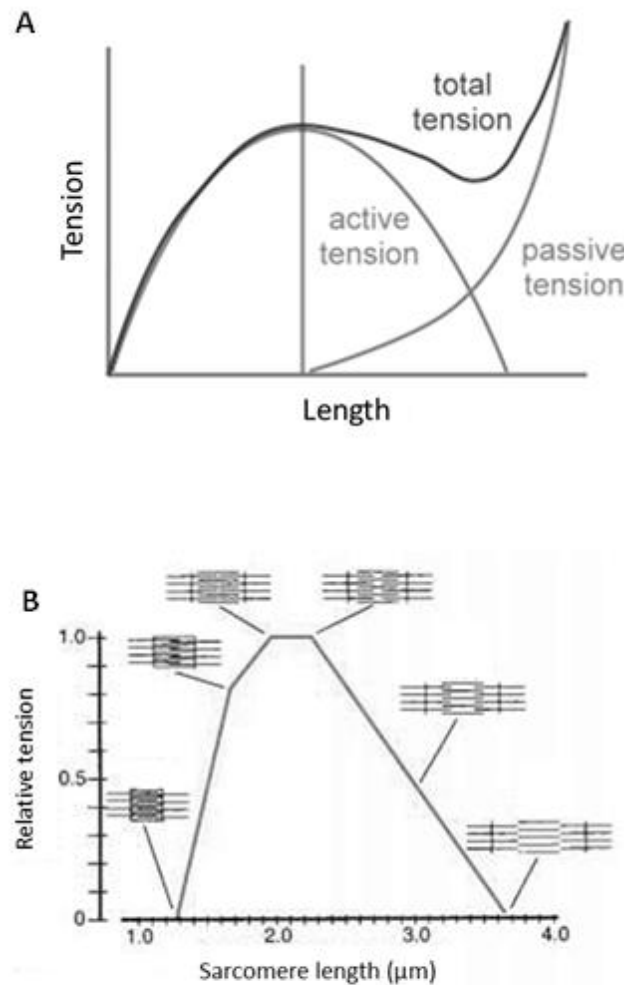


Figure 1.4 Typical Length - Tension relationship (A), with corresponding myofilament overlap (B).

In addition to maximal strength, other variables can be gleaned from MVC, to provide information regarding neural and muscular status. Rate of force/ torque development (RFD) is indicative of muscle activation, associated with MU firing rate (Maffiuletti et al., 2016). As such, changes in RFD during the early-phase (0–50 ms) of MVC (Figure 1.6) can be used as a surrogate for neural function (Casartelli, Lepers & Maffiuletti, 2014). Since first being reported (Thorstensson, Karlsson, Viitasalo, Luhtanen & Komi, 1976) RFD has been established as an important functional marker, due to temporal similarity to

both everyday and sporting activities (Aagaard, Simonsen, Andersen, Magnusson & Dyhre-Poulsen, 2010) and positive correlation with functional performance (Maffiuletti, Bizzini, Widler & Munzinger, 2010; Tillin, Pain & Folland, 2012).



Figure 1.5 Isometric MVC using a dynamometer (Biodex).

The time lapse between respective onsets of electrical activity and force production can inform regarding: action potential propagation, EC coupling and stretching of the series elastic component (Li & Baum, 2004; Muraoka, Muramatsu, Fukunaga & Kanehisa, 2004). This time lapse is referred to as electromechanical delay (EMD), which is ~~a~~ quantified, as depicted in Figure 1.7, through synchronised recording of force (i.e. MVC) and electromyography (EMG) and has reported very good reliability (Howatson, Glaister,

Brouner, & van Someren, 2009). Lengthened EMD has been observed during eccentric contractions, compared to concentric and isometric actions (Cavanagh & Komi, 1979). Elsewhere, impaired response has been reported following exercise-induced muscle damage (EIMD), with longer EMD 96h after damaging exercise (Howatson, 2010), and following endurance exercise (Gleeson, Reilly, Mercer, Rakowski, & Rees, 1998). These alterations were observed despite peak torque having recovered or remained unchanged.

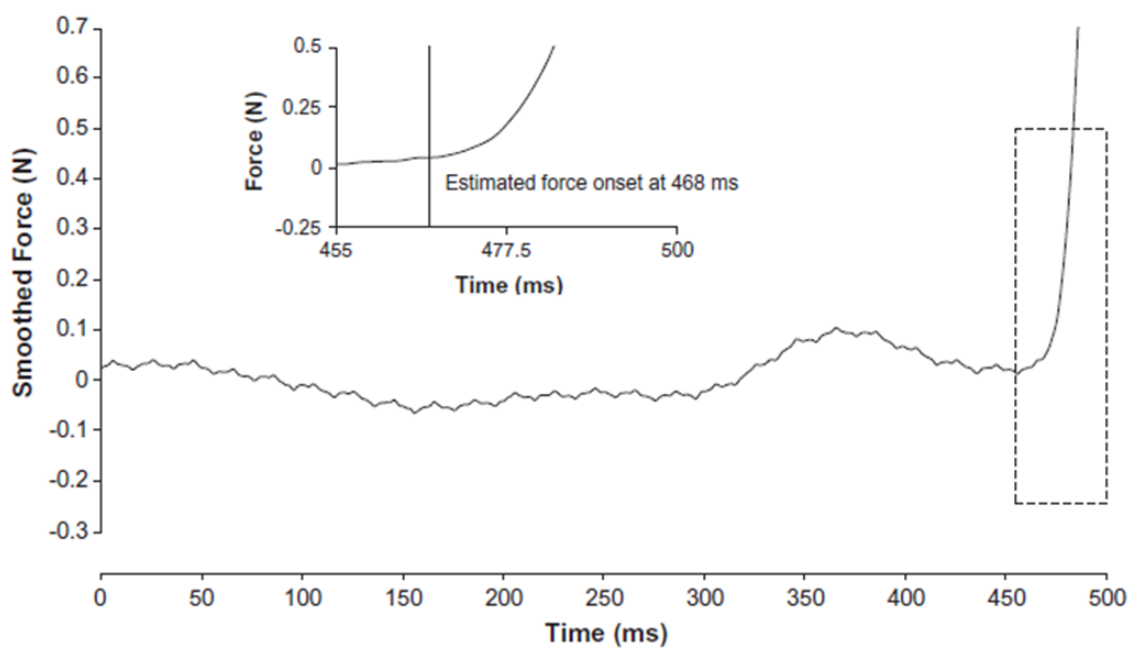


Figure 1.6 Determination of force onset and rate of force development (RFD). Adapted from Tillin, Pain & Folland (2013).

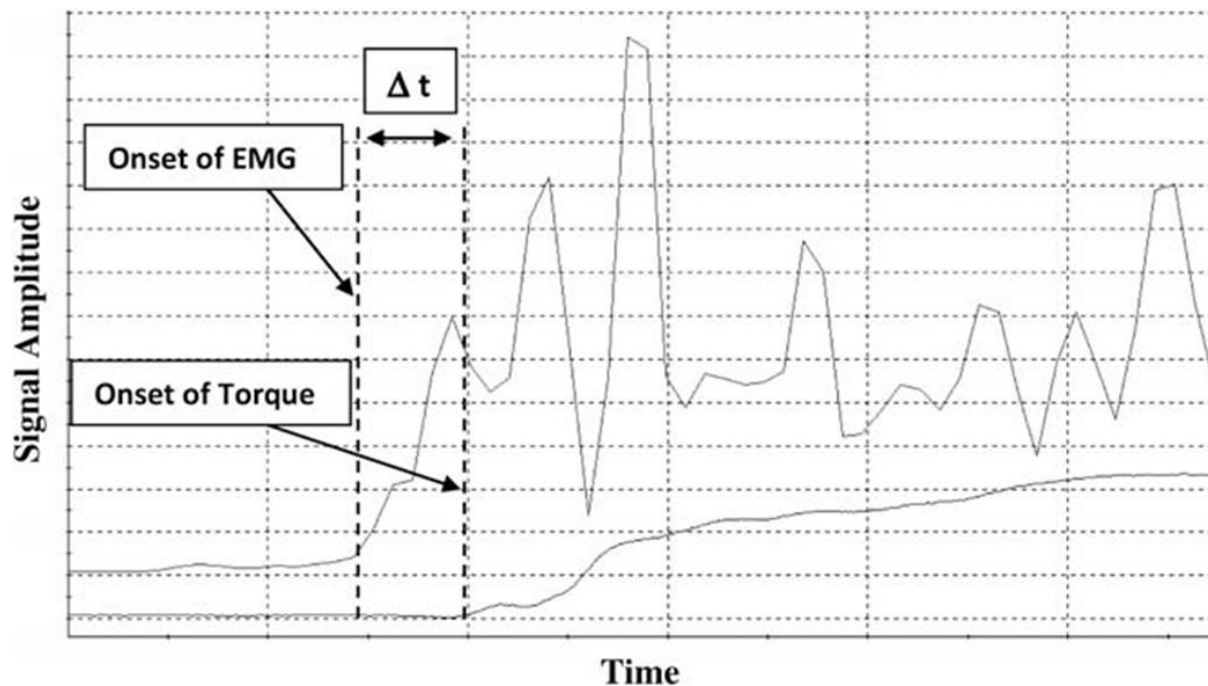


Figure 1.7 Schematic representation of the determination of EMD.  $\Delta t$  = time lapse between respective onsets of EMG activity and torque production. Adapted from Howatson (2010).

In athletic populations, it has been suggested that isometric assessments of muscle strength lack applicability. Isokinetic strength testing is performed concentrically or eccentrically at a predetermined angular velocity (Lund et al., 2005). Testing dynamically, particularly at higher speeds, has been proposed to better reflect functional demands during sporting activities (Iossifidou, Baltzopoulos, & Giakas, 2005). However, isokinetic assessments still occur across a restricted range of movement (ROM) and with a constant kinetic velocity, thereby limiting functional applicability. On the other hand, biomechanical analysis of vertical jump ability (Figure 1.8) has become a focus of interest in recent decades (Requena, Requena, García, de Villarreal, & Pääsuke, 2012). Variations in jump technique and assessment protocol have been developed, since the first reporting of a standing vertical jump test (Sargent, 1921). Multiple measurement devices also exist, including: force

platforms (Dowling & Vamos, 1993; Hatze, 1998), jump and reach devices (Isaacs, 1998) and video recording (Hatze, 1998). Vertical jump tests performed unilaterally or bilaterally have demonstrated sensitivity to training induced adaptations in muscular power, although significant control measures are advised (Young, MacDonald & Flowers, 2001).



Figure 1.8 Vertical jump test, performed as a countermovement jump (CMJ).

#### 1.4.2 Electromyography

Given that the primary role of any skeletal muscle is the production of force, often to initiate movement (Lieber & Bodine-Fowler, 1993); performance efficiency of skeletal muscle can be considered as a function of the mechanical properties of that muscle and of motor input (Enoka, 1988; Rutherford & Jones, 1986). Indeed, normal movement has been described as the culmination of complex interaction between the nervous system, muscles and joints. Broadly, these factors could be divided up as peripheral, pertaining to anything distal to the neuromuscular junction (Enoka & Duchateau, 2008), and spinal/ supraspinal,

relating to the central nervous system (CNS) (Gandevia, 2001). Central drive has been widely measured via the muscle as EMG since initial work by Weddell (1943), with research in recent years focussing on the impact of, for example, muscle temperature and aging (Dewhurst et al., 2010) and warm up strategies (Stewart, Macaluso & De Vito, 2003). Non-invasive surface EMG (sEMG) permits access to the activity of the motor control centre percutaneously (Hermens, 2000; Hug & Dorel, 2009). The properties of the EMG signal depend on the timing and force of contraction (Larsson et al., 1999). sEMG provides a global overview of neuromuscular activity, whilst intramuscular EMG, using indwelling electrodes, provides more specific information regarding firing patterns of (a small number of) individual MUs (Christie, Greig Inglis, Kamen & Gabriel, 2009). Recent studies (Waite, Brookham & Dickerson, 2010; Allen, Brookham, Cudlip & Dickerson, 2013) have intimated strong associations between measurements through both techniques. However, it is also now possible to decrypt specific MU firing from large pools by extracting action potential trains from sEMG. Principally, two disparate methods have been proposed for interpretation of motor unit action potential (MUAP) trains (De Luca, Lefever, Mccue, & Xenakis, 1982; Blok, van Dijk, Drost, Zwarts, & Stegeman, 2002; Zwarts, Lapatki, Kleine, & Stegeman, 2003; Adam & De Luca, 2005; De Luca, Adam, Wotiz, Gilmore, & Nawab, 2006; Kline & De Luca, 2014), high density EMG (HD-EMG) and decomposition EMG (dEMG). These methods differ primarily with regard to the electrode configuration and extraction process (Nawab, Chang, & De Luca, 2010).

#### **1.4.2.1 Decomposition Electromyography**

The main development has been towards non-invasive multi-channel sEMG recording systems (Merletti & Parker, 2004) which can comprise up to hundreds of

electrodes with the objective of identifying individual MU activity through spatial imagery of action potentials (Farina, Negro, Gazzoni & Enoka, 2008). Use of these multi-electrode arrays is known as HD-EMG (Blok et al., 2002; Zwarts et al., 2003). Evolution of multi-channel EMG sampling has led to innovation in algorithms to decompose EMG signals, applying latent component analysis approaches to identify MU activity (Negro, Muceli, Castronovo, Holobar, & Farina, 2014). Alternatively, multi-channel EMG recordings can be conducted using a single, four-channel sensor (Adam & De Luca, 2005; De Luca et al., 1982; De Luca et al., 2006; Nawab et al., 2010; Kline & De Luca, 2014). Due to the difference in recording methods, it is unsurprising that alternative decomposition algorithms are required. Single sensor multi-channel sampling and associated algorithmic decomposition is referred to simply as dEMG (Adam & De Luca, 2005; De Luca & Erifn, 1994). The unique dEMG system allows assessments of MU firing properties (Nawab et al., 2010) and facilitates evaluation of different MUs within the recruited pool, based on their recruitment threshold (De Luca & Contessa, 2012). dEMG has been specifically developed to investigate the independent and collective behaviour of MUs (Adam & De Luca, 2005; De Luca & Erifn, 1994). The merits of dEMG and validity of the associated decomposition algorithm have been debated at length (De Luca, Nawab, & Kline, 2015).

### **1.4.3 Mechanomyography**

Force produced through muscle twitch, in response to electrical stimulation, is frequently used to analyse muscle contractile properties (Bülow, Nørregaard, Danneskiold-Samsøel & Mehlsen, 1993; Degens, Sanchez Horneros, Heijdra, Dekhuijzen, & Hopman, 2005). However, interpretation of temporal characteristics of twitch force is challenging. Typically twitch force manifests at less than 10% of MVC (Prasartwuth, Allen, Butler,

Gandevia, & Taylor, 2006). Additionally, force must be conducted through connective tissue and recorded externally by a transducer; series compliance of both muscle and connective tissue influence the measurement. It is also difficult to assess individual muscle responses, as synergist and antagonist muscles integrate within the response (Huijing & Baan, 2001). Twitch force response is also delayed, relative to stimulus, due to kinematic dampening of contracting muscles by surrounding tissues (Stevens, Dickinson, & Jones, 1980; Koren, Simunic, Rejc, Lazzer, & Pisot, 2015).

Despite these challenges, intrinsic muscle contractile mechanics are key mediators of performance (Costill, Fink, & Pollock, 1975; García-García et al., 2015; Lattier, Millet, Maffiuletti, Babault, & Lepers, 2003; Loturco et al., 2015). Any unloaded (i.e. concentric) contraction will bring about pronounced muscle shortening, along the longitudinal axis. Given that skeletal muscle operates as a near constant-volume system (Baskin & Paolini, 1967), shortening on the longitudinal axis is interlinked to proportional expansion of transversal diameter (Hill, 1938), which is observed as radial deformation of the muscle belly. Deformation can be measured through a range of mechanomyographic methods. Phonomyography or soundmyography (Barry, Geiringer, & Ball, 1985; Maton, Petitjean, & Cnockaert, 1990; Orizio & Veicsteinas, 1992) use microphones to transform muscle fibre mechanical oscillations into audio signals. Vibromyography uses accelerometers and laser beams to record transverse widening and vibration of the muscle belly (Zhang, Frank, Rangayyan, Member, & Bell, 1992; Orizio, Baratta, Zhou, Solomonow, & Veicsteinas, 2000; Orizio, Liberati, Locatelli, De Grandis, & Veicsteinas, 1996). However, high variability due to low signal-to-noise ratio limits the interpretation of such data (Wong, 2001; Orizio, 2002). That being said, the non-invasive and passive nature of mechanomyographic assessments,



present an intriguing possibility for muscle contractile assessment within a high performance setting.

#### 1.4.3.1 Tensiomyography

Mechanomyography can be defined as direct measurement of muscle movement at the muscle belly (Križaj, Šimunič, & Žagar, 2008). An alternative method to measure radial deformation of muscle has been developed, primarily through pioneering work at the Faculty of Electrical Engineering, University of Ljubljana, Slovenia (Valenčič & Knez, 1997; Kersevan, Valenčič, Djordjevič, & Šimunič, 2002; Dahmane, Djordjevič, Šimunič, & Valenčič, 2005; (Dahmane, Djordjevič, & Smerdu, 2006). A high precision ( $4\mu\text{m}$ ) digital displacement sensor is applied to the muscle belly with a controlled pre-tension between the sensor tip and the muscle (Figure). It is this pre-tension from which the method has derived the name tensiomyography (TMG) (Valenčič & Knez, 1997). By providing controlled pre-tension the muscle twitch response is augmented, enhancing measurement of contraction dynamics (Križaj et al., 2008).

Muscle twitch is induced through single 1ms wide electrical stimulus. Stimulation amplitude is variable inter-individual, as the amplitude required to provide a maximal muscle response is not equal among all muscles or individuals. It would be inappropriate to apply similar stimulation amplitude universally as multiple factors influence muscle response, including: threshold, activation, skin conductivity, subcutaneous depth, water retention, temperature (Križaj et al., 2008). Typically studies report peak responses occurring at stimulation amplitudes between 60-100mA (Križaj et al., 2008). A number of parameters are extracted from twitch displacement curves (Figure). Peak radial displacement ( $D_m$ ) signifies the absolute spatial transverse deformation of the muscle.

Contraction time ( $T_c$ ) is measured as the time taken on the ascending curve between 10% - 90% of  $D_m$ . Delay time ( $T_d$ ) represents the time between delivery of electrical stimulus and 10% of  $D_m$ . Half-relaxation time ( $T_r$ ) is given as the time taken from 90% to 50% of  $D_m$  on the descending curve (Figure 1.9). The duration for which twitch is sustained ( $T_s$ ) is measured as the time between 50% of  $D_m$  on each side of the twitch curve (Tous-Fajardo et al., 2010).

Reliability of TMG measurement parameters has previously been investigated across a variety of conditions, in a number of studies. Križaj et al., (2008) analysed short-term repeatability of each parameter extracted from muscle twitch responses, by delivering thirty consecutive stimulations and capturing individual measurements from each twitch response (Figure 1.10). A strong intra-class correlation coefficient (ICC) of 0.86 or greater was recorded for all parameters (Table 1.1A). Inter-rater reliability has also been examined, with specific focus on the importance of transducer and electrodes positioning (Tous-Fajardo et al., 2010). In a similar finding, ICC was greater than 0.86 for each parameter, with exception of  $T_r$  (Table 1.1B). It was noted that deliberately altering the inter-electrode distance significantly impacted the measurement of  $D_m$ . Since muscle fibres do not shorten homogeneously, due to non-uniform sarcomeres, myofibres or fascicle lengths (Morgan, 1985), consideration is also necessary regarding sensor positioning, as morphological changes may occur in muscle when the sensor is repositioned (Rodríguez-Matoso et al., 2010). Inter-day reliability has returned comparatively favourable results (Šimunič, 2012), with  $D_m$  and  $T_c$  in particular displaying ICC of no less than 0.98, across three separate muscles. However, applicability of these findings must be queried, as measurements were performed following 60 minutes of bed-rest. As such, Ditroilo, Smith, Fairweather, & Hunter

(2013) evaluated the reliability of TMG in different muscle conditions – rested, exercised and fatigued; additionally assessing long-term (four week interval) stability of the contractile parameters. Although reliability was reported to suffer when measurements were carried out over longer intervals (ICC ranging from 0.86 – 0.95 for Dm), it is interesting that the measurements displayed greater reliability in exercised or fatigued state, compared to rested. Due to the reported findings of these four reliability studies, it has been recommended that assessment carried out through TMG should focus on the most stable parameters, namely peak displacement (Dm) and contraction time (Tc) (Benítez Jiménez, Fernández Roldán, Montero Doblas, & Romacho Castro, 2013).

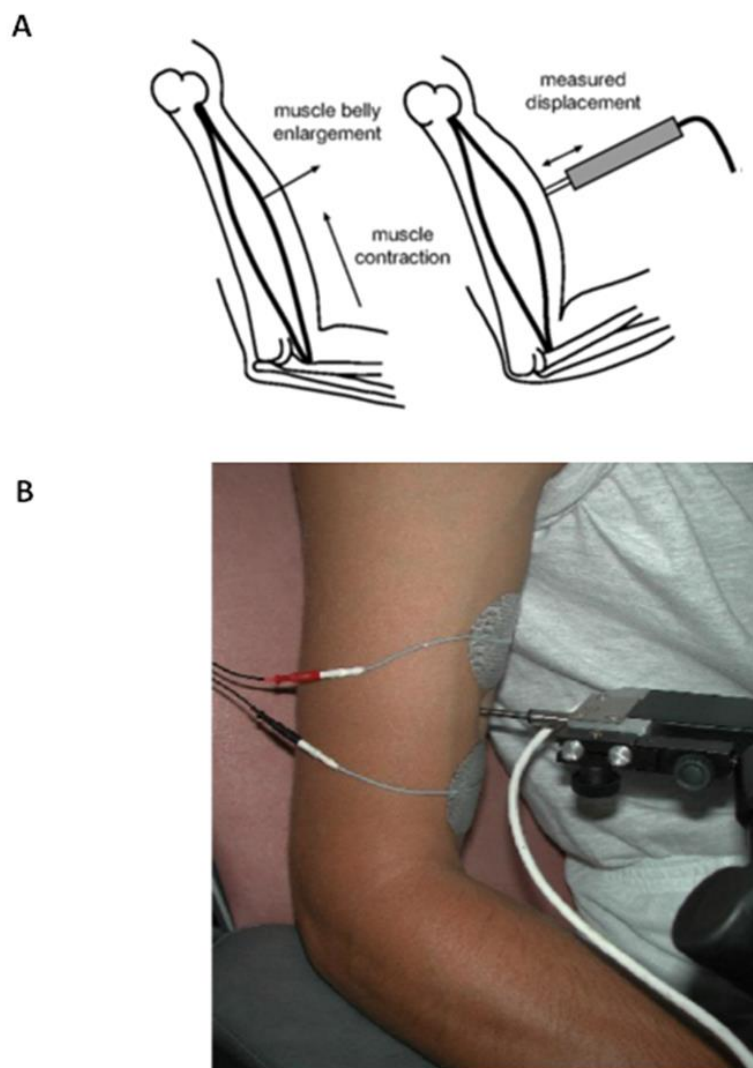


Figure 1.9 Positioning of the digital displacement transducer, perpendicular relative to the muscle belly (Biceps Brachii) (A) Dahmane et al (2001); (B) Dahmane et al (2005).

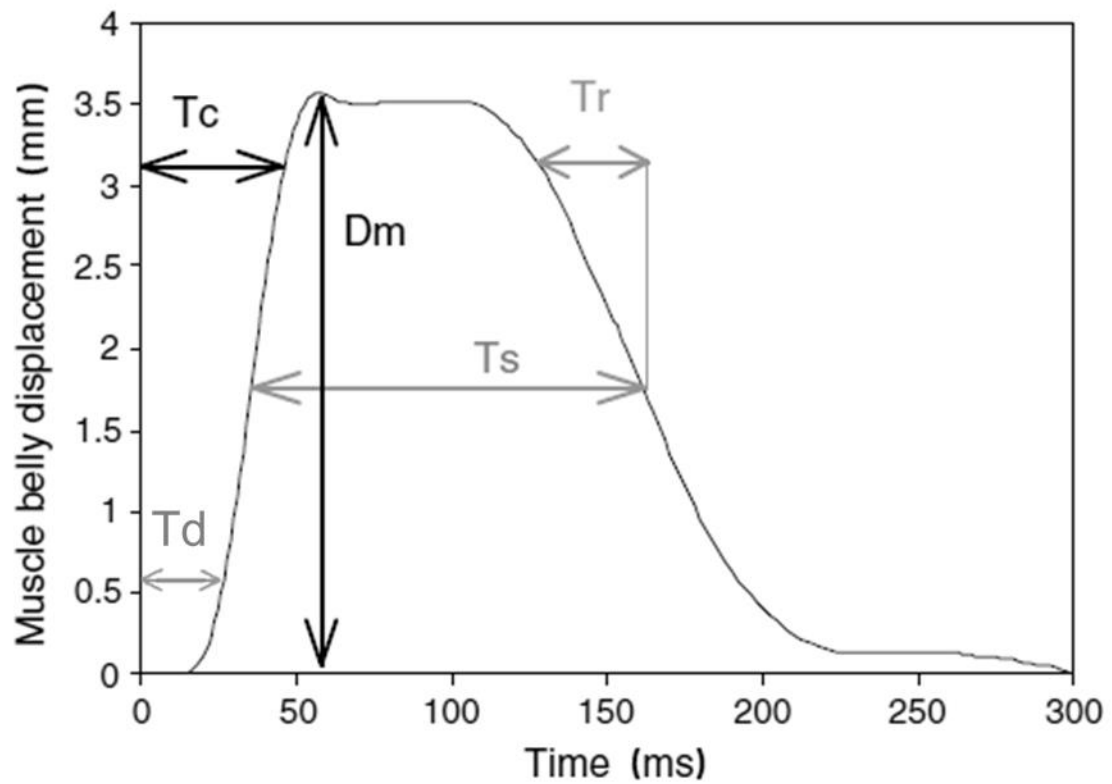


Figure 1.10 Parameters extracted from TMG twitch displacement-time curve. Adapted from Križaj et al (2008).

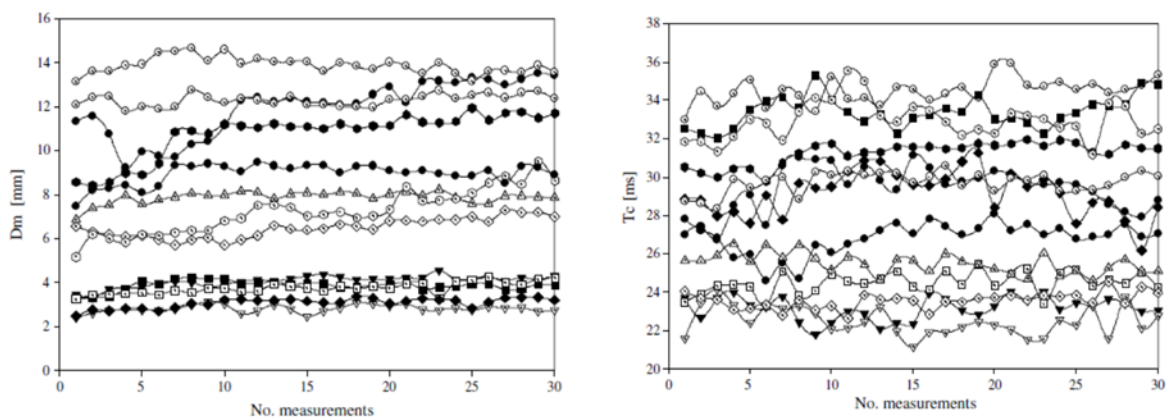


Figure 1.11 Results of an early reliability study for displacement ( $D_m$ ) and contraction time ( $T_c$ ).

Table 1.1 Reliability of TMG parameters. (A) Krizaj et al, 2008; (B) Tous-Fajardo et al, 2010.

<b>A</b>				
	<b>Min</b>	<b>Max</b>	<b>Mean</b>	<b>ICC</b>
<b>Dm</b>	2.42	14.66	7.47	0.98
<b>Tc</b>	21.14	35.92	28.18	0.97
<b>Td</b>	22.53	35.61	28.08	0.94
<b>Tr</b>	17.18	162.01	81.39	0.86
<b>Ts</b>	44.65	256.06	136.81	0.89

<b>B</b>				
	<b>Rater A</b>	<b>Rater B</b>	<b>ICC</b>	<b>95% CI</b>
<b>Dm</b>	7.3	7.0	0.97	0.92 – 0.99
<b>Tc</b>	21.1	21.4	0.92	0.81 – 0.97
<b>Td</b>	25.7	26.3	0.86	0.86 – 0.95
<b>Tr</b>	75.0	74.3	0.77	0.49 – 0.91
<b>Ts</b>	195.9	195.2	0.96	0.90 – 0.99

### 1.5 Application of TMG

In accordance with recommendations derived from the above mentioned reliability studies (Krizaj et al., 2008; Tous-Fajardo et al., 2010; Šimunič, 2012; Ditroilo et al., 2013) Dm and Tc have been the most thoroughly investigated parameters using TMG. The existing body of research suggests that TMG may provide a valid alternative to more invasive analyses of skeletal muscle properties. Dm is typically considered in association with muscle stiffness and tendon mechanical properties (García-García, Cancela-Carral, Martínez-Trigo, & Serrano-Gómez, 2013; García-García et al., 2015; Šimunič et al., 2011); it has also been shown that fluctuations in muscle size, such as following disuse atrophy (Pišot et al., 2008), are associated ( $r = 0.70$ ) with changes in Dm, such that increased Dm accompanied

reduction in muscle thickness. This decreased thickness would have led to diminished muscle stiffness (Reeves, Maganaris, Ferretti, & Narici, 2005), allowing greater Dm in response to electrical stimulation. Tc can be used as a non-invasive estimate of muscle fibre type composition and spatial distribution, and biomechanical properties (Dahmane, Valenčič, Knez, & Eržen, 2001; Dahmane et al., 2005; Šimunič et al., 2011), displaying significant correlation ( $r = 0.878$ ) with myosin heavy chain (MHC) I/ II content as assessed through histochemical analysis (Figure 1.12). Specifically, greater MHC I proportions are accompanied by longer Tc. The portable nature of the measurement hardware also lends itself aptly to field-based data capture. Measures of muscle Dm and Tc have been applied in acute settings. The impact of hypobaric, hypoxic conditions, associated with high altitude, were assessed (Morales-Artacho et al., 2015), however interpretation of findings is limited, as measurements of contractile mechanics were not overlaid with any other measurements. Alternatively, the acute response to EIMD was investigated (Hunter et al., 2012) with well understood physiological markers recorded alongside the use of TMG. Decreased radial displacement and extended contraction time were both linked to impairments in force production and rate of force development. In order to apply TMG effectively, it is important to integrate the measurements with physiologically functional variables. Previously, knee injury risk has been assessed, through measures of bilateral asymmetry among the knee extensors and flexors (Alentorn-Geli, Alvarez-Diaz, Ramon, Marin, Steinbacher, Boffa, et al., 2014), while plantar flexors have been shown to be poor predictors of knee injury risk (Alentorn-Geli, Alvarez-Diaz, Ramon, Marin, Steinbacher, Rius, et al., 2014). That being said, the relationship between contractile parameters (Dm and Tc) and muscle function has yet to be fully elucidated. Power assessments have shown weak associations with contractile parameters recorded through TMG (Gil et al., 2015). Comparisons between twitch torque/

force and TMG-recorded parameters (Šimunič, Križaj, Narici, & Pišot, 2010) have suggested that different mechanisms affect longitudinal and transversal skeletal muscle deformations. Measuring the temporal features of a muscles radial displacement may provide limited insight into contraction speed. As  $T_c$  is extracted as the duration of twitch between 10% and 90% of peak  $D_m$ ,  $T_c$  is highly dependent on the size of  $D_m$ . Therefore,  $T_c$  may not provide objective assessment of contraction velocity. As such,  $D_m$  assessment would appear to be the most advantageous mechanical measurement using TMG, assessment of temporal contractile parameters requires further investigation.

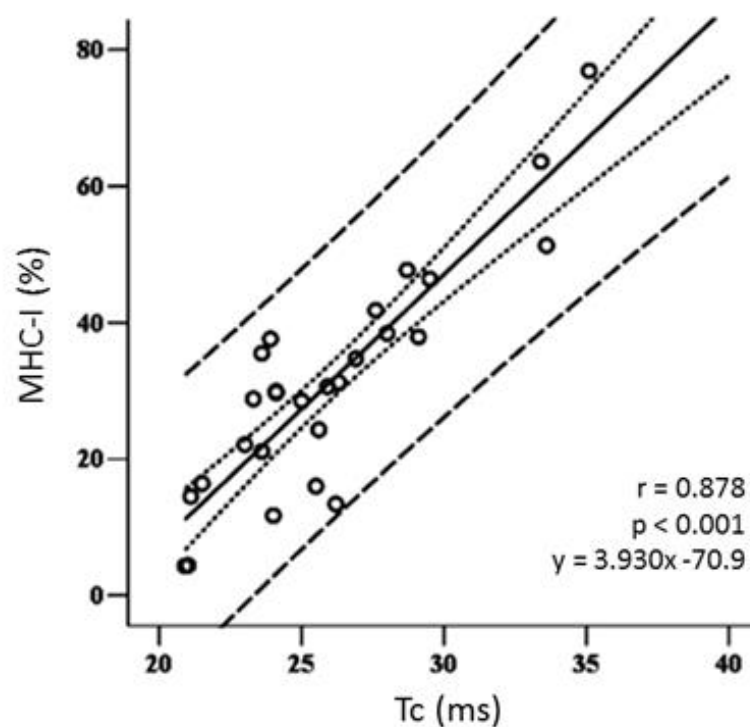


Figure 1.12 Correlation between myosin heavy chain I and contraction time. Adapted from Simunic et al (2011).

### 1.5.1 TMG within Sport

The use of TMG within a sports-setting has recently been discussed (Wiewelhove et al., 2015). In particular, interest surrounds the intensive demands of training and

competition. Where super-compensation (Olbrecht, 2007) transitions towards functional overreaching, performance may be negatively affected (Halson, 2014). It is therefore important to incorporate assessment of fatigue and recovery routinely, to help inform optimisation of training prescription and ensure competition readiness. Practically however, individuals will respond variably to given a training stimulus, hence inconsistencies in quantifiable fatigue or recovery markers are commonplace (Wiewelhove et al., 2015). Recommendations suggest that surrogate markers should be assessed with adequate regularity. Measurements should include performance markers, to monitor muscle function, as well as subjective assessments. However, neither individually nor grouped, do these markers cover all potential mechanisms that contribute to muscular adaptation (Wiewelhove et al., 2015). Muscle mechanical adaptations have been measured, through TMG, with strength (de Paula Simola et al., 2015; García-Manso et al., 2012) and endurance training (García-Manso et al., 2011). Also differences in mechanical properties between endurance and strength/ power athletes have been described (Loturco et al., 2015). Measurements of Dm were lower following fatigue resulting from workload matched high load or high volume resistance exercise (García-Manso et al., 2012), while Tc was longer, and Dm tended to be reduced, following 6 days of high-intensity interval training (Wiewelhove et al., 2015; Raeder et al., 2016). Both of these studies incorporated relatively short duration, high-intensity protocols; it is likely that observed reductions in Dm could be due to impaired propagation of electrical stimulus along the sarcolemma. This impairment is likely to occur from a pH driven alteration of the Na<sup>+</sup> and K<sup>+</sup> gradient across the muscle membrane resulting in reduced Ca<sup>2+</sup> and subsequent EC coupling (Brody, Pollock, Roy, De Luca, & Celli, 1991). Conversely, García-Manso et al. (2011) and Giovanelli et al. (2016) both demonstrated increased Dm following fatigue. In the cases of these studies, ultra-endurance



exercise was performed, specifically an Ironman® triathlon (García-Manso et al., 2011) and uphill-marathon (43 km, 3063m elevation gain). The incongruous change in Dm following this mode of exercise may be due to reduced muscle stiffness observed following ultra-endurance running possibly resulting from altered proprioception stimulated from elevated cytokine release (Morin, Tomazin, Edouard, & Millet, 2011); the findings from these studies are summarized in Table 1.2.

**Table 1.2 Summary of studies investigating changes in muscle mechanical parameters, as assessed by TMG, following fatigue. ↑= increase, ↓= decrease, – = unchanged.**

	<b>Dm</b>	<b>Tc</b>
<b>García-Manso et al (2011)</b>	↑	↑
<b>García-Manso et al (2012)</b>	↓	-
<b>Giovanelli et al (2015)</b>	↑	↓
<b>Wiewelhove et al (2015)</b>	↓ (trend)	↑

Among the few sport-specific applications of TMG assessment to be published, soccer has received the greatest attention. Studies focussing on injury have described how reconstructive surgery and subsequent rehabilitation, following anterior cruciate ligament injury, modified contractile mechanics of muscles in lower extremities on both the injured and uninjured soccer players (Alvarez-Diaz et al., 2014). Furthermore, (Alentorn-Geli, Alvarez-Diaz, Ramon, Marin, Steinbacher, Boffa, et al., 2014) proposed TMG as an appropriate screening tool to investigate knee flexor muscle stiffness as a risk factor for anterior cruciate ligament injury. Balance between contractile mechanics of knee flexors and extensors was also suggested as an important predictor for injury. Such studies have

strengthened the case for inclusion of TMG muscle contractile assessment within training (Rusu et al., 2013) and rehabilitation programmes (Alvarez-Diaz et al., 2014); although further integration of TMG with established physiological markers is required in order to validate the technique for use within high performance sport. Soccer has also been the basis for longitudinal research into contractile alterations across a ten week training cycle (García-García et al., 2015). Spatial and temporal parameters were influenced by the training period, furthermore, disparate responses were observed between soccer players and a control group. Greater insight could be provided by incorporating more regular measurements, as well as overlapping mechanical assessments with functional performance measures (e.g. MVC, CMJ).

## 1.6 Conclusions and Implications

Investigating skeletal muscle contractile function either through voluntary recruitment, by capturing firings of MUAPs from EMG, or through recording muscle deformation during a single electrically-elicited twitch, can provide non-invasive insight into muscle status in an acute setting or longitudinally. Therefore, such non-invasive approaches may have application within performance sport. In particular, neuromuscular adaptations to training and competition can be tracked, while data can also be used to inform best practice regarding preparation and recovery strategies. Further research is required to relate alterations in contractile and recruitment mechanics to functional adaptation, or indeed impairment. Additionally, the specific insights that can be garnered from single muscle assessments must be more deeply investigated, as *in vivo* muscles seldom work in isolation. Therefore the overall aim of this thesis is to explore critical components of high performance related preparation, exploiting non-invasive methodologies and techniques

which allow unhindered maintenance of regular training practices. The value of TMG in assessing muscle mechanics, in relation to adaptive responses and overall function will be investigated. Muscle recruitment behaviour will also be examined, to link central elements of the neuromuscular system to peripheral mechanics. The applicability of using TMG within a high performance sports setting will additionally be evaluated. In order to accomplish this, the experimental objectives of the thesis are:

- To evaluate the sensitivity of tensiomyography to quantify acute spatial and temporal alterations in skeletal muscle, in response to localised peripheral fatigue.
- To establish the applicable relationship between muscle twitch deformation measurements and muscle function, through examination of bilateral asymmetry.
- To assess acute alterations in motor unit recruitment and firing behaviour following eccentric exercise-induced muscle damage.
- To investigate both the acute and chronic influence of self-myofascial release on skeletal muscle contractile properties.
- To examine specific adaptations to high performance training programmes at the individual muscular level, in elite level athletes.

# Chapter 2

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## Radial Displacement Assessment of Skeletal Muscle Contractile Properties Following Electrically Elicited Fatigue

Lewis J Macgregor, Massimiliano Ditroilo, Iain J Smith, Malcolm M Fairweather, Angus M Hunter

## 2.1 Introduction

Before embarking on a series of studies designed to investigate muscle contractile mechanics within the context of high performance sport, it is first important to establish the validity of one of the main tools of assessment: TMG, within an appropriate setting. Fundamental to informed sports preparation is the ability to identify and quantify muscle fatigue. Muscle fatigue is characterised by a decrease in the external force or torque generating capacity (Garrandes, Colson, Pensini, Seynnes, & Legros, 2007), and/ or by impairment in peak power output (Ditroilo, Watsford, et al., 2011). The manifestation and magnitude of this reduced function depends upon multiple factors; including muscle contraction mode (Garrandes et al., 2007), the nature of fatigue protocol (Babault, Desbrosses, Fabre, Michaut, & Pousson, 2006) and the source of fatigue (Berchicci, Menotti, Macaluso, & Di Russo, 2013). Fatigue-related alterations of skeletal muscle can be observed, amongst other factors, by changes in contractile and mechanical properties.

Since fatigue is a condition that affects both athletic performance and recovery timeframe, the need for a valid monitor of muscle response is important to enable optimal management of athletes. In situations of muscle fatigue, or indeed musculoskeletal injury, it is impractical to assess muscle function through a measure which makes use of voluntary efforts (i.e. maximal voluntary contraction [MVC]), due to centrally mediated inhibition (Graven-Nielsen, Lund, Arendt-Nielsen, Danneskiold-Samsøe, & Bliddal, 2002). Furthermore, the potential for aggravation of any damage to the musculoskeletal unit cannot be ruled out. Having been developed over the last 20 years, tensiomyography (TMG) is a portable and non-invasive means of measuring muscle response through combined use of sub-maximal (below voluntary maximal activation) electrical stimulus and a highly sensitive

digital displacement sensor (Dahmane, Valenčič, Knez, & Eržen, 2001; Dahmane, Djordjevic, Šimunič, & Valenčič, 2005; Pišot et al., 2008), similar to that used in mechanomyography (Orizio, 2004). TMG records spatial and temporal parameters of the radial displacement of the muscle belly in response to electrical stimuli (Tous-Fajardo et al., 2010) and is reliable within (Križaj et al., 2008) and between days (Šimunič, 2012). Furthermore, TMG has also demonstrated good long-term stability following fatigue (Ditroilo et al., 2013) and has displayed significant interclass correlation coefficient with decline and recovery of MVC following exercise-induced muscle damage (Hunter et al., 2012).

TMG has successfully detected fatigue-associated changes, following ultra-endurance triathlon (García-Manso et al., 2011), and resistance exercise (García-Manso et al., 2012). However, these studies report inconsistent results in the fatigue-induced alteration of the TMG parameters, perhaps due to the vast differences in the fatigue protocols administered and the different muscles measured. Furthermore, previous studies have failed to relate TMG alterations to any valid functional measure, such as MVC or resting muscle tension (RMT), which leaves the physiological interpretation of the TMG data open to question. Therefore, in order to effectively provide meaningful validation of TMG measurement, to local fatigue, it is important to overcome this limitation. In practical terms, sub-maximal TMG could offer an attractive measure for athletes and sport practitioners in assessment of muscle response and status following fatigue based activity, without necessitating voluntary contractile effort.

Accordingly, the aim of the present investigation was to evaluate peripheral fatigue-induced alterations in mechanical and contractile properties of Gastrocnemius Medialis (GM), as measured by TMG. GM is one of the propulsive muscles, fundamental to different

types of human locomotion and is located superficially, making it clearly measurable by TMG. MVC and RMT were measured before and after intervention, to quantify the extent of muscle fatigue, and allow clearer interpretation of changes in TMG response; to our knowledge this has not been previously reported. The fatigue intervention used in the current investigation differs from previous studies in this area (García-Manso et al., 2011; García-Manso et al., 2012) in a number of key ways. Firstly, fatigue was induced locally with a low frequency stimulation that will necessitate a prolonged recovery, compared to higher frequency fatigue (Allen, Lamb, & Westerblad, 2008). Secondly, as motor unit discharge rarely exceeds 30Hz during voluntary contraction (Allen et al., 2008), low frequency stimulus can be considered a more functionally relevant intervention. Finally, as TMG is a passive and peripheral measurement it will minimise confounding variables such as the variability of central control factors.

## 2.2 Methods

### 2.2.1 Participants

Twenty-one healthy males with no history of neuromuscular or musculoskeletal disorders volunteered to undertake the study, after providing written informed consent. Volunteers were recruited from the University population and surrounding area. Participant characteristics included mean ( $\pm$  SD) age, height, and mass of  $21.3 \pm 3.4$  years,  $182.0 \pm 6.1$ cm, and  $79.5 \pm 10.0$  kg. The study was performed in accordance with the principles outlined in the *Declaration of Helsinki* and was approved by the University of Stirling Sports Studies Ethics Committee (SSEC).

### 2.2.2 Study Design

Mechanical and contractile properties of the right GM were monitored using TMG (BMC Ltd, Ljubljana). Participants were also tested for RMT and MVC of the right plantar flexors. Testing was carried out on two occasions, one week apart, as illustrated in Figure 2.1. Measurements were taken at a number of time points pre- and post- either control or fatigue intervention, according to the following order: TMG and RMT (measurement 1 [T1]); warm-up; TMG and RMT + MVC (T2); either control or fatigue intervention in random order; TMG and RMT + MVC (T3). Both TMG and RMT measurements were recorded three minutes after the warm-up, and after the control or fatigue intervention, to limit the effects of post activation potentiation in the GM muscle (Pääsuke et al., 2007). Participants reported to the laboratory on the morning of each experimental trial, in a fasted and rested state. Twenty-four hour dietary intake records were completed on the day preceding each trial, and participants were instructed to replicate their dietary intake before each visit.



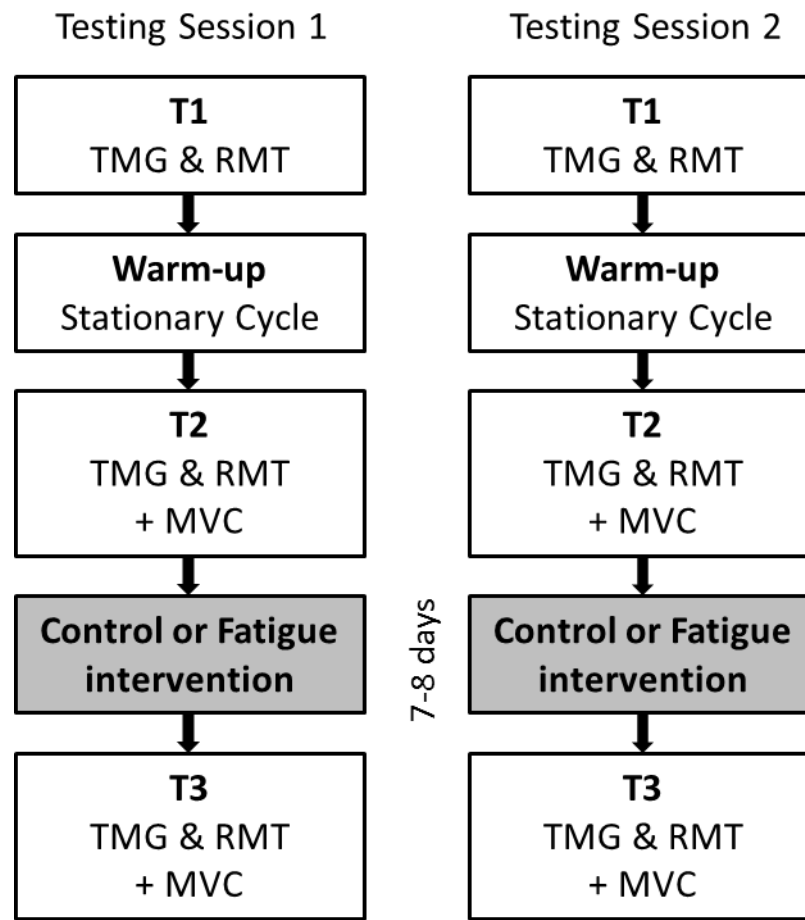


Figure 2.1 Schematic representation of the research design. TMG = Tensiomyography; RMT = resting muscle tension; MVC = maximal voluntary contraction

### 2.2.3 Warm-up

Participants warmed up by cycling at a low intensity (75 Watts) on an electromagnetically braked cycle ergometer (Lode Excalibur Sport V2, Lode BV, Groningen, Netherlands) for 5 minutes at a cadence between 80 and 90 rpm.

### 2.2.4 TMG Protocol

TMG measurements were performed in a prone position on a padded bench. A hemi-cylindrical foam pad, placed slightly proximal to the ankle joint, supported a knee flexion angle of around 5°. The digital displacement transducer (GK 40, Panoptik

d.o.o., Ljubljana, Slovenia), incorporating a 0.17 N/mm spring, was then positioned perpendicular to the muscle belly of the right GM with a controlled initial pressure of  $1.5 \times 10^{-2}$  N/mm<sup>2</sup>. This measuring position was selected by first manually palpating the GM at rest, to locate the thickest part of the muscle. If needed, the position was later adjusted slightly to obtain the highest mechanical response with the least amount of co-activation when externally stimulated; co-activation was typically identified by a second peak in the TMG response curve. Once the appropriate position was obtained, the skin was marked with semi-permanent ink to ensure exact uniformity when the sensor was repositioned for subsequent measurements. The centre point of each of the 2 stimulating electrodes (5 cm<sup>2</sup>) (Axelgaard, USA) was located approximately half way from the position of the sensor (~5cm) to the start of the respective GM proximal distal tendons. The anode was positioned distally and the cathode placed proximally. After each measurement these electrodes were left in place and unplugged to avoid any possible changes in muscle response via alterations in surface electrodes distance (Tous-Fajardo et al., 2010). A single 1 ms wide stimulation pulse was delivered, which applied initial current amplitude of 20 mA. This amplitude was progressively increased by 10 mA increments until peak twitch response was obtained. In order to minimize the effects of fatigue and potentiation, rest periods of 10 seconds were allowed between each stimulation pulse. Typical peak responses were observed at amplitude between 40 and 70 mA and only the output data for that particular stimulation intensity were used for analysis. Figure 2.2 shows a typical TMG displacement/ time curve before and after the administration of the fatigue protocol. Output parameters were extracted and analysed from each peak twitch response (Tous-Fajardo et al., 2010): *Displacement* (Dm), the extent of maximal radial deformation (mm) of the muscle belly during contraction; *Contraction velocity* (Vc), the rate (mm·s<sup>-1</sup>) of contraction between 10%

and 90% of maximal displacement [ $V_c = D_{m80}/T_c$ ] where  $T_c$  = contraction time between 10% and 90% of peak radial displacement of the muscle belly;  $D_{m80}$  = the radial displacement occurring during the time period of  $T_c$  (Valenčič & Knez, 1997). Muscle contraction time ( $T_c$ ) has been widely reported in previous studies (Tous-Fajardo et al., 2010; García-Manso et al., 2011; García-Manso et al., 2012) as the temporal change from 10% - 90% of muscle  $D_m$ , providing a value relative to the spatial characteristics of each muscle. However, when assessing intramuscular alterations, i.e. pre- and post- fatigue, the significance of calculating  $T_c$  in this manner should be questioned. Indeed, in the absence of signal latency or altered  $V_c$ , a decrease in  $D_m$  will always associate with a decrease in  $T_c$  when calculated as described above. Apparent decreases in  $T_c$ , suggesting a faster twitch response, would be reported simply as a result of reduced overall muscle contraction ( $D_m$ ). It was therefore proposed that assessment of  $V_c$  could provide greater insight, when monitoring the fatigue status of a muscle.

### *2.2.5 Resting Muscle Tension (RMT) Protocol*

Measurements of RMT of the right plantar flexors were recorded on an isokinetic dynamometer (Kin-Com, Chattanooga Group Inc, USA). With the knee fully extended, the ankle was flexed to  $90^\circ$  and secured, to maintain this fixed position (Figure 2.3). Participants were instructed to completely relax once in position, and the mean passive force was recorded during a period of 15 seconds, as a measure of resting muscle tension in the plantar flexors in a static position (Fowles, Sale, & MacDougall, 2000). A single measure was taken to determine RMT, as subsequent stretching of the ankle joint would cause an accumulative stretch effect.

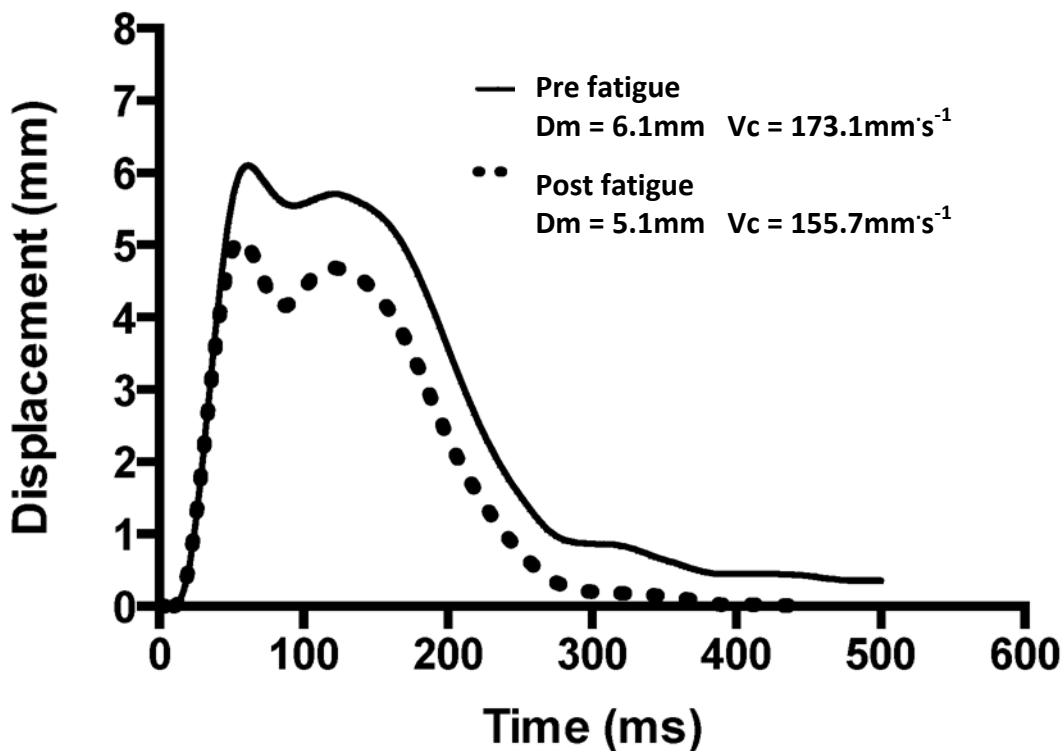


Figure 2.2 Typical displacement/ time curve of the tensiomyographic signal before and after the administration of the fatigue protocol. Dm = muscle displacement; Vc = contraction velocity

### 2.2.6 Fatigue Protocol

Whilst remaining secured in the same position as for the RMT measurement, participants received the fatigue intervention, which consisted of a 5 minute electrical stimulation of the right GM, to evoke fatigue. The stimulation protocol involved a train of 15 electrical pulses (1 every 100ms) with a 1 second gap before the start of each subsequent train. The protocol lasted 5 minutes and participants were asked to endure the maximum current they could, to ensure fatigue (~110 mA). The control intervention consisted of the same positioning but receiving no stimulation for a period of 5 minutes to account for the effect of time. Also in the same position, with the ankle placed at 90°, isometric MVC of the plantar flexors was measured, before and after both intervention and control, to assess whether fatigue occurred. Each participant performed three 5 second MVCs, with 60

seconds recovery between attempts. Participants were provided with consistent verbal motivation to ensure maximal effort throughout.



Figure 2.3 Isokinetic dynamometer setup for RMT and MVC assessment. Ankle flexed at 90° relative to the tibia

### 2.2.7 Statistical Analysis

All data are presented as mean ( $\pm$  SD). After testing for assumption of normality of the dependent variables and log-transforming where necessary, a 3 (measurements: before warm-up, T1; after warm-up, T2; after intervention, T3) x 2 (condition: control and fatigue intervention) ANOVA with repeated measures on both factors was used to detect differences in RMT and TMG parameters as a result of the fatigue/ control protocol. Where significance was detected a Tukey *post hoc* test was used to identify where any significant difference occurred. Paired *t-test* was conducted to compare the pre- / post-fatigue MVC difference between the control and fatigue intervention. Cohen's *d* effect sizes (ES) were calculated by:  $Cohen's\ d = \frac{Mean_1 - Mean_2}{SD_{pooled}}$ , where  $SD_{pooled} = \sqrt{[(SD_1^2 + SD_2^2) / 2]}$ ,

and 95% lower and upper confidence intervals (CI) were established relative to ES. ES were interpreted as  $< 0.2$  = trivial,  $0.2-0.6$  = small,  $0.6-1.2$  = moderate,  $1.2-2.0$  = large,  $> 2.0$  = very large (Cohen, 1988). The percentage differences between control and fatigue intervention were also calculated and interpreted based on the minimum detectable change as reported in a previous reliability study (Ditroilo et al., 2013). An alpha level of  $p < 0.05$  was considered statistically significant. Statistical analysis was performed using Minitab 16 statistical software (Minitab Ltd., Coventry, UK).

## 2.3 Results

### 2.3.1 MVC and RMT

Plantar flexor isometric MVC exhibited a significant ( $p < 0.001$ ) decline following the fatigue intervention ( $-122.6 \pm 104.0$  N) but not following control ( $p = 0.115$ ,  $-25.7 \pm 71.3$  N) (Table 2.1). The RMT exhibited a significant interaction of 'condition x measurement' ( $F_{(2,20)} = 5.9$ ,  $p = 0.005$ ). The *post-hoc* analysis revealed at T3 that fatigue caused significantly ( $p = 0.007$ ) more tension than control (CI [-0.05 to 1.18] ES = 0.64), ( $139.8 \pm 54.3$  vs.  $111.3 \pm 44.6$  N) and a percentage difference of 20.4% (Figure 2.4).

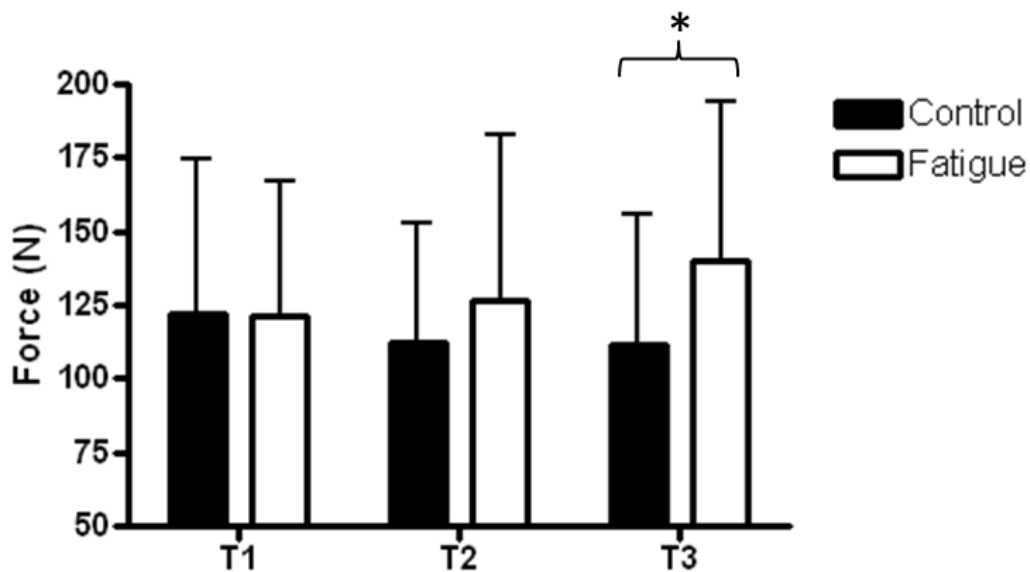


Figure 2.4 Mean values ( $\pm$  SD) of resting muscle tension (RMT) as assessed on the isokinetic dynamometer at the three measurement points. \* = significant difference between conditions at T3,  $p < 0.01$

Table 2.1 Mean ( $\pm$  SD) MVC in control and fatigue groups. T2 = pre-intervention, T3 = post-intervention.  
# = Significantly different from T2 in the fatigue condition,  $p < 0.001$

	Measurement	T2	T3
Control	Mean (N)	1236.6	1211.0
	$\pm$ SD	182.7	186.5
Fatigue	Mean (N)	1283.7	1161.1 #
	$\pm$ SD	238.2	232.1

Table 2.2 Mean ( $\pm$  SD) contraction velocity (Vc) as assessed by tensiomyography at the three measurement points

	Measurement	T1	T2	T3
Control	Mean ( $\text{mm}\cdot\text{s}^{-1}$ )	124.7	124.9	139.8
	$\pm$ SD	45.4	44.7	50.6
Fatigue	Mean ( $\text{mm}\cdot\text{s}^{-1}$ )	121.8	121.3	131.3
	$\pm$ SD	43.2	45.7	44.6



### 2.3.2 TMG Parameters

Dm demonstrated a fatigue-associated alteration. A significant main effect for 'condition' ( $F_{(1,20)} = 7.2$ ,  $p = 0.002$ ) was documented for Dm, along with a *post-hoc* difference at T3 demonstrating that the fatigue condition was significantly ( $p = 0.031$ ) lower than control condition ( $3.3 \pm 1.2$  vs  $4.0 \pm 1.4$  mm) (Figure 2.5). An ES of 0.5 was detected along with a percentage difference of 17.7%. No significant difference was found between conditions ( $F_{(1,20)} = 0.37$ ,  $p = 0.543$ ) or measurements ( $F_{(2,20)} = 1.02$ ,  $p = 0.365$ ) for Vc (T1  $121.8 \pm 43.2$  vs  $124.7 \pm 45.5$ , T2  $121.3 \pm 45.7$  vs  $124.9 \pm 44.7$ , T3  $131.3 \pm 44.6$  vs  $139.8 \pm 50.6$  mm·s<sup>-1</sup>). For condition, ES and percentage differences of 0.06, 2.3%; 0.08, 2.9%; 0.17, 6.1% (T1, T2 and T3 respectively) were detected (Table 2.2). No significant differences were encountered for any of the remaining muscle twitch parameters. All remaining parameters are presented in Appendix A.

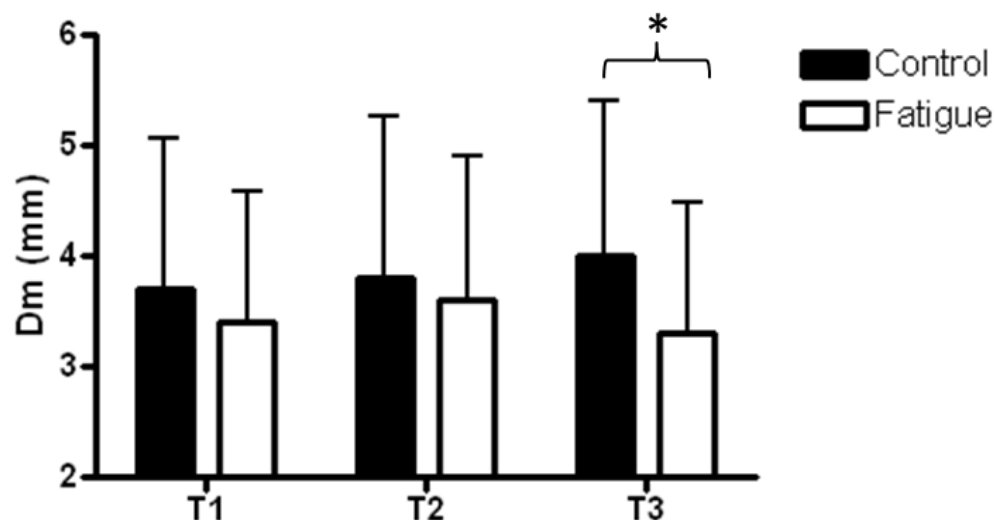


Figure 2.5 Mean values ( $\pm$  SD) of muscle displacement as assessed by tensiomyography at the three measurement points. \* = significant difference between conditions at T3,  $p < 0.05$

## 2.4 Discussion

This study was designed to evaluate the validity of TMG, as a sub-maximal assessment method, to detect local muscular fatigue, against functional physiological measures. Fatigue of GM was achieved, as evidenced by significant decline in peak force (MVC), which was absent following the control condition. This alteration in functional capacity of the muscle was associated with a significant decline in TMG Dm, similar to previous studies following dynamic fatigue (García-Manso et al., 2012; Carrasco, Sañudo, De Hoyo, Pradas, & Da Silva, 2011). In addition, plantar flexor RMT increased following the fatigue intervention suggesting that the Gastrocnemius skeletal musculotendinous unit became stiffer. Despite these alterations, muscle twitch Vc appeared to remain unaffected by fatigue.

It has been reported previously that a stiffer muscle will produce a reduced TMG Dm measurement (Pišot et al., 2008); however when considering the physiological effects of fatigue there are other important variables to examine. It has previously been demonstrated that during fatigued voluntary contractions muscle fibre conduction velocity declines, due to a reduction in extracellular pH (Hunter, De Vito, Bolger, Mullany, & Galloway, 2009). It is likely that this occurs due to a pH driven alteration of the Na<sup>+</sup> and K<sup>+</sup> gradient across the sarcolemma (Brody et al., 1991), impairing action potential propagation. Therefore, during TMG measurement the electrical stimulus applied to the surface of the fatigued muscle should result in reduced speed of the action potentials propagated, to reduce Ca<sup>2+</sup> release and subsequent excitation-contraction (EC) coupling. Low-frequency fatigue, as characterized by a disproportionate reduction in force at lower stimulation frequencies, has been associated with EC uncoupling (Hill, Thompson, Ruell, Thom, & White,

2001). It has been suggested that EC uncoupling is attributable to, amongst other factors, impaired  $\text{Ca}^{2+}$  transport via Ryanodine receptor channels in the triadic compartment (Balog, 2010). Furthermore, other contributing factors will be from increased  $\text{P}_i$  which can push the cross-bridge into a low force generating status (Nocella et al., 2011) and may also cause actin-myosin detachment (Takagi, Shuman, & Goldman, 2004). These altered characteristics of muscle function will inevitably impair its force generation capacity, as shown by the significant decline in MVC. Interestingly, García-Manso et al. (2011) showed an *increase* in Biceps Femoris TMG Dm following an ironman triathlon. The precise reasons for this disparity are unclear; however Morin, Tomazin, Edouard, & Millet (2011) showed a small decline in whole leg stiffness during a running task, following a 24-hour marathon. These authors also postulated that central fatigue would have been apparent which would have been linked to altered peripheral feedback from muscle afferents triggered from cytokine release. This, we suggest, may be why an increase in TMG Dm was observed following an ironman triathlon when a decline has been reported with other types of fatigue from far shorter contractile/ exercise durations.

As mentioned, increased muscle stiffness, as we have evidenced here by the rise in RMT (Figure 2.4), is another critical factor contributing to the decline in TMG Dm. Other studies have also demonstrated alterations in Dm alongside muscle architectural changes. Firstly, Pišot et al. (2008) showed that following 35 days of bed rest, TMG Dm increased alongside the reduction in muscle thickness which the authors suggested would have contributed to reduced muscle stiffness. Secondly, Ditroilo, Hunter, Haslam, & De Vito (2011) demonstrated that altering the length of the muscle, through altering joint angle, will determine the magnitude of TMG parameters. Thirdly, although not relating the decline in

TMG Dm to muscle stiffness changes, other studies (García-Manso et al., 2012; Carrasco et al., 2011) have also demonstrated a decline in TMG Dm following fatigue, suggesting that this is an important parameter when assessing the muscle status in this regard. In the present study we observed decreases in TMG Dm without significant alterations in Vc. Given previously described reductions in action potential propagation and muscle fibre conduction velocity associated with fatigue (Hunter et al., 2009), it may have been expected that TMG Vc would be observed to decline post-fatigue, in concurrence with Dm. It is plausible that the lack of significant alteration in Vc is due to the high degree of inter-individual variability associated with the measurement. Such an assertion may be reinforced by the small effect size that we have reported, associated with Vc (Cohen, 1988). The comparably low amplitude of the electrical stimulation used to elicit the peak TMG response, may perhaps render these data difficult to compare to existing conduction velocity findings. As such, it may be inappropriate to consider alterations in the speed/ time component of the TMG response when assessing muscle fatigue, with the focus instead being placed on spatial alterations (Dm), which we have shown here to be indicative of increased muscle stiffness.

The non-invasive nature of TMG assessment, coupled with its sub-maximal protocol, make for a preferable tool in measuring muscle fatigue in circumstances where a maximal muscle function test is inappropriate. Furthermore, TMG measurements are exempt from the bias of volitional effort and motivation, facilitating the incorporation of the procedure into existing programmes (Rey, Lago-Peñas, & Lago-Ballesteros, 2012). Yet, in any type of physiological measurement there will be a degree of variability. This variability has previously been accounted for, with TMG measured under different muscle conditions (Ditroilo et al., 2013) and Dm has been shown to be well within acceptable limits. Analogous

to this is establishing minimal detectable change so practitioners and researchers can be confident that the given magnitude of observed change following any intervention is real and physiologically significant. We have demonstrated in this study that the fatigue-altered Dm parameter (17.7%) clearly exceeds the minimal detectable change thresholds of 15.1% (Ditroilo et al., 2013).

## **2.5 Conclusion**

This is the first study to demonstrate that TMG was effective in detecting local muscular fatigue in gastrocnemius medialis. We propose that this response was directly related to increased stiffness of the muscle from impaired contractile capacity. It should be emphasised that, when assessing local muscular fatigue, Dm of the muscle, rather than Vc, is a valid measure. The current findings have important implications for researchers and practitioners seeking to establish fatigue status of skeletal muscle, with implications for training provision and injury management in performance sport. Given the nature of this type of measurement, TMG can also be successfully used to determine local muscular fatigue in cases where function assessments requiring maximal voluntary exertion are undesirable.

# Chapter 3

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## Radial Displacement Assessment of Bilateral Skeletal Muscle Asymmetry

Lewis J Macgregor, Thomas G Di Virgilio, Angus M Hunter

### 3.1 Introduction

In Chapter 2 we described the validity of skeletal muscle mechanical measurements, using TMG, in detecting physiologically relevant alterations associated with acute fatigue. It may also be pertinent to explore the sensitivity of TMG to subtler variations in contractile mechanics. Bilateral muscular asymmetry has previously been investigated in association with injury prevalence (Croisier, Forthomme, Namurois, Vanderthommen & Crielaard, 2002) and more recently, with regard to athletic ability (Bailey et al. 2013; Sugiyama et al., 2014). Indeed, bilateral asymmetry in strength has been proposed as a predictor of injury (Croisier, Ganteaume, Binet, Genty, & Ferret, 2008) and sporting performance (Menzel et al., 2013; Trivers et al. 2014). Most commonly, bilateral strength asymmetry has been assessed among the knee extensors and flexors, to quantify functional impairments resulting from knee injury (Clark, 2001). Monitoring of asymmetry has also been conducted to assess the effectiveness of rehabilitation programmes and to inform when an individual is able to return to their sport or activity (Wilk, Reinold, & Hooks, 2003). Additionally, bilateral asymmetry has been proposed as a risk factor for musculoskeletal injuries (Croisier et al., 2008; Ekstrand & Gillquist, 1983; Knapik, Bauman, Jones, Harris, & Vaughan, 1991; Yamamoto, 1993). Accordingly, incorporation of an effective screening tool into sports preparation programmes also may be useful to identify athletes at increased injury risk.

Numerous existing screening tools for lower extremity injury prediction have been reviewed (Dallinga, Benjaminse, & Lemmink, 2012). It has been proposed that any screening test should be as relevant as possible to the activities undertaken by the individual (Maloney, Fletcher, & Richards, 2015), however many such tests may not be applicable within a field based setting. This lack of applicability was highlighted by Impellizzeri et al.

(2007) who recently developed a dynamic vertical jump test, to measure asymmetries in lower limb force; this test may provide a valid and specific measure, incorporating relatively high-velocity, stretch-shortening cycle, closed chain movements. However, the complexity of a vertical jump task and the number of components involved pose difficulties when attempting to isolate the specific source of asymmetry. Alternatively, isokinetic or isometric strength tests provide information on isolated or synergist muscle groups, but cannot specifically pinpoint issues at the level of individual muscles. Therefore, it remains unclear whether bilateral asymmetry within individual muscles translates to functional asymmetry in complex movements; moreover, quantification of asymmetry, through functional assessment carries inherent limitations due to reliance on volitional motivation to achieve peak measures, which may inadvertently bias the outcomes (Graven-Nielsen et al., 2002). As such, the non-invasive, passive nature of TMG may present an objective option for athlete screening.

Muscle stiffness, as we have measured through radial displacement, has previously been associated with sprint performance (Chelly & Denis, 2001) as well as isometric and concentric force (Brughelli & Cronin, 2008). Greater stiffness appears to maximise isometric and concentric performance (Wilson, Murphy, & Pryor, 1994), although excessive stiffness may point to muscle fatigue (as demonstrated in Chapter 2) or exercise-induced muscle damage (Hunter et al., 2012). Acute changes in displacement ( $D_m$ ) have shown moderate relationships to alterations in peak isometric force (Hunter et al., 2012), however, the degree to which measurements of individual muscle contractile properties (i.e. radial displacement and contraction velocity) can predict functional outputs, such as strength and power remains to be fully established. The portable nature of the TMG displacement sensor,



which records muscle twitch response to a single submaximal electrical stimulus, provides obvious practical possibilities for field-based application. Furthermore, the technique is non-invasive and will not interfere with any existing skeletal musculotendinous injuries or impede recovery. TMG also is unaffected by covariates, inherent within assessments of muscle strength, such as motivation. However, it is important to establish whether there is a direct relationship between asymmetry at the level of spatial and temporal parameters of individual muscle twitch characteristics, and the functional capability of the muscle group/limb.

Accordingly, the aim of this study was to assess the relationship between bilateral asymmetry at the level of individual muscle twitch (radial displacement and contraction velocity) and contractile function of the muscle group/entire limb. It was anticipated that these findings could lead towards establishing the efficacy of a model for predicting functional asymmetry based on a passive, non-invasive and field based measurement. Counter movement jump (CMJ) has displayed good reliability as a measure of lower-limb power, particularly in younger individuals (Ditroilo, Forte, McKeown, Boreham, & De Vito, 2011), while knee extension isometric (MVC) and isokinetic peak force can be reliably measured using an isokinetic dynamometer (Maffiuletti, Bizzini, Desbrosses, Babault, & Munzinger, 2007). Meanwhile, although TMG has shown strong reliability under a range of conditions (Križaj et al., 2008; Tous-Fajardo et al., 2010; Šimunič, 2012), decreased reliability was shown when measurements were spread over longer periods of time (Ditroilo et al., 2013); with resting conditions producing diminished reliability compared to exercised or fatigued conditions. It was therefore an additional aim of this study to compare the reliability of TMG, MVC and CMJ under identical conditions. It was hypothesized that

bilateral asymmetry in radial and temporal contractile parameters of muscles within the knee extensors, as quantified by TMG, would predict asymmetry in knee extension strength and, to a lesser degree, lower-limb power. In particular it was predicted that asymmetry in  $D_m$  would relate to asymmetry in strength, whilst asymmetry in  $V_c$  would more closely relate to asymmetry in power. Given the greater objective control contained in the measurement, it was hypothesised that TMG would provide greater reliability than MVC or CMJ.

## 3.2 Methods

### 3.2.1 Participants

Twenty-five healthy individuals (10 = male, 15 = female) with no history of neuromuscular or musculoskeletal disorders volunteered to undertake the study, after providing written informed consent. Volunteers were recruited from the University population and surrounding area. Mean and standard deviation (SD) age, body mass and height are shown in Table 3.1, along with limb dominance. All aspects of the study were performed in accordance with the latest revision of the *Declaration of Helsinki* and the study was approved by the School of Sport, Research Ethics Committee (SSEC), University of Stirling.

### 3.2.2 Study Design

Participants were fully familiarised with all testing procedures  $\geq 48$ h prior to commencement of the experimental trials, with two familiarisation sessions carried out on separate days. Two identical trials were then completed, with 7d between each visit (Figure 3.1). Participants reported to the laboratory following an overnight fast and initially rested in a supine position for 30 minutes. Following this rest period, the mechanical and contractile properties of vastus lateralis (VL), rectus femoris (RF) and vastus medialis (VM) of both legs were measured using TMG (BMC Ltd, Ljubljana). The order in which the limbs were assessed was randomized. The randomized order was maintained for all subsequent testing with each participant. Peak isometric and isokinetic knee extension force were next assessed using an isokinetic dynamometer (Kin-Com Chattanooga Group Inc, Chattanooga, Tennessee, USA). Unilateral counter movement jumps (CMJ) were then performed to assess

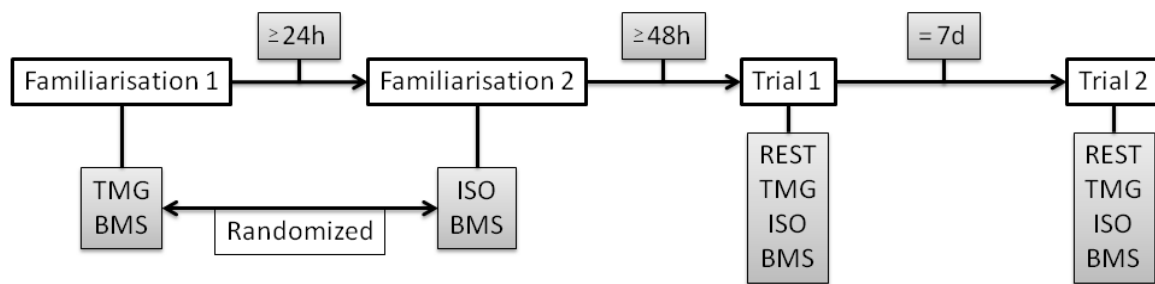
lower limb power. Finally, a series of thirty consecutive bilateral CMJs also were performed during which sEMG was recorded from the VL of each limb.

**Table 3.1 Participant characteristics. (n = 25). Limb dominance was determined as the leg favoured when kicking a ball. n = 15 male, 25 female.**

	<b>Age (y)</b>	<b>Mass (kg)</b>	<b>Height (cm)</b>	<b>Limb dominance (%)</b>	
<b>Mean</b>	23.67	67.96	170.28	<b>Right</b>	96
<b>± SD</b>	5.66	10.87	7.88	<b>Left</b>	4

### 3.2.3 TMG Protocol

Following 30 minutes rest, muscle twitch response was measured from VL, RF and VM of each leg, in a randomized order. From each twitch two parameters were recorded: Dm the peak radial deformation of the muscle belly during contraction (mm), and contraction velocity (Vc) the rate of contraction between 10% and 90% of Dm ( $\text{mm}\cdot\text{s}^{-1}$ ). Measurements were performed with the subject adopting a supine position. Knee joint angle of 60° from full extension was maintained with a triangular foam pad supporting the leg (Figure 3.2). A highly sensitive displacement transducer (TMG-BMC, Slovenia) and a pair of self-adhesive stimulating electrodes (30mm diameter) were located upon the muscle belly as described in Chapter 2. Data collection, processing and extraction were completed exactly as described in Chapter 2.



**Figure 3.1** Timeline of laboratory visits. TMG – tensiomyography, ISO – isokinetic dynamometer, BMS – ballistic measurement system, REST – 30min rest.

### 3.2.4 Isometric and Isokinetic Strength Assessment

Participants were positioned on an isokinetic dynamometer (Kin-Com Chattanooga Group Inc, Chattanooga, Tennessee, USA), and secured using nylon straps, according to the manufacturer's guidelines. Gravitational corrections were performed, in accordance with existing recommendations (Gleeson & Mercer, 1996). First a standardized isometric warm up was performed, consisting of 3 x 5s contractions at 50% effort and 3 x 5s contractions at 75% effort, with 30s recovery between each contraction (Balshaw & Hunter, 2012). Maximal isometric voluntary contraction (MVC) was subsequently measured at a knee joint angle of 60° (0° = full extension), the limb was secured by a Velcro strap proximal to the medial malleolus. The angle of 60° was chosen, as it lies within the well-established range of reported optimal knee joint angles, for peak isometric torque production (Knapik, Mawdsley, & Ramos, 1983). Participants were instructed to exert peak force as quickly as possible and to hold each contraction for 5s. Consistent verbal encouragement was provided by the investigator throughout, but no feedback was provided regarding force output. Participants performed three contractions with 60s recovery between each. The highest force output achieved was designate MVC and recorded for analysis. Peak isokinetic

concentric (CON) and eccentric (ECC) torque were next assessed at velocities of  $60^{\circ}\text{s}^{-1}$  and  $120^{\circ}\text{s}^{-1}$ . Three contractions were performed at each velocity for both CON and ECC, between knee angles of  $20^{\circ}$  and  $80^{\circ}$ . A recovery of 30s was allowed between each of the  $60^{\circ}\text{s}^{-1}$  contractions and between each of the  $120^{\circ}\text{s}^{-1}$  contractions. A further 60s recovery was permitted following the third  $60^{\circ}\text{s}^{-1}$  contraction, prior to the first  $120^{\circ}\text{s}^{-1}$  contraction.



Figure 3.2 TMG setup to measure rectus femoris.

### 3.2.5 Power Assessment

Prior to assessment of lower limb power, participants were fitted with 2 pairs of circular Ag/Aug self-adhesive electrodes (VERMED A10008-100 ECG, Vermont, USA), affixed to the muscle belly of each VL, with an inter electrode distance of 20mm (Hermens, 2000), a reference electrode was placed on the patella and secured with micro-pore tape. Participants first performed a maximal isometric squat with a knee joint angle of  $70^{\circ}$  ( $0^{\circ}$  = full extension). Participants completed 3 x 5s maximal efforts, with 60s recovery permitted between exertions. Ground contact force was recorded through a force platform (400S Force Plate, Innervations, Adelaide, SA, Australia) linked to data collection software

(AcqKnowledge® 3.8.1, Biopac Systems Inc, California, USA), and VL sEMG was recorded using BioPac MP100, linked to the same collection software. Peak EMG amplitude (root mean squared: RMS) was recorded from the isometric squat containing the highest ground contact force and all subsequent EMG signals were normalised to these values (Padulo et al., 2013). Three maximal CMJs were performed unilaterally, using each leg. A lightweight wooden beam (0.6kg) was held parallel to the ground, across the shoulders; the depth of squat phase during each CMJ was controlled by the wooden beam contacting wooden blocks positioned on both sides of the participant at a height which allowed the participant to achieve a knee joint angle of 70°. Each CMJ was performed in one continuous motion. Peak CMJ height was measured as the vertical displacement of the wooden beam, which was affixed to a linear transducer, connected to the data acquisition unit (Biopac Systems, Inc. Goleta, CA, USA). Force at the point of take-off was recorded through the force platform, which was also connected to the same data acquisition unit. The greatest displacement and force from the three CMJs were recorded and stored for each leg. Participants finally performed thirty consecutive CMJs using an identical setup to that described above, but CMJs were performed bilaterally. Thirty CMJs were continuous with no rest permitted until completion of the set. Participants were instructed to exert maximum effort from the first CMJ and maintain for as long as possible. Displacement and take-off force were recorded throughout the CMJ protocol, along with VL sEMG.

### *3.2.6 Data Analysis*

TMG parameters (Dm and Vc) were calculated for RF, VL and VM as described in Chapter 2. Parameters were recorded for each individual muscle and as the mean of all muscles for each limb. Average RMS was calculated for a moving window 200ms time period

across the entire waveform. RMS processing was used to analyse EMG based on previous recommendations for research investigating neuromuscular activation levels (Hägg, Melin, & Kadefors, 2004). RMS processing was conducted by the software used to operate the EMG system (AcqKnowledge® 3.8.1, Biopac Systems Inc, Goleta, CA, USA), in accordance with the manufacturer's guidelines. Symmetry index (S.I. %) of all parameters was calculated between limbs, in accordance with the formula described by (Herzog, Nigg, Read, & Olsson, 1989):

$$\frac{X_a - X_b}{0.5 (X_a + X_b)} * 100$$

As lower limb dominance has been shown not to be a determining factor in bilateral asymmetry (Carpes, Mota, & Faria, 2010; Alvarez-Diaz et al., 2014) all S.I. values were normalized to the greater measurement, not to the dominant limb. Symmetry was determined in line with thresholds set out by Herzog et al., 1989): S.I. > 10% = asymmetric, S.I. ≤ 10% = symmetric and participants were grouped accordingly.

### 3.2.7 Statistical Analysis

All data are presented as mean (± SD). Dependent variables were found to be normally distributed. Analysis was performed using Minitab 16 statistical software (Minitab Ltd., Coventry, UK). Dm, Vc, MVC, isokinetic peak force and unilateral CMJ height and CMJ force were analysed using paired T-tests to compare limbs. All S.I. were analysed using two-sample T-tests to compare groups. RMS was analysed by conducting a three factor (group [2] x limb [2] x time [3]) repeated measures ANOVA. Statistical significance was accepted at P<0.05. Where significant effects were observed, Cohen's d effect sizes (ES) were calculated



by: Cohen's  $d = \text{Mean1} - \text{Mean2} / \text{SD}_{\text{pooled}}$ , where  $\text{SD}_{\text{pooled}} = \sqrt{[(\text{SD}_{12} + \text{SD}_{22}) / 2]}$ , and 95% lower and upper confidence intervals (CI) were established relative to ES. ES were interpreted as  $< 0.2 = \text{trivial}$ ,  $0.2-0.6 = \text{small}$ ,  $0.6-1.2 = \text{moderate}$ ,  $1.2-2.0 = \text{large}$ ,  $> 2.0 = \text{very large}$ . Pearson's correlation coefficients ( $r$ ) were analysed between variables and between limbs for Dm, Vc, MVC and CMJ. Statistically significant correlation was accepted at  $P < 0.05$ . Coefficient of variation (CoV) was calculated for all dependant variables =  $\text{SD} / \text{mean}$  (Atkinson & Nevill, 1998).



Figure 3.3 Unilateral CMJ. The wooden beam was used to control the depth of the squat phase, and was attached to a linear transducer to record displacement. CMJs were performed on a force platform.

### 3.3 Results

### 3.3.1 Muscle Function

Significantly higher ( $F_{(1,24)} = 13.24$ ,  $p = 0.002$ ) MVC was observed in the knee extensors of the dominant leg compared to non-dominant (7.9%, CI [25.5 to 70.4] ES = 0.34,  $p < 0.001$ ) (Figure 3.4). Peak concentric force was also significantly higher ( $F_{(1,24)} = 18.76$ ,  $p = 0.001$ ) at  $60^{\circ}\cdot s^{-1}$  and ( $F_{(1,24)} = 10.47$ ,  $p = 0.018$ ) at  $120^{\circ}\cdot s^{-1}$  in the dominant leg (12.4%, CI [28.0 to 80.8] ES = 0.42,  $p < 0.001$ ) and (11.5%, CI [12.0 to 69.0] ES = 0.32,  $p = 0.006$ ) respectively (Figure 3.5A). The dominant leg also demonstrated significantly ( $F_{(1,24)} = 29.90$ ,  $p < 0.001$ ) higher peak eccentric force at  $60^{\circ}\cdot s^{-1}$  and ( $F_{(1,24)} = 19.15$ ,  $p < 0.001$ ) at  $120^{\circ}\cdot s^{-1}$  compared to the non-dominant leg (15.6%, CI [66.1 to 139.3] ES = 0.53,  $p < 0.001$ ) and (13.6%, CI [48.2 to 123.0] ES = 0.48,  $p < 0.001$ ) respectively (Figure 3.5B). There was no significant difference ( $F_{(1,24)} = 1.28$ ,  $p = 0.275$ ) between dominant and non-dominant legs for peak CMJ height or ( $F_{(1,24)} = 0.14$ ,  $p = 0.713$ ) relative force at the point of take-off (Table 3.2).

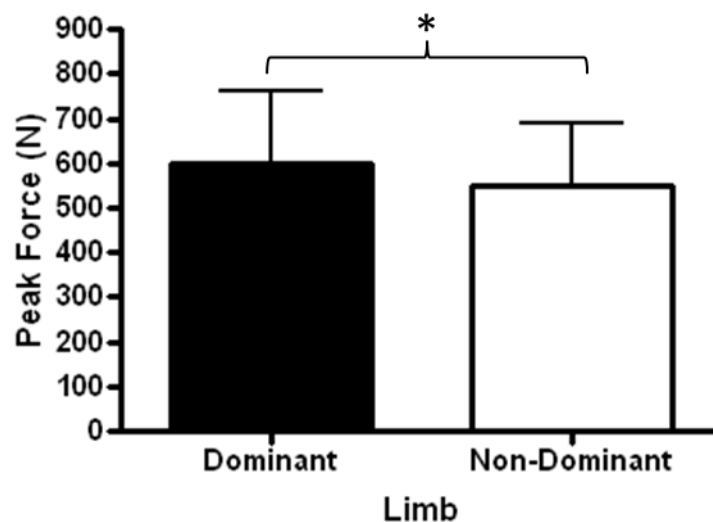


Figure 3.4 Maximal voluntary contraction (MVC) of the knee extensors in the dominant and non-dominant limbs. Values are mean  $\pm$  SD,  $n = 25$ . \* Significant difference between limbs,  $p < 0.001$

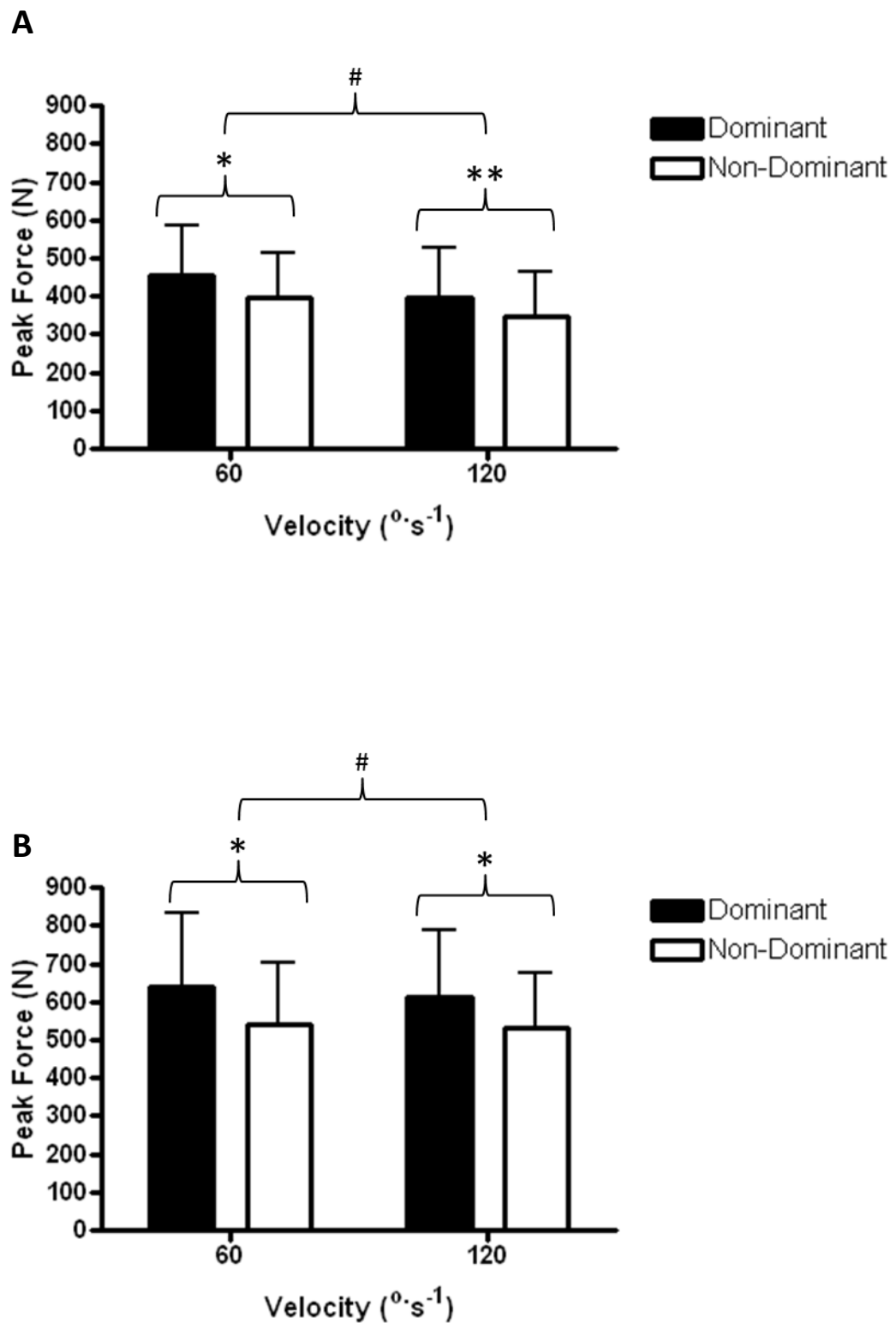


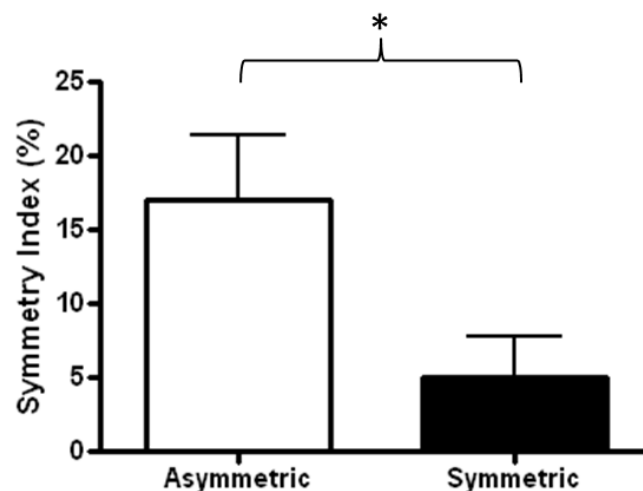
Figure 3.5 Peak concentric (A) and eccentric (B) force of the knee extensors at 60 and 120 $^{\circ}\cdot\text{s}^{-1}$ . Values are mean  $\pm$  SD, n = 25. \* Significant difference between limbs,  $p < 0.001$ . \*\* Significant difference between limbs,  $p < 0.05$ . # Significant difference between velocities,  $p < 0.05$ .

**Table 3.2** Unilateral countermovement jump (CMJ) peak height and peak take-off force relative to body mass, on the dominant and non-dominant limbs. Values are mean  $\pm$  SD, n = 25.

	Dominant		Non-Dominant	
	CMJ Height (cm)	CMJ Force (N.kg)	CMJ Height (cm)	CMJ Force (N.kg)
<b>Mean</b>	23.25	16.74	23.37	17.00
<b><math>\pm</math> SD</b>	2.40	1.90	2.07	1.68

### 3.3.2 Symmetry Indices

Nine participants were classified as bilaterally asymmetric (S.I. > 10%) based on S.I. of MVC, while sixteen participants were classified as bilaterally symmetrical (S.I.  $\leq$  10%). S.I. was significantly higher (CI [7.84 to 16.16] ES = 3.56,  $p < 0.001$ ) in the asymmetric group than in the symmetrical group ( $17.04 \pm 4.37$  vs  $5.04 \pm 2.69$  %) (Figure 3.6). S.I. was not significantly different between groups for peak concentric ( $p = 0.831$ ,  $p = 0.509$ ) and eccentric ( $p = 0.152$ ,  $p = 0.326$ ) force ( $60^{\circ}\text{s}^{-1}$  and  $120^{\circ}\text{s}^{-1}$ ), nor for peak CMJ height ( $p = 0.862$ ) (Table 3.3).



**Figure 3.6** Symmetry index (S.I. %) of MVC divided into (n = 9) asymmetric (S.I. > 10%) or (n = 16) symmetric (S.I.  $\leq$  10%) groups. Values are mean  $\pm$  SD. \* Significant difference between groups,  $p < 0.001$ .

**Table 3.3** Symmetry index (S.I. %) of peak concentric and eccentric knee extensors force at 60 and 120°s<sup>-1</sup> and of countermovement jump (CMJ) height, divided into (n = 9) asymmetric (S.I. > 10%) or (n = 16) symmetrical (S.I. ≤ 10%) groups. Values are mean ± SD.

		Con 60°s <sup>-1</sup>	Con 120°s <sup>-1</sup>	Ecc 60°s <sup>-1</sup>	Ecc 120°s <sup>-1</sup>	CMJ Height
<b>Asymmetric</b>	<b>Mean (%)</b>	13.72	13.14	22.05	18.49	5.95
	<b>± SD</b>	9.64	10.45	10.09	13.17	3.94
<b>Symmetric</b>	<b>Mean (%)</b>	14.75	15.23	17.01	14.48	3.92
	<b>± SD</b>	11.45	13.20	12.18	13.72	2.28

### 3.3.3 Contractile Properties

Knee extensor radial displacement was significantly ( $F_{(1,24)} = 5.69$ ,  $p = 0.018$ ) higher in the non-dominant leg compared to the dominant leg (6.5%, CI [0.113 to 0.768] ES = 0.29,  $p = 0.009$ ) (Figure 3.7A). VM displayed significantly ( $p = 0.013$ ) higher radial displacement in the non-dominant leg compared to dominant (10.4%, CI [0.173 to 1.416] ES = 0.34,  $p = 0.013$ ), RF showed a tendency ( $p = 0.068$ ) towards greater radial displacement in the non-dominant leg, while there was no significant difference ( $p = 0.729$ ) between legs in radial displacement of VL (Figure 3.7B). Knee extensor contraction velocity was significantly ( $F_{(1,24)} = 5.21$ ,  $p = 0.031$ ) higher in the non-dominant leg compared the dominant leg (6.8%, CI [3.57 to 22.40] ES = 0.30,  $p = 0.008$ ) (Figure 3.8A). RF displayed significantly ( $p = 0.009$ ) faster contraction velocity in the non-dominant leg compared to dominant (10.4%, CI [5.65 to 37.31] ES = 0.39,  $p = 0.009$ ), VM showed a tendency ( $p = 0.052$ ) towards faster contraction velocity in the non-dominant leg, while there was no significant difference ( $p = 0.912$ ) between legs in contraction velocity of VL (Figure 3.8B).

S.I. of radial displacement was significantly higher (CI [13.13 to 46.10] ES = 1.33,  $p = 0.003$ ) in the asymmetric group than in the symmetrical group for RF, there was no significant difference between groups for S.I. of radial displacement in VL ( $p = 0.097$ ) nor VM ( $p = 0.146$ ) (Figure 3.9A). S.I. of contraction velocity was significantly higher (CI [9.20 to 45.10] ES = 2.11,  $p = 0.008$ ) in the asymmetric group than in the symmetrical group for RF, there was no significant difference between groups for S.I. of contraction velocity in VL ( $p = 0.238$ ), VM displayed a tendency ( $p = 0.062$ ) towards greater S.I. of contraction velocity in the asymmetric group (Figure 3.9B). All individual data are presented in Figure 3.10 (Dm) and Figure 3.11 (Vc).

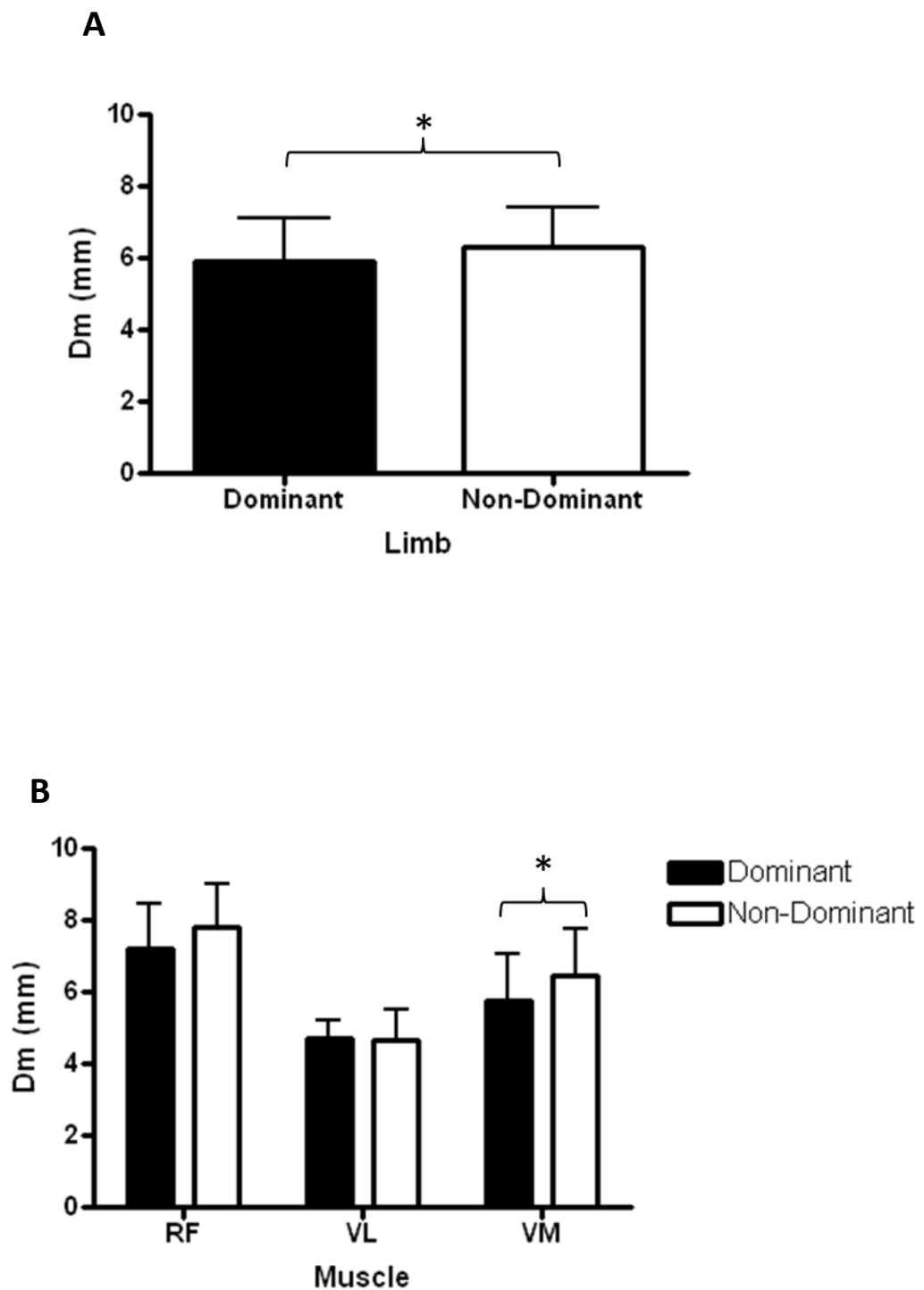


Figure 3.7 Peak radial displacement (Dm) of combined (A) and individual (B) RF, VL and VM in the dominant and non-dominant limbs. Values are mean  $\pm$  SD, n = 25. \* Significant difference between limbs,  $p < 0.05$ .

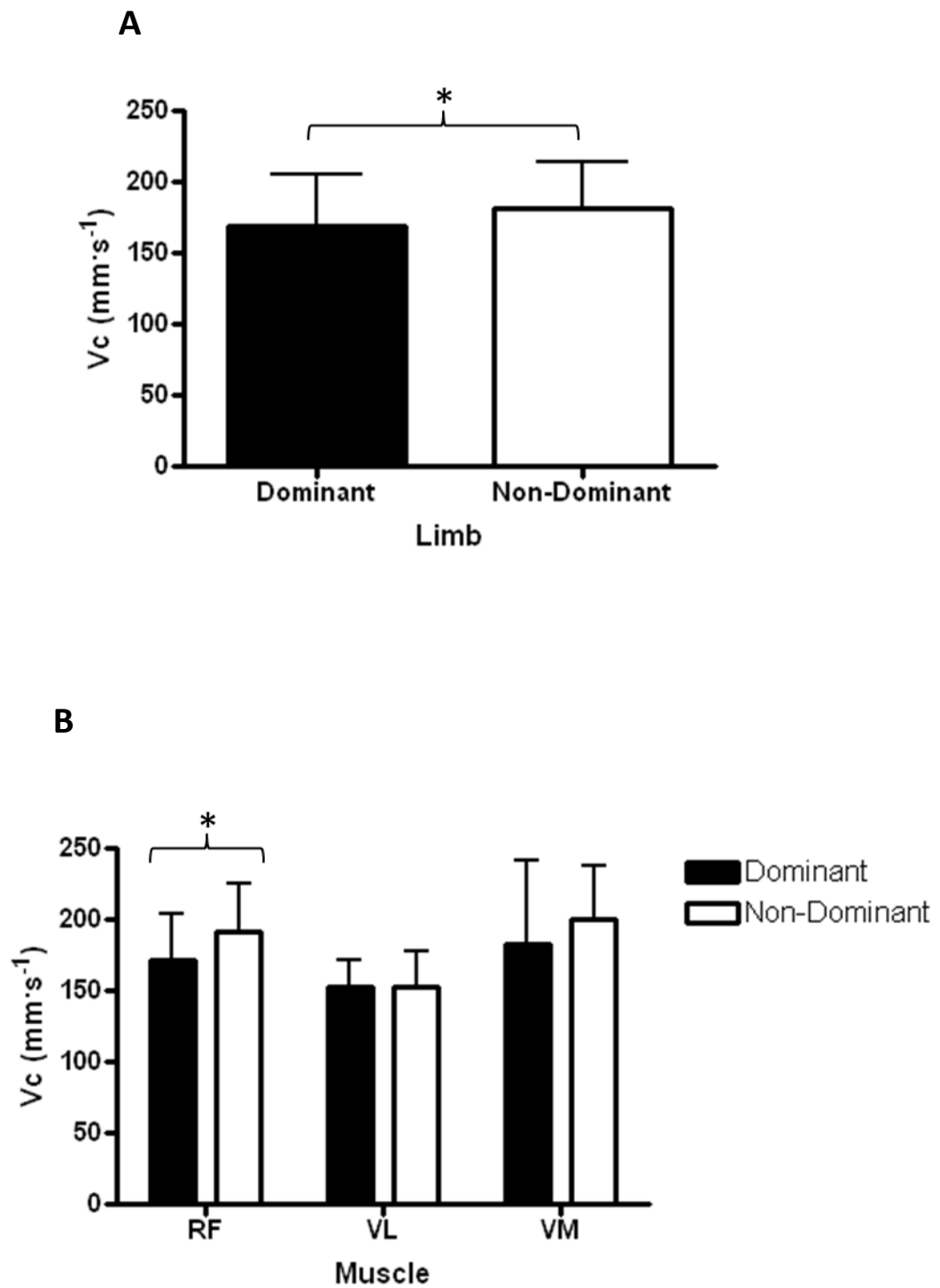


Figure 3.8 Peak contraction velocity ( $V_c$ ) of combined (A) and individual (B) RF, VL and VM in the dominant and non-dominant limbs. Values are mean  $\pm$  SD,  $n = 25$ . \* Significant difference between limbs,  $p < 0.05$ .



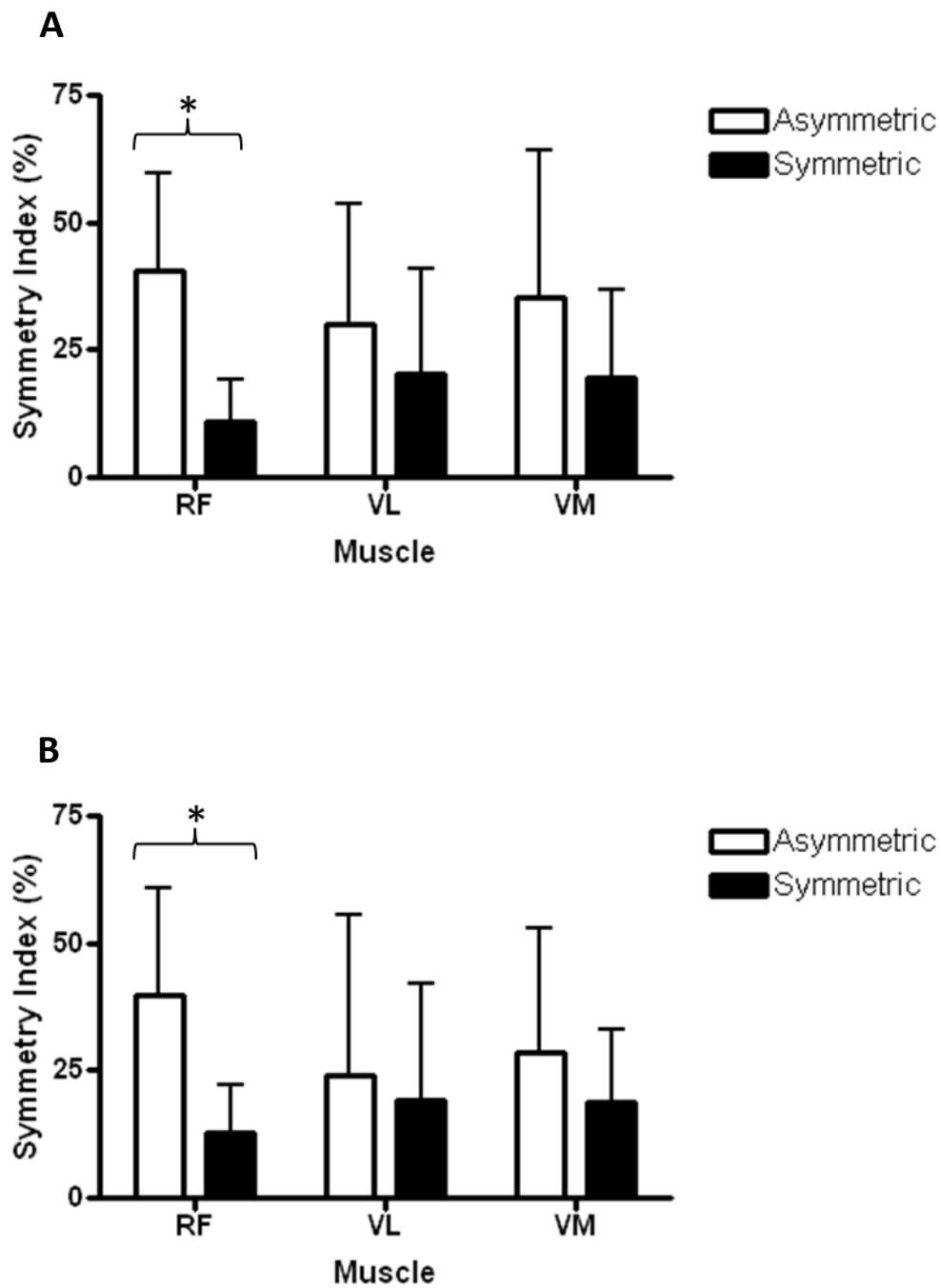


Figure 3.9 Symmetry index (S.I. %) of Dm (A) and Vc (B) of RF, VL and VM, divided into ( $n = 9$ ) asymmetric (S.I.  $> 10\%$ ) or ( $n = 16$ ) symmetric (S.I.  $\leq 10\%$ ) groups. Values are mean  $\pm$  SD. \* Significant difference between groups,  $p < 0.01$ .

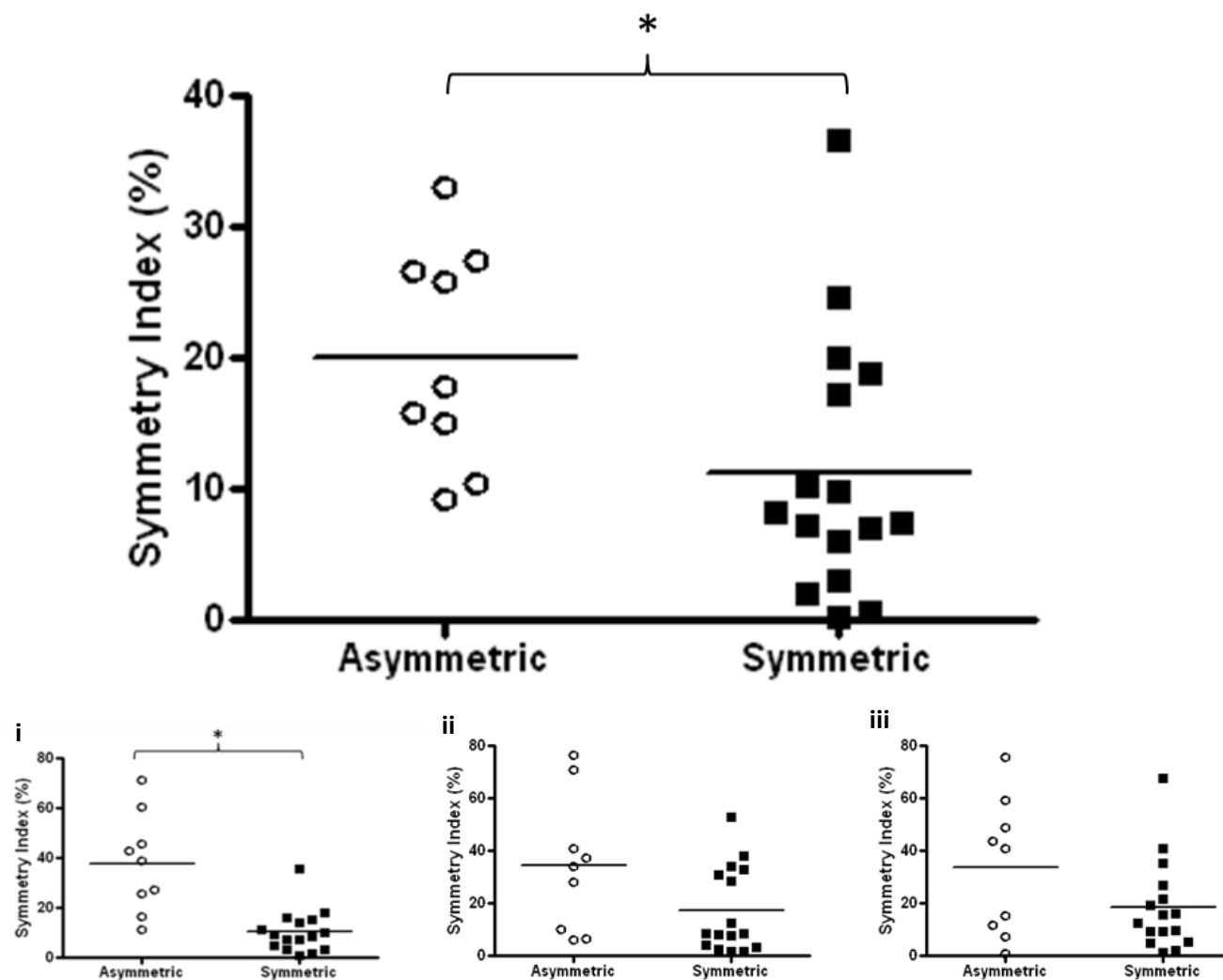


Figure 3.10 Individual participants symmetry index (S.I. %) of Dm of combined RF, VL and VM, divided into asymmetric (S.I. > 10%) or symmetric (S.I. ≤ 10%) groups. Insets show individual RF (i), VL (ii) and VM (iii). \* Significant difference between groups,  $p < 0.01$ .

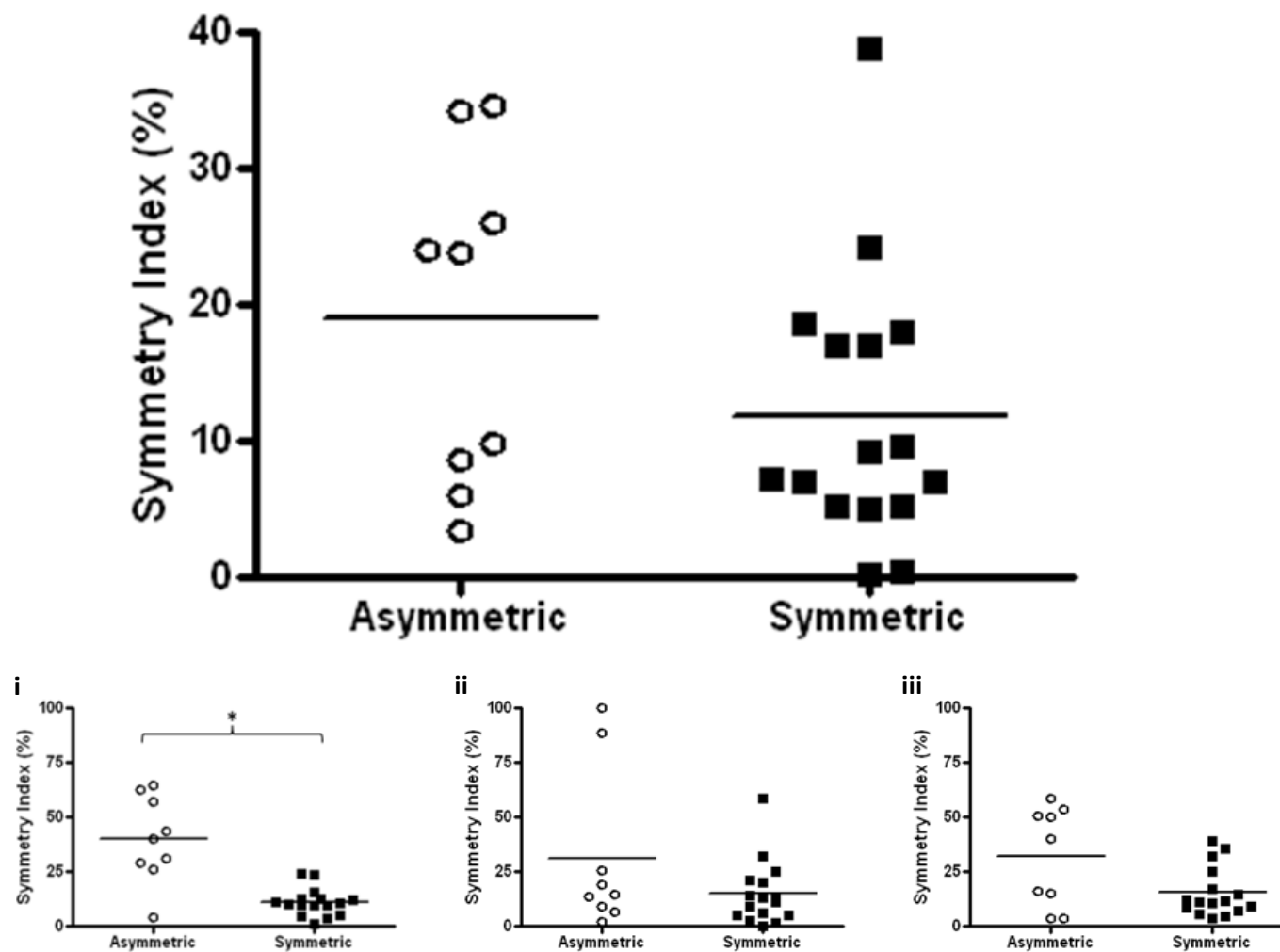


Figure 3.11 Individual participants symmetry index (S.I. %) of Vc of combined RF, VL and VM, divided into asymmetric (S.I. > 10%) or symmetric (S.I. < 10%) groups. Insets show individual RF (i), VL (ii) and VM (iii). \* Significant difference between groups,  $p < 0.01$ .

### 3.3.4 Correlations

There was no significant correlation between knee extensor muscle contractile properties (radial displacement and contraction velocity) and knee extension strength (Figure 3.12) nor lower limb power (Figure 3.13). Pearson's  $r$  values and associated  $p$ -values are presented in Table 3.4 (data for individual muscles is presented in Appendix B).

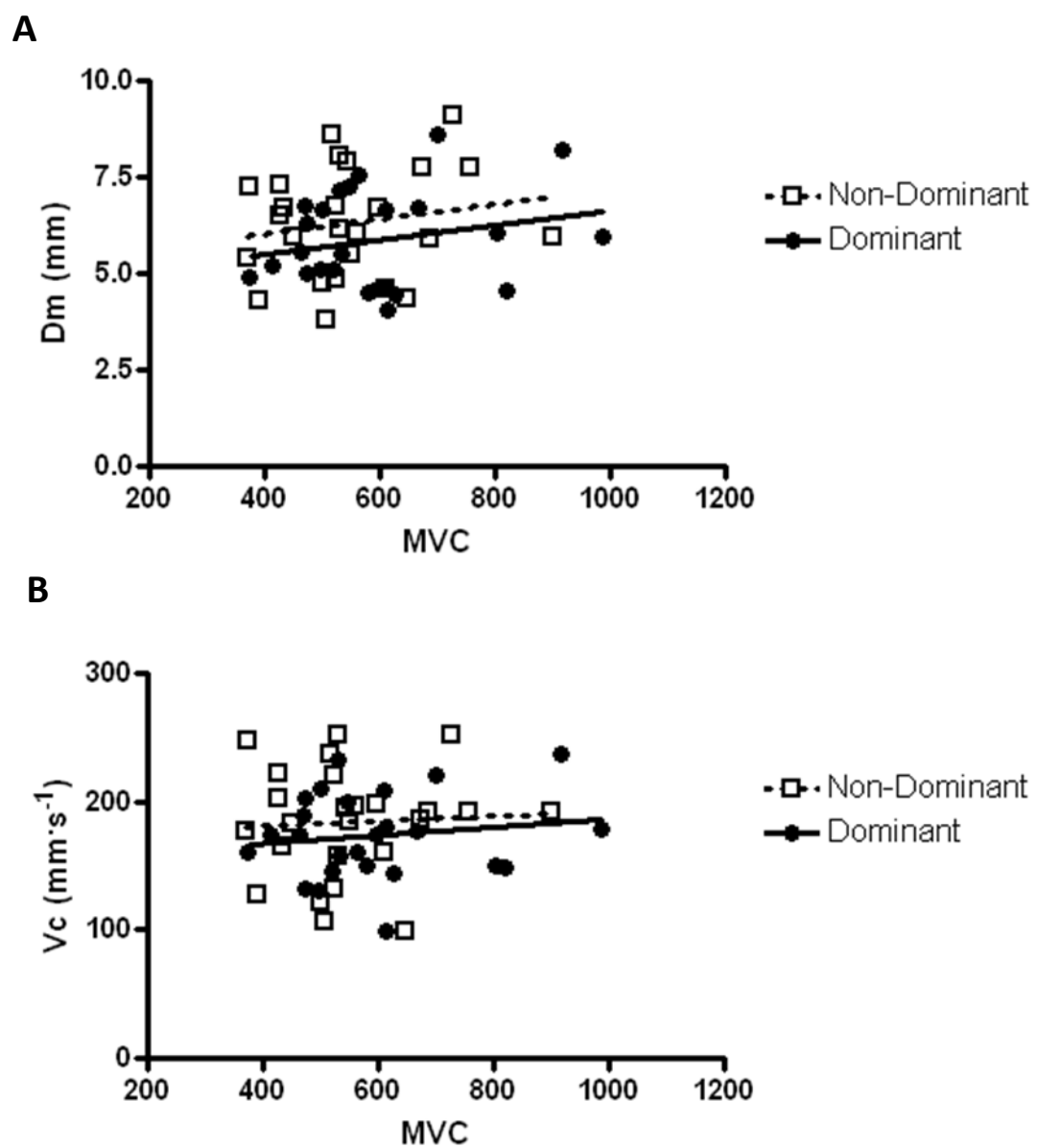


Figure 3.12 Relationship between MVC and peak radial displacement (A) and peak contraction velocity (B), averaged across RF, VL and VM, in dominant and non-dominant limbs.  $r$  and  $p$  values are shown in Table 3.4.

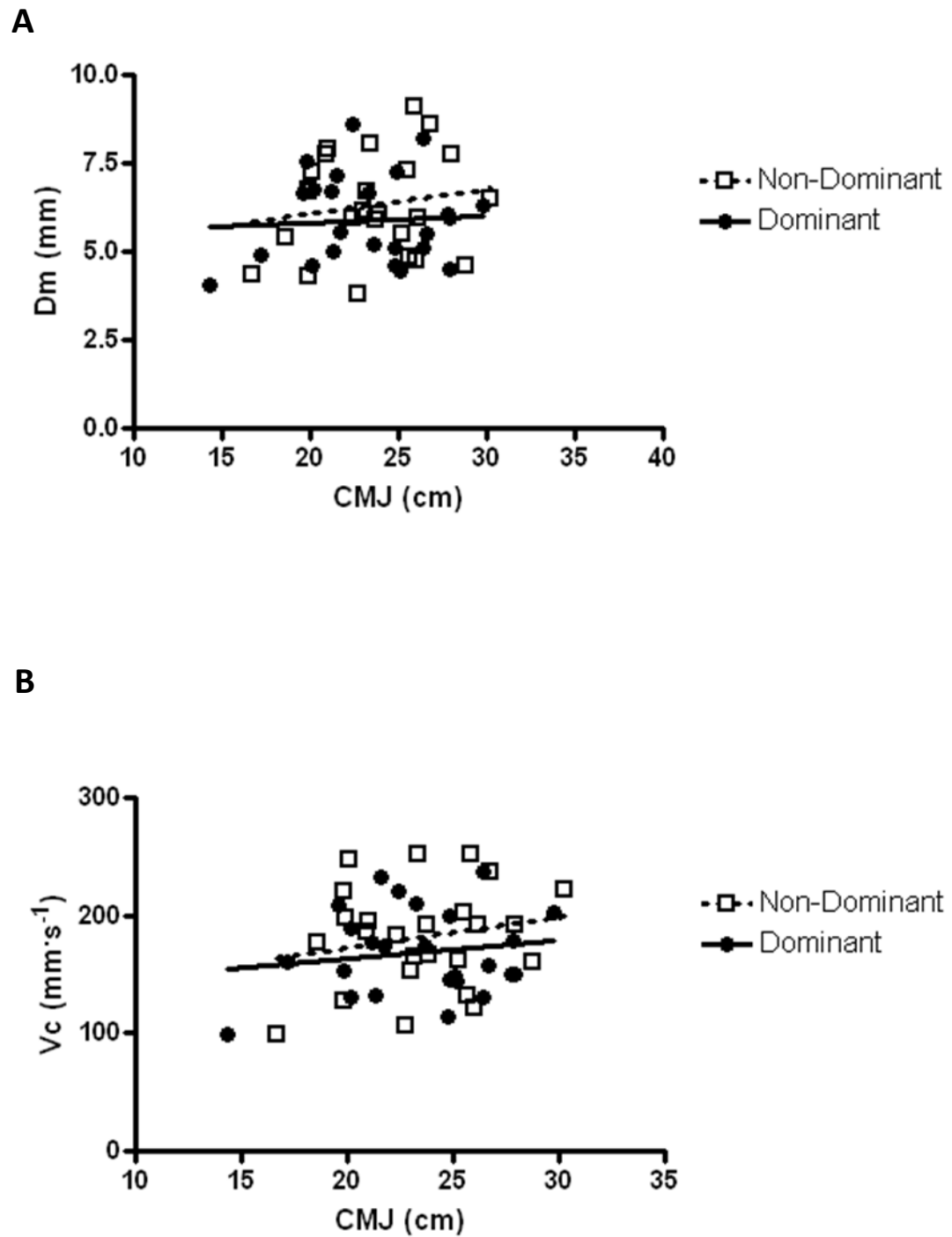


Figure 3.13 Relationship between unilateral CMJ height and peak radial displacement (A) and peak contraction velocity (B), averaged across RF, VL and VM, in dominant and non-dominant limbs.  $r$  and  $p$  values are shown in Table 3.4.

In both asymmetric and symmetrical groups the dominant leg demonstrated a positive relationship with non-dominant for MVC ( $r = 0.99$ ,  $p < 0.001$ ;  $r = 0.96$ ,  $p < 0.001$ ) and moderately-strongly for CMJ ( $r = 0.64$ ,  $p = 0.032$ ;  $r = 0.73$ ,  $p < 0.001$ ) (Table 3.5). Radial displacement and contraction velocity of dominant leg were positively related ( $r = 0.80$ ,  $p < 0.001$ ;  $r = 0.82$ ,  $p < 0.001$ ) with non-dominant in the symmetrical group, but not in the asymmetric ( $r = 0.27$ ,  $p = 0.56$ ;  $r = 0.62$ ,  $p = 0.135$ ) (Figure 3.14).

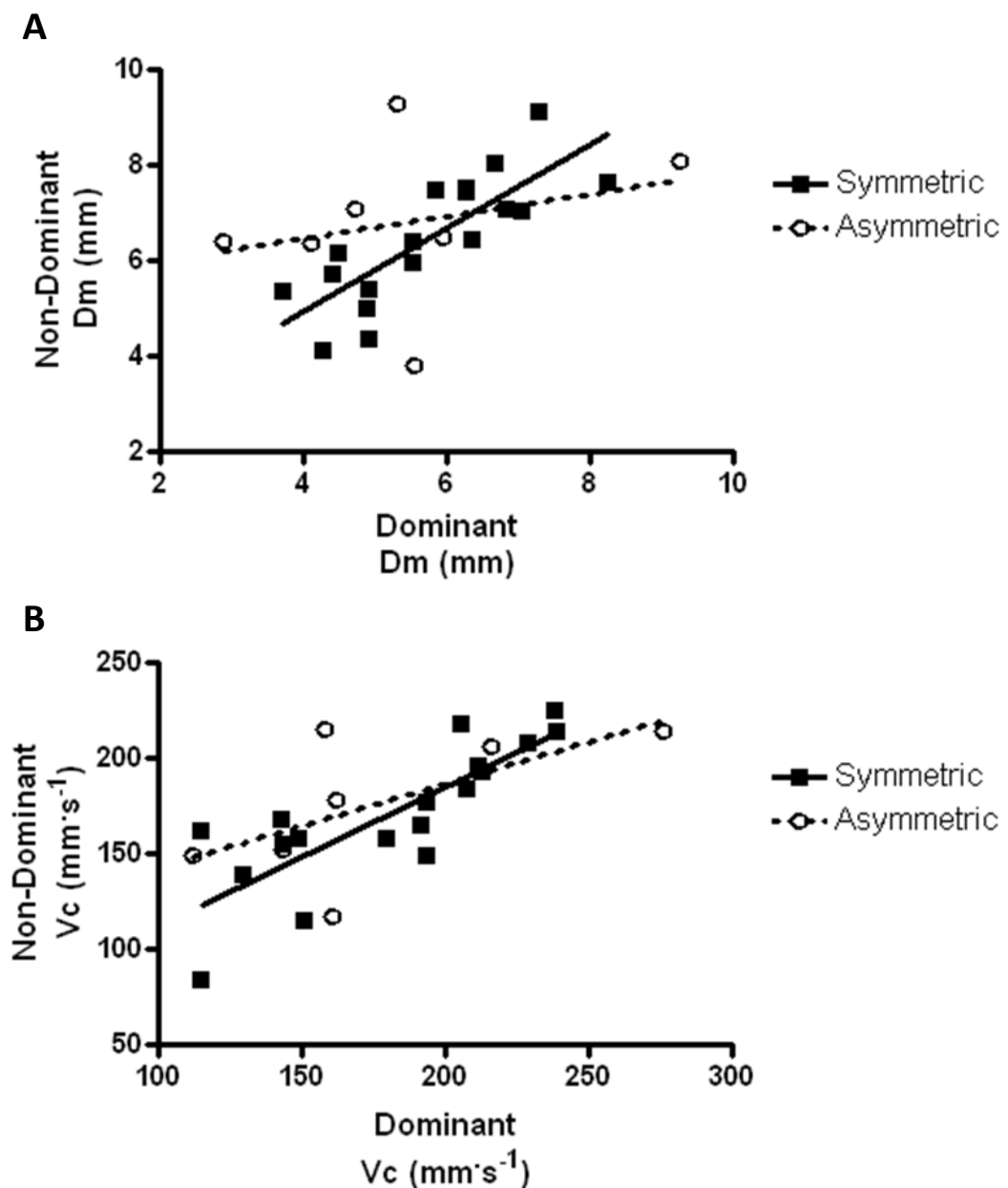


Figure 3.14 Relationship between dominant and non-dominant limbs for radial displacement (A) and contraction velocity (B).  $r$  and  $p$  values are shown in Table 3.5.

**Table 3.4** Relationship values for peak radial displacement (Dm) and peak contraction velocity (Vc), averaged across RF, VL and VM, with MVC and unilateral CMJ height. Pearson's correlation values for individual muscles are presented in Appendix B.

		Dominant		Non-Dominant	
		MVC	CMJ	MVC	CMJ
<b>Dm</b>	<b>r</b>	0.0493	0.0027	0.0300	0.0262
	<b>p</b>	0.286	0.803	0.408	0.440
<b>Vc</b>	<b>r</b>	0.0207	0.0265	0.00319	0.0413
	<b>p</b>	0.493	0.436	0.789	0.330

**Table 3.5** Relationship between dominant and non-dominant limbs MVC, unilateral CMJ height, radial displacement (Dm) and contraction velocity (Vc).  
\* Significant correlation between limbs.

	Asymmetric				Symmetric			
	MVC	CMJ	Dm	Vc	MVC	CMJ	Dm	Vc
<b>r</b>	0.990*	0.637*	0.271	0.623	0.958*	0.728*	0.804*	0.819*
<b>p</b>	< 0.001	0.032	0.557	0.135	< 0.001	< 0.001	< 0.001	< 0.001

Table 3.6 Relationship values for S.I. of radial displacement (Dm) of RF, VL and VM with S.I. of MVC, for symmetric and asymmetric groups. \* Significant correlation between limbs.

	Asymmetric			Symmetric		
	RF	VL	VM	RF	VL	VM
<b>r</b>	0.585*	0.212	0.186	0.009	0.056	0.003
<b>p</b>	0.045	0.299	0.334	0.724	0.376	0.855

Table 3.7 Coefficient of variation (%) values for all test parameters, in dominant and non-dominant limbs, and mean of both limbs.

	MVC	Con		Ecc		CMJ Height	CMJ Force	RF	Dm			Vc	
		60°·s <sup>-1</sup>	120°·s <sup>-1</sup>	60°·s <sup>-1</sup>	120°·s <sup>-1</sup>				RF	VL	VM	RF	VL
<b>Dominant</b>	2.82	0.83	0.21	1.61	2.45	8.46	3.08	3.17	1.66	13.34	3.49	0.44	15.55
<b>Non-Dominant</b>	2.65	2.32	1.42	1.00	2.42	6.95	4.64	6.59	11.47	0.91	5.33	8.60	2.02
<b>Mean</b>	2.74	1.58	0.82	1.31	2.44	7.71	3.86	4.88	6.57	7.13	4.41	4.52	8.79



### 3.3.5 30 Jumps

There was no significant difference ( $F_{(1,24)} = 0.88$ ,  $p = 0.353$ ) in VL RMS between dominant and non-dominant legs or between symmetrical and asymmetric groups ( $F_{(2,24)} = 0.07$ ,  $p = 0.931$ ) throughout thirty consecutive CMJs (Figure 3.16). The decline in CMJ height across thirty consecutive CMJs was not significantly different ( $F_{(1,24)} = 1.02$ ,  $p = 0.323$ ) between asymmetric and symmetrical groups (Figure 3.15).

### 3.3.6 Coefficient of Variation

The coefficient of variation (CoV %) for each measurement variable are presented in Table 3.7. CoV % were not significantly different ( $p = 0.974$ ) between dominant and non-dominant legs.

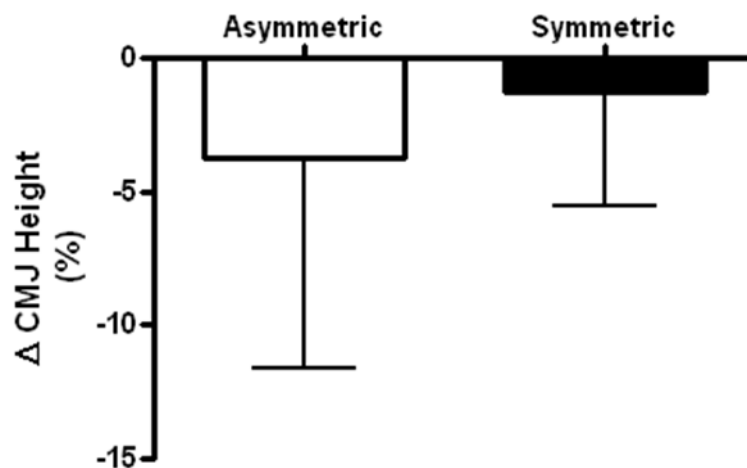


Figure 3.15 Percentage change in mean CMJ height between first 5 CJM and final 5 CMJ. Values are mean  $\pm$  SD,  $n = 25$ .

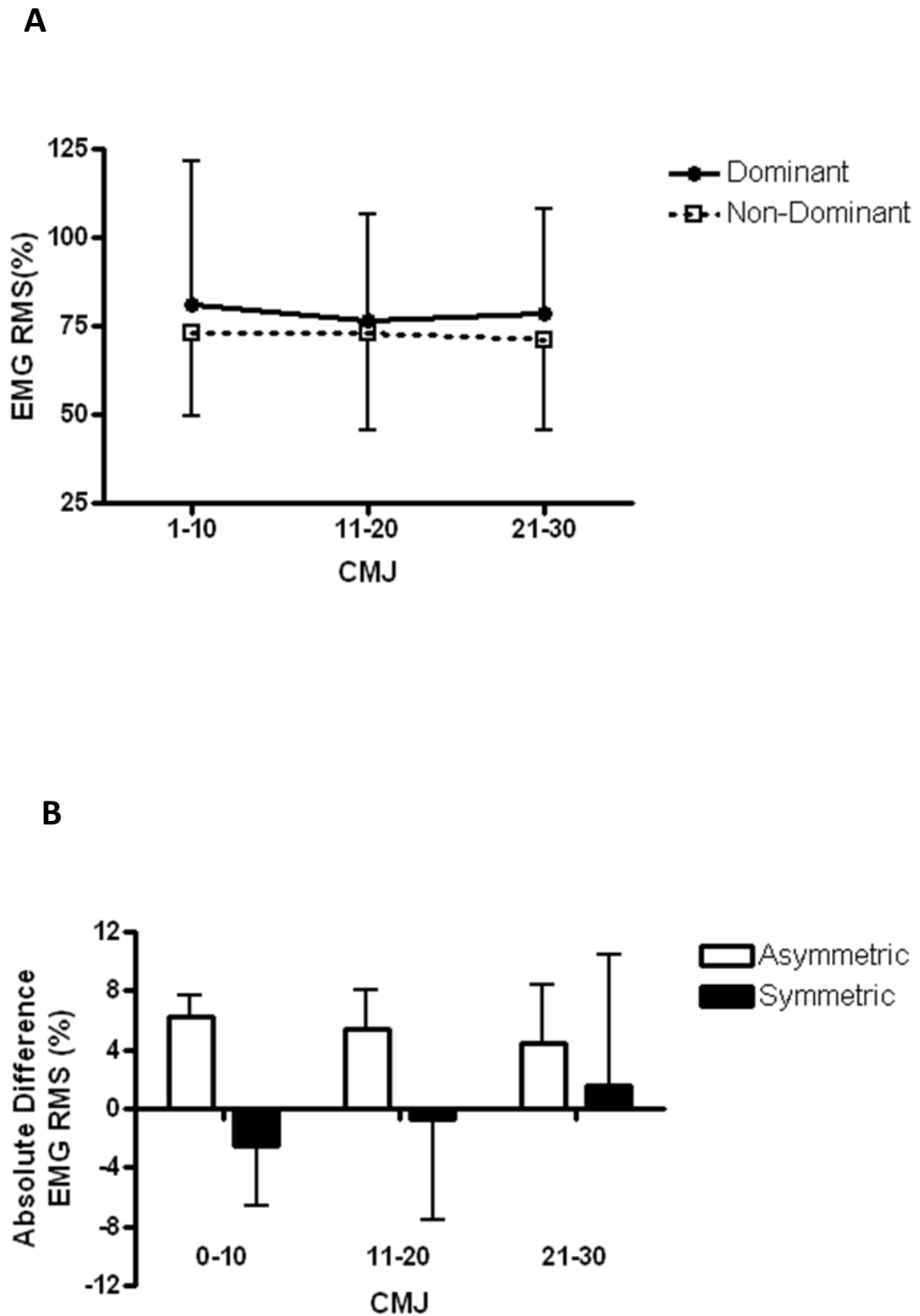


Figure 3.16 Normalised mean EMG root mean squared (RMS), during 1st, 2nd and 3rd tertile, on the VL of dominant and non-dominant limbs (A). Difference between dominant vs. non-dominant VL RMS (normalised), during 1st, 2nd and 3rd tertile, divided into asymmetric or symmetric groups (based on MVC S.I.) (B). Values are mean  $\pm$  SD, n = 25.

### 3.4 Discussion

This study investigated the association between bilateral asymmetry among contractile parameters of muscles within the knee extensors, and functional performance of the muscle group and limb as a whole. Strength measures were higher in the dominant limb compared to non-dominant, but there was no difference between limbs for unilateral CMJ. Dominant limb knee extensors displayed lower Dm and Vc than the non-dominant limb. Individuals were categorized as asymmetric (n = 9) or symmetric (n = 16) based on strength, and Dm showed greater asymmetry among individuals with strength asymmetry. There was no difference between groups in S.I. of the more complex functional tasks.

Strength symmetry index (S.I.) was established between dominant and non-dominant knee extensors, revealing that 9 individuals (4 male, 5 female) displayed asymmetric MVC (S.I. >10%) and 16 individuals (6 male, 10 female) displayed symmetric MVC (S.I. ≤10%). Mean S.I. of the asymmetric group was  $17.04 \pm 4.37\%$ , mean S.I. of the symmetric group was  $5.04 \pm 2.69\%$ , however there was no difference between groups in S.I. of isokinetic peak force or CMJ. Knee extensor Dm, taken as the mean across RF, VL and VM was less in the dominant than the non-dominant limb; this was driven by lower Dm in VM. These findings are in agreement with previous indications that increased muscle stiffness is associated with greater force (Brughelli & Cronin, 2008), while it has been suggested that VM contributes relatively greater load to knee extension force than RF or VL (de Ruiter, Hoddenbach, Huurnink, & De Haan, 2008). Mean Vc across the knee extensors was faster in the non-dominant than the dominant limb; driven by faster Vc in RF.

S.I. of knee extensor Dm was greater in the asymmetric group compared to symmetric, caused by greater Dm in RF. Overall S.I. of knee extensor Vc was not different

between groups, however RF Vc showed greater S.I. in the asymmetric group compared to symmetric. Despite this, Dm and Vc of the knee extensors, displayed no significant correlation with MVC. Neither did Dm nor Vc of any individual muscle correlate with MVC (Appendix B). Furthermore, CMJ performance was not correlated with Dm, Vc or MVC. However, within the asymmetric group only, S.I. of RF was correlated to S.I. of MVC. Furthermore, in both asymmetric and symmetric groups MVC and CMJ using the dominant limb was associated with the outcome for the non-dominant limb, yet in the symmetric group only, was there an association between limbs for Dm and Vc.

It was hypothesized that bilateral asymmetry in Dm would relate to asymmetry in strength; however, there was no relationship between MVC and either Dm or Vc of individual muscles nor as a mean across the muscle group. As such, it was not possible to quantify bilateral strength asymmetry through assessment of contractile properties. However, it was observed that individuals with strength asymmetry were more likely to present asymmetry in Dm. The group that displayed strength asymmetry (asymmetric MVC) presented significantly larger S.I. for Dm, averaged across RF, VL and VM, as well as larger S.I. in RF specifically. It is interesting that within this study, the biarticular RF appeared to be the principal driver of Dm asymmetry. Investigating knee flexor muscular injuries, (Croisier et al., 2002) suggest that recurrent muscle weakness, or asymmetry, likely promotes persistent injuries; with this in mind, identifying key drivers of impairment at individual muscle level could help inform personalized conditioning programming based around targeting detected muscular deficiencies.

It should perhaps be unsurprising that neither temporal nor spatial parameters of muscle twitch, even when averaged across the muscle group, relate quantifiably to

functional performance. Given the complex interaction of numerous component factors (Holsgaard Larsen, Caserotti, Puggaard, & Aagaard, 2007), prediction of variations in force generation based on a single variable involved is unlikely. This is perhaps demonstrated even more clearly by the lack of correlation between MVC and the more complex CMJ. Strength has long been associated with athletic performance, in particular sprint speed (Guskiewicz, Lephart, & Burkholder, 1993), however, despite obvious differences in the levels of strength asymmetry detected from MVC, there was no asymmetry detected in the more complex CMJ. Furthermore, despite similar overall differences in  $D_m$  and  $V_c$  between limbs when compared to differences in MVC, there was no difference in S.I. of symmetric and asymmetric group in either CON or ECC isokinetic peak force. These findings demonstrate that even in an isolated muscle group with only a single joint involved, the additional complexity of kinetics (shortening or lengthening) appears to hinder identification of variations between limbs. It has previously been demonstrated in sports such as swimming, that strength asymmetry is not always detectable when performing complex actions (Potts, Charlton, & Smith, 2002; Evershed, Burkett, & Mellifont, 2014), as such asymmetries are suggested to have been 'masked' due to the nature of the task performed.

It is interesting that no bilateral asymmetry was detected during the most complex task performed throughout the study (CMJ force and CMJ height). Higher S.I. values have been observed, in cycling, for crank torque, during moderate and low intensity exercise, with higher intensity efforts, close to maximal, shown to be more symmetric (Carpes et al., 2010); with this in mind, perhaps the workload or intensity is critical. However, during both CON and ECC assessments, at  $60^\circ\text{s}^{-1}$  and  $120^\circ\text{s}^{-1}$ , isokinetic peak torque was asymmetric in both groups, with no discernible difference between velocities. These findings are

comparable to previous data from (Newton et al., 2006), who reported S.I. of 13.51% at  $60^{\circ}\text{s}^{-1}$  and 13.06% at  $240^{\circ}\text{s}^{-1}$ . Perhaps, the intensity and velocity of the task are less significant than task complexity. Task performance becomes more difficult to predict during complex tasks, involving a greater number of agonist muscles (Holsgaard Larsen et al., 2007). It is possibly for this reason that lower-limb dominance plays only a small role in complex task performance (Fort-Vanmeerhaeghe, Montalvo, Sitja-Rabert, Kiefer, & Myer, 2015), which may further contribute to masking of strength asymmetry. The flexibility of muscle recruitment allows force, about a joint, to be produced by a number of combinations of muscle tensile forces (Bouillard, Jubeau, Nordez, & Hug, 2014). Such load sharing amongst muscles is controlled through central nervous system (CNS) mediated recruitment patterns (Erdemir, McLean, Herzog, & van den Bogert, 2007); greater understanding of CNS recruitment patterns in relation to functional asymmetry could be a key to targeted conditioning programmes (Zory, Boërio, Jubeau, & Maffiuletti, 2005).

During 30 consecutive bilateral CMJs, strength asymmetry appeared to have no detectable impact on muscle recruitment or fatigue. Bailey et al., (2013) reported impaired vertical jump performance among individuals who demonstrated strength asymmetry, consequently it might have been expected that, in the present study, the asymmetric group would have shown impaired ability within the 30 CMJ task, compared to the symmetric group, however this was not the case. Absolute strength has been proposed as a confounding variable in asymmetry assessment, (Bailey, Bazzyler, Chiang, Sato, & Stone, 2014) demonstrated that lower-limb performance was less influenced by strength asymmetry in stronger males than weaker. However, in the present study MVC ranged from 347.27 - 989.88N, demonstrating a large range of strength classification, but there was no

difference in mean strength between asymmetric and symmetric groups, in dominant ( $630.72 \pm 186.91\text{N}$  vs.  $603.42 \pm 174.68\text{N}$ ) or non-dominant ( $525.27 \pm 139.11\text{N}$  vs.  $583.66 \pm 159.75\text{N}$ ) limbs. It has also been proposed that gender may play a role (Bailey, Sato, Burnett, & Stone, 2015); however, no gender related differences were observed throughout the present study. It is possible, that relative strength differences overrode gender in the aforementioned study, whereas in the present study males ( $608.4 \pm 170.0\text{N}$ ) and females ( $549.2 \pm 136.5\text{N}$ ) did not differ in strength.

Although no differences were uncovered in RMS between dominant and non-dominant limbs, nor in the difference between limbs, between groups, we cannot rule out modified load sharing, resulting from altered recruitment patterns based on strength asymmetry. EMG was recorded from only one agonist muscle during 30 CMJs, providing no insight into synergist recruitment. Furthermore, VL, from which EMG was recorded, was not different between legs for Dm or Vc, nor was there a difference in VL S.I. between symmetric and asymmetric groups. Finally, CMJ involves multiple agonist groups across multiple joints, whilst strength asymmetry was only assessed within knee extensors, therefore whole limb asymmetry cannot be assumed. This final limitation is also likely to have contributed to the lack of difference in S.I. between groups for unilateral CMJ, which were similar to those previously reported by Newton et al., (2006) during unilateral jump tasks (0.92-4.93%).

The minimum detectable change for Dm measurements has been established as 15.05% (Ditroilo et al., 2013); within the symmetric group the difference in Dm between limbs for RF (5.24%), VL (4.20%) and VM (8.77%) all fall below minimum detectable change, while within the asymmetric group RF (16.71%) and VM (29.88%) are above minimum level,

although VL (4.77%) was not. Once again, it appears that recording EMG from VL alone may not have provided full insight into any potential differences in recruitment balance among agonists during a functional task. It has been proposed that unbalanced load sharing may stand as a critical source of knee joint injury (Cerny, 1995; Coqueiro et al., 2005), thus understanding varied muscle recruitment strategies, and how these interact with strength asymmetry, may shed light on the relationship between bilateral asymmetry and injury prevalence (Bouillard et al., 2014). It is likely that alternative recruitment patterns, designed to compensate for imbalanced strength may pose a heightened injury risk (Maupas, Paysant, Martinet, & André, 1999). With this in mind, the value of individual muscle assessments may be reinforced following injury; Jordan, Aagaard, & Herzog, (2015) reported larger bilateral asymmetries in muscle function following anterior cruciate ligament reconstruction, while Kuenze et al., (2015) revealed that pre-existing asymmetries persisted following similar reconstruction surgery. Perhaps there is scope, within injury rehabilitation programmes, for single muscle assessments, in order to target conditioning exercises for optimal adaptation. Further research is needed to compare contractile mechanics of individual muscles with muscle recruitment patterns across synergist groups.

It is unclear exactly why, within the current study, differences between limbs were observed only in VM for Dm and in RF for Vc. It could be reasoned that greater relative muscle mass of VM, compared to RF or VL (Wickiewicz, Roy, Powell & Edgerton, 1983) contributed to heightened sensitivity for quantifying differences in Dm. However, further investigation is required to elucidate whether there exists a positive relationship between muscle mass and sensitivity of TMG to detect Dm. On the other hand, perhaps the positioning of sensor and electrodes may merit consideration; in the case of Vc, RF displays



between-limb differences, which are absent in VL or VM. It is possible that the lineation of the electrode-sensor arrangement more closely associated with the direction of myofibres in RF, compared to VL and VM. Similarly, only RF, among the three muscles, displayed differences in S.I. of Dm and of Vc, between symmetric and asymmetrical groups. Once again, it could be speculated that the positioning of the measurement apparatus, relative to myofibre pennation angle may influence the sensitivity, and therefore the reliability, of measurements.

### 3.5 Conclusion

This study compared bilateral asymmetry across a whole limb, within an isolated muscle group and at the individual muscle level. As has been described elsewhere, strength asymmetry is often masked during more complex tasks, involving multiple muscle groups and/ or multiple joints. However, this is the first study to demonstrate that individual contractile properties also do not relate directly to strength of a muscle group. Individuals with strength asymmetry were more likely to display asymmetry in at least one muscle, it may be that underlying differences in muscle contractile mechanics lead to altered recruitment patterns to load share among synergist muscles. It remains to be established whether such alterations could be detrimental long-term, by increasing the likelihood of injury. Assessment of individual muscle contractile properties, using TMG, may be advantageous in identifying inadequate muscles, to allow targeted training interventions, which could lead to preferable recruitment strategies. Having established the validity of TMG against established physiological parameters in Chapter 2, and now, in the current chapter, explored the application of TMG within the context of performance assessment, a series of descriptive studies and case-studies were next carried out among an elite athlete

population. The first of these studies will describe bilateral asymmetry within a population of high performance swimmers, and is presented below. Findings from the remaining case-studies are presented in Chapter 7.

### **3.6 Bilateral Asymmetry within Elite Scottish Swimmers: A Descriptive Study**

#### **3.6.1 Introduction**

Previously in this chapter, we discussed the potential role of muscle contractile assessment, to identify bilateral asymmetry within performance sport. Asymmetry has long been a topic of interest within competitive swimming; in particular the relationship between bilateral symmetry and stroke biomechanics, or technique, has prompted equivocal inferences (Jones & Bampouras, 2010; Shorter, Polk, Rosengren, & Hsiao-Weckler, 2008). As discussed previously, a threshold of 10% (Herzog et al., 1989) is typically used to classify asymmetry, since inherent variances within the human body mean that a degree of asymmetry (i.e. < 10%) can be deemed acceptable (Jaszczak, 2008). It has been noted that bilateral asymmetry can be magnified within unilateral sports (Ellenbecker, Roetert, Piorkowski, & Schulz, 2002; Brushøj, Bak, Johannsen, & Faunø, 2007; Saccol et al., 2010); as such, asymmetry is often considered to be adaptive. However, within swimming, which can be broadly described as bilateral (Evershed et al., 2014), athletes do not necessarily appear to adapt bilaterally concerning force or power outputs (Potts et al., 2002). Measuring propulsive force in swimming is inherently challenging, due to the aquatic environment, which results in complex propulsive and resistive force relationships (Formosa, Mason, & Burkett, 2011). Forces increase exponentially in association with velocity, and factors such as body position, act as confounding variables (Blanch, 2004; Kologorov, Rummyantseva, Gordon, & Cappaert, 1997). That being said, it may be predicted that the masking of

asymmetric performance, as outlined above, could likely be prevalent within swimming. Indeed, land-based strength and power measurements have shown strong association with in-water performance (Morouço et al., 2011). And yet, there is substantial evidence of functional asymmetry, within complex movements, in highly trained swimmers (Evershed et al., 2014; Potts et al., 2002), which may be linked to breathing laterality or arm dominance (Seifert, Chollet, & Allard, 2005). Critically, asymmetry in force and power, is intrinsically linked to asymmetry in stroke co-ordination (Tourny-Chollet, Seifert, & Chollet, 2009), and could therefore dramatically impinge on swimming technique, and ultimately, performance. Therefore, this descriptive study will evaluate levels of contractile asymmetry across a range of muscles, in isolation, which are critical to functional kinematics within swimming.

### **3.6.2 Methods**

Contractile properties of 30 elite level swimmers were recorded from vastus lateralis (VL), gastrocnemius medialis (GM), bicep femoris (BF) and latissimus dorsi (LD) of both the left and right flank. An overview of athlete characteristics is presented in Table 3.8. Measurements were repeated on 3 occasions, on consecutive weeks and in similar temporal and environmental conditions. Dm and Vc of each muscle were calculated as described in Chapter 2. Data were assessed to ensure normality, and parameters were analysed using paired T-tests to compare flanks. S.I. (%) was calculated for both groups as described previously in this chapter.

**Table 3.8 Athlete characteristics of the cohort of elite swimmers. Values are mean  $\pm$  SD, n = 30. (PB = personal best time, relative to world record).**

<b>Athlete characteristics</b>	
<b>Age (y)</b>	21.0 ( $\pm$ 2.4)
<b>Competition history (y)</b>	11.0 ( $\pm$ 3.2)
<b>Height (cm)</b>	178.3 ( $\pm$ 6.6)
<b>Arm span (cm)</b>	185.9 ( $\pm$ 7.9)
<b>Leg length (cm)</b>	84.4 ( $\pm$ 3.5)
<b>Body mass (kg)</b>	72.9 ( $\pm$ 8.5)
<b>PB (%)</b>	+ 5.2 ( $\pm$ 2.9)

### 3.6.3 Results

Significant differences in Dm between flanks were observed in BF ( $p = 0.002$ ) and GM ( $p = 0.044$ ), BF displayed larger Dm on the left flank ( $4.6 \pm 1.2$  vs  $2.1 \pm 0.8$ mm, CI [1.158 to 3.925]), GM displayed larger Dm on the right flank ( $1.8 \pm 0.6$  vs  $1.4 \pm 0.3$ mm, CI [0.012 to 0.773]). Significant difference in Vc between flanks was observed in BF ( $p = 0.004$ ), Vc was greater on the left flank ( $98.7 \pm 27.4$  vs  $71.3 \pm 25.1$ mm s<sup>-1</sup>, CI [10.61 to 44.16]). No significant differences were detected in Dm between flanks in VL ( $p = 0.927$ ) or LD ( $p = 0.231$ ), nor in Vc in VL ( $p = 0.397$ ), GM ( $p = 0.07$ ) or LD ( $p = 0.103$ ) (Figure 3.17). CoV (%) for Dm and Vc of each muscle are listed in Table 3.9. S.I. of Dm and Vc, in VL, GM, BF and LD are presented in Table 3.10.

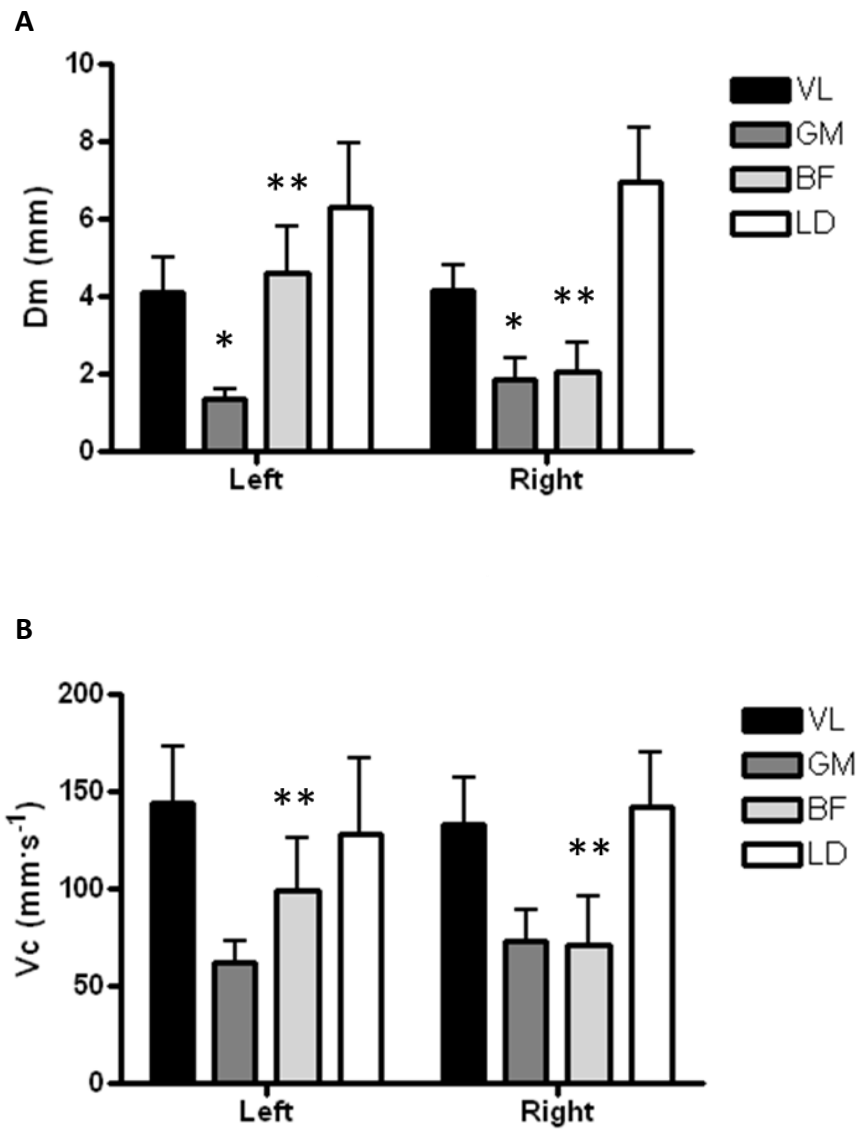


Figure 3.17 Dm (A) and Vc (B) of VL, GM, BF and LD. Values are mean  $\pm$  SD, n = 30. \* Significant difference between flanks,  $p < 0.05$ . \*\* Significant difference between flanks,  $p < 0.01$ .

Table 3.9 Coefficient of variation (%) values for Dm and Vc, in left and right flanks.

	Dm				Vc			
	VL	GM	BF	LD	VL	GM	BF	LD
Left	21.1	23.7	36.4	35.7	19.8	20.4	30.8	37.0
Right	18.5	31.2	43.7	26.6	19.5	23.5	38.9	23.6

Table 3.10 Symmetry index (S.I. %) of Dm and Vc in VL, GM, BF and LD. Values are mean  $\pm$  SD, n = 30.

		VL	GM	BF	LD
Dm	Mean (%)	24.3	38.5	69.9	23.6
	$\pm$ SD	19.4	34.9	44.5	20.6
Vc	Mean (%)	28.6	24.4	39.6	22.2
	$\pm$ SD	17.6	28.1	22.9	18.3

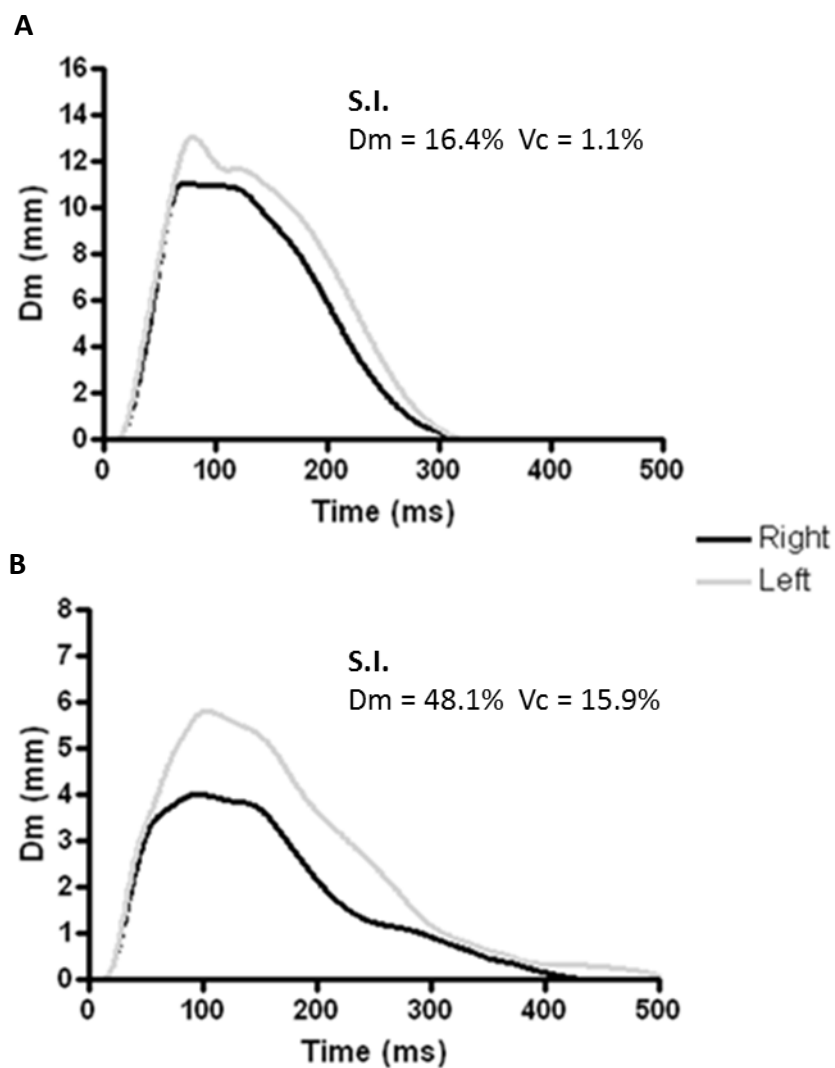


Figure 3.18 Example displacement/ time curves of the TMG signal in LD (A) and BF (B). Symmetry indices (%) of Dm and Vc are presented.

### 3.6.4 Discussion

This descriptive study examined the prevalence of bilateral muscular asymmetries within a cohort of elite swimmers. Despite the predominantly bilateral nature of biomechanics involved in swimming, it appears that asymmetries are commonplace within the sport, even in experienced, high-level performance athletes. The group examined during the current study comprised on average 11.0 ( $\pm$  3.2) years of competition experience and had produced personal best times within  $\leq$  5.2 ( $\pm$  2.9) % of the world record in their preferred event. Specifically, asymmetry in muscle contractile mechanics was detected in the posterior muscles of the lower limbs, with BF displaying higher Dm and faster Vc in the left limb ( $4.6 \pm 1.2$  vs  $2.1 \pm 0.8$  mm and  $98.7 \pm 27.4$  vs  $71.3 \pm 25.1$  mm s<sup>-1</sup> respectively), and GM displaying greater Dm in the right limb ( $1.8 \pm 0.6$  vs  $1.3 \pm 0.3$  mm). Asymmetry was not evident within VL or LD. These differences were associated with high S.I. in BF Dm ( $69.9 \pm 44.5\%$ ) and Vc ( $39.6 \pm 22.9\%$ ), and GM Dm ( $38.5 \pm 34.9\%$ ). S.I. for all other muscle parameters assessed were  $< 30\%$ .

Although these data are preliminary, and have not been associated with functional measures of force or technical asymmetry, it might be speculated that a higher threshold than previously suggested (i.e. 10%) may be more appropriate when categorizing asymmetry at the level of individual muscles. It would be interesting to analyse the relationship between contractile asymmetry  $\geq 30\%$  and asymmetry in swimming technique (Tourny-Chollet et al., 2009). As discussed previously, performance of complex motor tasks may mask underlying muscular asymmetry, due to interactions of agonist muscles; with this in mind, it is necessary to investigate contractile asymmetries in other muscles involved in knee flexion and plantar flexion. That being said, it is interesting that greater Dm observed

in the left limb BF is somewhat offset by greater Dm in the right limb GM. Future research should explore intra-limb asymmetries, among muscle agonists and relate findings to performance measures, in particular stroke co-ordination, as a marker of technique.

To date, research into asymmetry and stroke biomechanics within swimming has predominately focused on front crawl/ freestyle (Evershed et al., 2014; Potts et al., 2002; Seifert et al., 2005; Tourny-Chollet et al., 2009); with the upper body being of particular focus (Virag, Hibberd, Oyama, Padua, & Myers, 2014). Given the differing biomechanics required for each stroke (Barbosa, Marinho, Costa, & Silva, 2011), it might be speculated that there could be differences in levels of asymmetry between athletes who specialise in different events; however the current cohort was not divided according to event, due to the resultant lack of statistical power. As such, the investigation of potential differences in asymmetry between varying events is another valuable direction for future research. Finally, it should be noted that CoV were considerably higher among the muscles assessed in this elite cohort (Dm = 26.6%; Vc = 26.7%), compared to CoV of muscles assessed in previously in this chapter (Dm = 6.2%; Vc = 5.9%) in non-elite individuals. On the other hand, S.I. of VL were comparable between this case-study and the asymmetric group presented earlier in the chapter ( $29.9 \pm 23.6\%$  vs  $24.3 \pm 19.4\%$  and  $23.9 \pm 31.8\%$  vs  $28.6 \pm 17.6\%$ , for Dm and Vc respectively). These observations suggest, that despite maintenance of extrinsic conditions during testing, the variability encountered when measuring elite athletes is greater than encountered elsewhere. It seems reasonable to suggest that such elevated variability arises from individualised responses to high training loads and volumes performed on a daily basis.



# Chapter 4

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## Impaired Firing Rates of High-Threshold Motor Units Following Eccentric Exercise-Induced Muscle Damage of the Vastus Lateralis

Lewis J Macgregor, Katerina MA Kay, Angus M Hunter

## 4.1 Introduction

In Chapter 6 we will present a series of case-studies which demonstrate the acute changes in skeletal muscle contractile mechanics which are associated with training and competing in elite level sport. Wiewelhove et al. (2015) recently proposed TMG for tracking changes associated with a prescribed high-intensity interval training programme; other markers indicated that significant exercise-induced muscle damage (EIMD) arose from high-intensity training. Hunter et al. (2012) have previously demonstrated the viability of TMG to track muscular impairments associated with EIMD, however, the deeper underlying mechanisms involved in EIMD still require further investigation. In Chapter 3 we reported that deficiencies in individual muscles may potentially lead to altered muscle recruitment patterns, which are likely associated with injury incidence (Maupas et al., 1999). It is possible therefore, that examining neuromuscular recruitment patterns may provide further insight into the impaired muscle function which accompanies EIMD.

Since early investigations into muscle soreness by Theodore Hough in the early 1900s it has been established that performing exercise which is unaccustomed in terms of either volume (Nosaka et al. 2002; Newton et al. 2008), intensity (Warren et al. 2001; Paschalis et al., 2005) or mode (Mchugh et al., 2002; Ye et al., 2015) may cause EIMD, which impairs muscular force capacity (Jackman et al., 2010; Hunter et al., 2012). One of the main causes of EIMD is eccentric exercise due to muscle lengthening during contraction, which places additional strain on the muscle fibres (Byrne et al., 2004; Clarkson, 1997; Proske & Morgan 2001). Following EIMD maximal force production can be impaired for up to ten days (Clarkson, 1997; Clarkson et al., 1992), which could have significant implications for athletic performance.

EIMD force decrements are often accompanied by delayed onset muscle soreness (DOMS) (Clarkson et al., 1986) and inflammation, identified by elevated blood born cytokines (Macintyre et al., 2001) or localised edema (Evans et al., 2002) which may, in turn, feedback to further contribute towards force impairment. This feedback will alter afferent signalling to the central nervous system (CNS) (Dartnall et al., 2008; Gauche et al., 2009; Racinais et al., 2008) in order to modify neuromuscular recruitment strategy preventing further damage from occurring, in accordance with the central governor hypothesis first proposed by (Hill et al., 1924). Specifically, recruitment threshold of motor units (MUs) has been seen to drop (Dartnall et al., 2009) alongside increased synchronisation of MU firing. However, interpretation of these data is limited as indwelling electrodes were used which enabled recording of only ~10 motor units, a very small representation of the complete MU pool. In order to decipher details of motor unit behaviour from larger pools, constituent motor unit action potential trains (MUAP) can now be extracted from surface electromyographical (sEMG) signals. This can be done by either high density EMG (Blok et al., 2002; Zwarts et al., 2003) or precision decomposition EMG (dEMG) (Adam & De Luca, 2005; De Luca, Lefever, Mccue, & Xenakis, 1982; De Luca et al., 2006; Nawab et al., 2010; Kline & De Luca 2014). dEMG has been designed to investigate the behaviour of motor units at the individual level, as well as the collective control (termed common drive) of the motor unit pool (Adam & De Luca, 2005; De Luca & Erim, 1994), following capture from a single multi-channel sEMG electrode array sensor. The unique dEMG system allows assessments of MU firing properties (Nawab et al., 2010) and for the first time facilitates evaluation of different MUs within the recruited pool, based on their recruitment threshold (De Luca & Contessa, 2012). By condensing the electrode array into a single sensor, it becomes easier to

achieve the quality skin-electrode interface required to capture highly accurate sEMG signals from which to extract MUAPs.

High force producing type II (fast oxidative/ glycolytic) muscle fibres are more susceptible to EIMD than type I (slow oxidative) fibres (Vijayan et al., 2001; Macaluso et al., 2012). It is therefore likely that there will also be preferential impairment of specific motor units. Therefore, when applying the principle of size ordered recruitment (Henneman, 1985) it seems plausible that later recruited motor units would become affected, although this is yet to be shown. Recently, it was shown that eccentric exercise caused an alteration in the relationship between motor unit recruitment threshold and mean firing rate (Ye et al., 2015). This suggests that EIMD might disrupt the later-recruited motor units, which in all likelihood are associated with type II muscle fibres (Henneman, 1985).

Therefore, the aim of the present study was to investigate the acute effects of eccentric EIMD on alterations in motor unit firing rate and common drive in the knee extensor muscles with dEMG. Specifically, motor unit firing rates will be analysed by dividing the motor unit pool into early-recruited, mid-recruited and later recruited units. In this way, for the first time, the firing behaviour of high-threshold motor units will be investigated, in isolation from the entire motor unit pool. It was hypothesized that the firing rate would be altered in high-threshold motor units only, and that common drive would be elevated following EIMD, pointing towards a centrally mediated compensatory mechanism for impairments in muscle function.

## 4.2 Methods

### 4.2.1 Participants

Fourteen healthy, recreationally active, male participants with no history of neuromuscular or musculoskeletal disorders were recruited. Mean and standard deviation ( $\pm$ SD) age, height, body mass and knee extension strength (MVC) at baseline are shown in Table 4.1. All participants were deemed to be unaccustomed to eccentric resistance exercise, for at least the six months prior to their participation in the study. Participants refrained from: 1) any unaccustomed physical activity for the duration of the trial and 2) any strenuous exertion for at least 24h prior to each testing session. Volunteers provided written consent, having been informed of any potential risks involved in their participation. The study was performed in accordance with the standards set by the latest revision of the *Declaration of Helsinki* and was approved by the University of Stirling Sports Studies Ethics Committee (SSEC).

**Table 4.1 Participant characteristics (n = 14). MVC is taken at baseline of the dominant leg.**

	<b>Age (y)</b>	<b>Height (m)</b>	<b>Mass (kg)</b>	<b>MVC (Nm)</b>
<b>Mean</b>	25.4	1.8	79.0	233.2
<b><math>\pm</math> SD</b>	5.4	0.08	12.0	47.7

### 4.2.2 Study Design

Following full familiarisation of the testing procedures, participants reported to the laboratory on five occasions, over a 14 day period. Participants recorded food intake for three consecutive days, prior to beginning the trial. Participants reported to the laboratory

following an overnight fast. Baseline measures were recorded for knee extensor muscle soreness, before isometric maximal voluntary contraction (MVC) and neuromuscular measures were performed using an isokinetic dynamometer (Biodex System 3, Medical Systems, USA). In all cases, measurements were carried out for the non-dominant (control) leg prior to the dominant (intervention) leg. The order of measurements was consistent across all trials.

#### *4.2.3 Protocol*

Participants were coupled to the isokinetic dynamometer for assessment of MVC, neuromuscular measures and muscle soreness as well as for performing eccentric contractions to induce EIMD. The lateral femoral epicondyle of the testing leg was visually aligned with the axis of rotation of the dynamometer, and seat positions were adjusted to suit each individual participant's anthropometric characteristics. In accordance with the manufacturers' instructions straps (across the chest, pelvis and resting leg) were used to secure the participant in the required position, and to reduce mechanical assistance from other body parts. During contractions participants were instructed to cross their arms in front of their chest. The final positioning of each participant was recorded on the initial visit and replicated throughout the experimental period, to ensure constancy.

Participants rated perceived muscle soreness while positioned in the isokinetic dynamometer. Soreness was measured while the knee was fully extended (joint angle of  $0^{\circ}$ ). Pressure was applied to the midpoint on the lateral and transverse planes of the Quadriceps Femoris, using a custom-built, spring loaded algometer. The investigator applied 1kg/cm pressure. Participants rated their level of soreness using a 200mm visual analogue scale (VAS) which ranged from 'no pain' at the extreme left to 'most pain imaginable' at the

extreme right (Howatson & van Someren, 2007). The two ends of the VAS were anchored by perpendicular lines, but there were no increments between the end markers. Participants were instructed to mark a point along the line which represented the perceived soreness felt as pressure was applied to the muscle. For each measurement a fresh scale was used, with no reference to previous measurements. Muscle soreness was quantified by measuring the distance (to the nearest 0.1cm) from the left anchor point to the point marked by the individual. During pre-exercise testing, muscle soreness, of the dominant leg, was rated twice, once before baseline measures and once immediately following the cycling warm-up.

With the participant secured in the dynamometer gravitational corrections were performed, in accordance with existing recommendations (Glesson & Mercer, 1996), in order to account for the effect of limb weight on torque measurements. A knee joint angle of  $60^{\circ}$  was set and the limb was secured by a Velcro strap proximal to the medial malleolus. The angle of  $60^{\circ}$  was chosen, as it lies within the well-established range of reported optimal knee joint angles, for peak isometric torque production (Knapik et al., 1983). Participants performed a standard submaximal warm-up, consisting of two sets of 3 x 5s isometric contractions; with 30s rest between repetitions and 60s recovery between sets. For the first set participants contracted at an intensity perceived to be 50% of maximum effort; for the second set the intensity of contraction was 75% of perceived maximum (Balshaw & Hunter, 2012; Hunter et al., 2012), visual feedback was available on a monitor positioned in front of the dynamometer seat, as an output guide.

Immediately following the warm up, participants performed 3 x 5s isometric maximal voluntary contractions (MVC). Participants were required to react to an audio prompt and were instructed to exert as much force as possible, as quickly as possible, in response to the

prompt. The gap between prompts was randomized, such that participants could not anticipate their next contraction. The contraction containing the highest peak torque was designated MVC. From this contraction RTD was calculated over 0-300ms from the onset of contraction ( $\pm 2SD$  from baseline) using MATLAB version 7.11.0.584 (R2010b) software (The MathWorks, Inc.). Participants were instructed not to hold back any effort for subsequent contractions. The same investigator provided standardized verbal commands and encouragement, to assist the participants in achieving maximal effort for every contraction.

Following determination of baseline MVC, for each leg, the subjects performed a submaximal isometric muscle action following a trapezoidal template. Participants linearly increased the magnitude of isometric contraction, tracing the shape of the template, from 0 - 60% of MVC for 6s, at a rate of  $\sim 10\% \cdot s^{-1}$ ; the contraction level was held steady at 60% of MVC for 10s, then linearly decreased from 60% - 0 at the same rate as above. Participants were instructed to completely relax the knee extensor muscles at the start and end of each action; this was confirmed by visual inspection of the quiescent portions before and after the signal. The template and output feedback trace were visible on a monitor positioned directly in front of the dynamometer. Participants were required to follow the template as closely as possible with their output trace (Appendix C).

A surface array dEMG sensor (Delsys, Inc., Boston, Massachusetts) was used to detect bipolar surface EMG signals, on four separate channels, from the VL of each leg in turn, during isometric MVCs and submaximal trapezoid contractions. The sensor (Figure 4.1) consisted of five cylindrical pin electrodes, each 0.5mm in diameter, protruding from the housing (2x3cm). The pins are blunted, such that they make an indentation when pressed against the skin, but do not puncture the surface. Four of the five pins are arranged at the



corners of a 5x5mm square; the fifth (reference) pin is in the center of the square, equidistant from each of the other four, such that the inter-electrode distance is 3.6mm. For more detailed information regarding the surface EMG sensors used in this study, the reader is advised to refer to Nawab et al. (2010). Before placement, the skin over the distal region of the muscle was prepared by carefully shaving and then cleansing with rubbing alcohol, the skin was then abraded in accordance with SENIAM recommendations (Hermens, 2000). The sensor was first cleaned with rubbing alcohol, before fixing to the prepared skin with adhesive tape. The sensor was located over the belly of the VL - 25% of the distance from the Gerdy prominence to the AIS (Blanc & Dimanico, 2010). A reference electrode was affixed to the patella; if it was deemed necessary, the investigator also shaved the skin over the patella before attaching the reference electrode.



**Figure 4.1** The surface array dEMG sensor (Delsys, Inc., Boston, Massachusetts) used to detect bipolar sEMG signals, on four separate channels. The sensor consists of five cylindrical pin electrodes, each 0.5mm in diameter, protruding from the housing (2x3cm).

Visual inspection of the signal, on all four channels, was carried out, prior to recording, to ensure that excessive background noise and artifact were not present. All analog EMG signals were low-pass (fourth-order Butterworth, 24 dB/octave slope, 1900-HZ cut-off) and high-pass (second-order Butterworth, 12 dB/octave slope, 20-HZ cut-off) filtered prior to sampling at a rate of 20,000 Hz (Appendix C). The four separate filtered EMG signals from the array were entered into the Precision Decomposition III (PD III) algorithm and decomposed into constituent motor unit action potential trains. Precision Decomposition techniques were originally described by De Luca & Adam (1999), having been in development since the 1970s. The technique has subsequently been refined by Nawab et al. (2010). PD III uses artificial intelligence to identify action potentials and assign them to individual motor units. This technique was specifically developed for decomposing surface EMG signals into their constituent MUAPs. The resulting output contains the firing instances for each motor unit. The mean firing rate, for each active motor unit, can then be calculated and plotted as a function of time. Mean firing rate curves were smoothed using a Hanning window; in this case all curves were filtered using a 800ms Hanning window, as recommended by the Software manufacturer. For analysis a long enough portion of the mean firing rate curves was needed to allow fluctuations in firing rate to be analyzed, however excessively long portions are not desirable, as the period should include minimal fluctuations in force or EMG RMS. A 3s portion has previously been deemed, by our group, to be appropriate. The 3s period at the distal end of the contractions steady-phase has also been found to be the region of greatest reliability (Balshaw, 2013). Prior to calculating mean motor unit firing rates, MUAPs were separated into three equal groups (where MUAPs could not be equally divided by three, the third group contained any additional MUs), such that MUAPs could be isolated into tertiles containing either early recruited, mid-recruited or

later recruited Mus (Figure 4.2). To assess the accuracy of the decomposed signal a Decompose-Synthesize-Decompose-Compare test, as described by De Luca & Hostage (2010) was performed over the selected 3s window (Appendix D). The level of common drive was quantified by performing cross-correlation analysis of all of the mean firing rates during the constant firing rate portions of the curves. The same 3s portion of the isometric contraction was analyzed for common drive as for mean firing rate. All possible combinations of motor units were cross-correlated with one another (Beck, Kasishke, Stock, & Defreitas, 2012) using the equation:

$$(f * g)[n] \stackrel{\text{def}}{=} \sum_{m=-\infty}^{+\infty} f^*[m]g[n+m]$$

where  $f$  and  $g$  are the mean firing rates of two motor units;  $f^*$  is the complex conjugate of  $f$ ,  $(f * g)$  is their cross-correlation; and  $m$  and  $n$  are temporal indices. The peak cross-correlation coefficient was calculated from each cross-correlation to determine common drive (Appendix E).

#### 4.2.4 Eccentric Exercise

Following baseline measurements on Day 0, subjects used their dominant leg to perform an eccentric exercise protocol, as outlined below, designed to induce temporary muscle damage. Repeat measures of all baseline characteristics were then taken on Days +2, +3, +7 and +14. The eccentric exercise was performed on one day only. During familiarization a maximum of one full set of twelve eccentric contractions was practiced. Prior to the eccentric exercise participants performed a brief warm-up, consisting of cycling for 5min at a cadence of 70rpm with power output of 50w (Lode Excalibur Sport V2 electrically-braked cycle ergometer, Lode BV, Groningen, Netherlands), to our knowledge no

evidence has been reported for this type of warm-up to cause muscle damage in healthy males. Immediately following the warm-up, participants were secured in the isokinetic dynamometer, (as described above) and measurements of muscle soreness were repeated, exactly as before, to ensure that no significant muscle soreness had resulted from the baseline measures and warm-up cycling.

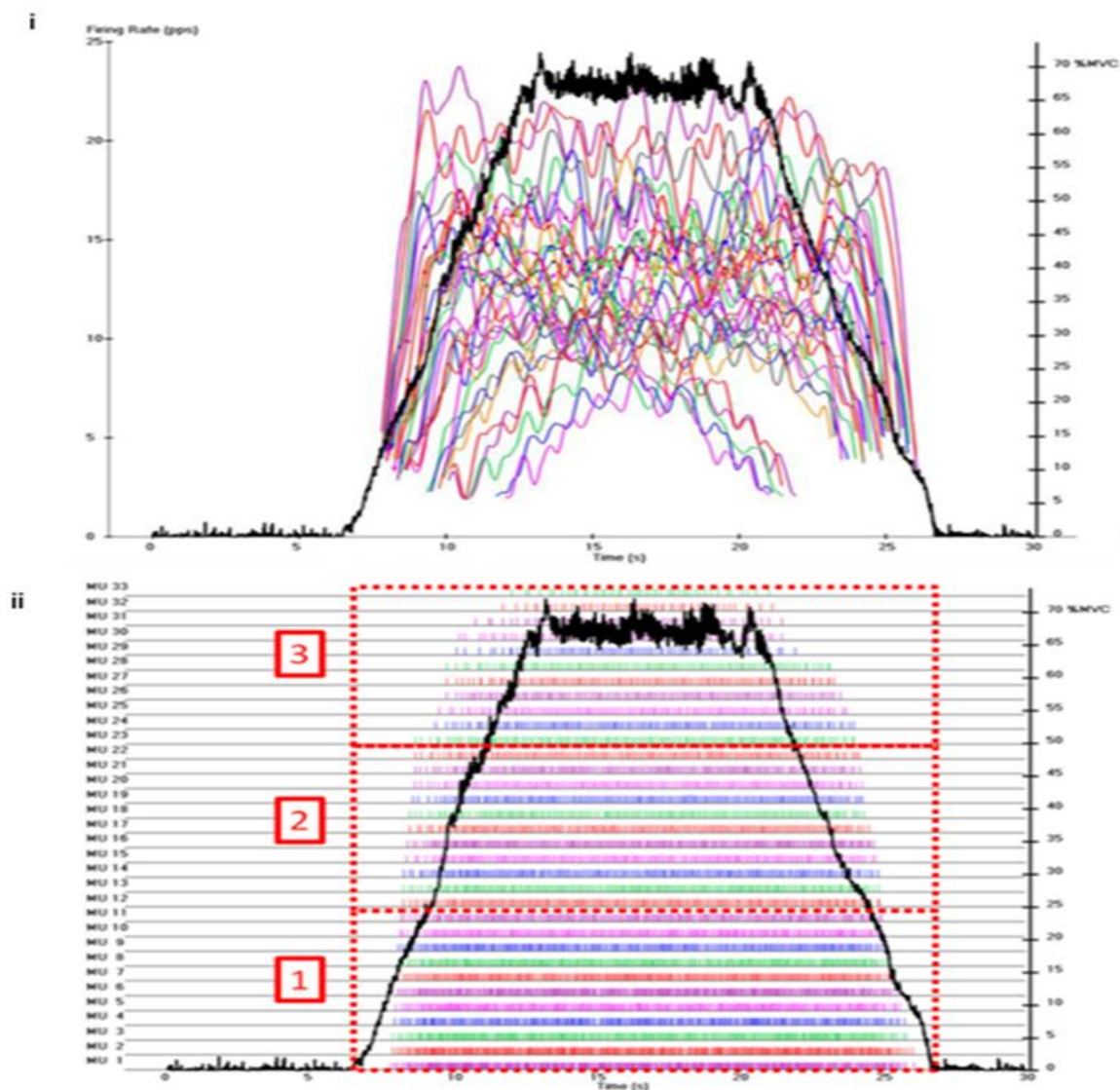


Figure 4.2 Example of mean firing rate curve plot (i) and firing rate bar plot (ii) from one participant. Each individual curve (i) represents the mean firing rate of a single detected motor unit; vertical bars (ii) represent the firings of each motor unit. The black line indicates the force output trace. Tertile groupings (ii) indicate earlier-recruited (1); mid-recruited (2); and later-recruited (3) motor units.

Participants then performed sets of twelve maximal eccentric contractions, until exhaustion; a minimum of 120 seconds recovery was permitted between each set. The ROM of these contractions was  $90^\circ$ , participants were instructed to provide maximum resistance from knee extension angle  $20^\circ$  to  $110^\circ$  (full extension being  $0^\circ$ ). The velocity of contraction was  $60^\circ\text{sec}^{-1}$ . Each eccentric contraction was followed by a passive return to start angle, at a velocity of  $180^\circ\text{sec}^{-1}$ , such that each set lasted for 24 seconds, with participants actively contracting for 75% of that time. This protocol, adapted from a number of previously published studies (Paschalis et al., 2005; Molina & Denadai, 2012) is designed to maximize eccentric workload whilst concomitantly minimizing concentric work and metabolic demand. Subjects were verbally encouraged to generate maximum force during each eccentric contraction, throughout the whole ROM. Participants were instructed to drink sufficient water following the eccentric exercise protocol to avoid a possible risk of acute renal failure due to rhabdomyolysis (Warren, 2002), but they were instructed to abstain from any therapeutic treatments, designed to ameliorate the symptoms of EIMD, prior to and throughout the trial period. Therapies to avoid included, but were not restricted to: whole-body vibration, massage, cryotherapy, NSAIDs and BCAAs (Aminian-far, Hadian, Olyaei, Talebian, & Bakhtiary, 2011; Connolly et al, 2003; Howatson & Someren, 2008; Howatson et al., 2012).

#### 4.2.5 Statistical Analysis

MVC, RTD muscle soreness, motor unit firing rate and common drive were analyzed using two-way (group, 2 x time, 5) repeated measures analysis of variance (ANOVA) with Tukey *post hoc* analysis performed where appropriate (Minitab 16 statistical software, Minitab Ltd., Coventry, UK). Data are presented throughout as mean ( $\pm$  SD), with statistical

significance set at  $p < 0.05$ . Where significant effects were observed, Cohen's  $d$  effect sizes (ES) were calculated by:  $Cohen's\ d = Mean_1 - Mean_2 / SD_{pooled}$ , where  $SD_{pooled} = \sqrt{[(SD_1^2 + SD_2^2) / 2]}$ , and 95% lower and upper confidence intervals (CI) were established relative to ES. ES were interpreted as  $< 0.2 =$  trivial,  $0.2-0.6 =$  small,  $0.6-1.2 =$  moderate,  $1.2-2.0 =$  large,  $> 2.0 =$  very large.

### 4.3 Results

EIMD significantly ( $F_{(4,13)} = 11.77$ ,  $p < 0.001$ ) reduced peak MVC (Figure 4.3A) and showed significant interaction effects ( $F_{(4,13)} = 18.49$ ,  $p < 0.001$ ) with the control leg, post hoc testing revealed significantly reduced MVC by 31.4% in the exercised leg at 48 hours compared to baseline (CI [0.58 to 2.24] ES = 1.45,  $p < 0.01$ ) which had fully recovered by day 7. RTD responded similarly by showing a significant ( $F_{(4,13)} = 9.96$ ,  $p < 0.001$ ) reduction of 67.04% in the exercised leg post-EIMD, with significant interaction effects ( $F_{(4,13)} = 9.49$ ,  $p < 0.001$ ), post hoc testing revealed significantly reduced RTD compared to baseline in the exercised leg at 48 and 72 hours with peak reduction at 48 hours (CI [1.21 to 3.07] ES = 2.20,  $p < 0.01$ ) which had recovered by day 7 (Figure 4.3B). PMT and EMD were unaffected by EIMD (Table 4.5).

The high-threshold motor units, in the third tertile fired significantly ( $F_{(4,13)} = 4.81$ ,  $p < 0.01$ ) slower following EIMD demonstrating a significant interaction ( $F_{(4,13)} = 4.81$ ,  $p < 0.01$ ) with the control leg and post hoc testing showed a significant decline at 48 hours (CI [1.01 to 2.79] ES = 1.96,  $p < 0.05$ ) which had returned to baseline levels after 72 hours (Figure 4.4C). The mid recruited motor units as shown by the second tertile demonstrated a tendency ( $F_{(4,13)} = 2.16$ ,  $p = 0.093$ ) towards lower mean firing rates in the exercised leg post-EIMD (Figure 4.4B). The mean firing rate of early recruited motor units, within the first tertile, was not significantly different ( $F_{(4,13)} = 1.19$ ,  $p > 0.05$ ) between days or groups (Figure 4.4A).

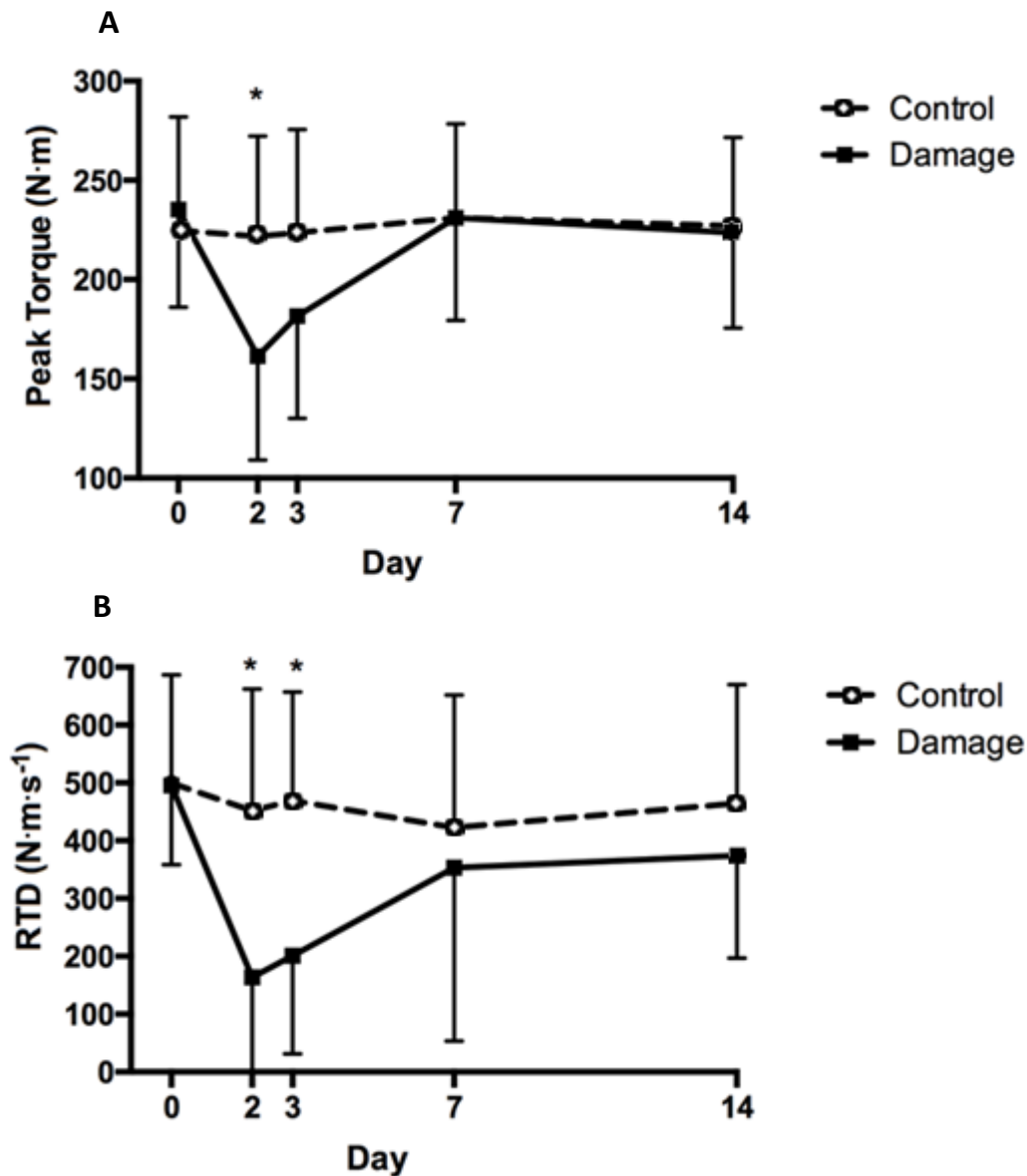


Figure 4.3 A) Maximal isometric voluntary contraction (MVC) of the exercised and control knee extensors. Values are mean  $\pm$  SD,  $n = 14$ . \* Significantly lower than baseline in the exercised leg,  $p < 0.001$ . B) Rate of torque development (RTD) of the exercised and control knee extensors. Values are mean  $\pm$  SD,  $n = 14$ . \* Significantly lower than baseline in the exercised leg,  $p < 0.001$ .

Common drive, as shown by the cross correlation coefficient of active motor units, was significantly ( $F_{(4,13)} = 8.52$ ,  $p < 0.05$ ) elevated from 0.36 to 0.56, and displayed significant interaction ( $F_{(4,13)} = 22.34$ ,  $p < 0.001$ ) with the control leg, post hoc analysis revealed a significant increase 48 hours post-EIMD (CI [4.41 to 7.98] ES = 6.39,  $p < 0.001$ ) which returned to baseline after 72 hours (Figure 4.6).



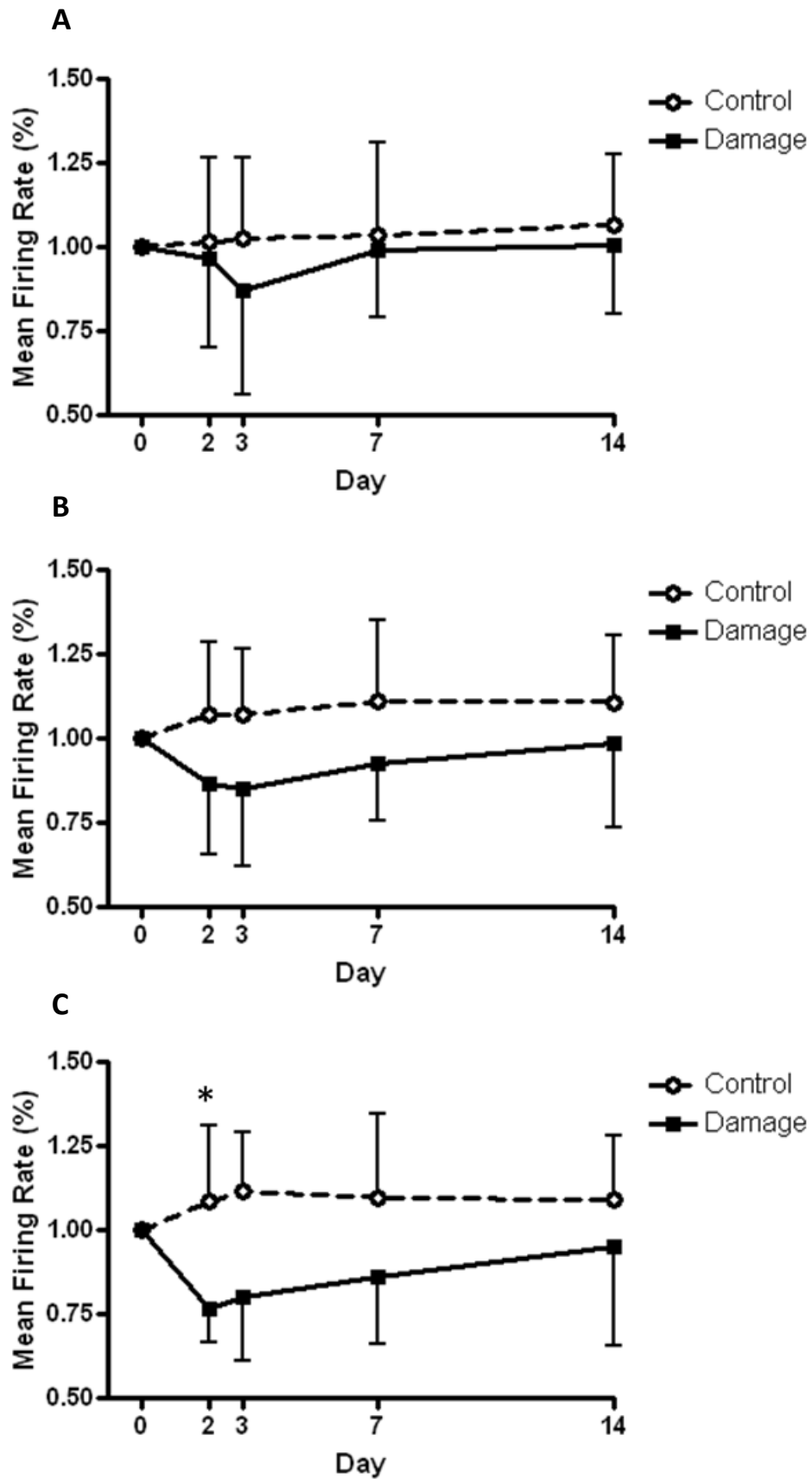


Figure 4.4 Mean motor unit firing rates of the exercised and control knee extensors. A) Early recruited motor units, B) mid recruited motor units and C) late recruited motor units. Values are % change  $\pm$  SD,  $n=14$ . \* Significantly lower than baseline in the exercised leg,  $p < 0.05$ .

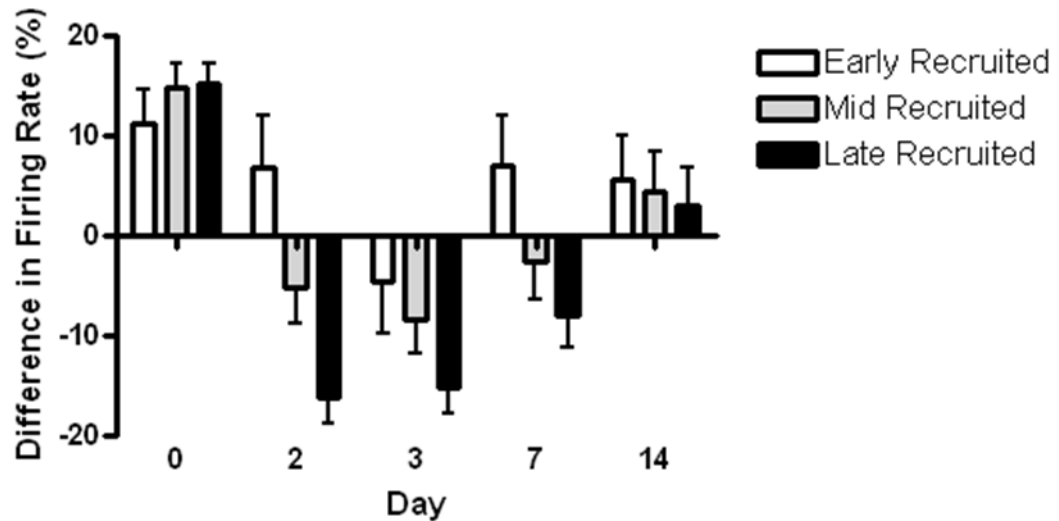


Figure 4.5 Percentage difference between mean motor unit firing rates of the control and exercised knee extensors, divided into low-threshold (early recruited), mid-threshold (mid recruited) and high-threshold (late recruited) motor units.

Muscle soreness (Table 4.3) was significantly greater ( $F_{(4,13)} = 5.35$ ,  $p < 0.01$ ) in the exercised leg following EIMD, post hoc testing revealed a main effect of time at 48 and 72 hours with peak elevation at 72 hours (CI [0.05 to 1.60] ES = 0.85  $p < 0.01$ ). The number of motor units detected by dEMG did not differ significantly ( $F_{(4,13)} = 0.62$ ,  $p > 0.05$ ) across time or group (Table 4.4).

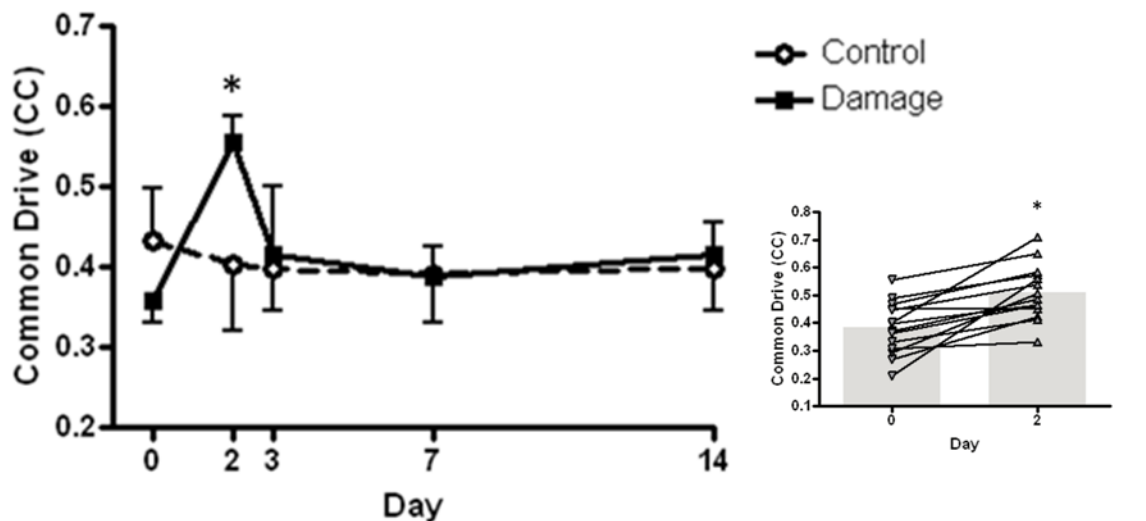


Figure 4.6 Cross correlation coefficient of active motor units in the exercised and control knee extensors. Cross correlation coefficients of active motor units for each individual participant between baseline (Day 0) and peak EIMD (Day 2) are presented inset. Values are mean  $\pm$  SD,  $n = 14$ . \* Significantly higher than baseline in the exercised leg,  $p < 0.05$ .

Table 4.2 Mean firing rate of early, mid and late recruited motor units of VL, during 60% MVC isometric contraction.

		Early Recruitment					Mid Recruitment					Late Recruitment				
Day		0	2	3	7	14	0	2	3	7	14	0	2	3	7	14
Control	Mean (Hz)	19.25	19.52	19.75	19.92	20.54	15.91	17.04	17.24	17.65	17.63	13.86	14.59	15.06	15.21	15.09
	± SD	3.44	4.91	4.77	5.53	4.32	2.55	3.68	3.36	4.31	3.45	2.10	3.28	2.53	3.76	2.88
Damage	Mean (Hz)	21.69	20.93	18.90	21.45	21.76	18.71	16.21	15.92	17.20	18.45	16.35	12.55	13.07	14.09	15.58
	± SD	3.45	5.51	5.86	4.20	4.40	2.19	3.45	3.66	3.29	4.60	2.17	1.69	2.37	2.84	4.62

Table 4.3 Visual analogue scale (VAS) scores for knee extensor muscle soreness in flexed (90°) and extended (0°) positions. \* Significantly higher than baseline in the exercised leg,  $p < 0.01$ .

Day		Flexed					Extended				
		0	2	3	7	14	0	2	3	7	14
Control	Mean (cm)	3.93	3.64	3.54	2.89	2.89	4.17	4.29	4.64	4.25	2.61
	± SD	4.22	3.40	2.80	2.46	2.81	4.59	3.34	3.65	2.39	2.19
Damage	Mean (cm)	4.17	4.39	3.89	3.04	2.68	3.25	5.93*	6.43*	3.00	2.50
	± SD	2.66	2.72	2.69	3.77	2.32	3.28	3.83	4.16	1.98	2.28

Table 4.4 Number of motor units identified by PD III decomposition algorithm.

Day		0	2	3	7	14
Control	Mean	22.80	26.36	20.64	22.09	20.46
	± SD	11.72	9.06	10.58	10.49	8.26
Damage	Mean	19.82	22.27	20.18	21.00	21.64
	± SD	8.89	7.55	9.09	8.07	8.88

Table 4.5 Pre-motor time (PMT) and electromechanical delay (EMD) calculated from VL during MVC.

Day		0	2	3	7	14	
PMT	Control	Mean (ms)	323.41	387.05	647.41	385.59	255.91
		± SD	90.37	359.84	469.46	146.85	158.75
	Damage	Mean (ms)	402.27	732.32	425.14	408.14	373.14
		± SD	346.36	432.66	235.79	232.29	186.23
EMD	Control	Mean (ms)	26.55	27.23	59.45	27.41	238.82
		± SD	19.78	25.87	50.73	25.13	211.69
	Damage	Mean (ms)	20.65	513.27	30.41	168.58	104.73
		± SD	26.18	546.81	57.70	464.08	307.83

#### 4.4 Discussion

This study aimed to investigate the firing rates of individual motor units, specifically high-threshold motor units, following a bout of EIMD. Additionally, the collective control (common drive) of the motor unit pool was examined, before and after damaging exercise. EIMD was successfully induced as shown by the 31.4% MVC decline at 48 hours post-exercise. This MVC force decrease was accompanied by diminished rate of torque development also at 48 hours post-exercise. Taken together these findings are clear indicators of EIMD derived muscle contractile impairment (Hunter et al., 2012; Molina & Denadai, 2012; Peñailillo et al. 2015). Coinciding with these functional impairments, the mean firing rate of high-threshold/ later recruited motor units dropped by 22.3% 48 hours post-exercise. Firing rates of these units returned to baseline levels by 7 days post-exercise, in line with MVC. The cross correlation coefficient or synchronization of the motor unit pool increased from 0.36 at baseline to 0.56 after 48 hours, indicating increased common drive.

As hypothesized we showed that later recruited motor units (i.e. higher-threshold) would be specifically impaired following EIMD, as we know that later recruited motor units associate with type II muscle fibres (Henneman, 1985) and that it is these fibres that are most susceptible to EIMD (Macaluso et al., 2012). It seems likely that the observed impairment stems from a feedback mechanism resulting from elevated III/IV afferent signaling following EIMD, perhaps as a result of proprioceptive inhibition (Komi, 2000) or through pain-associated elevation of nociceptor activity (Kennedy et al., 2015). Mediation by III/IV afferents also appears a plausible explanation given the lack of any impairment in the contralateral limb (Kennedy et al., 2015).

Ye et al. (2015) previously examined the relationship between motor unit firing rate and recruitment threshold, reporting a decreased slope in the regression following eccentric exercise, which the authors suggested to indicate disruption to the higher threshold motor units. Although we cannot say for certain, it seems likely that during an isometric contraction at 60% of MVC the latest recruited motor units will be associated with type II muscle fibres. The current results reveal a clear decline in firing rate in high-threshold motor units. The motor units belonging to the group of midpoint recruitment showed a tendency toward impaired firing. It seems plausible that in most cases this middle group would be composed of a mix of type I and type II fibres, in this case we could speculate that those units comprising type II fibres may be exhibiting reduced firing rates, while the firing rates of the type I fibre-associated units are preserved, as we see in the early recruited motor unit group.

Despite the decline in high-threshold motor unit firing rate the 60% (of baseline) MVC target was successfully achieved during EIMD suggesting that compensation had occurred. We propose this came from the increased common drive we showed, which typically increases when higher forces are required (Semmler & Nordstrom, 1998) and during muscle fatigue (Contessa et al., 2009). However, Beck et al. (2012) surprisingly reported no alteration in common drive following EIMD in the biceps brachii, despite a 19.5% drop in peak force; it should be noted that the isometric contractions during which common drive was assessed were based on feedback provided by EMG (RMS) and not on a predetermined target force output, with this in mind, objective comparison cannot be made between pre- and post-exercise conditions. Nevertheless, the most likely mechanism causing the increased common drive in our study emanates from impaired proprioception

which can occur following EIMD (Torres et al., 2010). It has been previously demonstrated (Hunter et al. 2012; Murayama et al. 2000) that EIMD transiently alters skeletal muscle architecture which is likely to alter proprioception from muscle spindles which has been suggested to influence common drive (De Luca et al., 2008) Furthermore, Contessa et al. (2009) observed a relationship between the number of newly recruited motor units and the common drive with contraction endurance time, leading them to propose a decreased muscle spindle influence would result in increased common drive.

This is the first study to investigate motor unit firing rate, through decomposed EMG, by segregating the motor unit pool into early recruited (low-threshold) units, and later recruited (high-threshold) units. This was done using robust decomposition methodology (Nawab et al., 2010) with 95% accuracy result following Decompose-Synthesize-Decompose-Compare validation test. In summary we have shown that EIMD provides an acute insult to the muscle, leading to altered firing behavior in later recruited motor units. Specifically, we observed reduced firing rates among these high-threshold motor units suggesting an EIMD derived impairment. However the concomitant increase in common drive likely points to a centrally mediated compensatory effect, allowing force to be maintained at higher than otherwise possible levels. We propose that these alterations stem from type III/IV afferent feedback, namely through impaired proprioception.

#### **4.5 Conclusion**

In conclusion, this study provides new evidence that exercise-induced muscle damage causes associated decreases in mean motor unit firing rate of later recruited motor units with higher force thresholds, whilst low-threshold units, recruited early after the onset of contraction remain unaffected. In response to the impaired firing of these later motor



units, common drive is elevated in order to counteract the impaired force output suffered by the damaged muscle. These findings increase our understanding of the central mechanisms associated with EIMD and may in turn lead to development of novel management and preventative procedures.

# Chapter 5

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## Self-Myofascial Release Increases Skeletal Muscular Efficiency without Affecting Range of Motion

**Lewis J Macgregor, Ryan M Bennett, Angus M Hunter**

## 5.1 Introduction

We have demonstrated that TMG provides a useful method to assess muscle contractile mechanics. However, as outlined in Chapter 3, the extent to which these contractile mechanics relate to muscle function remains unclear. An intervention which may affect muscle function, through altered muscle mechanics, may provide further insight. As discussed in Chapter 4, exercise-induced muscle damage (EIMD) is associated with impaired muscle function, which can be detrimental to athletic performance. As such, effective interventions to combat deleterious symptoms of EIMD are strongly sought after within performance sport. Among recent innovations, foam rolling (FR) has become increasingly popular (Kim, Park, Goo, & Choi, 2014; Okamoto, Masuhara, & Ikuta, 2014). Indeed, research suggests that FR performed following a bout of physical activity can reduce delayed onset muscle soreness (DOMS) (Jay et al., 2014; Pearcey et al., 2015). MacDonald, Button, Drinkwater, & Behm, (2014) also reported reductions in the decrement of peak force, twitch force and voluntary activation subsequent to a bout of eccentric exercise when followed by FR. To date there has been little research into the application of FR as a prophylactic as opposed to a recovery tool (Sullivan et al., 2013; Macdonald et al., 2014). Moreover, there remains much conjecture around the overall efficacy of FR, with equivocal findings regarding the acute effect on muscle function (Janot et al., 2013; Peacock, Krein, & Silver, 2014) and range of motion (Roylance et al., 2013; Škarabot, Beardsley, Hons, & Štirn, 2015).

Although evidence remains inconclusive, FR is widely seen, within sport, as an attractive alternative to massage and is commonly used to supplement, or in some cases replace, traditional warm up protocols (Healey, Hatfield, Blanpied, Dorfman, & Riebe, 2014). FR was developed to mimic myofascial release (MFR) (Barnes, 1997), a procedure in which

connective tissues are manipulated through application of sustained pressure and stretching, in order to reduce restriction (Galloway, Hunter, & Watt, 2012). MFR is typically administered by therapists, with the aim of restoring muscle function, by reducing fibrous adhesions formed within the soft tissue surrounding skeletal muscle (MacDonald et al., 2013). In recent years FR has been developed as a novel self-MFR technique (Curran, Fiore, & Crisco, 2008) with the aim of enhancing recovery and performance (Cheatham, Kolber, Cain, & Lee, 2015). It involves an individual applying their own body weight to a neoprene coated cylinder, using small repetitive undulating movements to exert pressure on the muscle (Paolini, 2009). Suggested mechanisms of action for MFR have been reviewed (Schleip, 2003; Simmonds et al. 2012), although to date there is a lack of consensus surrounding these mechanisms. Potential mechanisms can be divided into two categories: first, mechanical, mostly focussed around alterations in the structure or state of fascial tissue; or second, neurophysiological, focussing on afferent signalling from mechanoreceptors (Beardsley & Škarabot, 2015). As the majority of FR research has focused on functional outcomes, or has involved FR subsequent to EIMD, it remains difficult to ascertain which, if any, of these proposed mechanisms holds true.

Given the improvements in joint ROM often observed with FR (Bradbury-Squires et al., 2015; Škarabot et al., 2015), obvious comparisons have been drawn with stretching protocols, which may shed more light on the mechanisms behind FR. Static stretching (SS), for example, is believed to be effectual mainly through increasing perceived tolerance to muscle stretch (Wepppler & Magnusson, 2010). Indeed, SS and FR have been demonstrated to be similarly successful in improving ROM (Halperin, Aboodarda, Button, Andersen, & Behm, 2014). However, given that SS (Bandy et al. 1997; Kay & Blazeovich, 2012; Power et al.

2004), but not FR (MacDonald et al., 2013), leads to decreased muscle force production, and considering that combined FR and SS is more effective at increasing ROM than SS or FR alone (Mohr, Long, & Goad, 2014; Škarabot et al., 2015) it would appear that disparate mechanisms underlie SS and FR.

As mentioned previously, potential mechanisms of action for FR can be divided into the category of mechanical or neurophysiological (Beardsley & Škarabot, 2015). Combining massage with SS results in reduced spinal reflex excitability without affecting twitch contractile properties; SS alone can prolong EMD, which remains unaffected by massage (Behm et al., 2013). Therefore, whilst SS increases ROM through both neural and mechanical factors, massage-induced alterations can be attributed solely to reflex inhibition (Behm et al., 2013). However, massage also has been linked to muscle force impairment while maintaining neuromuscular recruitment (Hunter, Watt, Watt, & Galloway, 2006); as a result of this these authors suggested the observed force impairment was due to changes in muscle architecture and resulting alterations in series compliance. As such these architectural changes are likely to be associated with alterations in muscle stiffness (Ditroilo et al., 2011), which can be measured from the extent of muscle radial displacement. MFR, the basis behind FR, involves sustained pressure applied to muscle, it has been speculated that FR acts fundamentally on connective tissue (Macdonald et al., 2014) whilst leaving muscle architecture (i.e. series compliance and length-tension relationship) unaltered. However, due to the scarcity of evidence, there still remains no definitive consensus as to the mechanisms underlying FR, and as such its efficacy cannot be determined.

If FR is to be effectively used to provide benefits for high performance sport, it is necessary to first fully elucidate the mechanisms of action, in order to establish optimal

methodology. Therefore, the aim of this study was to investigate the impact of a well-controlled bout of MFR using FR on functional muscle performance, specifically static and dynamic strength and ROM. Impacts on muscle contractile properties and neural drive to muscles were examined to provide insight into the mechanisms underlying any functional adaptations. The time course was examined by performing repeated measurements of all variables post-treatment and repeating the treatments on three consecutive days. It was hypothesised that FR would increase ROM; however this would be accompanied by a reduction in force production.

## 5.2 Methods

### 5.2.1 Participants

Sixteen healthy, recreationally active, male participants with no history of neuromuscular or musculoskeletal disorders were recruited to complete this study. Participant characteristics are shown in Table 5.1. Participants were not currently undertaking any form of MFR at the time of their participation in the study. Participants refrained from: 1) any unaccustomed physical activity for the duration of the trial and 2) any strenuous exertion for at least 24h prior to each testing session. Volunteers provided written informed consent prior to being included in the study. The principles outlined in the latest revision of the *Declaration of Helsinki* were adhered to and the study was approved by the School of Sport, Research Ethics Committee (SSEC), University of Stirling.

**Table 5.1 Participant characteristics (n = 16). MVC is taken at baseline of the control condition.**

	<b>Age (y)</b>	<b>Height (cm)</b>	<b>Mass (kg)</b>	<b>MVC (Nm)</b>
<b>Mean</b>	24.5	180.0	82.4	222.0
<b>± SD</b>	4.4	5.4	8.0	36.5

### 5.2.2 Study Design

Following full familiarisation of the testing procedures (utilizing the non-dominant leg), participants reported to the laboratory for 2 separate trials. Each trial consisted of 3 testing sessions on 3 consecutive days, with 7 days separating the start of each trial (Figure 5.1). Participants reported to the laboratory following an overnight fast and initially rested in a supine position for 30 minutes. Following this rest period, mechanical and contractile properties of the vastus lateralis (VL) and rectus femoris (RF) were measured using TMG

(BMC Ltd, Ljubljana) as has been described in Chapter 3. Participants were then tested for knee flexion range of movement (ROM) before isometric and isokinetic assessments, using an isokinetic dynamometer (Kin Com, Chattanooga, Hixson, TN, USA). Following baseline measures participants either rested for 2 minutes or performed 2 minutes of self-MFR (FR). All measurements were then repeated, in an identical order to pre-intervention: immediately-, 15 minutes- and 30 minutes-post rest/ FR. All measurements were performed on the dominant leg. Dietary intake records were completed on the day preceding each session of the first trial, and participants were instructed to replicate their dietary intake before each visit for the second trial.

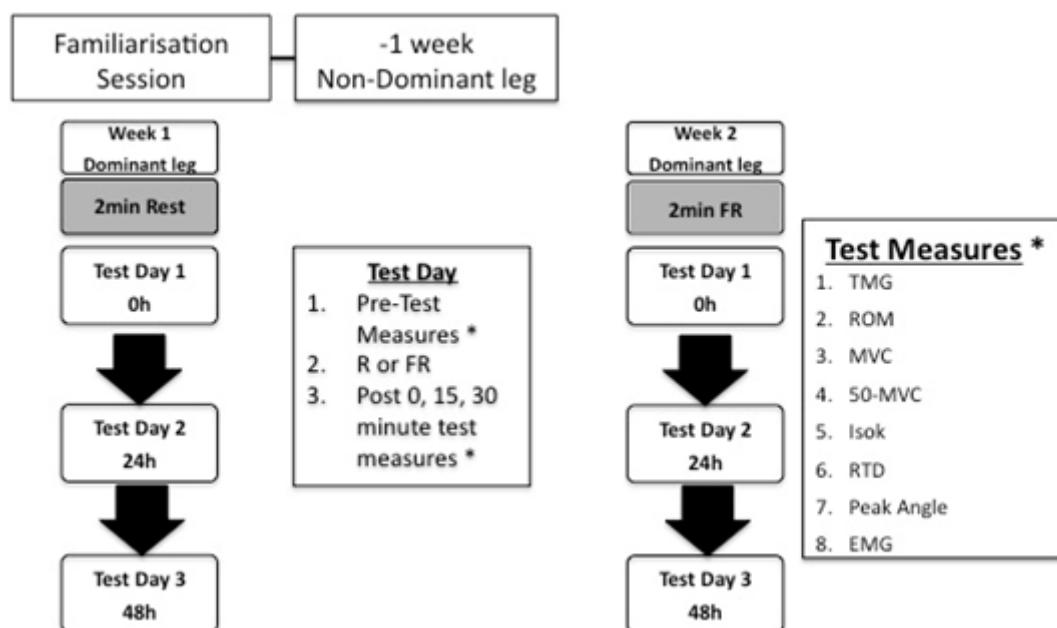


Figure 5.1 Timeline of the testing period (R = 2 minutes rest, FR = 2 minutes self-MFR).

### 5.2.3 Protocol

After resting in a supine position for 30-minutes participants then adopted a knee joint angle of  $60^\circ$  ( $0^\circ$  = full extension), which was maintained by the use of a foam support



placed beneath the popliteal fossa. Two pairs of self-adhesive electrodes (5cm<sup>2</sup>) (Axelgaard, USA) were affixed to the skin; one pair over the VL and one pair over the RF. Electrode positioning and measurement protocol were conducted as described in Chapter 3. The sites over each muscle, of the sensor and the electrode pair, were marked with semi-permanent ink to enable exact relocation following FR treatment and on subsequent days. Following FR, electrodes were unplugged but remained attached to skin for the remainder of the trial.

Knee flexion ROM was measured in accordance with previous literature (MacDonald et al. 2014). Participants adopted a modified kneeling lunge position. The non-dominant leg was positioned with the sole of the foot flat on the floor and the knee flexed to 90°, participants were permitted to place their hands on this knee for support, but were instructed to angle their torso perpendicular to the floor throughout the ROM assessment. With the hip of the dominant leg extended as far as possible the foam roller was placed under the ankle in order to standardize the starting position (Figure 5.2). Internal knee angle was recorded, using a goniometer, and then the knee was flexed as far as voluntarily possible. Maximal knee flexion was held only as long as was required to measure the internal angle for a second time. Total ROM was taken as the starting knee angle subtracted from the end knee angle. ROM assessment was performed only once at each time point, in order to limit the impact of repeated stretching of the knee extensors.



**Figure 5.2 Measurement of knee flexion range of motion (ROM).**

Participants were coupled to the isokinetic dynamometer (Kin-Com, Chattanooga Group Inc, USA) as described previously, in Chapter 3. Participants performed a standardized warm up (Balshaw & Hunter, 2012; Hunter et al., 2012) prior to baseline measurements only. A pair of Ag/Agu self-adhesive electrodes (PNS Dual Element Electrode; Vermed, Vermont, USA) were affixed to the skin  $\frac{1}{3}$  of the distance from the greater trochanter to the lateral femoral epicondyle, following thorough preparation of the skin in accordance with SENIAM guidelines (Hermens, 2000). A reference electrode was affixed to the patella. Surface electromyography (sEMG) was recorded during all isometric contractions and was synchronized with torque output to allow determination of pre motor

time (PMT) and electromechanical delay (EMD) (Howatson et al., 2009). sEMG was captured at 2KHz, anti-aliased and filtered automatically using a 1Hz band pass filter.

Isometric assessments were performed exactly as described in Chapter 3. Participants next performed a submaximal isometric contraction (50-MVC) at 50% of their baseline MVC, for 30s. Torque was to be increased gradually until the target output was reached then held as steady as possible for 30s. Participants were provided with visual and verbal feedback throughout 50-MVC but verbal feedback only throughout MVC. Peak isokinetic torque as well as angle of peak torque were assessed at  $60^{\circ}\text{s}^{-1}$ , starting at  $80^{\circ}$  and finishing at  $20^{\circ}$  ( $0^{\circ}$  = full extension). Three contractions were performed with 30s rest between each contraction, verbal feedback was provided to ensure maximum effort during each contraction.

#### *5.2.4 Self-Myofascial Release*

Following baseline measurements participants performed self-MFR, using a commercially available foam roller (TriggerPoint Performance, Austin, Texas, USA) constructed of a hollow PVC pipe surrounded by a thin layer of neoprene as described by Curran et al. (2008). The foam roller was positioned initially at the mid-point between the anterior inferior iliac spine (AIS) and the upper border of the patella, participants supported themselves upon their forearms. To ensure that there were no other ground contact points, participants were instructed to plantar flex, whilst also positioning their feet and knees together, in order to focus the pressure of the foam roller upon the lateral-frontal aspect of the thigh (Figure 5.3). The length of area that was treated with MFR was  $\frac{2}{3}$  of the distance between the AIS and the upper border of the patella. A custom-built metal frame positioned beneath the participant ensured that the correct area was treated. Once in position,

participants rolled backwards and forwards in an undulating motion, the rate of movement was controlled by a metronome set at a predetermined rate based on one complete roll of the treated area (proximal-to-distal or distal-to-proximal) per 1-second. The foam roller was exclusively in contact with a force platform (400S Force Plate, Innervations, Australia) and force was recorded throughout the 2-minute FR treatment using Acqknowledge software (Acqknowledge 3.9.1, Biopac Systems Inc) at a sampling frequency of 2KHz (Table 5.2). For the control, participants adopted a supine position with the popliteal fossa of their dominant leg resting upon the foam roller in order to maintain a knee joint angle of  $\sim 60^\circ$  ( $0^\circ$  = full extension). The duration of the rest period was identical to the duration of MFR.



Figure 5.3 A participant performing self-MFR using the foam roller.

Table 5.2 Force applied during FR treatment. Values are mean  $\pm$ SD (Coefficient of Variation %), n = 16.

	<b>D0</b>	<b>D24</b>	<b>D48</b>	<b>Overall</b>
<b>Average Force rolled (N)</b>	48.2 $\pm$ 54.4 (1.13)	45.3 $\pm$ 50.6 (1.12)	48.0 $\pm$ 81.6 (1.70)	49.3 $\pm$ 62.9 (1.28)
<b>Average Force/kg rolled (N/kg)</b>	7.0 $\pm$ 1.3 (0.19)	6.6 $\pm$ 1.0 (0.15)	6.0 $\pm$ 0.9 (0.15)	6.5 $\pm$ 1.1 (0.17)

### 5.2.5 Data Analysis

Peak isometric torque was analysed as described in previous chapters, along with rate of torque development (RTD) over time windows of 0-50ms, 0-100ms, 0-200ms and 0-300ms, and PMT and EMD using Matlab version 7.11.0.584 (R2010b) software (The MathWorks, Inc.). Synchronous capture of sEMG and torque output allowed for assessment of pre-motor time (PMT) (the time delay between the audio prompt and the initiation of EMG) and electromechanical delay (EMD: the time delay between the initiation of EMG and an increase in torque  $\geq$  2SD above baseline). sEMG was captured for 30s during 50-MVC, signals were RMS converted using the data collection software (Acqknowledge), and normalized to sEMG RMS from the MVC. Normalized RMS were divided into 5 x 6s epochs for analysis. During 50-MVC participants were required to maintain torque equivalent to 50% of their baseline MVC. Peak isokinetic torque was recorded from the highest of the three contractions, the angle at which peak torque occurred was also recorded from this contraction.

### 5.2.6 Statistical Analysis

All dependent variables were tested for assumption of normality and analyzed using a three factor analysis of variance (ANOVA) with repeated measures (group [2] x time [4] x day [3]). Tukey *post hoc* analysis was performed where appropriate (Minitab 16 statistical software, Minitab Ltd., Coventry, UK). Statistical significance was accepted at  $P < 0.05$ . All values were reported as mean  $\pm$  standard deviation (SD). Where significant effects were observed, Cohen's d effect sizes (ES) were calculated by:  $\text{Cohen's } d = \frac{\text{Mean1} - \text{Mean2}}{\text{SD}_{\text{pooled}}}$ , where  $\text{SD}_{\text{pooled}} = \sqrt{\frac{(\text{SD}_1^2 + \text{SD}_2^2)}{2}}$ , and 95% lower and upper confidence intervals (CI) were established relative to ES. ES were interpreted as  $< 0.2 = \text{trivial}$ ,  $0.2-0.6 = \text{small}$ ,  $0.6-1.2 = \text{moderate}$ ,  $1.2-2.0 = \text{large}$ ,  $> 2.0 = \text{very large}$ .

## 5.3 Results

### 5.3.1 Range of Movement and Maximal Isometric Voluntary Contraction

A significant interaction effect ( $F_{(2,15)} = 9.20$ ,  $p < 0.001$ ) was detected in MVC with time and condition (Figure 5.4), this was caused by significantly elevated MVC on D48 in the FR treatment condition (CI [2.52 to 4.78] ES = 3.75,  $p < 0.001$ ). ROM was not significantly different between conditions or over time ( $F_{(3,15)} = 1.93$ ,  $p = 0.125$ ) (Table 5.3).

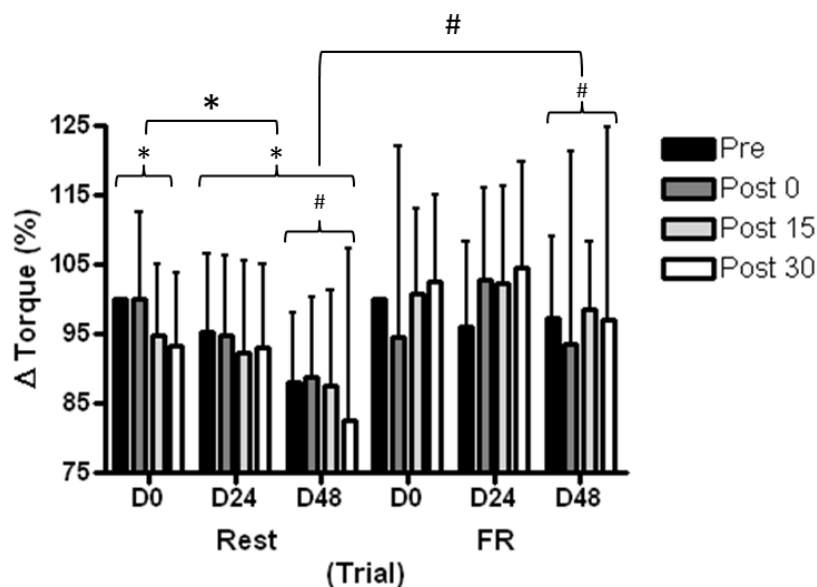


Figure 5.4 Maximal isometric voluntary contraction (MVC) of the knee extensors in the control (Rest) and intervention (FR) conditions. Values are mean  $\pm$  SD,  $n = 16$ . \* Significant within condition differences,  $p < 0.001$ . # Significant difference between conditions,  $p < 0.001$ .

### 5.3.2 Neuromuscular Recruitment

Foam rolling significantly ( $F_{(1,15)} = 9.32$ ,  $p < 0.01$ ) reduced RMS amplitude in the first 20% (0-6 seconds) of a 30 second contraction compared to control (Appendix F), with significant interaction effect ( $F_{(3,15)} = 5.24$ ,  $p < 0.01$ ) between time and condition, post hoc testing revealed significantly reduced RMS immediately, 15- and 30-minutes post-FR, compared to post-Rest (CI [1.77 to 3.70] ES = 2.81,  $p < 0.01$ ). Between 13-18 seconds (Figure

5.5), foam rolling significantly ( $F_{(1,15)} = 8.78$ ,  $p = 0.01$ ) reduced RMS compared to control, with significant interaction effect ( $F_{(3,15)} = 6.56$ ,  $p < 0.001$ ) between time and condition, post hoc testing revealed significantly reduced RMS immediately, 15- and 30-minutes post-FR, compared to post-Rest (CI [1.01 to 2.66] ES = 1.88,  $p < 0.001$ ). During the final 20% (24-30 seconds) of the 30 second contraction, foam rolling significantly ( $F_{(1,15)} = 8.35$ ,  $p < 0.05$ ) reduced RMS compared to control (Appendix F), with significant interaction effect ( $F_{(3,15)} = 7.29$ ,  $p < 0.001$ ) between time and condition, post hoc testing revealed significantly reduced RMS immediately, 15- and 30-minutes post-FR, compared to post-Rest (CI [0.85 to 2.46] ES = 1.7,  $p < 0.001$ ). Rate of perceived exertion (RPE) was not significantly different between conditions or over time ( $F_{(3,15)} = 0.66$ ,  $p = 0.578$ ) (Table 5.9).

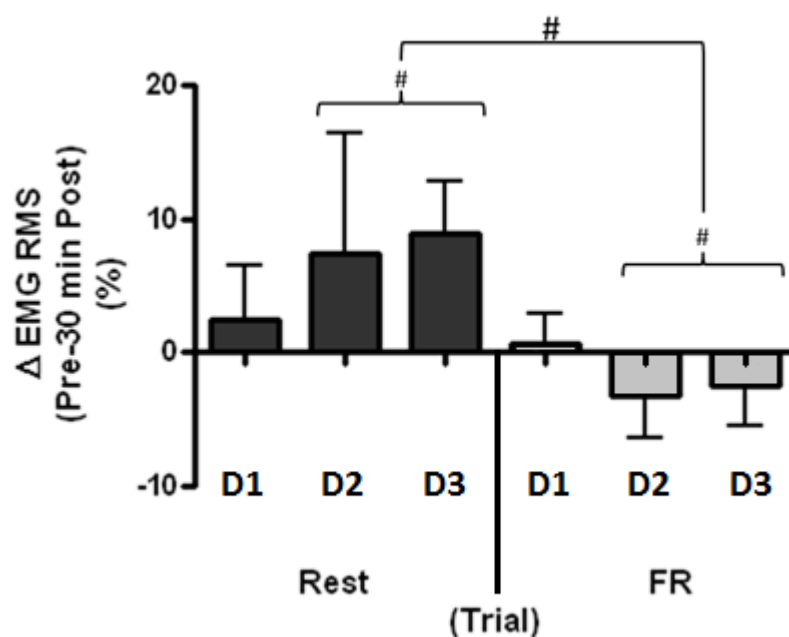


Figure 5.5 Change in normalized RMS (13-18 seconds) from pre- to 30 minutes post-treatment in the control (Rest) and intervention (FR) conditions. Values are mean  $\pm$  SD,  $n = 16$ . # Significant difference between conditions,  $p < 0.001$ . (0-6 seconds and 24-30 seconds are shown in Appendix F).



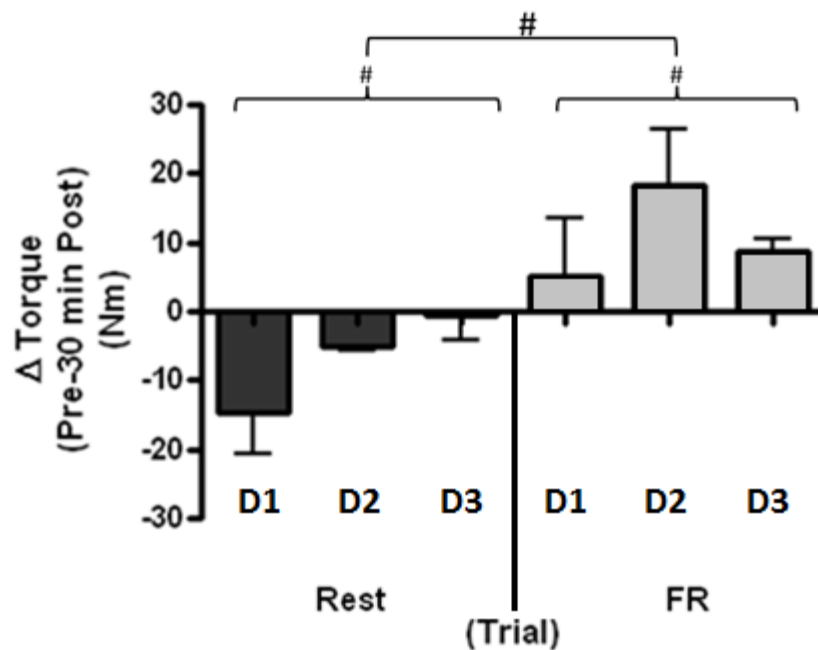


Figure 5.6 Change in MVC between pre- and 30 minutes post-treatment in the control (Rest) and intervention (FR) conditions. Values are mean  $\pm$  SD,  $n = 16$ . # Significant difference between conditions,  $p = 0.003$ .

### 5.3.3 Maximal Isokinetic Strength and Angle of Peak Torque

A significant interaction effect ( $F_{(2,15)} = 8.22$ ,  $p < 0.001$ ) was detected in isokinetic ( $60^\circ\text{s}^{-1}$ ) peak torque with time and condition (Figure 5.7), which was caused by significantly reduced peak torque, compared to baseline, on D48 in the Rest condition and D24 and D48 in the FR treatment condition (CI [2.63 to 4.95] ES = 3.89,  $p < 0.001$ ). There was a significant increase ( $F_{(3,15)} = 4.23$ ,  $p = 0.010$ ) in knee joint angle of peak torque with time (Table 5.4), post hoc testing revealed a significantly larger joint angle 15 and 30 minutes post-treatment compared to baseline, independent of condition (CI [0.96 to 2.59] ES = 1.82,  $p < 0.05$ ).

Table 5.3 Range of motion (ROM) of the knee extensors in the control (Rest) and intervention (FR) conditions. Values are mean  $\pm$  SD, n = 16.

	Day	D0				D24				D48			
	Time	Pre-	0	15	30	Pre-	0	15	30	Pre-	0	15	30
Rest	Mean (°)	32.8	33.3	36.7	35.8	35.6	35.6	37.3	36.7	34.2	35.1	34.4	34.9
	$\pm$ SD	16.1	14.6	17.2	15.4	15.1	17.2	16.6	16.8	16.0	16.4	16.4	17.8
FR	Mean (°)	36.3	36.1	35.7	37.8	35.5	34.6	35.3	34.0	36.3	35.7	34.4	34.3
	$\pm$ SD	15.5	15.1	13.6	16.7	15.3	12.7	13.6	14.4	15.6	17.4	16.6	18.1

Table 5.4 Knee joint angle of peak torque production in the control (Rest) and intervention (FR) conditions (0° = full knee extension). Values are mean  $\pm$  SD, n = 16.

	Day	D0				D24				D48			
	Time	Pre-	0	15	30	Pre-	0	15	30	Pre-	0	15	30
Rest	Mean (°)	60.7	60.6	62.6	64.1	62.0	63.1	62.4	64.1	62.2	61.2	64.9	63.6
	$\pm$ SD	5.0	5.8	5.9	5.0	3.7	5.2	6.5	5.4	5.1	6.0	4.1	5.2
FR	Mean (°)	61.2	63.3	62.8	63.6	62.4	63.2	64.3	61.8	62.3	63.9	63.3	63.7
	$\pm$ SD	5.9	4.6	6.8	4.8	8.2	4.6	5.8	6.9	6.9	5.8	5.3	6.5

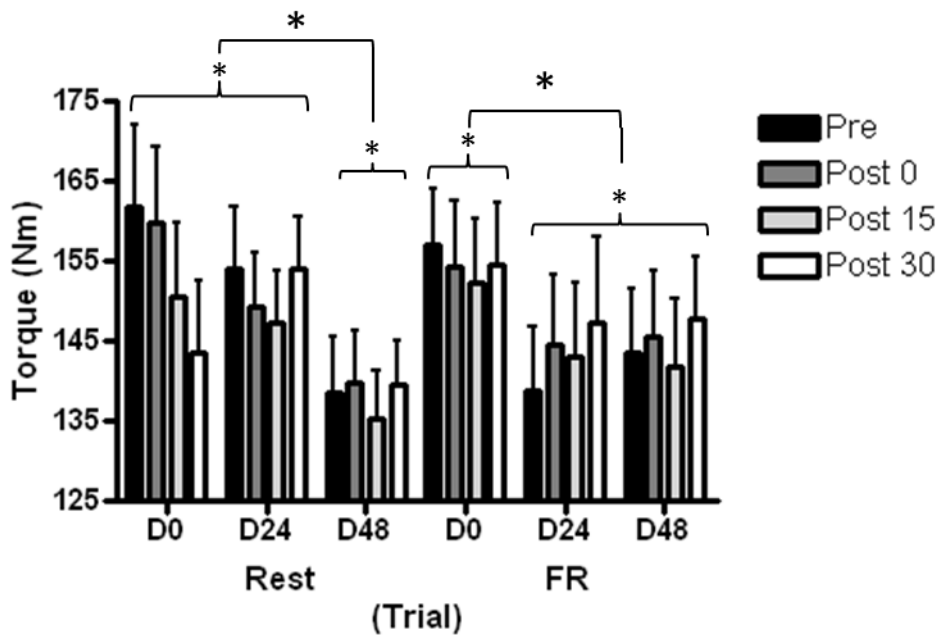


Figure 5.7 Maximal isokinetic torque ( $60^{\circ}\text{s}^{-1}$ ) of the knee extensors in the control (Rest) and intervention (FR) conditions. Values are mean  $\pm$  SD,  $n = 16$ . \* Significant within condition differences,  $p < 0.001$ .

#### 5.3.4 Contractile Properties

A significant interaction effect ( $F_{(2,15)} = 6.92$ ,  $p = 0.001$ ) was detected in peak displacement of VL with time and condition (Figure 5.8), caused by significantly greater displacement on D48 in the FR treatment condition compared to the Rest condition (CI [2.64 to 4.96] ES = 3.90,  $p = 0.001$ ). There was a non-significant trend in RF displacement ( $F_{(2,15)} = 2.79$ ,  $p = 0.063$ ) (Table 5.5). Velocity ( $V_c$ ) of VL ( $F_{(2,15)} = 0.33$ ,  $p = 0.801$ ) and RF ( $F_{(2,15)} = 2.44$ ,  $p = 0.089$ ) were not significantly different between conditions or over time (Table 5.6).

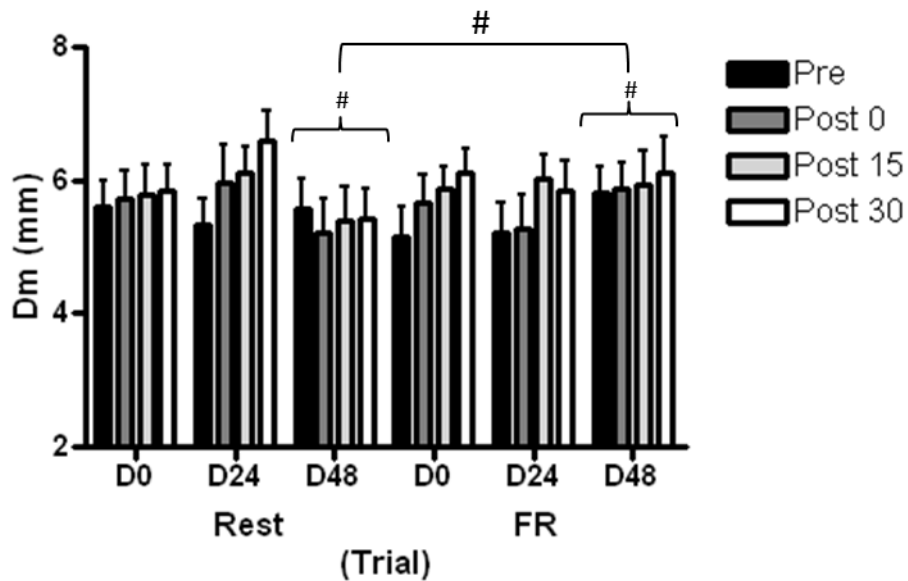


Figure 5.8 Maximal radial displacement (Dm) of vastus lateralis (VL) muscle belly. Values are mean  $\pm$  SD,  $n = 16$ . # Significant difference between conditions,  $p < 0.001$ .

### 5.3.5 Electromechanical Delay and Rate of Torque Development

PMT ( $F_{(3,15)} = 1.73$ ,  $p = 0.162$ ) and EMD ( $F_{(3,15)} = 0.33$ ,  $p = 0.807$ ) were not significantly different between conditions or over time (Table 5.7). RTD was not significantly different between conditions or over time during 50ms ( $F_{(3,15)} = 0.29$ ,  $p = 0.830$ ) or 300ms ( $F_{(3,15)} = 0.54$ ,  $p = 0.658$ ) following onset of MVC (Table 5.8).

Table 5.5 Maximal radial displacement of rectus femoris (RF) muscle belly. Values are mean  $\pm$  SD, n = 16.

	Day	D0				D24				D48			
		Pre-	0	15	30	Pre-	0	15	30	Pre-	0	15	30
Rest	Mean (mm)	7.6	7.3	7.1	7.4	8.6	7.4	7.5	7.2	6.9	6.9	7.0	7.2
	$\pm$ SD	3.6	3.7	3.3	3.5	3.0	2.8	2.8	3.0	3.0	2.9	3.2	3.4
FR	Mean (mm)	8.0	7.3	7.8	7.5	7.6	6.7	7.4	7.1	7.7	6.5	8.0	7.3
	$\pm$ SD	3.5	3.0	2.9	2.8	3.1	3.0	3.1	2.5	2.9	2.8	2.8	2.8

Table 5.6 Contraction velocity (Vc) of vastus lateralis (VL) and rectus femoris (RF) muscle. Values are mean  $\pm$  SD, n = 16.

		Day	D0				D24				D48			
		Time	Pre-	0	15	30	Pre-	0	15	30	Pre-	0	15	30
VL	Rest	Mean (mm·s <sup>-1</sup> )	175.4	185.0	193.0	190.6	164.2	192.4	203.3	229.0	169.4	165.2	175.7	184.7
		$\pm$ SD	54.6	51.2	59.4	49.5	56.6	71.9	51.6	76.3	60.6	55.7	66.6	58.3
	FR	Mean (mm·s <sup>-1</sup> )	152.4	177.9	194.9	199.7	180.4	167.7	201.5	196.3	180.9	188.3	201.0	206.9
		$\pm$ SD	61.7	55.4	40.2	46.7	90.6	62.6	49.2	54.9	48.3	53.5	63.3	63.9
RF	Rest	Mean (mm·s <sup>-1</sup> )	208.5	204.4	206.6	208.2	220.1	207.7	208.5	207.8	184.4	192.5	200.4	198.3
		$\pm$ SD	104.4	104.4	99.4	95.7	91.5	96.7	76.2	93.2	76.9	79.4	85.6	86.7
	FR	Mean (mm·s <sup>-1</sup> )	207.8	199.5	217.7	218.9	198.4	187.1	206.5	195.0	204.8	176.1	210.5	204.1
		$\pm$ SD	89.1	76.5	87.4	87.3	85.	93.4	87.7	85.3	83.2	82.8	78.2	86.4

Table 5.7 Pre motor time (PMT) and electromechanical delay (EMD) of vastus lateralis (VL) muscle during MVC. Values are mean  $\pm$  SD, n = 16.

		Day	D0				D24				D48			
		Time	Pre-	0	15	30	Pre-	0	15	30	Pre-	0	15	30
PMT	Rest	Mean (ms)	0.09	0.18	0.11	0.16	0.19	0.11	0.09	0.18	0.12	0.14	0.06	0.16
		$\pm$ SD	0.08	0.23	0.08	0.12	0.15	0.13	0.08	0.16	0.13	0.19	0.08	0.19
	FR	Mean (ms)	0.12	0.16	0.09	0.12	0.11	0.17	0.10	0.10	0.09	0.15	0.16	0.13
		$\pm$ SD	0.10	0.17	0.08	0.11	0.09	0.24	0.13	0.11	0.07	0.13	0.16	0.14
EMD	Rest	Mean (ms)	1.9	2.8	2.6	3.3	2.6	2.5	1.8	2.2	2.7	2.1	2.2	2.9
		$\pm$ SD	1.7	3.0	2.2	3.8	2.3	2.9	1.8	2.2	2.7	2.4	2.4	2.8
	FR	Mean (ms)	2.4	1.6	0.57	2.8	0.93	2.6	2.2	0.8	1.2	0.90	1.7	1.2
		$\pm$ SD	2.5	2.2	0.8	3.1	1.5	2.6	2.5	1.7	2.1	1.9	2.1	1.8

Table 5.8 Rate of torque development (RTD) during 50ms and 300ms of MVC. Values are mean  $\pm$  SD, n = 16.

Day		D0				D24				D48				
Time	Pre-	0	15	30	Pre-	0	15	30	Pre-	0	15	30		
0-50	Rest	Mean (Nm·s <sup>-1</sup> )	4047.4	4388.5	3841.7	4074.5	3870.6	3541.3	4022.4	4137.1	3835.5	3114.1	3735.7	3114.3
		$\pm$ SD	1785.6	2222.0	2119.7	2002.2	1783.6	1789.1	2035.8	1564.1	1952.8	1874.4	1783.9	1862.9
	FR	Mean (Nm·s <sup>-1</sup> )	3876.6	3375.6	4458.0	4369.4	3936.2	3764.7	3891.4	3716.6	3450.7	3394.8	3505.2	3811.0
		$\pm$ SD	1834.1	1749.1	2396.1	2075.9	1910.3	1695.9	1619.8	2049.6	1283.5	1734.1	1679.9	1880.1
0-300	Rest	Mean (Nm·s <sup>-1</sup> )	1427.0	1418.9	1195.6	1208.4	1172.1	1315.1	1324.0	1323.8	1249.0	997.6	1150.2	1162.9
		$\pm$ SD	445.4	550.0	457.9	491.2	306.9	465.2	387.7	425.9	469.2	424.2	310.4	353.5
	FR	Mean (Nm·s <sup>-1</sup> )	1243.7	1148.4	1305.5	1359.6	1066.9	1269.9	1231.7	1214.7	1097.1	1056.1	1118.4	1181.4
		$\pm$ SD	317.7	429.9	388.8	383.3	387.5	455.1	353.5	410.3	286.1	351.2	291.9	347.4



Table 5.9 Rate of perceived exertion (RPE) during 30s submaximal isometric contraction, in the control (Rest) and intervention (FR) conditions (6-20 scale (Borg, 1982)).  
Values are mean  $\pm$  SD, n = 6.

	Day	D0				D24				D48			
		Time	Pre-	0	15	30	Pre-	0	15	30	Pre-	0	15
Rest	Mean	13.8	14.7	15.3	15.7	14.5	14.5	15.3	15.5	15.3	14.8	15.3	15.3
	$\pm$ SD	1.3	1.9	1.8	2.1	1.5	1.4	1.9	1.6	1.5	1.6	2.2	1.8
FR	Mean	13.2	14.0	14.0	14.0	13.7	13.8	14.7	14.2	13.3	13.7	14.2	14.3
	$\pm$ SD	1.2	1.1	0.9	1.1	1.0	1.5	1.4	0.9	1.4	1.2	0.9	1.2

## 5.4 Discussion

This study demonstrated reduced RMS, during sustained submaximal contraction, following FR when compared to increased RMS following rest. Surprisingly, ROM remained unchanged over time and did not differ between conditions. On the other hand, MVC was maintained with FR, avoiding decreases observed throughout the rest condition. The contractile characteristics of RF were unaffected, as shown by unaltered TMG, however VL displayed reduced stiffness on the third day following FR. Dynamic strength was lower on the second and third day of FR compared to the first. It had been hypothesized that elevated  $D_m$  (i.e. reduced stiffness) would be indicative of altered muscle architecture and thus be associated with impaired MVC and/ or augmented ROM; however these change in function were not observed.

Perhaps the greatest insight into the mechanisms behind the effects of FR has been provided by investigating neuromuscular recruitment during static fatiguing contractions; RMS was lower at all time points following FR compared to the same time points in the control condition. Within the control condition RMS increased from baseline up to 15 and 30 minutes post-rest, indicating that the muscle required greater neural drive in order to maintain 50% of MVC (Dideriksen et al., 2011), which suggests submaximal fatigue occurred (Moritani, Muro, & Nagata, 1986). Increased neural drive is indicated by increased RMS which occurs as higher threshold motor units (MU) are recruited to compensate for earlier recruited fatigued MUs (Adam & De Luca, 2005; Dias da Silva & Gonçalves, 2006), to sustain force production. This finding suggests that FR was able to reduce the impact of fatigue during this submaximal task. Furthermore, on the 3rd day RMS of the control condition was higher than the previous 2 days in addition to being elevated

vs. FR condition; indicating chronic adaptation to repeated bouts of FR on consecutive days. Despite the observed differences in fatigue between conditions, a sub-group of participants ( $n = 6$ ) reported no change in RPE during the submaximal task, suggesting there was no perceived facilitation of submaximal force maintenance associated with the FR treatment. It would perhaps be interesting to repeat such subjective measures with a larger sample. Future research should also aim to examine whether subjecting muscles to FR prior to submaximal exercise leads to greater task duration.

It seems plausible that sustained muscle lengthening under pressure, applied during FR, will stimulate muscle spindles and Golgi tendon organ (Bradbury-Squires et al., 2015; Behm et al., 2013). Stimulation of these mechanoreceptors would lead to increased activity of type Ib afferents, thereby leading to greater proprioceptive feedback from the muscle in question, to the CNS (Kay, Husbands-Beasley, & Blazevich, 2015). However, Bradbury-Squires et al., (2015) observed a change in EMG activity only during dynamic activity, with no difference observed during isometric contraction. Perhaps the reason for the difference in findings relates to the timings of post-treatment measurements or the nature of the FR protocol. Increased ROM can be facilitated by alterations in muscle-spindle length or stretch perception, as with proprioceptive neuromuscular facilitation (PNF) (Feland & Marin, 2004). As such, employing different rest periods between repeated bouts of FR (Bradbury-Squires et al., 2015) may result in disparate alterations in muscle compared to the neural adaptations observed in the present study, following a single continuous bout of FR.

Peak torque (MVC) was elevated 30 minutes after FR, which was also significantly higher than 30 minutes post-rest. The decline in MVC on the second and third days of the rest condition can be interpreted as another marker of the fatiguing effect of the test

protocol; it follows that lower neural drive required to complete the submaximal task following FR, compared to rest, indicates a lesser demand placed on the muscle and as such, has enabled maintenance of MVC following consecutive days of FR. Previous studies have reported no alteration in strength or performance (Healey, Hatfield, Blanpied, Dorfman, & Riebe, 2014; Halperin et al., 2014) or improved performance through combining FR with a dynamic warm up compared to a dynamic warm up alone (Peacock et al., 2014). The present study is the first to report an improvement in MVC following FR alone. Previous studies have measured strength or performance  $\leq 10$  minutes after FR, in accordance with this we found no change in MVC immediately and 15 minutes post-FR. It is not entirely clear why increased force production is delayed in this manner, following a bout of FR; however these findings suggest that to achieve optimal results, FR should be performed at least 30 minutes prior to exercise. Meanwhile, FR did not appear to have the same impact on dynamic strength. Indeed peak isokinetic torque ( $60^{\circ}\cdot\text{s}^{-1}$ ) was reduced on the second and third days in the foam rolling condition compared to the first day.

As FR has previously been reported to increase ROM without decrements in force production, one of the main aims of this study was to examine the contractile mechanics of muscles subjected to FR. As discussed in Chapter 2, TMG can accurately detect acute alterations in muscle stiffness. It was hypothesized that muscle displacement would have increased following FR, indicative of reduced muscle stiffness. Despite the lack of improvement in ROM, significantly higher displacement was observed in VL on the third day of FR treatment compared to the third day of the control condition. Furthermore, Dm was 15.7% greater than baseline, 15 minutes post-FR, and 19.0% greater than baseline 30 minutes post-FR on the third day (compared to 3.7% lower and 2.8% lower at 15 and 30

minutes post-rest). Although no differences were revealed in RF, there was a tendency towards greater displacement following FR. Given that displacement only differed between groups on the third day, it may be speculated that continuing with FR treatment for a more extended period of time may lead to more pronounced decreases in muscle stiffness. Indeed, Ebrahim & Abd Elghany, (2013), Mohr et al., (2014) and Bushell, Dawson, & Webster, (2015) have all presented improvements in ROM following FR protocols lasting between 1-3 weeks. It must be noted however, that in the present study, independent of day, VL displacement was greater 15 and 30 minutes post treatment, in both the control and FR conditions. Therefore, it seems that the test protocol itself may have impacted upon muscle stiffness. In order to fully elucidate the impact of FR on muscle stiffness, investigation of the impact of FR on muscle contractile properties in isolation from other measurements may be required, in order to avoid additional stretch and potentially fatigue.

It was hypothesized that flexibility would be improved following FR, in line with previous research (Bradbury-Squires et al., 2015; MacDonald et al., 2013). However, we observed no change in ROM following 1, 2 or 3 days of FR treatment. Studies utilizing repeated bouts of FR have reported positive changes to ROM (MacDonald et al., 2013; Sullivan et al., 2013; Bradbury-Squires et al., 2015; Škarabot et al., 2015), whereas those applying a single bout, as adopted in the present study, remain unchanged (Mikesky, Bahamonde, Stanton, Alvey, & Fitton, 2002; Roylance et al., 2013; Peacock et al., 2014). Interestingly, total time spent undergoing FR is fairly consistent across all studies mentioned, therefore dividing treatment bout into a series of repetitions separated by brief rest periods may result in increased ROM. Furthermore, prolonged periods of treatment, lasting perhaps  $\geq 1$  week, may prove more effective with regards to flexibility. Further

research is required to explore this element and to investigate the optimal duration and distribution of FR treatment.

One potential mechanism, which was not controlled for, was muscle temperature in response to FR. It has not been established whether the friction generated during FR (Barnes, 1997) is sufficient to effectively elevate muscle temperature; indeed, surface temperature also has not been assessed following FR. However, Okamoto et al (2014) demonstrated increased arterial blood flow associated with FR. It has long been established that there exists a positive relationship between peripheral temperature and blood flow (Barcroft & Edholm, 1943), so it is certainly plausible that muscle temperature could be elevated following FR. Petrosfsky & Laymon (2005) reported increased muscle fibre conduction velocity, with increased temperature, which may permit reduced MU firing rate (Fuglsang-Frederiksen & Rønager, 1988), resulting in reduced RMS. Future research should incorporate measurements of muscle temperature, when investigating FR.

## **5.5 Conclusion**

This is the first study to illustrate elevated strength 30 minutes after a 2 minute bout of FR, as well as improved muscular efficiency during submaximal activity following FR, which was likely a result of increased proprioceptive afferent activity. This increased efficiency protected the muscles from the fatiguing effects of the protocol, observed through the control condition. Muscle stiffness was reduced after 3 consecutive days of FR, however this did not translate into improved ROM. It seems that the structure of FR protocol adopted is important, with multiple bouts separated by brief rest periods potentially leading to greater flexibility adaptations, while a single constant bout may lead to alterations in neural drive. Such alterations potentially enhance strength and

performance, and delay the onset of fatigue. Performance adaptations appear to peak at least 30 minutes after FR treatment. This time course should be taken into account when incorporating FR routines into performance sport programmes.

# Chapter 6

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## Skeletal Muscle Contractile Mechanics of High Performance Athletes

Lewis J Macgregor, Neil Donald, Angus M Hunter



## 6.1 Introduction

In Chapter 2 we discussed the validity of radial displacement measures in assessing acute alterations in skeletal muscle. Fatigue-associated elevations in resting muscle tension were detected by decreased  $D_m$  in gastrocnemius medialis. As such, it was concluded that TMG is well suited for incorporation into high performance sport monitoring, to provide portable and non-invasive assessment of peripheral neuromuscular function. Subsequently, in Chapter 3 we quantified bilateral asymmetry in muscles of the knee extensor group, revealing no apparent relationship between lower-limb power, static and dynamic strength, and either spatial ( $D_m$ ) or temporal ( $V_c$ ) characteristics of the involved muscles. This absence of relationship is attributed to a masking of imbalanced muscle function, by specific load-sharing recruitment patterns, during complex actions. As such, individuals who displayed strength asymmetry also presented greater occurrences of bilateral asymmetry among concerned muscles, however such asymmetry was not detected during whole-limb, ballistic tasks. We proposed that screening of individual muscles contractile mechanics may provide greater insight into strength imbalances and inform targeted conditioning interventions. We also demonstrated, in Chapter 5, the sensitivity of TMG to detect acute changes in muscle stiffness, following a controlled intervention (foam rolling). In this chapter a series of case-studies will be presented, involving use of TMG incorporated into high performance training programmes, with elite athletes. These studies focused on evaluation of asymmetry in relation to injury and to training, the challenge of incorporating reliable assessment into training cycles, and responses to preparation for competition. The overall aim of the chapter is to explore the practical application of muscle contractile assessment within high performance sports programmes.

## 6.2 Case-study 1

### *Radial Displacement of Knee Extensors and Flexors, Before and After Anterior Cruciate Ligament Surgery*

Anterior cruciate ligament (ACL) injuries are widespread within team sports (Erickson et al., 2014; Sikka, Kurtenbach, Steubs, Boyd, & Nelson, 2015). ACL injuries occur in both contact and non-contact situations, with the majority (~70%) sustained through non-contact (Griffin et al., 2000); it has also been reported that ACL injuries are more prevalent in female athletes than male (Agel, 2005; Stanley, Kerr, Dompier, & Padua, 2016), which may be linked to reduced muscle stiffness in females (Wang et al., 2015). Neuromuscular risk factors for ACL injury in athletes have been reviewed, it has been suggested that dynamic knee joint stability is one of the key factors in ACL injury risk (Griffin, 2006; Posthumus, Collins, September, & Schwellnus, 2011; Serpell et al., 2015). Imbalances between contralateral muscle groups (i.e. bilateral asymmetry), or within synergist groups, may well contribute to joint instability (Jordan et al., 2015; Kuenze et al., 2015). This case-study examined contractile mechanics of knee extensor and knee flexor muscles of a female international hockey player over a 21 week period (prior to, and following ACL surgery).

#### **6.2.1 Methods**

The athlete's physical characteristics are presented in Table 6.1. Muscle contractile mechanics were assessed through recording radial deformation of muscle belly, using TMG (BMC Ltd, Ljubljana). Measurements were captured from vastus lateralis (VL), vastus medialis (VM), rectus femoris (RF) and bicep femoris (BF) on the injured and uninjured limb. Peak  $D_m$  and  $V_c$  were calculated from twitch measurements for each muscle.

Measurements were performed, and data analysed using the protocols described in Chapter 2. Measurements were recorded on 3 occasions: measurement 1 (T1), 1 week prior to ACL surgery; measurement 2 (T2), 6 weeks after surgery; measurement 3 (T3), 20 weeks after surgery. CoV (%) of knee extensor Dm of each limb was calculated, for T1, T2 and T3, to provide relative standard deviation (rSD) of the muscle group. Ratio of VL to VM, and knee extensors (as the mean of VL, RF, VM) to knee flexors (BF) was calculated using S.I. equation, as detailed in Chapter 3.

**Table 6.1 Baseline physical characteristics for the athlete.**

<b>Athlete characteristics</b>	
<b>Age (y)</b>	17
<b>Height (cm)</b>	146.0
<b>Body mass (kg)</b>	62.0
<b>BMI (kg/m<sup>2</sup>)</b>	23.1

### **6.2.2 Results**

The injured limb showed sizeable alterations in Dm from T1 to T2 in both VL (-47.7%) and VM (+85.3%), and from T1 to T3 in VL (+70.5%) and VM (+114.7%). RF showed no change from T1 to T2 and negligible change (+2.7%) from T1 to T3 (Figure 6.1A). Dm of the uninjured limb showed changes from T1 to T2 in VL (+41.2%), RF (+40.9%) and VM (-55.4%), and from T1 to T3 in VL (+17.7%), RF (+19.7%) and VM (+16.1%) (Figure 6.1B). Dm and Vc of knee extensors and knee flexors of each limb at T1 are shown in Table 6.2. Vc of the injured limb showed alterations from T1 to T2 (Table 6.3) in VL (-29.1%), RF (-14.9%) and VM (-33.2%), and from T1 to T3 (Table 6.4) in VL (+76.8%), with negligible changes in

RF (- 1.0%) and VM (- 3.1%). The uninjured limb showed alterations in Vc from T1 to T2 (Table 6.3) for VL (+ 28.0%), RF (+ 65.1%) and VM (- 41.6%), and from T1 to T3 (Table 6.4) for RF (+ 30.5%), with negligible changes in VL (+ 3.2%) and VM (- 3.4%). The rSD of knee extensor Dm in injured and uninjured limbs, at each time point, are presented in Table 6.5A. The injured limb showed greater rSD than the uninjured limb at T1 (65.9 vs 28.5%), but rSD of each limb was similar at T2 (66.7 vs 58.1%) and T3 (25.9 vs 29.0%). The ratio of VL : VM Dm in the injured limb, at each time point, is shown in Table 6.5B. VL : VM ratio improved to - 2.7% at T3, compared to 25.6% (T1) and -93.0% (T2). Knee extensor : knee flexor ratio improved to 5.9% at T3, compared to 16.6% (T1) and 40.0% (T2).

**Table 6.2 Radial (Dm) and temporal (Vc) contractile properties of knee extensors and flexors in injured and uninjured limbs 1 week pre surgery (T1).**

		<b>Muscle</b>	<b>Dm</b>	<b>Vc</b>
<b>Injured</b>	<b>Knee Extensors</b>	<b>VL</b>	4.4	124.8
		<b>RF</b>	11.0	237.2
		<b>VM</b>	3.4	132.7
	<b>Knee Flexors</b>	<b>BF</b>	7.4	118.9
<b>Uninjured</b>	<b>Knee Extensors</b>	<b>VL</b>	3.4	127.1
		<b>RF</b>	6.1	127.1
		<b>VM</b>	5.6	188.2
	<b>Knee Flexors</b>	<b>BF</b>	7.0	124.2

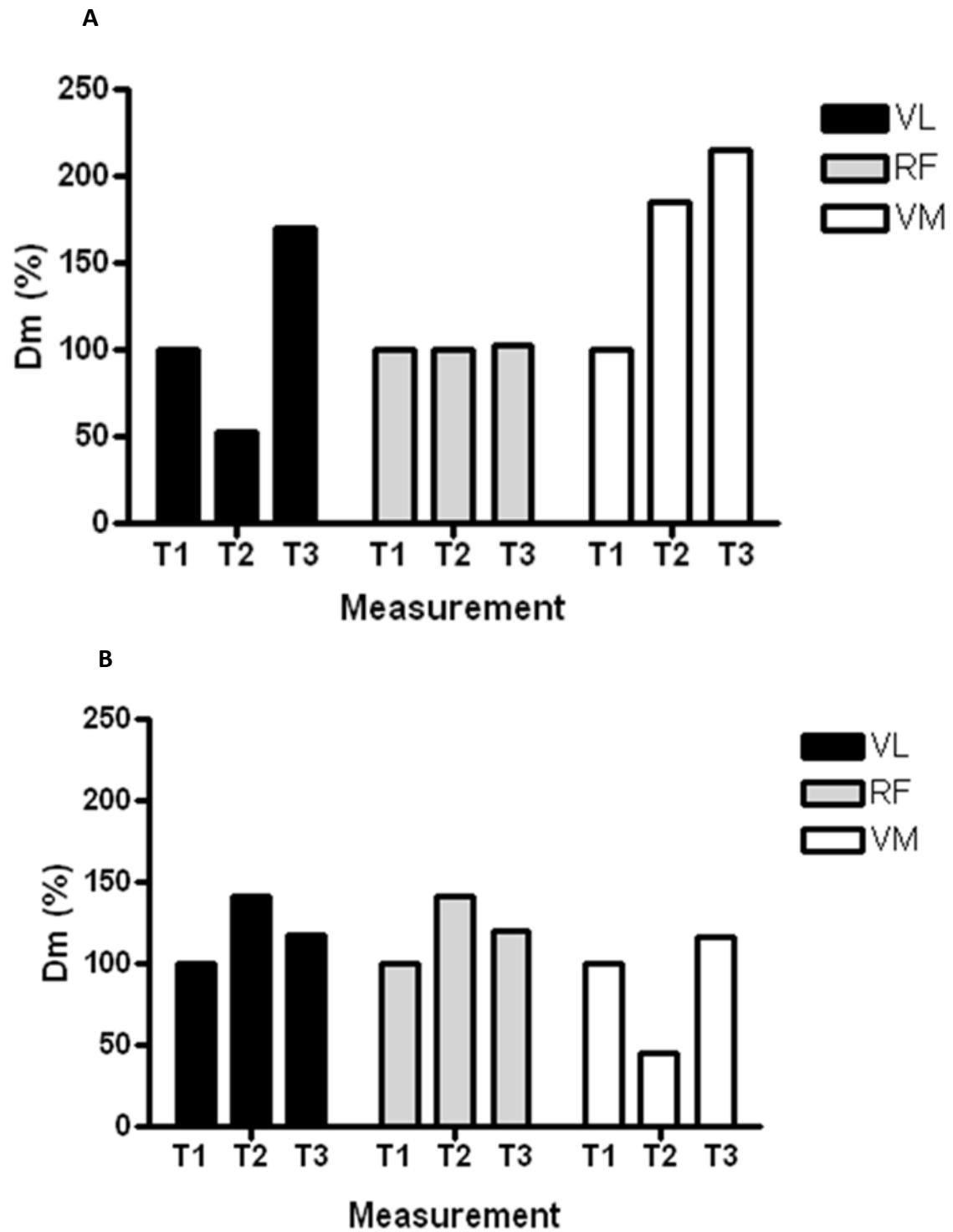


Figure 6.1 Change in Dm of VL, RF and VM relative to 1 week prior to surgery (T1), 6 weeks post surgery (T2) and 20 weeks post surgery (T3) in injured (A) and uninjured (B) limbs.

**Table 6.3 Radial (Dm) and temporal (Vc) contractile properties of knee extensors and flexors in injured and uninjured limbs 6 weeks post surgery (T2).**

		<b>Muscle</b>	<b>Dm</b>	<b>Vc</b>
<b>Injured</b>	<b>Knee Extensors</b>	<b>VL</b>	2.3	88.5
		<b>RF</b>	11.0	201.8
		<b>VM</b>	6.3	88.6
	<b>Knee Flexors</b>	<b>BF</b>	9.8	332.2
<b>Uninjured</b>	<b>Knee Extensors</b>	<b>VL</b>	4.8	162.7
		<b>RF</b>	8.6	209.8
		<b>VM</b>	2.5	109.9
	<b>Knee Flexors</b>	<b>BF</b>	6.0	77.1

**Table 6.4 Radial (Dm) and temporal (Vc) contractile properties of knee extensors and flexors in injured and uninjured limbs 20 weeks post surgery (T3).**

		<b>Muscle</b>	<b>Dm</b>	<b>Vc</b>
<b>Injured</b>	<b>Knee Extensors</b>	<b>VL</b>	7.5	220.6
		<b>RF</b>	11.3	234.8
		<b>VM</b>	7.3	128.6
	<b>Knee Flexors</b>	<b>BF</b>	8.2	108.1
<b>Uninjured</b>	<b>Knee Extensors</b>	<b>VL</b>	4.0	131.2
		<b>RF</b>	7.3	165.9
		<b>VM</b>	6.5	181.8
	<b>Knee Flexors</b>	<b>BF</b>	8.9	112.5

**Table 6.5 A) Relative standard deviation (CoV %) of knee extensor Dm in injured and uninjured limb, pre (T1), 6 weeks post (T2) and 20 weeks post (T3) surgery. B) Ratio (S.I. %) of VL to VM in injured limb at T1, T2 and T3**

A	CoV (%)		B	VL : VM S.I. (%)	
	Injured	Uninjured			
T1	65.9	28.5	T1	25.6	
T2	66.7	58.1	T2	-93.0	
T3	25.9	29.0	T3	-2.7	

### 6.2.3 Discussion

This case-study demonstrates the transient alterations in skeletal muscle contractile mechanics, in a female hockey player, following ACL reconstruction surgery. Dm of VL and VM were markedly increased 20 weeks following surgery, Vc of VL also was elevated. Six weeks after surgery, VL Dm and Vc were reduced, while VM Dm increased, but Vc decreased. RF remained relatively stable for Dm throughout, although there was a slight reduction in Vc 6 weeks post-surgery. It should be noted that 6 weeks post-surgery, elevations in Dm were evident in VL and RF of the uninjured limb, and diminished Dm was observed in VM. Contractile mechanics of the knee extensors displayed greater balance after 20 weeks, compared to pre-surgery, as indicated by reduced rSD. Additionally, symmetry of the vasti was improved in the injured limb, and knee extensor to knee flexor ratio was more closely balanced, 20 weeks post-surgery.

This is the first investigation, utilising TMG, to compare muscle contractile properties before ACL reconstruction surgery and during the acute rehabilitation period. (Alvarez-Diaz

et al., 2014) previously reported a variety of changes in temporal parameters of muscle twitch from pre-surgery to 12 months post-surgery; with changes in Dm observed only in BF. In accordance with the current findings, knee extensor : knee flexor ratio was improved, however, they reported no change in VL : VM ratio. Interestingly, (Alvarez-Diaz et al., 2014) reported alterations in contractile mechanics of the uninjured limb, as well as the injured, 12 months post-surgery; while we observed changes in Dm and Vc 6 weeks post-surgery, these alterations had mostly returned to pre-surgery levels following 20 weeks of rehabilitation.

Contractile mechanics of the knee flexors and knee extensors may be key determinants of ACL injury (Alentorn-Geli, Alvarez-Diaz, Ramon, Marin, Steinbacher, Boffa, et al., 2014); as such this case-study has provided valuable insight into expected alterations in temporal and spatial parameters of individual muscles, following surgery. It is important to note, when dealing with rehabilitation, that the uninjured leg is likely to display altered mechanics also, whether through potential contralateral adaptation (Pairot de Fontenay, Argaud, Blache, & Monteil, 2014) or through compensatory mechanisms (Colné & Thoumie, 2006). In this case-study we observed improvements in the balance of contractile mechanics across the knee extensors, and specifically between the vasti, as well as improved balance between knee joint agonist-antagonist groups, following ACL surgery. These findings, combined with the findings reported in Chapter 3, point towards the applicability of TMG measurements as a screening tool, to identify individuals at heightened risk of injury, as well as an objective monitor of post-injury recovery. Potentially, non-invasive TMG could inform training practices among players recovering from injury.



Future research should explore the efficacy of targeted interventions, designed to combat injury risk, by tracking changes in contractile mechanics over time. The full time-course of recovery of contractile mechanics, following ACL surgery, also remains to be elucidated; we have demonstrated impaired mechanics 6 weeks post-surgery with improvements apparent following 20 weeks. Finally, as this is a single-athlete case-study, the findings necessitate replication in order to support our conclusions.

## **6.4 Case-study 2**

### ***Intra-individual Reliability within an Elite Athlete Training Cycle***

In Chapter 3 it was observed that greater variability in TMG parameters was encountered when assessing elite athletes. It was suggested that such elevated variability arises from individualised responses to high training loads and volumes performed on a daily basis. As such, the next case-study will describe an assessment of intra-individual variability, when dealing with high performance athletes. As demonstrated in Chapter 2, TMG provides a valid measure of muscle twitch, which can be applied to detect changes in muscle status, as may be encountered during fatigue, for example. However, understanding the day-to-day repeatability of TMG measurements is crucial for appropriate interpretation and application. Reliability of TMG under various conditions has been discussed in Chapter 1; however, to date reliability has not been assessed in an elite athlete population. As discussed previously, greater CoV may be expected in highly trained individuals, likely due to heavy training loads and periodization of training cycles.

Ditroilo et al., (2013) provided evidence that greater reliability is achieved among measurements performed on exercised, or indeed fatigued muscle, compared to resting

state; however, in that study, participants performed a standardized warm up, followed by isometric MVC and an electrically stimulated fatigue protocol. In practice exercise protocols lack such levels of control. Furthermore, contractile responses to fatigue have been shown to vary, depending on the mode, or perhaps more specifically the duration, of exercise, with longer, endurance-based activity leading to decreases in muscle tone (García-Manso et al., 2011; Giovanelli et al., 2016), while higher intensity, shorter duration activities have resulted in increased muscle stiffness (García-Manso et al., 2012; Wiewelhove et al., 2015). As high performance athletes, in sports such as swimming (Aspenes, Kjendlie, Hoff, & Helgerud, 2009), incorporate large volumes of strength and endurance training within their weekly programmes, day to day variability will likely exceed levels described previously (Križaj et al., 2008; Rodríguez-Matoso et al., 2010; Šimunič, 2012; Tous-Fajardo et al., 2010). Demanding training volumes may interfere with the ability to capture meaningful measures. As such, this case-study sought to assess intra-individual reliability within the specific challenges posed by performing measures among an elite athlete population.

#### **6.4.1 Methods**

Muscle contractile parameters (Dm and Vc) were measured, as described previously, from VL, GM, BF and LD of 4 athletes from within the cohort described in Chapter 3 (Table 3.8). All measurements were performed on the muscles of the right flank. Dm and Vc were recorded immediately before and immediately after training sessions for 3 consecutive days. Within each 3 day measurement period, athletes completed 4.5 ( $\pm$  0.6) training sessions. All environmental conditions were maintained constant throughout the 3 days of assessment; TMG sensor and electrode positions (as described in Chapter 3) were marked with semi-permanent ink, to ensure accurate repositioning for each measurement.

### 6.4.2 Results

Mean Dm and Vc of each muscle, across the 3 day period are presented in Table 6.6, for each individual athlete (A - D). An example of measurement parameters extracted from TMG recordings is presented in Figure 6.4; data were captured from VL of Athlete D. Intra-individual variability (CoV) of Dm ranged from 7.9% to 42.0%, and of Vc from 8.6% to 31.8% (Table 6.6). Mean CoV of VL was 15.4% (Dm) and 18.1% (Vc), GM was 13.8% (Dm) and 12.5% (Vc), BF was 24.7% (Dm) and 20.4% (Vc), and LD was 20.2% (Dm) and 17.9% (Vc). Measurement CoV separated by time of day, and exercised or rested status, are presented in Table 6.7.

Table 6.6 Individual athlete mean and coefficient of variation of Dm and Vc for VL, GM, BF and LD.

		Dm (mm)				Vc (mm·s <sup>-1</sup> )			
		VL	GM	BF	LD	VL	GM	BF	LD
A	Mean	6.2	1.4	1.6	11.3	172.5	65.9	76.9	260.4
	CoV (%)	20.3	12.4	20.6	11.5	21.2	8.6	14.5	11.9
B	Mean	2.4	2.2	2.5	8.1	64.3	83.6	52.6	149.1
	CoV (%)	17.3	12.7	20.9	16.9	16.1	13.5	24.4	20.5
C	Mean	5.8	4.2	3.5	13.2	201.8	131.7	84.9	289.9
	CoV (%)	15.9	18.0	42.0	15.3	24.8	18.2	31.8	9.2
D	Mean	5.9	1.2	3.4	8.4	203.8	58.3	145.4	192.0
	CoV (%)	7.9	12.1	15.2	37.0	10.2	9.7	10.8	29.9

Table 6.7 Individual athlete coefficient of variation of Dm and Vc for VL, GM, BF and LD, separated by time point (AM vs PM) and rested (Pre) or exercised (Post) state.

			Dm (mm)				Vc (mm·s <sup>-1</sup> )				
			VL	GM	BF	LD	VL	GM	BF	LD	
A	CoV (%)	AM	Pre	15.9	14.5	19.2	11.7	15.9	8.3	11.9	4.5
			Post	26.7	10.0	14.6	4.5	25.3	7.9	11.8	6.2
		PM	Pre	6.7	7.6	19.7	1.6	9.8	6.9	11.0	6.2
			Post	31.1	2.7	16.9	6.3	35.5	0.9	1.1	13.9
B	CoV (%)	AM	Pre	8.2	1.9	29.5	19.5	20.6	2.5	28.8	29.3
			Post	28.3	8.3	12.3	11.7	21.1	3.2	9.9	8.8
		PM	Pre	7.4	16.4	29.5	9.3	10.3	27.2	40.1	14.6
			Post	24.2	14.6	8.1	0.1	23.6	14.4	7.7	7.6
C	CoV (%)	AM	Pre	32.3	9.1	3.8	0.1	36.1	2.8	43.1	0.3
			Post	13.5	8.7	16.1	13.1	33.1	12.8	33.5	14.4
		PM	Pre	13.2	28.4	70.6	15.4	22.1	21.3	26.3	5.3
			Post	17.2	21.3	58.2	8.6	25.0	33.3	29.1	17.0
D	CoV (%)	AM	Pre	7.9	3.4	18.5	30.2	3.7	3.7	6.1	4.2
			Post	3.3	3.3	14.9	36.0	4.5	4.9	3.2	12.3
		PM	Pre	9.2	15.4	17.2	11.2	10.4	4.7	6.1	6.7
			Post	9.7	15.6	22.5	2.9	0.9	7.6	8.7	4.4

Athlete mean CoV for VL was lowest in PM measurements, pre-exercise (9.1% and 13.15% for Dm and Vc respectively). For GM, CoV was lowest in AM measurements, pre-exercise (7.2% and 4.3% for Dm and Vc respectively). In BF, CoV of Dm was lowest in AM measurements, post-exercise (14.5%) and for Vc in PM measurements, post-exercise (11.7%). The CoV for LD was lowest, for Dm, in PM measurements, post-exercise (4.5%) and for Vc in PM measurements, pre-exercise (8.2%).

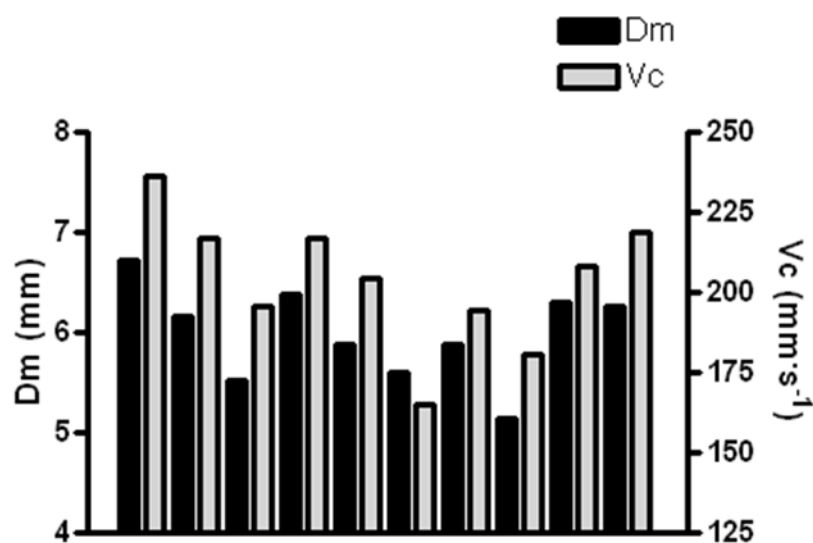


Figure 6.4 Example TMG parameters captured from VL of a single athlete (D) across 3 consecutive days.

### 6.4.3 Discussion

This case-study sought to examine the disconnect between established reliability ratings of TMG, in controlled laboratory settings, and measurements applied in the field of high performance sport. Coefficient of variation ranged from 7.9 - 42.0% and 8.6 - 31.8% for Dm and Vc respectively. Of the 4 muscles examined, each returned greatest reliability (i.e. lowest CoV) at a different time point, relative to training sessions and time of day.

Comparing means across all muscles, there are negligible differences between measurements performed in the morning (Dm: 14.1%, Vc: 13.6%) or afternoon (Dm: 16.8%, Vc: 14.4%). Similarly, rested state (before training) measurements resulted in comparable variability (Dm: 15.7%, Vc: 16.8%) to exercised (post-training) measures (Dm: 15.2%, Vc: 13.8%).

Across the athletes assessed, Dm of VL ( $15.4 \pm 5.3\%$ ) and of GM ( $13.8 \pm 2.8\%$ ) showed the lowest mean CoV, which also were comparable to CoV reported for GM (8.0 – 14.8%) previously (Ditroilo et al., 2013). Both VL and GM displayed their lowest variability in rested state (pre-training); although this observation is in contrast to Ditroilo et al., (2013), the more erratic nature of training performed throughout the current testing is distinct from the controlled exercise and fatigue interventions employed in previous reliability investigations. As such the demands placed upon each muscle group likely varied between training sessions, leading to unpredictable levels of peripheral fatigue. Indeed, it has been demonstrated that muscle activation patterns vary between individuals, even among elite athletes, executing the same stroke (Guignard et al., 2015). BF has previously demonstrated greater variability (19.9 – 43.1%) compared to other muscles that have been investigated (Ditroilo, Hunter, Haslam, & De Vito, 2011), a similar observation was noted in this case-study ( $24.7 \pm 11.8\%$ ); additionally, LD presently displayed high variability ( $20.2 \pm 11.4\%$ ). It may be suggested that during training, BF and LD were more active, compared to other muscles measured, resulting in greater fluctuations in contractile status throughout the testing period. This suggestion is perhaps supported by observations of greatest reliability in BF and LD in exercised state (post-training); taken together with evidence from Ditroilo et

al., (2013), it seems plausible that these muscles have been exposed to more consistent training loads than VL and GM across the testing period.

While Vc of both VL and GM exhibited strongest reliability at the same time point as Dm (pre-training PM and pre-training AM, respectively), there were discrepancies over the most reliable measurement point, between parameters, for BF and LD. This observation may provide further evidence as to the greater demand placed on these particular muscles during swimming training, compared to VL and GM; a disconnect between spatial and temporal responses of muscle contraction may be symptomatic of fatigue. It is also noteworthy that Vc has demonstrated heightened variability (VL =  $18.1 \pm 6.4\%$ , GM =  $12.5 \pm 4.3\%$ , BF =  $20.4 \pm 9.5\%$ , LD =  $17.9 \pm 9.4\%$ ) compared to previously reported measurements of contraction time (Tc), in GM (3.8 – 9.4%) although not in BF (16.5 – 33.3%). Taken together, these findings may support the proposal presented in Chapter 2, that Vc, rather than Tc, is a more appropriate temporal measure of skeletal muscle contractile properties.

This case-study has demonstrated the need for care when assessing individual muscle contractile mechanics in highly trained athletes, with regard to specific muscles chosen for assessment and the timing of measurements in relation to training. Muscles which are subjected to high, but potentially consistent, demands may be most reliably assessed following exercise, while those muscles which are more sporadically active would be best assessed in a rested state. Timing of assessments may be critical in optimising reliability of the measurement, and thus increasing the potential sensitivity to changes in muscle contractile properties.



### 6.5 Case-study 3

#### ***Response to Pre-competition Taper in Swimming***

The final case-study aimed to investigate acute alterations in muscle contractile mechanics, which may have performance related benefit. To promote optimal performance, athletes may utilise periods of reduced training volume, known as tapering, prior to competition (Hellard, Avalos, Hausswirth, & Pyne, 2013). Gradual reduction in training volume, for 7 - 21 days, is seen to result in 2 - 4% improvements in performance (Costill et al., 1985; Johns et al., 1992; Luden et al., 2010). Reported elevations in muscle glycogen and oxidative enzymes augment endurance capacity (Neary, Martin, Reid, Burnham, & Quinney, 1992; Shepley et al., 1992). Swimming power has also been noted to improve by ~25% following a period of tapering (Costill et al., 1985). Single muscle fibres display increased isometric peak force and contraction velocity following taper; while muscle fibre diameter, specifically of type II fibres, has been shown to increase by ~11% (Trappe, Costill, & Thomas, 2000; Neary, Martin, & Quinney, 2003). Reduction in training stress is thought to be key to inducing these adaptations (Mujika, Padilla, Pyne, & Busso, 2004), as marked by decreased levels of plasma creatine kinase (CK). As such, hypertrophic and contractile adaptations in single muscle fibres are thought to drive observed improvements in muscle function (Mujika et al., 2004). Given the findings previously reported from *in vitro* studies, it was hypothesised that following a period of taper, characterised by reduced training volume with maintained intensity,  $D_m$  would decrease, as expected with greater muscle fibre thickness (Pišot et al., 2008), whilst  $V_c$  would increase,  $s$  has been observed in single fibres.

### 6.5.1 Methods

Contractile properties (Dm and Vc) of 5 elite level swimmers, from within the cohort outlined in Table 3.8, were recorded from gastrocnemius medialis (GM), bicep femoris (BF) and latissimus dorsi (LD). All measurements were performed on the muscles on the right flank. Measurements were recorded on consecutive days, in the weeks prior to National Summer Championships. Athletes reported to the laboratory at the same time each day, following completion of a training session; in the previous case-study it was demonstrated that BF and LD show greater reliability in exercised state. Although, in the previous case-study, GM displayed improved reliability rested, measurements were all performed at the same time, to reduce the number of laboratory visits required by athletes, Ditroilo et al., (2013) previously demonstrated improved reliability of GM in exercised state.

TMG sensor and electrode positions (as described in Chapter 2) were marked with semi-permanent ink, to ensure accurate repositioning on each day. Measurements were recorded for  $8.6 (\pm 3.9)$  days prior to commencing taper, and for  $8.2 (\pm 3.8)$  days between the start of taper and competition. On alternate days, capillary blood samples were collected, for analysis of CK, and counter-movement jump (CMJ) height was assessed, as described in Chapter 3, to assess changes in lower-limb power. Training load (km) and session RPE, on a scale of 1-10 (Foster et al., 2001), were recorded for all training sessions. Mean Dm and Vc of each muscle, and mean CMJ were calculated prior to, and during taper. Data were tested to ensure normal distribution, and contractile parameters and CK concentration were analysed using paired T-tests to compare pre-taper with during taper.

### 6.5.2 Results

During taper, training workload decreased by ~25% with intensity maintained, as indicated by negligible difference in session RPE (Table 6.8). CK concentration was reduced ( $p = 0.001$ , CI [-191.4 to -57.6]) during taper. Dm was significantly reduced ( $p = 0.001$ ) during taper in GM (21.5%, CI [-0.706 to -0.198]) and ( $p < 0.001$ ) in BF (39.5%, CI [-2.341 to -1.153]), but was not altered ( $p = 0.498$ ) in LD (Figure 6.5A). Vc was significantly reduced ( $p = 0.002$ ) during taper in GM (17.0%, CI [-28.22 to -6.58]) and ( $p < 0.001$ ) in BF (36.6%, CI [-80.89 to -44.89]), but was not altered ( $p = 0.665$ ) in LD (Figure 6.5B). CMJ did not improve during taper (Table 6.9).

Table 6.8 Training workload and RPE prior to, and during taper. Values are mean  $\pm$  SD,  $n = 5$ .

	Pre-taper	Taper
<b>Workload (km)</b>	7033.3 ( $\pm$ 2786.9)	5250.0 ( $\pm$ 2349.4)
<b>RPE</b>	4.8 ( $\pm$ 1.3)	4.1 ( $\pm$ 1.9)
<b>CK (IU/L)</b>	319.0 ( $\pm$ 139.0)	187.0 ( $\pm$ 61.0)

Table 6.9 Individual athlete counter-movement jump height prior to, and during taper. Values are mean  $\pm$  SD,  $n = 5$ .

	CMJ (cm)	
	Pre-taper	Taper
<b>A</b>	55.7 ( $\pm$ 1.9)	54.0 ( $\pm$ 0.1)
<b>B</b>	54.0 ( $\pm$ 1.6)	57.0 ( $\pm$ 1.0)
<b>C</b>	48.3 ( $\pm$ 1.4)	48.0 ( $\pm$ 1.4)
<b>D</b>	49.8 ( $\pm$ 0.9)	51.3 ( $\pm$ 1.7)
<b>E</b>	55.0 ( $\pm$ 2.5)	55.5 ( $\pm$ 2.1)

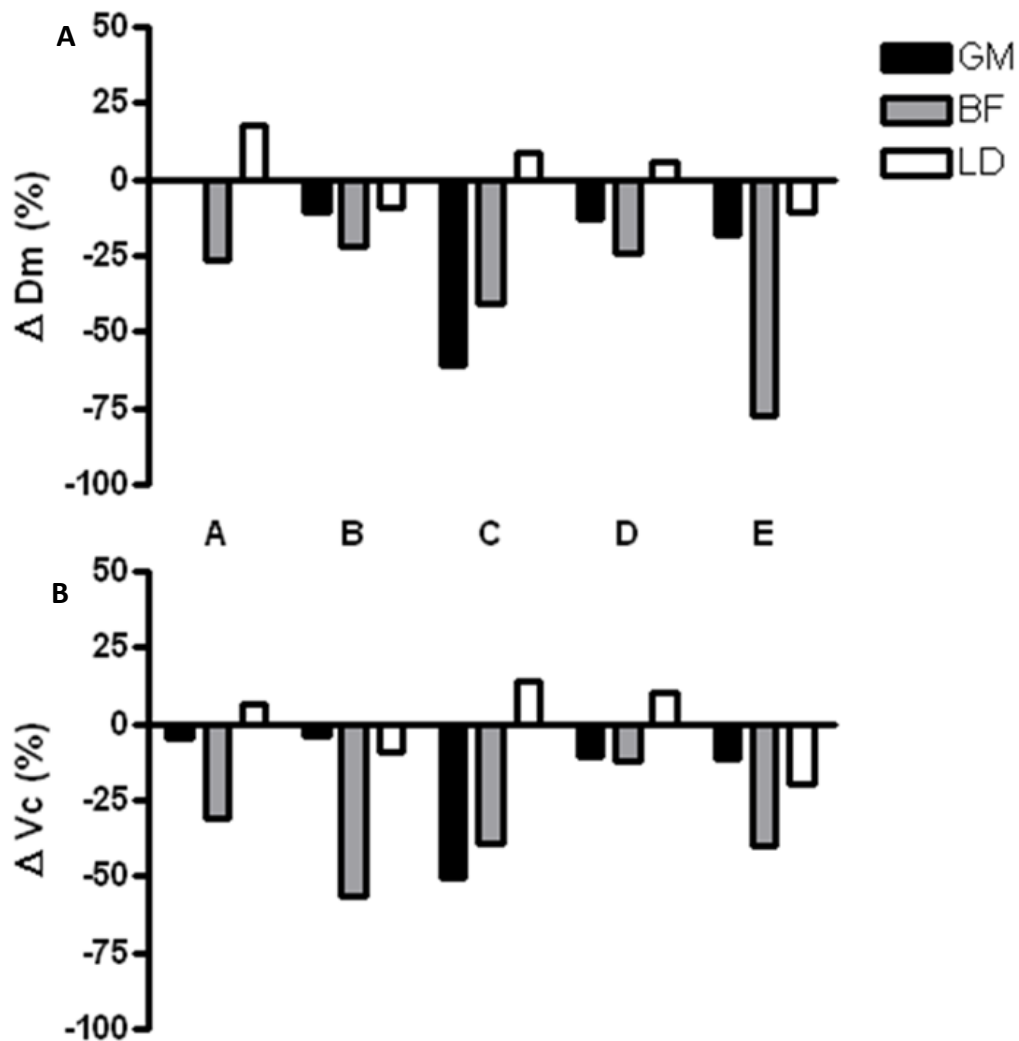


Figure 6.5 Individual athlete changes (%) in Dm (A) and Vc (B) from pre-taper to during taper. n = 5.

### 6.5.3 Discussion

A period of reduced training volume tapering, lasting ~8 days, resulted in reduced Dm of GM and BF, with no detectable alteration in LD. However, Vc of GM and BF was also reduced during taper. Additionally, no improvement was observed in CMJ performance. *In vitro* investigations into taper response have demonstrated increased cross-sectional area of single muscle fibres, specifically type II (Trappe et al., 2000), as well as augmented Vc (Neary et al., 2003). Decreased fibre diameter, through muscle atrophy, has been recorded using

TMG, as increased Dm, symptomatic reduced muscle tension (Pišot et al., 2008). It follows therefore, that an increase in fibre diameter would be detected through lower Dm, as observed in this case-study. It appears that reduced training volume and stress, has resulted in an increase in muscle resting tension due to fibre hypertrophy (Mujika et al., 2004). Reduced training stress also is corroborated by lower CK concentration during taper, compared to pre-taper.

Acute increases in muscle tension following taper could be thought to be related to increased force production (Costill et al., 1985), as has been reported in single fibres (Trappe et al., 2000; Neary et al., 2003), however, no improvement (nor detriment) was detected in CMJ performance. Perhaps concomitant decreases in Vc have resulted in maintenance, rather than enhancement of muscle power, as both BF and GM, prime movers in CMJ, have presented similar adaptive responses. It is interesting that while both lower limb muscles displayed similar alterations, LD was unaffected by taper. Since it has been noted that specifically type II muscle fibres respond to reduced training volume, one explanation could be the fibre type composition of LD compared to GM and BF, however mean LD velocity is greater ( $241.8 \pm 20.0 \text{mm}\cdot\text{s}^{-1}$  and  $243.2 \pm 43.6 \text{mm}\cdot\text{s}^{-1}$ , pre-taper and during taper, respectively) than GM ( $92.3 \pm 18.7 \text{mm}\cdot\text{s}^{-1}$  and  $76.6 \pm 22.4 \text{mm}\cdot\text{s}^{-1}$ ) and BF ( $143.8 \pm 35.7 \text{mm}\cdot\text{s}^{-1}$  and  $91.3 \pm 25.9 \text{mm}\cdot\text{s}^{-1}$ ), suggesting higher type II fibre composition (Dahmane et al., 2001; Šimunič, 2012). Dm in LD is also greater ( $11.1 \pm 1.6 \text{mm}$  and  $11.5 \pm 2.9 \text{mm}$ , pre-taper and during taper, respectively) than GM ( $2.1 \pm 0.3 \text{mm}$  and  $1.6 \pm 0.5 \text{mm}$ ) and BF ( $5.1 \pm 1.3 \text{mm}$  and  $3.1 \pm 1.4 \text{mm}$ ); perhaps due to lesser muscle tension pre-taper, LD requires a longer period of reduced training volume, in order for de-loading to be effective.

In this case-study, acute muscle contractile adaptations were detected via TMG assessment, namely decreased  $D_m$ , related to increased muscle tension due to type II fibre hypertrophy. However, these adaptations were not associated with improved performance in a power-related CMJ task, as had been expected. A decline in  $V_c$ , alongside increased muscle tension, is the likely explanation for the observed lack of change in power. Given that LD has displayed no adaptation in the current case-study, it is possible that the duration of taper was too brief to allow adequate response.

## 6.6 Conclusion

The aim of this chapter was to apply TMG within an elite sports setting. The specific demands of performance sport were addressed. We demonstrated potential applications of individual muscle contractile assessments in injury screening and rehabilitation; it is possible to identify imbalances among agonist muscles which may result in abnormal recruitment patterns, heightening the risk of injury. Imbalances were also assessed with a view to aiding performance, as asymmetric muscle mechanics may impair movement coordination and skill performance. Increased day-to-day variability has been detected among highly trained athletes, promoting the critical importance of appropriate measurement timing. Finally, acute contractile adaptations have been detected, however further research is required to link such adaptations to functional changes.

# Chapter 7

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## Summary & Conclusions

## 7.1 Conclusion

The objective of this thesis was to take advantage of non-invasive and non-disruptive measurement techniques to examine neuromuscular components of physical activity and training, which are fundamentally related to preparation for sporting performance at the highest level. As such, peripheral fatigue and exercise-induced muscle damage were among the primary focus for the research. Functionally, fatigue is associated with muscular failings as regards producing the expected force output (Edwards, 1983). At an isolated muscle level, fatigue leads to contractile responses, to stimulation, that are less than expected or anticipated (MacIntosh & Shahi, 2011). We detected acute alterations in the spatial parameters of isolated muscle contraction, following fatigue, which we deemed to be related to increased stiffness of the muscle, due to impaired contractile capacity.

Following examination of acute deficiencies in contractile mechanics, we next explored intrinsic imbalances in muscle symmetry, and potential functional impairments associated with unbalanced contractile capacity. Although difficult to apply direct relationships between isolated muscle contraction and overall functional performance, we highlighted the possibility of centrally mediated alterations in muscle recruitment patterns, to offset imbalances in muscle contractile mechanics. As such, asymmetries between muscles are often masked during whole-body, or limb, functional assessment, as has been demonstrated in a sports setting (Potts et al., 2002; Evershed et al., 2014). It remains to be established whether alterations in recruitment could be detrimental long-term, however, we explored this area further, in a case-study of an athlete undergoing ACL reconstruction surgery. We demonstrated that, following surgery and rehabilitation, the knee extensor muscles demonstrated a more balanced ratio, at least in contractile capacity. We also



present a case that muscular asymmetries are, at least, as high, and potentially even higher, in an elite athlete population, compared to a recreationally active group. This suggests that athletic performance may not be strongly affected by muscular imbalances, of the type we have investigated. This dissociation between athletic ability and levels of asymmetry provides further evidence that general function can mask imbalances at the individual muscle level. However, the relevance of assessing individual muscles, particularly within high performance sport, may still merit some support.

Screening for muscle contractile imbalances may offer a beneficial option, within performance sport, for both injury management and risk reduction, and for enhancement of functional performance. However, applying such measurements to high performance athletes incorporates specific obstacles; not least, we reported diminished reliability among elite athletes, compared to recreationally active individuals. We attribute the greater variability in muscle contractile responses to the high, and inconsistent, demands placed upon muscles through sport-specific training. Appropriate choices must be made regarding the timings of assessment; while it appears that more reliable measurements of contractile mechanics are achieved with muscles in an exercised (or indeed, fatigued) state, certain muscles that are not critical to the type of training being performed, may display greater irregularity. Therefore, rested measurements are advantageous, when seeking reduced day-to-day noise. Within swimming, for example, we present evidence that bicep femoris and latissimus dorsi respond more consistently to training sessions than do vastus lateralis or gastrocnemius medialis. These observations are significant when assessing specific acute responses in contractile mechanics; we demonstrated that ~8 day reduced training volume taper was sufficient to elevate muscle tension in bicep femoris and gastrocnemius medialis,

however longer duration may be required to affect latissimus dorsi. While a 'whole body' approach is typically taken, as judged through performance, individually isolating specific segments, to bring about optimal adaptation, or change, may provide further improvements in overall functional performance.

To more deeply understand the mechanisms involved in alterations in muscle recruitment and firing patterns, motor unit firing rates were explored, along with the central control of the active motor unit pool. Acute alterations in muscle function were induced through application of an eccentric exercise protocol, resulting in exercise-induced muscle damage. Muscle force capacity was reduced, peaking 48 hours after eccentric exercise; such decrements have been demonstrated to be associated with similar decrements in contractile mechanics (Hunter et al., 2012). Attenuated motor unit firing rate was observed, specifically in later recruited motor units, with higher force thresholds, which are suggested to be associated with less resilient muscle fibres. Concomitantly, motor unit common drive was elevated, representing more synchronous firing. Alterations in motor unit firing coincided with peak muscle function impairment; it is suggested that acute muscle damage induced centrally mediated alterations in motor unit firing behaviour, to compensate for reduced muscle function. In future it may be interesting to explore the possibility of correlating changes in firing behaviour, and centrally driven muscle recruitment, to altered muscle mechanics. An approach which combines both TMG and dEMG could provide extremely detailed insight into the neuromuscular system *in vivo* at an isolated muscle level.

Finally, both muscle contractile mechanics and muscle recruitment were investigated, following a short-term intervention of self-myofascial release. The extensive testing procedures resulted in fatigue-induced decrements in the control condition, which

were countered by the intervention. As such, peak muscle force was maintained by self-myofascial release. Fatigue was counteracted by improvement in muscular efficiency, displaying a reduction in firing, for a given force output; this was attributed to an elevation in proprioceptive afferent activity. Mechanically, muscle stiffness was lower towards the end of the intervention period, compared to control; it seems likely that this mechanical difference is due to the fatigued state of muscle following control, rather than a direct result of the intervention. Overall, the research presented in this thesis promotes the case for non-invasive assessments of muscle contractile mechanics among athletic populations. Understanding the link between mechanical properties of individual muscles, and how these relate to central recruitment is critical to optimise the neuromuscular component of high performance sport.

We have demonstrated that TMG, as a means to assess muscle mechanical properties, incorporates a degree of variability, particularly within an athletic population with high training volumes. However, we have presented a case for the value of individual muscle assessments, and there is currently no alternative method of such assessment, which has demonstrated greater reliability. Further study could perhaps optimise methodologies of testing, in order to enhance the capture of data using TMG. Perhaps performing measurements in truly isometric circumstances, in order to completely restrict movement artefact, would augment the measurement of muscle twitch. This methodological consideration remains to be tested however.

We made the case for TMG as a non-invasive assessment tool, which can be utilised without adversely impacting the status of the muscle, or the individual as a whole. In Chapter 5, it was clearly demonstrated that tests of muscle function, whether maximal

efforts, or sustained at a submaximal level, can lead to fatiguing of the muscles in question. And while we have not confirmed, within this body of work, that continual testing with TMG does not result in a fatiguing effect, seminal studies into the reliability of TMG (as described in Chapter 1) used repeated assessments, and presented consistent muscle responses, indicating that the muscle status has remained constant.

Although it is not possible, with the existing evidence, to propose that individual muscle contractile mechanics are directly associated to outcome performance, perhaps we can offer a means to evaluate the function of a muscle within the system as a whole. Future insight may be gained from studying the adaptive responses of individual muscle mechanics in conjunction with system functional performance. The provision of greater insight into the constituent parts of a muscle group may optimise the enhancement of that group. In conclusion, within high performance sport, the requirements to assess muscular adaptations in response to training, or injury rehabilitation, necessitate an objective means to test isolated muscle *in vivo*, and in a manner that does not directly affect the muscle itself. Mechanical assessments using TMG have the potential to provide valuable insight into muscle status. It must now be ascertained exactly what links can be drawn between muscle mechanical status and functional performance.

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# Appendices

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Appendix A Parameters of muscle twitch response as measured through tensiomyography at three measurement time points, in control (5 minute rest) and fatigue (5 minute electrical stimulation) conditions,

T1 = pre-warm up; T2 = post-warm up; T3 = post-intervention.

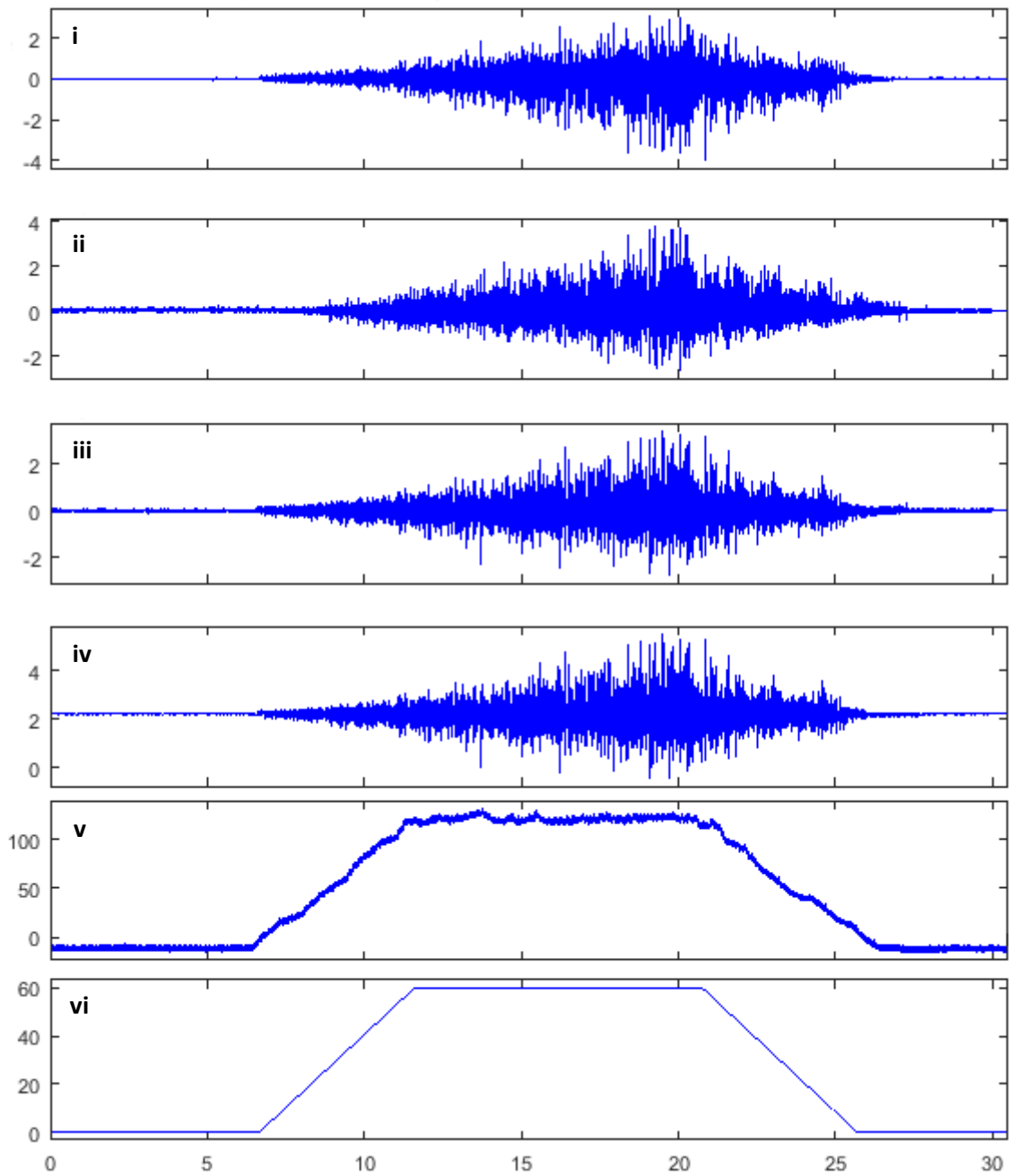
Tc = contraction time; Td = time delay; Ts = sustain time; Tr = half-relaxation time

		T1	T2	T3	
Control	Tc	Mean (ms)	24.0	24.2	25.1
		± SD	2.7	3.3	3.3
	Td	Mean (ms)	21.8	21.6	21.8
		± SD	2.1	2.5	2.3
	Ts	Mean (ms)	167.5	167.4	173.1
		± SD	18.3	16.7	13.7
	Tr	Mean (ms)	78.0	78.1	58.0
		± SD	50.2	49.5	36.9
Fatigue	Tc	Mean (ms)	23.5	24.7	22.9
		± SD	3.2	3.2	3.1
	Td	Mean (ms)	22.5	22.4	20.7
		± SD	3.1	3.3	2.7
	Ts	Mean (ms)	162.3	166.6	148.9
		± SD	21.5	24.5	14.9
	Tr	Mean (ms)	68.0	77.5	58.4
		± SD	41.5	45.4	34.8

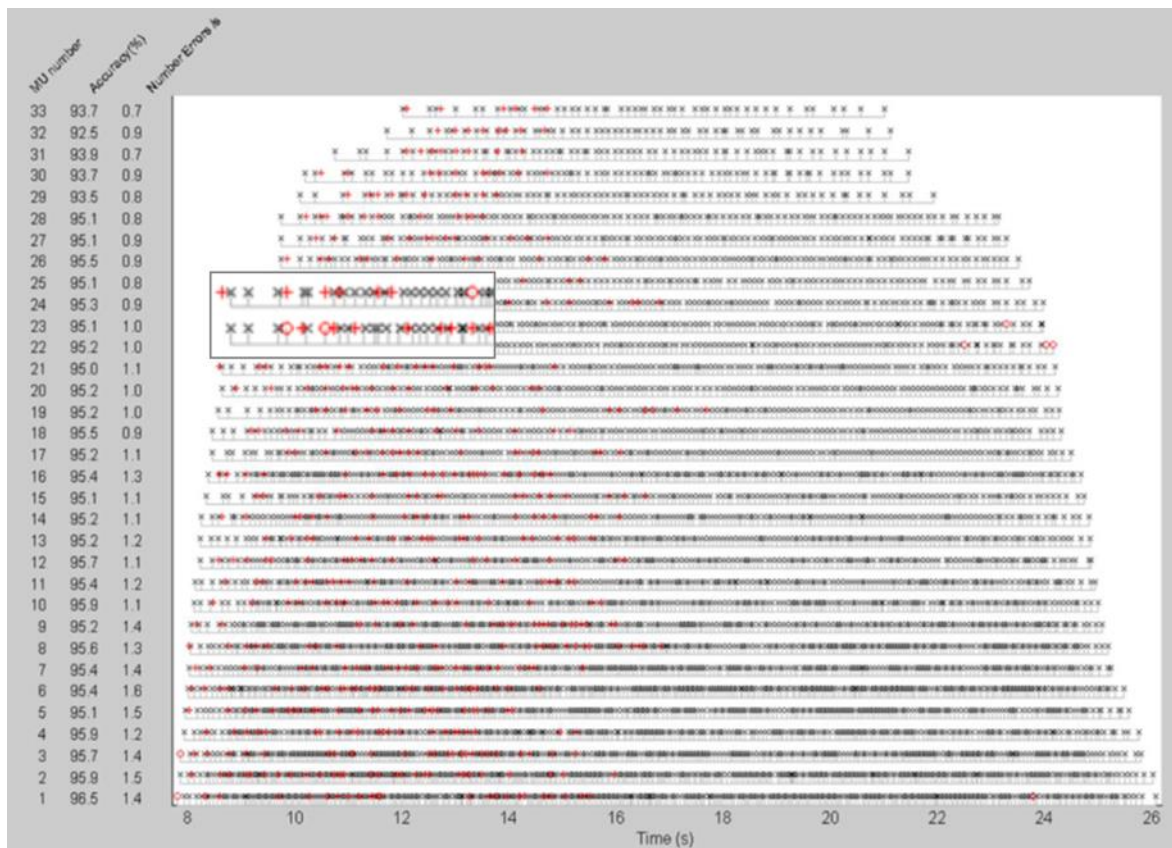
Appendix B Pearsons correlation  $r$  values, and associated  $p$ -values, for peak radial displacement (Dm) and peak contraction velocity (Vc) of RF, VL and VM, with MVC and unilateral CMJ height.

			Dominant		Non-Dominant	
			MVC	CMJ	MVC	CMJ
<b>Dm</b>	<b>RF</b>	<b>r</b>	0.298	-0.034	-0.196	-0.033
		<b>p</b>	0.177	0.873	0.383	0.879
	<b>VL</b>	<b>r</b>	-0.038	-0.057	0.253	0.207
		<b>p</b>	0.866	0.791	0.256	0.331
	<b>VM</b>	<b>r</b>	0.227	0.176	0.191	0.291
		<b>p</b>	0.309	0.411	0.394	0.168
<b>Vc</b>	<b>RF</b>	<b>r</b>	0.118	0.184	-0.138	0.12
		<b>p</b>	0.601	0.388	0.539	0.578
	<b>VL</b>	<b>r</b>	-0.161	-0.015	0.306	0.212
		<b>p</b>	0.474	0.946	0.166	0.319
	<b>VM</b>	<b>r</b>	0.27	0.193	0.037	0.221
		<b>p</b>	0.225	0.366	0.871	0.3





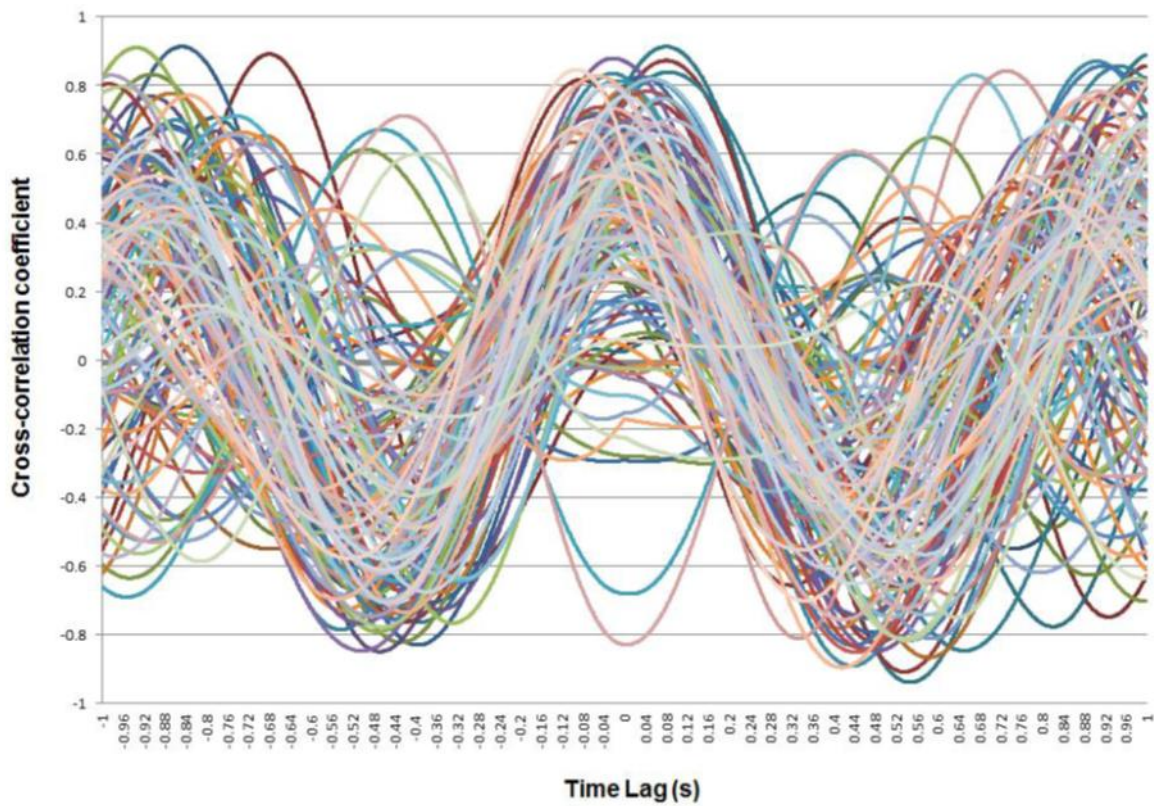
Appendix C Example of sEMG captured concurrently on 4 channels (i - iv), force output (Nm) and target (% of MVC) are also shown (in v and vi, respectively).



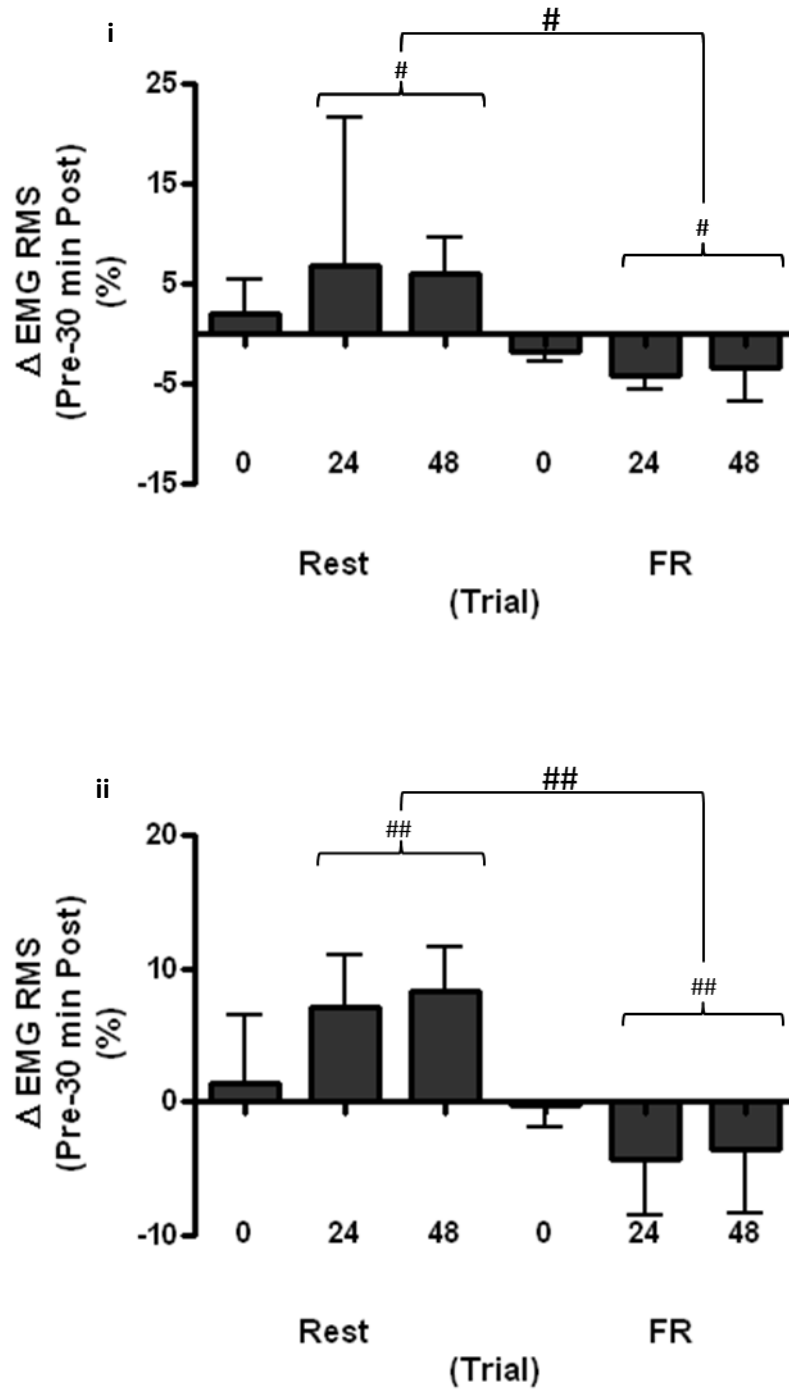
Appendix D (i) Example output of Decompose-Synthesize-Decompose-Compare reliability test used to determine decomposition accuracy for one participant. Motor unit number, accuracy rate (%), and number of errors s<sup>-1</sup> are displayed on the y-axis. Vertical spikes on the figure represent each motor unit firing, firings with a circle denote a false positive, and firings with crosses denote a false negative.

Appendix D (ii) Results of Decompose-Synthesize-Decompose-Compare reliability test. Values are mean  $\pm$  SD, n=14.

Day	0	2	3	7	14	
Control	Mean (%)	95.1	95.7	95.7	95.1	94.1
	$\pm$ SD	2.6	2.5	2.3	2.7	1.8
Damage	Mean (%)	96.1	96.3	96.3	94.3	95.2
	$\pm$ SD	2.3	2.1	2.1	4.8	2.3



**Appendix E Cross-correlation coefficient function output for a single time-point for one participant. Each curve displayed on the figure represents the output of the cross-correlation between two motor unit mean firing rate curves in which peak cross-correlation coefficients occurred within the specified constant force time period of the isometric trapezoid force trace effort.**



Appendix F Change in normalized RMS (i = 0-6 seconds; ii = 24-30 seconds) from pre- to 30 minutes post-treatment in the control (Rest) and intervention (FR) conditions. Values are mean  $\pm$  SD, n = 16. # Significant difference between conditions,  $p < 0.01$ . ## Significant difference between conditions,  $p < 0.001$ .