Kingston
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# FACULTY OF SCIENCE, ENGINEERING AND COMPUTING

## School of Life Sciences, Pharmacy and Chemistry

# The evaluation of muscle damage and muscle recovery following exercise in elite youth soccer players

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**Doctor of Philosophy**

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The thesis is being submitted as partial fulfilment of the requirements of the University for the award of Doctor of Philosophy of Kingston University.

# Declaration

I hereby declare that this thesis, submitted in partial fulfilment of my requirements for the degree of Doctor of Philosophy and entitled 'The evaluation of post-match recovery strategies for the attenuation of muscle damage in elite youth soccer players, represents my own work and has not been previously submitted to this or any other institution for any degree, diploma or qualification. In accordance with the Kingston University regulations, I also confirm that this thesis is the one upon which I expect to be examined for the above degree.

Samuel Pooley

April 2020

# Abstract

In elite youth soccer, coaches aim to optimise player training time to maximise learning opportunities and enhance athlete development with the end goal of preparing youth soccer players for a career in the adult game. As such, determining the effects of soccer matches on muscle damage, and the efficacy of recovery interventions to reduce damage and accelerate muscle recovery is of great importance to practitioners. Previous studies have assessed the effects of competition on muscle damage, and the effects of interventions on recovery, yet the accessibility to the elite youth soccer population is limited. As such, many implemented applied interventions are based on laboratory-based studies using sub-elite athletes and simulated eccentric exercise protocols. The primary aim of this thesis investigated the post-match muscle damage of the elite youth soccer population in applied settings to gain a greater understanding of these areas and determine the effectiveness of commonly used recovery interventions in this setting.

Due to the vast amount of muscle damage research utilising controlled eccentric exercise protocols to inflict muscle damage and assess the effects of recovery interventions thereafter, firstly (chapter 4) it was necessary to determine the extent to which muscle damage is elicited following elite youth soccer matches (n=10 players) within an English Premier League professional soccer academy in comparison to an isokinetic eccentric exercise protocol. The findings established that, although both exercise conditions inflicted significant muscle damage, soccer matches induced greater damage than isolated eccentric protocols raising questions on the applicability of previous non-applied studies utilising eccentric exercise protocols to the elite soccer environment. As such, further research on the effects of applied recovery interventions following such exercise in elite youth soccer was required.

Chapter 5 identified that soccer matches induced muscle damage that remained for up to 48 hours post-match following no, or ineffective recovery interventions. Having established that competitive soccer matches significantly induce muscle damage, it was necessary to define the physical components of soccer matches that correlate to indicators of muscle damage. Chapter 6 identified that high-intensity accelerations and decelerations, and absolute high speed running significantly correlated to an increase in creatine kinase (CK) post-match, providing a new and alternative indirect indicator or predictor of muscle damage.

Furthermore, chapter 5 established that static stretching, a commonly used recovery intervention in applied settings, provided no recovery benefit (n=10 players), at 48 hours post-match and as such further research on alternative recovery modalities was necessary. Chapter 7 assessed the effectiveness of active recovery (AR) and cold-water immersion (CWI) with a randomised cross-over design (n=15 players). The findings revealed significant recovery benefits when compare to static stretching, aiding markers of muscle damage in their return to baseline measures, and thus promotes the use of AR and CWI in applied elite settings as recovery interventions following soccer matches.

This thesis provides the applied industry with evidence of a muscle-damaging effect of elite youth soccer matches that remains for up to 48 hours post-match. It also provides recommendations for the use of effective (AR and CWI) and ineffective (static stretching) recovery interventions, and the high intensity, eccentric components of soccer matches that correlate to muscle damage indicators.

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## Publications and presentations arising from this thesis

*Manuscripts in peer-reviewed journals*

**Pooley, S.,** Spendiff, O., Allen, M. and Moir, H.J. (2017). Static stretching does not enhance recovery in elite youth soccer players. *BMJ open sport & exercise medicine*, 3(1): .e000202. doi: 10.1136/bmjsem-2016-000202

*Refers to chapter 5*

**Pooley, S.,** Spendiff, O., Allen, M. and Moir, H.J. (2019). Comparative efficacy of active recovery and cold water immersion as post-match recovery interventions in elite youth soccer. *Journal of Sports Sciences:*1-9. doi: 10.1080/02640414.2019.1660448.

*Refers to chapter 7*

*Conferences abstracts and other presentations*

**Pooley, Sam,** Allen, Matt, Spendiff, Owen and Moir, Hannah (2016). The effect of static stretching on muscle recovery following competitive soccer matches. In: *European College of Sport Science (ECSS) 21st Annual Congress*; 6 - 9 Jul 2016, Vienna, Austria.

*Refers to chapter 5*

# Table of Contents

## List of Tables

## List of Figures

## Operational Definitions

| | |
|---|---|
| **AccelsZ4-6** | Accelerations > 3m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **AccelsZ5-6** | Accelerations > 4m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **ADT** | Adenosine diphosphate |
| **ANOVA** | Analysis of variance |
| **AR** | Active recovery |
| **ATP** | Adenosine Triphosphate |
| **$Ca^{2+}$** | Calcium |
| **CI** | Confidence intervals |
| **CK** | Creatine Kinase |
| **CMJ** | Countermovement jump |
| **CMJA** | Countermovement jump with arm swing |
| **CoV** | Coefficient of variance |
| **CRP** | C-reactive Protein |
| **CWI** | Cold water immersion |
| **CWT** | Contrast water therapy |
| **DecelsZ4-6** | Decelerations > 3m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **DecelsZ5-6** | Decelerations > 4m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **DOMS** | Delayed onset muscle soreness |
| **E-C** | Excitation contraction |
| **ECC** | Eccentric exercise protocol |
| **ECM** | Extra cellular matrix |
| **EIMD** | Exercise induced muscle damage |
| **Ex.D** | Explosive distance |
| **GPS** | Global positioning system |
| **$H^{+}$** | Hydrogen ions |
| **HMLD** | High metabolic loading distance |
| **HSRAb** | Absolute high-speed running |
| **HSRRe** | Relative high-speed running |

| | |
|---|---|
| **IL-1β** | Interleukin 1 beta |
| **IL-10** | Interleukin 10 |
| **IL-6** | Interleukin 6 |
| **Match%** | Match percentage |
| **MP** | Minutes played |
| **mPTP** | Mitochondrial permeabilisation transition pore |
| **MRI** | Magnetic resonance imaging |
| **MVC** | Maximal voluntary contraction |
| **OedemaG1** | Lower 1/3 gastrocnemius oedema |
| **OedemaG2** | Upper 1/3 gastrocnemius oedema |
| **OedemaQ** | Quadriceps oedema |
| **PCr** | Phosphocreatine |
| **$PLA_2$** | Phospholipase $A_2$ |
| **PMS** | Perceived muscle soreness |
| **PR** | Passive recovery |
| **RBE** | Repeat bout effect |
| **ROS** | Reactive oxygen species |
| **SD** | Standard deviation |
| **SR** | Sarcoplasmic reticulum |
| **SS** | Static stretching |
| **TE** | Typical error |
| **TGF-β1** | Transforming growth factor beta 1 |
| **TNF-α** | Tumour necrosis factor alpha |
| **TQR** | Total quality recovery |
| **VAS** | Visual analogue scale |
| **$[Ca2^+]_c$** | Cytosol calcium concentrations |
| **$\eta^2_p$** | Partial $ETA^2$ |

# 1. Introduction

Professional soccer academies house some of the brightest talents in youth soccer, providing the opportunity for athletes to develop into professional soccer players – an occupation with rewards, financial and otherwise, that are far beyond many alternative sports and professions. It is the role of the academy coaching staff to develop professional soccer players through exposure to training and matches. To allow the greatest opportunity for success in an elite sporting career, youth soccer players are required to train and compete at exceptionally high physical, technical and psychological intensities, resulting in high stress loads being placed on their bodies (Akenhead, 2014; Bowen, *et al.*, 2017).

In order to maintain and improve training and performance levels, athletes require sufficient time to recover following intense training and games to allow muscular adaptations to take place. However, in order to accelerate the muscle recovery process and reduce the risk of overuse and overreaching injuries in the days following competitive soccer matches, athletes and their coaches may seek recovery interventions beyond rest, such as active recovery techniques and cold water immersion, allowing optimal training performance in the days surrounding competitive games, providing the greatest opportunity for players to improve and develop. It may also be the case that in an attempt to increase training exposure, recovery of athletes following competitive soccer matches is overlooked by coaches, placing more emphasis on the importance of accelerating recovery.

Research has revealed that throughout a 90 minute soccer match, players can cover between 9 and 14 km (Bradley & Ade, 2018), of which up to 3km may consist of high-intensity running, and up to 0.5 km sprinting (Devrnja & Matković, 2018; Mohr, *et al.*, 2017). In addition, players can complete 1000 – 1500 discrete movement changes throughout soccer matches, occurring every 4 to 6 seconds (Bloomfield, *et al.*, 2007), and a substantial number of the explosive activities with eccentric components comprising jumps, accelerations, decelerations and changes of direction which have been argued to be causally related to post-match muscle damage (Devrnja & Matković, 2018). Despite this, at present there is a dearth of evidence evaluating the impact of elite youth soccer matches on athlete experiences of muscle damage and the efficacy of the current recovery interventions on subsequent performance, and therefore this requires further research.

Therefore, the aims of this thesis are as such:

- Investigate to what extent elite youth soccer matches elicit muscle damage in the hours and days following match exposure.
- Investigate the effects of commonly used recovery interventions for the attenuation of muscle damage in elite youth soccer players following soccer match exposure.

In an attempt to fulfil these aims, this thesis will:

1. Compare the effects of muscle damage elicited from elite youth soccer matches to that of an eccentric exercise protocol previously inflicting significant muscle damage post-exercise.
2. Compare the effects of static stretching as a recovery intervention on attenuating muscle damage of elite youth soccer matches.
3. Establish whether physical locomotion parameters of elite youth soccer matches significantly correlate to indicators of muscle damage.
4. Assess the effects of commonly used recovery interventions such as active recovery and cold-water immersion on the attenuation of muscle damage following elite youth soccer match exposure.

Throughout this thesis, the following topics will be discussed and investigated; 1. In comparison to previously used eccentrically-induced muscle damaging protocols, how much muscle damage (if at all) do soccer matches inflict on the elite youth population; 2. To what extent does static stretching affect the recovery of elite youth soccer players following competitive soccer matches; 3. To what extent do the physical locomotion demands of competitive soccer matches impact on the indicators of muscle damage in elite youth soccer players; 4. In comparison to static stretching, to what extent do more conventional recovery interventions improve muscle recovery following competitive soccer matches of elite youth soccer players.

To investigate the objectives of this thesis, hypotheses were generated in relation to each research question:

Hypothesis 1 (chapter 4):

- $H_1$: Soccer matches of elite youth soccer players induce muscle damage.
- $H_0$: Soccer matches of elite youth soccer players do not induce muscle damage.

Hypothesis 2 (chapter 4):

- $H_2$: Soccer matches of elite youth soccer players induce greater muscle damage than an eccentric muscle damaging protocol.
- $H_0$: Soccer matches of elite youth soccer players do not induce greater muscle damage than an eccentric muscle damaging protocol.

Hypothesis 3 (chapter 5):

- $H_3$: Static stretching reduces markers of muscle damage following soccer matches of elite youth soccer players when compared to passive recovery.

- $H_0$: Static stretching has no effect on markers of muscle damage following soccer matches of elite youth soccer players when compared to passive recovery.

Hypothesis 4 (chapter 6):

- $H_4$: Physical locomotive match outcomes correlate to increases in markers of muscle damage following elite youth soccer matches.
- $H_0$: Physical locomotive match outcomes do not correlate to increases in markers of muscle damage following elite youth soccer matches.

Hypothesis 5 (chapter 7):

- $H_5$: Active recovery improves indicators of muscle damage following elite youth soccer matches when compared to static stretching.
- $H_0$: Active recovery does not improve indicators of muscle damage following elite youth soccer matches when compared to static stretching.

Hypothesis 6 (chapter 7):

- $H_6$: Cold water immersion improves indicators of muscle damage following elite youth soccer matches when compared to static stretching.
- $H_0$: Cold water immersion does not improve indicators of muscle damage following elite youth soccer matches when compared to static stretching.

# 2. Review of Literature

## 2.1 Muscle Damage

The physiological effects of eccentric, high-intensity exercise have been well documented, with research indicating such exercise may lead to exercise-induced muscle damage (EIMD) (Owens, *et al.*, 2019; Lavender & Nosaka, 2008; Peake, *et al*., 2005; Byrne, *et al*., 2001; Clarkson, *et al*., 1992), causing skeletal muscle microtrauma (Baumert, *et al*., 2016) and resulting in perforations in the sarcolemma and damage to the sarcomeres (Russell, *et al*., 2016) often causing delayed onset muscle soreness (DOMS). Although the exact mechanisms of muscle damage remain unclear, research evaluating muscle damage has shown significant reductions in post-exercise performance markers such as jump performance (Roberts, *et al*., 2014; Elias, *et al*., 2013; Duffield, *et al*., 2010; Ingram, *et al*., 2009; Howatson, *et al*., 2009; Tessitore, *et al*., 2007; Dawson, *et al*., 2005), and significant elevations in markers of myoproteins (Ascensão, *et al*., 2011; Ingram, *et al*., 2009; Howatson, *et al*., 2009; Rowsell, *et al*., 2009; Howatson & van Someren, 2008; Gill, *et al*., 2006) and subjective assessments of soreness (Ascensão, *et al*., 2011; Ingram, *et al*., 2009; Howatson, *et al*., 2009; Rowsell, *et al*., 2009; Howatson & van Someren, 2008; Dawson, *et al*., 2005). With this in mind, and the commonly accepted theory that DOMS peaks between 48 and 72 hours post-exercise (Cheung, *et al*., 2003; Nosaka, *et al*., 2002; Cleak & Eston 1992), it may be hypothesised that eccentric, high-intensity exercise significantly inhibits performance in the days following such exposure, and time to optimal performance following said exercise may be accelerated should sufficient recovery modalities be administered. It has been suggested that eccentric muscle contractions can be attributed to muscle damage due to the greater tension per cross-sectional area of activated muscle in comparison to concentric contractions (Thorpe & Sunderland, 2012), and the overstretching of sarcomeres and myofibrils (Peake, *et al*., 2017a). As such when considering the effects of elite soccer performance on muscle damage, it is these eccentric contractions such as accelerations, decelerations, sprints, jumps and unpredictable changes in intensity (Scott, *et al*., 2016; Nédélec, *et al*., 2014; Devrnja & Matković, 2008) that should be considered.

### 2.1.1 Theories for the Aetiology of Muscle Damage

Despite the potential effects of muscle damage inhibiting performance of professional soccer players, and although the exact mechanisms of muscle damage remain unclear, a number of theories have been presented. Armstrong (1990) proposed an integrated theory of muscle damage, consisting of four stages of the muscle damaging process; 1) initial events, 2) autogenic processes, 3) phagocytic stage, and 4) the regenerative stage. Although the muscle damaging process can be divided in to these four definite stages, it must be considered that these stages overlap significantly throughout the process. It should also be considered that there are events that occur throughout the injury and repair process that are not confined to one of the four stages as suggested by Kendall and Eston (2002), involving the presence of reactive oxygen species (ROS).

Armstrong (1990) presented all four theoretical stages, however, discusses in detail the initial two stages of the muscle damaging process. More recent reviews have also discussed the initial events and autogenic stages (Kendall & Eston, 2002), whilst other authors (Gissel, 2005; Morgan & Proske, 2004; Proske & Morgan, 2001) have presented theoretical models of specific stages within the proposed process presented by Armstrong (1990). Significant research has been conducted and presented relating to the specific stages of the muscle damage process as published by Armstrong (1990), however to date no study has proposed a detailed model for the entire muscle damage and repair process. As such, a suggested model has been presented in the present thesis, taking inspiration from previously proposed work (Armstrong, 1990; Proske & Morgan, 2001; Morgan & Proske, 2004; Gissel, 2005;

Tee, *et al*., 2007) and findings from relevant literature to display all four stages of the muscle damage process, and the intricacies of each stage (Figure 2.1).

#### 2.1.1.1 *Initial Causes of Damage*

Two main theories surrounding the initial causes of muscle damage have been elucidated, consisting of mechanical stress and metabolic stress (Owens, *et al.*, 2019; Peake, *et al*., 2017a; Peake, *et al*., 2005; Close, *et al*., 2004; Kendall & Eston, 2002; Armstrong, 1990). Although the exact mechanisms of muscle damage are yet to be identified, it has been suggested that metabolic and mechanical stress may occur separately or simultaneously (Kendall & Eston, 2002), however Close and co-workers (2005) suggest that due to fewer motor units being recruited in eccentric contractions in comparison to concentric contractions for a given work load, and energy costs of eccentric contractions being lower than that of concentric contractions, it is likely that mechanical stress as opposed to metabolic stress results in muscle damage. In contrast to this, Tee and colleagues (2007) propose a model in support of metabolic stress as the initial cause of EIMD due to metabolic deficiencies, or that these metabolic deficiencies increase the vulnerability of the muscle fibre to mechanical stress.

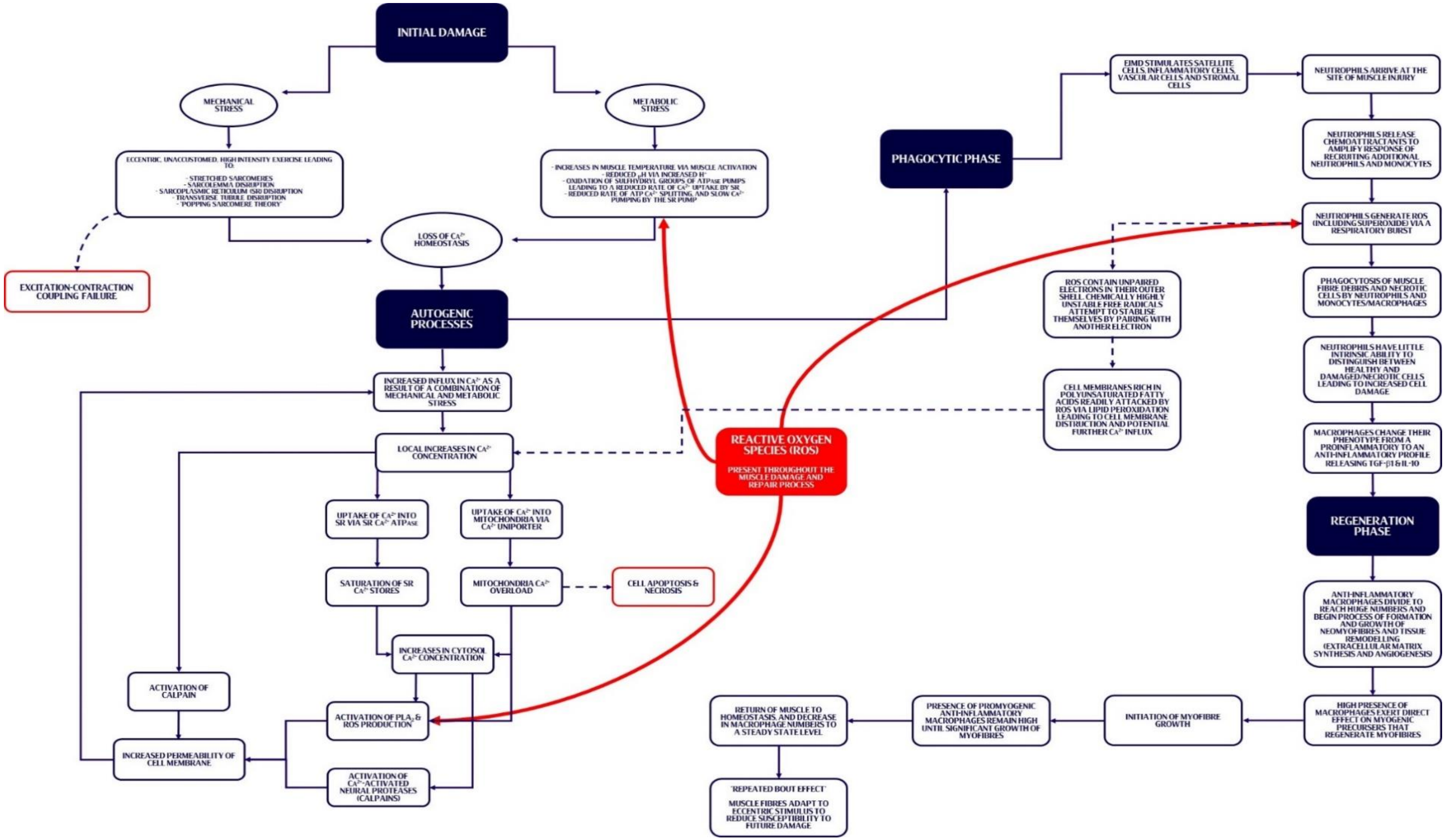


**Figure 2.1** Postulated series of events displaying the processes involved in the muscle damage and regeneration phase following eccentric, high intensity and unaccustomed exercise. Developed from the work of Armstrong, 1990; Proske & Morgan, 2001; Morgan & Proske, 2004; Gissel, 2005; Tee, *et al.*, 2007.

##### 2.1.1.1.1 *Mechanical Stress*

Mechanical stress is suggested to be the result of eccentric muscle contractions (Douglas, *et al.*, 2017; Peake, *et al.*, 2017a; Kendall & Eston, 2002), whereby muscle fibres lengthen under tension, forming one part of the sliding filament theory (Butterfield, 2010). Unlike concentric exercise, where actin and myosin filaments overlap to shorten the muscle and form a contraction, eccentric exercise involves tension whilst the muscle is lengthening (Douglas, *et al*., 2017), resulting in filaments being pulled away from each other by the external forces acting upon them (Peake, *et al.*, 2017a; Kendall & Eston, 2002). When myofibrils are stretched under contraction, some weaker sarcomeres may be less tolerant to the tension, and myofilaments may be forced into a state of no overlap (Douglas, *et al*., 2017; Peake, *et al.*, 2017a; Peake, *et al.*, 2005). On completion of the eccentric action, the majority of myofilaments of overstretched sarcomeres return to their normal function however some fail to reconnect and become disrupted. Repetitive eccentric contractions may initially lead to overstretching of the weaker sarcomeres, followed by the overstretching of the stronger sarcomeres resulting in structural disruption and ‘popped sarcomeres’ (Peake, *et al.*, 2017a). This damage may spread to adjacent areas of the muscle potentially resulting in damage to the sarcoplasmic reticulum (SR) membranes, transverse tubules and sarcolemma (Peake, *et al*., 2005). Within soccer, numerous actions exist that may inflict the mechanical stress through eccentric muscle contractions, such as accelerations (Scott, *et al.*, 2016; Arnason, *et al*., 2008), decelerations (Scott, *et al.*, 2016; de Hoyo, *et al.*, 2015), high speed running and sprinting (de Hoyo, *et al.*, 2015; Arnason, *et al.*, 2008) and explosive ball striking (de Hoyo, *et al*., 2015; Arnason, *et al*., 2008).

The events of mechanical stress have been suggested to cause openings in stretch-activated channels, membrane disruption and excitation-contraction coupling dysfunction (Peake, *et al*., 2017a), potentially allowing calcium to enter the cytosol through stretch-activated channels, or permeable sections of the sarcolemma, stimulating activation of calpain enzymes, Phospholipase $A_2$ ($PLA_2$) and ROS production leading to a 'vicious cycle' of increased cell permeability, increases in cytosol calcium concentrations $[Ca^{2+}]_c$ and cell necrosis (Howatson & van Someren, 2008; Gissel, 2005).

#### 2.1.1.1.2 *Metabolic Stress*

Metabolic stress is characterised by a number of events that occur at the site of muscle cells involved in muscle contractions, including increased muscle temperature, lowered pH, a reduction in mitochondrial respiration and production of reactive oxygen species (ROS). An increase in muscle temperature to greater than 38°C has been suggested to uncouple the calcium stimulated ATPase activity from calcium transport to the SR, thus increasing $[Ca^{2+}]_c$ (Kendall & Eston, 2002). Furthermore, the increase in hydrogen ions ($H^+$) causing a decrease in pH as a result of strenuous exercise has been suggested to reduce the SR uptake of calcium, potentially due to hydrogen and calcium competing for the calcium binding site on the ATPase pump (Kendall & Eston, 2002), again increasing $[Ca^{2+}]_c$. Also, a reduction in localised ATP due to exercise, and an increase in ADP could reduce the rate of ATP splitting, and slow the calcium pumping by the SR pump (Kendall & Eston, 2002). These theories are supported by Tee and colleagues (2007), suggesting that due to glycogen depletion within muscle cells, ATP levels are reduced, therefore limiting the capability of calcium removal via calcium-ATPase, resulting in increases in cytosol calcium concentrations, leading to a cascade of metabolic events culminating in muscle fibre degradation. This theory has been supported by Krustrup and others (2006) demonstrating that following a soccer match, 47% of muscle fibres assessed were almost or completely depleted of glycogen. This glycogen depletion may impact on the rate of calcium removal via ATPase, increasing cytosol calcium concentrations and leading to a loss of calcium homeostasis resulting in $PLA_2$ and ROS production therefore increasing cell membrane permeability, whilst Mohr and others (2017) support the suggestion that glycogen depletion may be associated with muscle damage and a severe inflammatory response.

#### 2.1.1.2 *Reactive Oxygen Species*

Reactive oxygen species (ROS), also termed free radicals throughout literature (Close, *et al.*, 2005), are any species capable of independent existence containing one or more unpaired electron in their outer shell. Due to their unpaired electron attempting to stabilise itself by pairing with other electrons, free radicals are chemically highly reactive (Steinbacher & Eckl, 2015; Sachdev & Davies, 2008). They are produced by neutrophils via an oxidative burst to react with lipids, initiating lipid peroxidation and assisting in the destruction and removal of damaged and necrotic cells. As demonstrated by Close and others (2005), the extent of their reactivity depends on the environment the radical is presented with, for example if one radical ($A^{\bullet}$) meets another radical ($B^{\bullet}$), a termination reaction occurs and a non-radical is formed ($A^{\bullet}+B^{\bullet}\rightarrow AB$). However, Close *et al.* (2005) also explained that if a radical meets a non-radical, a new radical is formed ($A^{\bullet}+B\rightarrow A+B^{\bullet}$), and the process continues until a non-radical is formed, or until the radical meets an antioxidant. It has been proposed that ROS production is accelerated by intense muscular activity (Powers, *et al.*, 2016; Steinbacher and Eckl, 2015; Nikolaidis, *et al.*, 2008; Close, *et al.*, 2004), with evidence suggesting free radicals produced by neutrophils and macrophages may contribute to muscle damage (Sachdev & Davies, 2008; Peake, *et al.*, 2005). Oxygen-derived free radicals are a common feature of the damage and repair process, and it has been suggested that there are a number of sites throughout the cycle where they can be produced (Kendall & Eston, 2002). ROS are capable of attacking all major classes of biomolecules, although lipids are particularly susceptible, and since cell membranes are rich in polyunsaturated fatty acids, cell membranes are readily attacked by free radicals (Close, *et al.*, 2005). This supports suggestions of Niess, *et al.*, (1999) that the oxidising effect against lipids and proteins play a fundamental role in the destructive nature of ROS. Lipid peroxides are a form of ROS that cause lipid peroxidation – the result of a damaging radical chain reaction (Sachdev & Davies, 2008). The damaging radical, in this case lipid peroxyl, is highly unstable and reacts with neighbouring fatty acids causing damage to cell membranes, mitochondrial membranes, endoplasmic reticulum membranes and nuclear membranes (Sachdev & Davies, 2008).

Free radicals would appear to act as a recovery mechanism to the damage sustained to muscle cells as a result of exercise. As suggested by Close and colleagues (2004), it is possible that ROS are involved in the phagocytosis of damaged tissue in the days following exercise. This supports Kendall and Eston (2002), who suggest that free radicals play a vital role in the removal of damaged muscle tissue as a result of exercise, however the existence of free radicals in the cells may result in increased inflammatory responses such as leukocyte infiltration and upregulation of pro-inflammatory cytokines (Tumor necrosis factor-α (TNF-α), Interleukin-1β (IL-1β), IL-6) in damaged muscle tissue (Steinbacher & Eckl, 2015; Peake, *et al*., 2005), and increased toxins which ultimately result in additional cell damage (Best, *et al*., 1999). The demand for oxygen during eccentric exercise would appear to be reduced in comparison to that of equivalent concentric exercise (Peñailillo, *et al*., 2017; Howatson & van Someren, 2008; Perrey, *et al*., 2001; LaStayo, *et al*., 1999), and may therefore reduce the occurrence of free radicals following eccentric exercise, suggesting this would not be the primary mechanism for muscle damage (Close, *et al*., 2005). However, as muscle tissue becomes damaged due to repetitive eccentric activity, the destruction of these damaged cells by free radicals in the phagocytosis process may promote further inflammatory responses. Furthermore, it is possible that an unwanted leakage of ROS from the damaged site to surrounding cells may occur resulting in peroxidation of non-damaged, healthy lipid membranes, exacerbating muscle damage (Close, *et al*., 2005). This is supported by Chazaud (2016), suggesting it may be possible for healthy cells to be attacked during the process of phagocytosis, leading to further cell damage.

#### 2.1.1.3 *Autogenic Processes*

The autogenic process, also referred to as the ‘calcium ($Ca^{2+}$) overload’ (Gissel, 2005) is the process whereby the loss of calcium homeostasis has been suggested to play a primary role in the muscle damaging cycle. In order to understand the process of calcium overload during exercise, it is important to examine the calcium concentration in a healthy cell whilst resting. The cytosol calcium concentration ($[Ca^{2+}]_c$) in a resting state is maintained at ~50nM, with an extracellular concentration of calcium of ~1mM (Gissel, 2005), creating a very high chemical gradient. As a result of this chemical gradient, there is a passive diffusion of calcium into the muscle cell, despite the low permeability of the sarcolemma for calcium (Hidalgo, *et al.*, 1986). In order to maintain calcium homeostasis, the muscle cell must remove calcium via calcium-ATPase in the sarcoplasmic reticulum (SR) and sarcolemma/T-tubule membrane (Gissel, 2005).

The use of calcium is vital for muscle cell function, as it binds to troponin allowing tropomyosin to reveal binding sites for myosin heads on the actin myofilament. As exercise progresses, so does the $[Ca^{2+}]_c$, allowing contractions to occur. However, as displayed in the initial metabolic events of the muscle damaging process, a loss of calcium homeostasis may occur due to increases in muscle cell temperature, reduced pH, a reduction in mitochondrial respiration, and an increase in ROS production, coupled with myofibrillar disruption allowing and opening of stretched-activated channels due to eccentric mechanical stress. In response to the increases in localised calcium concentrations, mitochondria and the SR increase their uptake of calcium respectively via the calcium uniporter and calcium-ATPase, resulting in saturation of the mitochondria and SR calcium stores, possibly leading cell necrosis in the case of mitochondria (Gissel, 2005). Full saturation of the mitochondria and SR calcium stores result in increased intracellular calcium (Owens, *et al.*, 2019), and thus the loss of calcium homeostasis.

Due to the calcium, Gissel (2005) suggests increased $[Ca^{2+}]_c$ may result in calpain activation. Calpain splits a variety of protein substrates including cytoskeletal, myofibrillar and membrane proteins, and is closely associated with I and Z band regions of muscle fibres. Activation of calpain may therefore result in degradation of cytoskeletal, myofibrillar and membrane proteins, leading to alterations in muscle structure. Additionally, increases in calcium loads are said to increase ROS production (Gissel, 2005), consequently increasing lipid peroxidation of cell membranes (Sachdev & Davies, 2008). As suggested by Owens and others (2019), mitochondrial calcium overload can lead to inner mitochondrial membrane permeabilisation, and opening of the mitochondrial permeabilisation transition pore (mPTP), resulting in a large efflux of stored calcium from the mitochondria, increasing intracellular calcium concentrations causing cell apoptosis and necrosis. Gissel (2005) also suggested an increase in $[Ca^{2+}]_c$ may activate Phospholipid $A_2$ ($PLA_2$), which will subsequently attack mitochondrial and other membrane phospholipids causing cell membrane disruption.

As shown in the Figure 2.1 and as described by Gissel (2005), the autogenic process resulting in a loss of calcium homeostasis and increases in cell permeability appear to form a ‘vicious cycle’ (Gissel, 2005), further exacerbating the muscle damage inflicted by initial mechanical and metabolic events. This process, coupled with the initial causes of muscle damage results in a subsequent inflammatory cascade (phagocytic process), a vital process in the removal of damaged cells and initiating the repair and recovery process (Owens, *et al.*, 2019).

#### 2.1.1.4 *Phagocytic Stage*

The phagocytic stage of the muscle damage and repair process signals the initiation of an inflammatory cascade (Owens, *et al*., 2019), as immune cells including mast cells, neutrophils, macrophages and T-lymphocytes, along with satellite cells and stromal cells infiltrate the damaged tissue (Peake, *et al*., 2017a) following EIMD. As suggested by Chazaud (2016) it would appear that the inflammatory response seen within the phagocytic phase consists of three stages; 1) the mounting of the inflammatory response characterised by infiltration of immune cells to the damaged tissue and the release of pro-inflammatory cells, 2) the resolution of inflammation, characterised by a shift from a pro-inflammatory to an anti-inflammatory environment, 3) the regeneration of muscle fibres, a reduction in the number of anti-inflammatory cells, and the return of the muscle cell to homeostasis.

It would appear that neutrophils are rapidly mobilised immediately following EIMD (Peake, *et al*., 2005), activated by calcium-stimulated proteolysis (Owens, *et al*., 2019), where they release chemoattractants to amplify the inflammatory response by recruiting additional neutrophils and mononuclear cells (Kendall & Eston, 2002) circulating in blood (Chazaud, 2016). Following the arrival of neutrophils and monocytes to the injury site, the breakdown of damaged cell debris and necrotic cells commences via phagocytosis, and the release of proteolytic enzymes and ROS (Peake, *et al*., 2017a; Close, *et al*., 2005) via a respiratory burst (Howatson & van Someren, 2008; Toumi, *et al*., 2006; Kendall & Eston, 2002). It has been suggested that neutrophils are programmed for overkill (McCord, 1995), and have little intrinsic ability to distinguish between healthy and damaged or necrotic cells (Kendall & Eston, 2002), potentially leading to further cell damage. It is also possible that macrophages produce ROS, adding to the inflammation whilst causing potential further cell damage. Cytokines such as IL-1β and TNF-α are present in the early stages of phagocytosis

(Chazaud, 2016), creating a pro-inflammatory phenotype and playing a role in the initial breakdown of damaged muscle tissue (Peake, *et al.*, 2005).

Pro-inflammatory cytokines are said to be present in muscle cells for up to five days (Peake, *et al*., 2005), however following the phagocytosis of the damaged and necrotic tissue macrophages are able to change their phenotype to create an anti-inflammatory environment (Owens, *et al*., 2019; Chazaud, 2016; Peake, *et al*., 2005). Cytokines such as transforming growth factor-1β (TGF-1β), insulin-like growth factor-1 and IL-10 are involved in the promyogenic effect of anti-inflammatory macrophages (Chazaud, 2016), and the subsequent regeneration of muscle fibres.

### 2.1.1.5 *Regeneration*

The process of regeneration and repair is activated as a direct result of the inflammation of cells following EIMD, with Peake and others (2017a) suggesting that the notion that inflammation is a key process in muscle repair and regeneration is gaining acceptance. This is supported by Peake and co-workers (2017b), stating that reducing inflammation impedes the process of muscle repair and recovery, whilst Chazaud (2016) stated inflammation should not be considered a bad or detrimental process, and as such techniques utilised to impede the inflammatory process may impact on the muscle repair process.

Following the transition from a pro-inflammatory to anti-inflammatory environment, macrophages bearing an anti-inflammatory phenotype divide to reach huge numbers (Chazaud, 2016), with these macrophages responsible for the release of growth factors such as TGF-β1 (Owens, *et al*., 2019). The high number of macrophages exert a direct effect on myogenic precursors that regenerate myofibers (Chazaud, 2016), whilst satellite cells migrate as myoblasts, recruited due to the initial cause of muscle damage, fusing to form myotubes (Paulsen, *et al*., 2012). Kendall and Eston (2002) suggest that the stimulation of satellite cell activation may occur as a result of macrophage infiltration, whilst Paulsen and others (2012) suggest a high satellite cell presence may occur within 24 hours post-exercise. The high number of anti-inflammatory macrophages present throughout the regeneration process would appear to remain until significant growth of myofibres occurs (Chazaud, 2016), at which point the number of macrophages decreases and the muscle cell returns to homeostasis.

It has become an accepted theory that, following an initial bout of EIMD, muscle becomes tolerant to future eccentric exercise (Owens, *et al.*, 2019; Peake, *et al.*, 2017a), termed the ‘repeated bout effect’ (McHugh, *et al.*, 1999). Furthermore, Close and co-workers (2005) suggest skeletal muscle is able to adapt to the stresses of oxidative stress. Although the reasoning behind the repeated bout effect is not wholly understood, mechanisms have been postulated including neural adaptations, extracellular matrix (ECM) remodelling, and a modified inflammatory response (Hyldahl, *et al.*, 2017). Hyldahl and colleagues (2017) suggest that following an initial bout of eccentric activity, an increased motor unit synchronisation may be observed in subsequent eccentric bouts, displaying a strategy of the central nervous system to promote coordination between synergist muscles, as such reducing the muscle damage inflicted. Furthermore, adaptations to the ECM via increased collagen expression and TGF-β post-eccentric exercise providing a greater buffering effect of the ECM to myofibers by increased passive tension, and a more targeted acute inflammatory response following a secondary bout of eccentric exercise in comparison to a primary bout, accelerating the recovery speed of damaged muscle, may all contribute to the repeated bout effect (Hyldahl, *et al.*, 2017). Hyldahl, *et al*. also suggested that the greater the potential for muscle damage in the initial bout, the greater the repeated bout effect, and as such it can be expected that untrained individuals experience a greater repeated bout effect than elite, trained athletes potentially due to a lower initial muscle damage elicited. It has also been suggested that children are less susceptible to muscle damage than adults (Chen, *et al*., 2014), therefore reducing the possible repeated bout effect post-exercise, and therefore it can be expected that elite, youth athletes experience a reduced repeated bout effect. It may also be possible that due to the training status of elite athletes, and the reduced repeated bout effect they experience, that the repeated bout effect has a limit beyond which it does not provide a protective mechanism, which is an area that requires future research.

## 2.2 Muscle Recovery

The regeneration stage of the muscle damage and repair model (Figure 2.1) demonstrates the natural course of events in the muscle recovery process following EIMD. It has been suggested that the unaided return to muscle homeostasis of eccentrically damaged muscle tissue can take 7-9 days (Baired, *et al*., 2012), however during in-season elite soccer, competitive games typically occur on a weekly basis, and in some cases may occur more frequently. As physical conditioning, technical and tactical preparation is often required prior to soccer matches, and is demanded by coaching staff, there is a need to assist in the acceleration of the muscle recovery process. It may be argued that the use of recovery interventions aimed at minimising muscle oedema immediately post-exercise and the effects of resulting inflammatory response, hinder the natural recovery process of skeletal muscle (Peake, *et al*., 2017b), however in times of fixture congestion and high training loads, such recovery interventions may be implemented to prevent cumulative muscle damage and potential soft tissue injury, allowing for optimal physical performance.

### 2.2.1 Recovery Aids

Throughout literature, many muscle recovery aids have been discussed, compared and evaluated, however a review of all recovery methods and their merits and limitations is beyond the scope of this thesis. This section of the report will discuss muscle recovery interventions static stretching, active recovery and cold-water immersion which are commonly used in elite youth soccer (Nédélec, *et al*., 2013), the mechanisms behind their use, and the effects they may have as reported in research.

### 2.2.1.1 *Static Stretching*

Static stretching (SS) is an intervention previously used in sports to aid muscle recovery from exercise. The process involves elongating skeletal muscle into a stretched position and holding statically for a prescribed period of time. As a result of its popularity, research assessing its effects on recovery and performance has been conducted (Kinugasa & Kilding 2009; Montgomery, *et al*., 2008; Dawson, *et al*., 2005; Jayaraman, *et al*., 2004; Smith, *et al*., 1993; Buroker & Schwane, 1989). It has been suggested that the use of static stretching following exercise may have beneficial effects on muscle recovery and reducing DOMS, with researchers recommending its implementation (Wessel & Wan, 1994). Wessel and Wan (1994) suggested that static stretching may relieve the muscle spasms described in de Vries' muscle spasm theory (de Vries, 1966; Cheung, *et al*., 2003), and the use of static stretching may assist in the dispersal of muscle oedema caused as a result of muscle activity (Lund, *et al*., 1998). As aforementioned, muscle inflammation may promote further cell damage by increased leukocyte infiltration and ROS generation, potentially resulting in lipid peroxidation of healthy muscle cells and exacerbating skeletal muscle damage (Sachdev & Davies, 2008; Close, *et al*., 2005). Furthermore, the oedema that occurs as a result of exercise has been suggested to increase perceived muscle soreness and the sensation of DOMS, potentially due to increased sensitisation of nociceptors due to inflammation (Owens, *et al*., 2019).

As mentioned by Cheung and others (2003), static stretching has been recommended as an intervention to reduce muscle soreness (Wessel & Wan, 1994). This early research has been supported by the more recent findings of Dawson and co-workers (2005) when comparing the effects of static stretching on semi-professional Australian league footballers. Dawson and others (2005) reported a significant improvement in peak power determined by a six second maximal cycle sprint when comparing the effects of static stretching to a control of no recovery intervention. These findings may support the use of static stretching as a recovery intervention; however, the findings may not be directly applicable to the elite professional, in particular elite soccer players due to their use of semi-professional participants. Additionally, the participants of Dawson and colleagues (2005) were males aged 24.2 ± 2.9 years, and therefore these results may not reflect the true outcomes of elite youth soccer players. Furthermore, the baseline assessments of peak power were taken 45 hours prior to competitive performance, and no assessments were taken immediately following exercise. As a result, it is difficult to determine the effects of the soccer game on muscle damage, and whether the soccer match was sufficient in inflicting significant muscle damage.

Research of Kinugasa and Kilding (2009) compared the effects of static stretching, contrast water therapy, and cold-water immersion and active recovery on muscle damage in young male footballers (14.3 ± 0.7 years) following three, 90 minute competitive soccer matches. Their findings show no significant difference as a result of static stretching when compared to a control intervention on recovery at 24 hours post-exercise. Interestingly, they also found no significant difference between the three recovery interventions, suggesting static stretching was no less effective than contrast water therapy or a combination of cold-water immersion and active recovery. Despite finding static stretching to be no less effective than alternative recovery interventions, the participants used were non-elite, and therefore these results may not be directly applicable to an elite youth population. Additionally, a number of assessment methods of muscle damage were subjective (comprising total quality recovery (TQR) scale, perceived thermal sensations and perceived leg heaviness), and as a result the published findings must be considered carefully before making assumptions of the effectiveness of these recovery interventions as an element of bias in the reporting of muscle soreness may present itself with all subjective assessments. Finally, the participants were required to complete three competitive soccer matches within one week, and an average of all pre, post, and post-recovery assessments of muscle damage for all three games calculated and used in the statistical analysis. Baired and colleagues (2012) suggest muscle recovery can take up to 9 days, and as a result, it may be hypothesised that baseline measures for assessments of muscle damage used by Kinugasa and Kilding (2009) would increase for games two and three following the initial bout of exercise due to reduced recovery time, also increasing muscle damage assessments post-match, and potentially reducing the impact of recovery interventions. This would therefore suggest participants may experience an element of cumulative muscle damage which may alter the overall findings from this study. Gill and others (2006) support this, reporting indicators of muscle damage are yet to return to baseline at 84 hours post-match despite the implementation of recovery interventions.

There has been limited research (Delextrat, *et al*., 2014; Dawson, *et al*., 2005) conducted that supports the use of static stretching following exercise for the reduction of muscle damage, however a number of reports have been published that suggest its use provides no recovery benefit (Calleja-González, *et al*., 2016; Kinugasa & Kilding, 2009; Montgomery, *et al*., 2008). When considering the non-elite population, review reports taking into consideration between five and ten studies using healthy participants published findings of no significant effect of static stretching post-exercise on muscle soreness (Herbert, *et al*., 2011; Anderson, 2005; Herbert & Gabriel, 2002). This would suggest that the static stretching intervention provides no recovery benefits to the non-elite population. These review studies used subjective assessments as markers of muscle soreness and therefore may be subject to bias opinions. Additionally, the level of quality in one review article is reported as moderate (Herbert & Gabriel, 2002), leading to doubts over the validity of their findings.

In support of these findings by Anderson (2005), Herbert and Gabriel (2002) and Herbert, and co-workers (2011), Jayaraman and others (2004) compared the effects of static stretching and heat therapy using magnetic resonance imaging (MRI) evaluation on 32 males from a university community following eccentric knee extension contractions to failure. Their findings suggest that static stretching had no effect on muscle damage or recovery following eccentric exercise. Despite the findings being consistent with many other research articles (Herbert, *et al.*, 2011; Kinugasa & Kilding, 2009; Montgomery, *et al.*, 2008; Anderson, 2005; Herbert & Gabriel, 2002), it must be considered that the static stretching protocol was initiated at 36 hours post-completion of eccentric exercise, and as a result, any benefits that may be observed could be nullified due to a reduction in time for potential recovery benefits to occur. Additionally, the population used were untrained individuals of university age, and therefore findings may not represent those of elite youth soccer players, as it can be expected that athletes of an elite population are less susceptible to eccentrically induced muscle damage due to previous exposure (Owens, *et al.*, 2019; Peake, *et al.*, 2017a; Close, *et al.*, 2005). Finally, when considering the application of static stretching in an applied environment, practitioners may wish to implement interventions that have proven effective within applied settings, for example muscle damage inflicted due to competitive soccer matches. The eccentric activity utilised by Jayaraman and co-workers (2004) does not necessarily replicate the muscle damaging effects of soccer matches and therefore caution must be taken when considering disregarding the use of static stretching in an applied soccer setting based on these findings.

In agreement with the study produced by Jayaraman and others (2004), Montgomery and co-workers (2008) also suggest static stretching provides no significant improvement on recovery when assessing 29 male basketball players (19.1 ± 2.1 years) compared to pre-competition values. Participants used in this study were trained basketball players regularly competing in state competitions. Again, these findings represent those of the majority of research articles on the static stretching effect on muscle recovery, however it does have its limitations when comparing them to the effects of static stretching in elite youth soccer players. Firstly, the study was conducted on a tournament basis of three games in three days and therefore the results are more representative of cumulative muscle damage rather than the typical effects of static stretching. Additionally, the physical outcomes of basketball are significantly different to those seen in competitive soccer – firstly the game time is much shorter in basketball, and physical outcomes such as sprints, accelerations and decelerations are likely to differ to soccer due to the smaller playing area and higher intensity of the sport. Although research arguing against the use of static stretching as a recovery intervention exists, Dadebo and others (2004) reported English Premier League football clubs dedicate almost 40% of training time to flexibility training, with the most common technique being static stretching, although this study did not specify the context in which static stretching was implemented (warm-up or cool-down), demonstrating its use in applied settings.

Despite the vast amounts of research into the effects of static stretching on muscle recovery, it would appear that there is limited research into its effects on elite youth soccer players and therefore a need for soccer focused research, especially given its prevalence in post-match recoveries. It is also apparent that static stretching is a technique often used post-match in elite youth soccer as a recovery intervention, yet with a dearth of applied research in this area, there is a need to establish a relationship between research and practice.

#### 2.2.1.2 *Active Recovery*

A widely accessible recovery intervention, active recovery (AR) is often implemented in cool-downs post-exercise to assist in the process of muscle recovery (Peake, *et al.*, 2017b; Reilly & Ekblom, 2005). Comprising low-intensity exercise, active recovery can be completed using a variety of exercise modes, including light aerobic jogging, and cycling. Although still not completely understood, it has been reported active recovery assists in the muscle recovery process by restoring glycogen stores to their pre-exercise state (Darani, *et al.*, 2018), whilst accelerating the clearance of metabolic by-products following exercise (Martin, *et al.*, 1998) via increased cardiac output and limb blood flow (Peake, *et al.*, 2017b) following the cessation of exercise, reducing the potential pooling effects of pro-inflammatory cytokines, ultimately preventing further cell damage (Chazaud, 2016).

It has been suggested that the use of AR post-exercise is more efficient than static stretching (Reilly & Ekblom, 2005) particularly when comparing the clearance rate of blood lactate (Spierer, *et al.*, 2004; Monedero & Donne, 2000). As reported by Martin and others (1998) this may be due to AR increasing metabolism and systemic blood flow expediting the metabolism of lactate. This hypothesis is supported by Cochrane (2004), suggesting AR may assist in the clearance of lactate post-exercise, although may depend on numerous factors including AR exercise intensity. Despite support for the use of AR as an intervention in the clearance of metabolic by-products, specifically lactate, it may be argued that lactate is not an indicator of muscle damage, and rather is a more accurate indicator of muscle fatigue (Monedero & Donne, 2000). Nevertheless, the mechanisms presented may have some merit, however further research may be required to support these claims. In order to determine whether the clearance of metabolic by-products is key to the recovery effects of AR, such by-products involved as a result of exercise that indicate muscle damage should be assessed.

To date, research has shown that AR is an efficient method for assisting in muscle recovery following exercise (Peake, *et al.*, 2017b; Tessitore, *et al.*, 2007; Gill, *et al.*, 2006; Reilly & Ekblom, 2005; Coffey, *et al.*, 2004; Spierer, *et al*., 2004). Tessitore and colleagues (2007) reported significantly reduced muscle soreness upon completing an AR intervention in comparison to passive recovery and shallow-water aerobic exercise following soccer training of 12 elite male soccer players (18.1 ± 1.2 years), however no performance differences were determined. These findings suggest AR may provide recovery benefits for reducing perceived soreness of elite soccer players following training, but as this study did not assess its effects following competitive soccer matches, caution must be taken when considering its transferability to soccer match related muscle damage which is likely to induce greater muscle damage than training sessions.

In comparison, Gill and co-workers (2006) reported a significant improvement in recovery after 84 hours (88.2%) when comparing results of an indirect biomarker of muscle damage, creatine kinase (CK), following AR to a control of passive recovery in elite rugby players post-competitive rugby matches. Participants (25 ± 3 years) were highly trained, suggesting these findings are transferable to alternative elite sports such as soccer where similar recovery effects may be found. However, the recovery effects reported in this study were found at 84 hours post-match and it has previously been suggested that the effects of DOMS would peak at 48 hours post-exercise (Kendall & Eston, 2002) and as a result the natural repair processes of the damaged muscle may contribute to the 88.2% recovery reported in this study. Additionally, it may be suggested that the physical effects of elite adult rugby matches are significantly different to elite youth soccer, and so comparisons between studies are unreliable. Finally, the use of adult participants would suggest that these findings are not directly transferable to elite youth soccer players, despite the reported successful implementation of active recovery for improving recovery time.

Despite the findings of these studies suggesting active recovery may be a useful intervention to reduce muscle soreness and DOMS post-exercise, the dearth of research of applied studies would suggest that there is a need for future assessment of its effects in practical settings. Additionally, there is limited research assessing the effects of active recovery following elite soccer matches, and in particular elite youth soccer matches, again emphasising the necessity for future research.

#### 2.2.1.3 *Cold Water Immersion*

Cold water immersion (CWI) is a cryotherapy technique involving the immersion of skeletal muscle in cold water conditions for a prescribed period of time. Numerous physiological mechanisms have been proposed suggesting possible benefits of CWI, including the acceleration of venous return due to vasoconstriction inflicted by exposure to cold temperatures (Cochrane, 2004) and hydrostatic pressure (Ingram, *et al.*, 2009), reductions in perceived pain due to dampening of neural signalling (Broatch, 2015), and decreased inflammation therefore reducing osmotic pressure of exudate on pain signalling nociceptors (Broatch, 2015). Furthermore, it is plausible to suggest that the reduction in oedema due to the aforementioned mechanisms of CWI, may reduce the potential damaging effects of inflammation such as lipid peroxidation of healthy cell membranes (Close, *et al.*, 2005) caused by phagocytosis (Kendall & Eston, 2002) and the production of ROS (Close, *et al.*, 2005). It has also been suggested that CWI will reduce capillary permeability due to vasoconstriction, limiting the acute inflammatory response (Broatch, 2015), potentially reducing the unwanted leakage of ROS and neutrophils to healthy cells (Close, *et al*., 2005) and preventing further cell damage.

As with AR, many CWI protocols exist, and can differ in submersion time and water temperature. Machado and colleagues (2016) completed a meta-analysis suggesting CWI significantly improved immediate and delayed recovery effects when compared to passive recovery. The analysis also suggested water temperatures between 11 and 15$^{o}$C, and submersion times between 10 and 15 minutes were most effective for both immediate and delayed recovery effects.

Research concerning elite youth soccer players, implementing CWI intervention protocols that fall within the recommended guidelines of 10-15 minutes submersion time in water temperature of 11-15$^{o}$C as suggested by Machado and other (2016) are limited. Ingram and co-workers (2009) reported a significant improvement in performance indicators at 48 hours post-exercise as a result of CWI when compared to contrast water therapy (CWT) and passive recovery, when implementing a CWI protocol comprising a submersion time of 2x5 minutes at 10$^{o}$C. However, the application of this study to elite youth soccer players presents limitations, as the participants used by Ingram, *et al.*, (2009) consisted of non-elite participants, whilst an exhaustive exercise protocol was used, which cannot exactly replicate competitive soccer matches of elite youth athletes. Peake and others (2017a) suggested that mechanical stress as a result of eccentric exercise may cause opening of stretch-activated channels, allowing calcium to enter the cytosol causing activation of calpain enzymes and the resulting degradation of contractile proteins leading to a reduction in force production (Morton, *et al*., 2005). It may be possible that the improvements in performance indicators of muscle damage as a result of CWI in comparison to CWT and passive recovery interventions can be attributed to a reduced calcium efflux in to the cytosol due to greater vasoconstriction (Cochrane, 2004) reducing inflammation and improving venous return, potentially limiting the effects of the loss of calcium homeostasis (see autogenic processes section). However, the calcium concentrations, stimulation of calpain enzymes and degradation of contractile proteins were not measured by Ingram and others (2009) and may be an area for future investigation.

In support of the findings of Ingram and colleagues (2009), Elias and others (2013) reported a significant reduction in perceived soreness, and significantly improved performance measures at 48 hours post-match following a CWI protocol consisting of 14 minutes submersion in 12˚C water when compared to CWT and passive recovery interventions. Their findings support the CWI protocol recommendations (10 to 15 minutes submersion in 11-15˚C water) suggested by Machado and colleagues (2016), although limitations arise when considering the transferability of this study to elite youth soccer players, as differences in physical load of Australian professional football and elite youth soccer would likely alter the muscle damage elicited.

Although Machado and co-workers (2016) suggest greater recovery benefits are achieved when CWI protocols fall within recommended optimal ranges for temperature and time, it is possible that studies concerning youth soccer players that implement protocols outside of Machado’s recommended ranges may result in similar positive effects. Rowsell, *et al.*, (2009) reported a significant improvement in perceived soreness when comparing a CWI protocol (consisting of 1 minute submersion at 10˚C followed by 1 minute seated at room temperature repeated 10 times) to a thermoneutral protocol, over a 4 day soccer tournament using 20 high-performance male soccer players. Despite the improvement in perceived soreness, no other significant findings were identified when analysing countermovement jump (CMJ) height, repeated sprint ability, CK, interleukin-1b, interleukin-10, interleukin-6 and myoglobin. It is possible that due to participants being subjected to four 90 minute soccer matches in four consecutive days, that physiological and performance improvements were unaffected by recovery interventions due to cumulative fatigue and muscle damage.

Ascensão and others (2011) compared the effects of a CWI intervention (10 minutes submersion at 10˚C) to a thermoneutral water immersion intervention following a one-off soccer match using male, junior national league players. Results demonstrated a significant reduction in CK at 24 hours and 48 hours post-match, and a significant reduction in C-reactive protein (CRP) at 24 hours post-match as a result of CWI, supporting its use as a recovery intervention. Ascensão and others (2011) also reported a significant improvement in maximal isometric voluntary contraction, and perceived calf and quadriceps soreness at 24 hours post-match when comparing CWI to thermoneutral water immersion. No significant differences were found between squat jump, CMJ and sprint performance. The findings of improved perceived soreness may be attributed to a reduced firing rate of pain sensory receptors in the skin after cooling, and therefore a reduced sensation of pain (Broatch, 2015). Additionally, an increased vasoconstriction as a result of CWI may result in reduced inflammation and osmotic pressure of exudate, therefore reducing pressure on pain signalling nociceptors, further reducing perceived pain (Broatch, 2015).

It may also be possible that, unlike alternative recovery interventions, the mechanisms of CWI may remain even upon cessation of the recovery intervention, providing additional recovery benefits. As suggested by Peiffer and others (2009), CWI results in a reduction in femoral venous diameter, increasing vasoconstriction and possibly accelerating the clearance of metabolic by-products of exercise. However, unlike alternative interventions where the recovery mechanism stops on completion of the protocol, Peiffer and colleagues (2009) report a significant reduction in femoral venous diameter for up to 45 minutes post-intervention, at which point sample collection was terminated. This may suggest that although the intervention itself has concluded, the mechanisms of vasoconstriction remain for a substantial period of time, providing an enhanced recovery benefit in comparison to alternative interventions.

## 2.3 Summary

Despite the substantial research (Tessitore, *et al*., 2007; Ingram, *et al*., 2009; Rowsell, *et al*., 2009; Ascensão, *et al*., 2011; Bahnert, *et al*., 2013; Machado, *et al*., 2016) into recovery interventions, and their effects on muscle recovery following exercise-induced muscle damage, there remains limited scientific evidence of such effects on elite youth soccer players following competitive soccer matches, and further research is therefore required to strengthen the knowledge of the industry and support current practice. Additionally, some research assessing the effects of recovery interventions has considered eccentric exercise protocols, however as yet, there is no evidence to suggest these exercise protocols induce muscle damage similar to that expected from elite youth soccer matches, and consequently it cannot be said with confidence that the findings from such research are transferable to applied, elite soccer environments. It is therefore the purpose of this thesis to investigate to what extent isolated eccentric exercise protocols induce muscle damage in comparison to elite youth soccer matches, the physical parameters of elite youth soccer matches that are correlated to markers of muscle damage, and the effects of recovery interventions following elite youth soccer.

# 3 General Methodology

## 3.1 Introduction

The present chapter describes the participant cohort, general methodologies, and rationale for equipment and protocols used within this thesis. For clarity, this chapter includes an overview of all protocols and equipment used throughout the entire thesis, whilst each individual study (chapters 4 to 7) contain the relevant information reported in this chapter so the study can be considered in isolation.

## 3.2 Participants

A total of 38 athletes (U16/U17 age groups) from an elite soccer academy of an English Premier League soccer team (Tottenham Hotspur Football Club) were voluntarily recruited. Due to the elite level of participants and the limited access to such participants for research purposes, small sample sizes of individual studies were considered acceptable, and in line with previous literature (Redden, 2019). Data was collected across three competitive seasons initiating at the very beginning of the 2015-16 season and terminating following completion of the 2017-18 season. The physical characteristics of participants concerned with each study are contained within each individual chapter. Prior to participation in the research project, all players were given an information sheet explaining the procedures, and all provided written informed consent form. Due to the age of the participants, parental consent was also obtained (see appendices). All players recruited were playing elite academy soccer for a minimum of 12 months prior to testing. The research ethics committee of Kingston University approved all studies and procedures (ethics registration number: 1415/016), and all procedures were conducted in accordance with the Declaration of Helsinki.

## 3.3 Experimental Design

Development of research for applied, elite environments often utilise non-elite participants and simulated exercise protocols to replicate the expected physical conditions of elite settings, from which, practical recommendations on interventions are made. As such, applied research assessing elite youth soccer players is lacking, and therefore this sample population was selected for the assessment of muscle damage and muscle recovery. The access to these participants as granted by the soccer club due to the researcher’s role within the club’s sport science department allowed for an assessment of muscle damage sustained following soccer matches, a comparison of damage sustained via soccer matches to eccentric exercise protocols, and establishing an understanding of what physical components of elite youth soccer matches contribute to muscle damage, and recovery interventions to aid in the recovery process.

## 3.4 General Procedures

### 3.4.1 Testing Environments

As previously established, much research to date has assessed participants in non-applied, laboratory-based settings using non-elite athletes and simulated protocols to replicate the environments experienced in elite applied settings (Bieuzen, *et al.*, 2013; Hill, *et al.*, 2013; Leeder, *et al.*, 2012). Findings from such research often provide recommendations on interventions for the elite, applied settings, yet it is likely that the physical exposure and stress of the elite athletes in their natural training and competition environment is not replicated in such non-applied research. In order to address these issues, and to assess elite athletes in their natural environment, it was necessary for the studies of this thesis to reflect the applied settings.

#### 3.4.1.1 *Eccentric Protocol*

The eccentric exercise protocol was completed in a Premier League soccer club rehabilitation gym using a CSMi Humac Norm Dynamometer (Stoughton, MA, USA), and conducted by a trained physiotherapist. For consistency and to minimise assessor error, the same assessor was used for all eccentric exercise trials (Hopkins, 2000). All pre- and post-exercise assessments of muscle damage were completed within the rehabilitation gym and were taken by the same sports scientist for consistency and to minimise statistical error.

#### 3.4.1.2 *Soccer Match-Play*

In order to reduce disruption to the soccer season and to ensure a controlled, applied research project, soccer matches were completed at ‘home’ (Tottenham Hotspur Training Centre) and ‘away’ (Category 1 academy training centres) depending on where the soccer match was scheduled to be played. It was decided that home fixtures were utilised to assess recovery interventions (active recovery and cold-water immersion; chapter 7) where access to gym and pool facilities were required, whilst away fixtures were utilised for the assessment of recovery interventions requiring no equipment (static stretching and passive recovery; chapter 4 and 5). Both home and away fixtures were considered for the assessment of physical parameters inducing muscle damage (chapter 6), as the requirements of recovery interventions was removed.

### 3.4.2 Equipment and Procedures

#### 3.4.2.1 *Assessment Methods*

The development of research on muscle damage and the effects of eccentric and unaccustomed exercise has led to substantial assessment of objective markers of muscle tissue injury. Due to the assessment of muscle fibres, muscle biopsies can provide a direct indicator of muscle damage, however their use in elite sport is impractical and has limitations. Peake *et al.* (2005) report the local inflammatory response to exercise can be similar to that of the biopsy alone, whilst Peake and others (2017a) support their findings suggesting a local inflammatory response may be caused by repeated muscle biopsies. Further limitations to muscle biopsies exist, with Peake and colleagues (2005) suggesting measurements of leukocyte numbers and cytokines in blood may be easier than immunohistochemically staining and gene expression of muscle samples, whilst blood sampling is less invasive.

Due to the potential detrimental effects of muscle biopsy on elite athletes, many applied studies have looked to use more practical, indirect objective assessments of muscle damage, such as myoprotein assessment (Scott, *et al*., 2016; Thorpe & Sunderland, 2012; Ascensão, *et al*., 2011; Duffield, *et al*., 2010; Ingram, *et al*., 2009; Gill, *et al*., 2006), exercise performance (Bahnert, *et al*., 2013; Ascensão, *et al*., 2011; Rowsell, *et al*., 2009; Ingram, *et al*., 2009; Kinugasa & Kilding, 2009; Tessitore, *et al*., 2007), range of motion (Lavender & Nosaka, 2008; Dawson, *et al*., 2005), and muscle oedema (Ingram, *et al*., 2009; Lavender & Nosaka, 2008; Montgomery, *et al*., 2008). Despite these markers being indirect, their use in a vast number of studies allows a broad comparison of the effects of exercise on muscle damage. Additionally, when using these methods in applied settings with elite athletes their explanation, simplicity, and non-invasiveness is more likely to be preferable than muscle biopsy.

#### 3.4.2.1.1 *Countermovement Jump Performance*

Countermovement jump (CMJ) performance has been used extensively throughout research as an indirect marker of muscle damage and muscle recovery (Bahnert, *et al*., 2013; Ascensão, *et al*., 2011; Kinugasa & Kilding, 2009; Rowsell, *et al*., 2009). Jump performance has been shown to be a reliable assessment of lower body strength and power (Redden, *et al*., 2018; Nuzzo, *et al*., 2011), and as such, a reduction in jump performance post-exercise may indicate a loss of strength and muscle function due to muscle damage.

Applied studies using CMJ performance as an indicator of muscle damage in soccer have revealed reduced CMJ performance as a result of competitive soccer matches when compared to baseline measures (Ascensão, *et al*., 2011; Kinugasa & Kilding, 2009; Rowsell, *et al*., 2009). Although the exact mechanisms may not yet be identified, it is possible that as a result of eccentric actions performed in soccer, myofilaments are forced into a state of no overlap (Peake, *et al*., 2017a) resulting in a loss of strength due to a reduced capacity for fibre recruitment. It is also possible that as a result of opening of stretch-activated channels due to eccentric muscle contractions, the influx of calcium stimulates calpain enzymes leading to degradation of contractile proteins, further reducing physical performance (Peake, *et al*., 2017a). These theoretical suggestions, alongside the reliability of jump performance for demonstration of lower limb strength and power and its use in previous applied soccer research resulted in the use of countermovement jump performance as an indirect assessment tool of muscle damage in this thesis. It was also considered necessary to allow arm-swing whilst performing the countermovement jump (CMJA), as this was deemed more sport specific (Richter, *et al*., 2012), limiting systematic error (Hopkins, 2000).

#### 3.4.2.1.2 *Muscle Oedema*

Muscle inflammation and oedema has been suggested to be present following physical exercise due to the increase in blood flow to damaged muscles (Chazaud, 2016; Peake, *et al*., 2005; Kendall & Eston, 2002; Sjøgaard, *et al*., 1985), however this alone does not provide a possible explanation for the suggested increase in muscle swelling as a result of eccentric and unaccustomed exercise. As aforementioned, the body’s immune system reacts to the eccentric and unaccustomed muscle damaging exercise by mobilising neutrophils, lymphocytes and killer cells as a protective mechanism (Peake, *et al*., 2005). Additionally, the accumulation of neutrophils, and the arrival of macrophages has been suggested to produce free radicals which promote the inflammatory response of the injured area (Peake, *et al*., 2017a). Peake and others (2005) suggest that the mobilisation of these cells is in response to the damage caused to muscle fibres as a result of exercise, and assists in the regeneration of muscle cells. As an increase in eccentric and unaccustomed exercise may lead to an increase in muscle damage and therefore the mobilisation of these protective cells by the immune system – muscle oedema has previously been used as an indirect marker of muscle damage (Montgomery, *et al*., 2008).

Previous research has assessed limb circumference with a constant tape measure to determine muscle oedema post-exercise, and averages of measured points given as the circumference (Lavender & Nosaka, 2008). An increase in muscle damage leading to inflammatory responses has shown to increase limb circumference significantly following eccentric exercise (Lavender & Nosaka, 2008). The increase in limb circumference following eccentric exercise would suggest that this is a suitable assessment of muscle oedema. To date, no study has assessed the effects of competitive soccer matches on limb circumference, and it may be recommended for future research as an indirect marker of muscle damage and intracellular inflammation.

#### 3.4.2.1.3 *Creatine Kinase*

Creatine kinase (CK) is a myoprotein commonly used in research assessing muscle damage and recovery interventions. It is an enzyme that is significant to the performance of muscle cells. CK is an important catalyst in the Phosphocreatine (PCr) cycle, and plays a critical role in the production and maintenance of energy supply to respiring muscle cells (Baird, *et al*., 2012). As a result, an increase in muscular activity due to exercise leads to increased CK levels in muscle cells. A rise in CK levels in blood serum has been suggested to be linked to the level of muscle damage sustained from exercise, and has therefore been used as an indirect marker of muscle damage throughout literature (Ascensão, *et al*., 2011; Magal, *et al*., 2010; Saka, *et al*., 2009; Clarkson, *et al*., 2006; Gill, *et al*., 2006; Gunst, *et al*., 1998).

Despite its use throughout literature, findings on CK as a marker of muscle damage have been inconclusive. Large inter-variability of CK levels have been reported (Magal, *et al*., 2010) reducing the reliability of comparisons between participants, however it may be possible to analyse CK levels of participants individually for a valid assessment of muscle damage. Additionally, modes of exercise, gender, genetic predispositions and training status are all factors that have been suggested to affect the level of CK generated as a result of exercise (Lee & Clarkson, 2003). Nevertheless, a number of studies have used CK as an indirect marker of muscle damage, with results showing significant increases in CK following exercise (Ascensão, *et al*., 2011; Ingram, *et al*., 2009; Gill, *et al*., 2006).

When studying the effects of post-match recovery interventions on elite male rugby players (25 ± 3 years), Gill and others (2006) report a significant increase in CK levels when comparing pre- (308.3 U/L) and post-competition (833.7 U/L) blood CK values. In comparison to those results, Ingram and co-workers (2009) report a significant increase from pre- (166 U/L) to post-exercise (410 U/L) CK levels following simulated team sport performance in non-elite males of a similar age (27.5 ± 6 years). As aforementioned, research has criticised the use of CK as a marker of muscle damage due to its inter-individual variability and therefore comparisons between the two studies are difficult. However, the elevated CK level reported in the study of elite male rugby players in comparison to the non-elite athletes may be a result of cumulative muscle damage from pre-competition training from an elite environment. Additionally, comparisons in the post-exercise CK levels may be attributed to the higher intensity at which elite sport is played, resulting in elevated CK levels. When comparing the results of Gill and others (2006) to research assessing the effects of soccer matches on CK levels of elite soccer players (18 ± 2 years) in a study by Ascensão and others (2011) reporting pre- (~120 U/L) and post-competition (~400 U/L), again CK levels reported by Gill and colleagues of both pre and post-exercise are much higher. Once more, comparisons between studies are difficult to make due to the suggested inter-individual variability of CK, although it may be hypothesised that the greater physical exertion of rugby could lead to higher muscle damage, and therefore increased CK levels in comparison to others sports such as soccer. In addition, it has been reported that differences in age may alter CK levels observed and may have influenced results in both studies compared above. It is common practice in these studies to compare a pre- and post-exercise CK value in addition to values collected 24 to 48 hours following exercise. In order to reduce the inter-individual variability, it may be advised that future research uses percentage recovery (as used by Gill, *et al.*, 2006) which would allow comparisons between studies to take place. As such, percentage change in CK levels recorded was used in the present thesis (Chapter 5 & 7).

#### 3.4.2.1.4 *Perceived Soreness*

All participants were required to provide a subjective assessment of lower limb muscle soreness at 2.5 hours prior to exercise, immediately after exercise, and 48 hours post-exercise. All assessments were recorded privately on an online system (The Sports Office, Wigan, UK) which was password protected and not accessed until the game had been completed to give the participants assurance that their perceived muscle soreness would not influence their selection in competitive matches. Perceived muscle soreness (PMS) was indicated using a 10-point visual analogue scale (VAS) from 0.5 to 5, with 0.5 increments. Players were familiar with this method, having previously completed muscle soreness assessments as a requirement of the soccer club, thus minimising error.

### 3.4.2.2 *Establishing Reliability*

Reliability refers to the ability to reproduce an observed value in repeat trials. A greater reliability reduces variability in observed measures (Hopkins, 2000), reducing the probability of false positive findings. The two main factors, as described by Hopkins (2000), affecting the reproducibility of a measure are random error and systematic change. Random error or ‘noise’ of a measure alters the means of trials, and as such, greater sample sizes reduce random error by cancelling out noise when calculating means. Systematic change, also referred to as systematic bias is a non-random change in a means between two trials and can be observed in trials where a learning or training effect is present (Hopkins, 2000). It may also be evident in trials where performance reduces due to fatigue or reduced motivation. Hopkins (2000) describes the three most important measures of reliability as within-subjects variations, changes in the mean, and retest correlation.

#### 3.4.2.2.1 *Within-Subjects Variation*

Hopkins (2000) argued that within-subjects variation is the most important measure of reliability for researchers as it affects the precision of estimates of change of a variable within a study – the greater the within-subjects variation, the less precise the estimates of change. Hopkins describes this as the most important type of reliability measure for practitioners and coaches using tests for monitoring performance in athletes. In applied settings such as elite soccer, the smaller the variation, the easier it becomes to measure a true change in performance. Within multiple tests, the standard deviation of an individual’s values represents the random variability between measures. This within-subjects standard deviation can be referred to as standard error and represents the typical error (TE) of a measurement (Hopkins, 2000). Hopkins describes the main source of TE as biological, such as alterations in physical and mental state, however equipment can also contribute ‘noise’, and a combination of biological and technological error is often unavoidable in experimental studies. It is, however, possible to reduce and minimise noise from equipment and changes in assessors, and as such throughout this thesis the equipment used, and assessors remained consistent to minimise TE. TE is often displayed as coefficient of variation (CoV) and is expressed as a percentage of the mean.

###### 3.4.2.2.2 *Change in the Mean*

Hopkins (2000) describes this measure of reliability as the change in means observed between two trials and consists of two components; random change and systematic bias. Random change has been suggested to be due to sampling error, and occurs inevitably from random measurement error (Hopkins, 2000). Systematic change is a non-random change between two trials such as a learning effect or reduced performance due to fatigue. Hopkins (2000) notes that the magnitude of change between individuals is likely to differ, with the differences making the test less reliable and increasing TE.

###### 3.4.2.2.3 *Retest Correlation*

Hopkins (2000) explains that retest correlation is a reliability measure that represents how closely related the values of one trial are to another. The closer the correlation coefficient is to 1, the greater the reliability, whilst a correlation coefficient approaching 0 shows less reliability.

###### 3.4.2.2.4 *Reliability of CSMi Humac Norm Dynamometer*

The eccentric muscle damaging protocol of this thesis (chapter 4) utilised the CSMi Humac Norm isokinetic Dynamometer to induce muscle damage. Previous research utilising such isokinetic dynamometers have demonstrated successfully inflicting significant muscle damage post-exercise (Tsatalas, *et al*., 2013; Hody, *et al*., 2013; McKinnon, *et al*., 2012; Tsatalas, *et al*., 2010; Saka, *et al*., 2009). Additionally, research of Habets and others (2018) assessing the reliability of the CSMi Humac Norm isokinetic dynamometer report 'excellent' intraclass correlation coefficient (knee extensor [R,L]: 0.849, 0.787; knee flexor [R,L]: 0.891, 0.840), and CoV (knee extensor [R,L]: 14.8, 19.3; knee flexor [R,L]: 13.0, 16.0), and as such the CSMi Humac Norm isokinetic dynamometer was deemed a suitable tool for eccentric exercise of the knee extensor and flexor muscle groups. In order to minimise assessor error, the same practitioner was used for delivery of all eccentric protocols for all participants. Additionally, to minimise systematic error, participants were required to have completed two prior exercise protocols using the CSMi Humac Norm dynamometer within 6 months prior to testing. Furthermore, all participants completed a unilateral warm-up for familiarisation, comprising three eccentric contractions of the knee extensor and flexor muscle groups of both legs prior to completing the eccentric protocol. Before testing the CSMi Humac Norm dynamometer was calibrated to minimise technological error.

#### 3.4.2.2.5 *Reliability of Smart Speed Jump Mat*

Previous research assessing the reliability of double leg jumps including countermovement jumps have demonstrated ‘good’ or ‘excellent’ retest reliability (Redden, *et al.*, 2018; Nuzzo, *et al.*, 2011) reporting a CoV between 1.8% and 6.0%, and intraclass correlation coefficient values between 0.88-0.93. Research of Lloyd and others (2009) assessing the Smart Speed jump mat (Fusion Sport, Australia) indicated a CoV of 12.88% for countermovement jump assessments, and an intraclass correlation coefficient value of 0.83. As such, the Smart Speed jump mat was deemed suitable for the assessment of countermovement jump performance.

#### 3.4.2.2.6 *Reliability of i-STAT Analyser*

This thesis assessed CK as an indirect marker of muscle damage, taken approximately 2.5 hours pre-exercise, immediately post-exercise and at 48 hours post-exercise. CK was measured using the i-STAT 1 Analyser (Abbott Point of Care, IL, USA) using fingertip blood samples. Although the i-STAT 1 analyser reported values of CK-MB, it was determined that in the absence of myocardial damage (Brancaccio, *et al.*, 2010), and as a result of prolonged, strenuous exercise (Brancaccio, *et al.*, 2010; Totsuka, *et al.*, 2002) and damage to skeletal muscle (Archer, 1982), increased CK-MM would be observed and reported, therefore indicating muscle damage. All assessments of CK were taken by the same assessor who had been trained in the use of the i-STAT 1 Analyser. CK results were recorded and results kept anonymously.

Previous research assessing the reliability of the i-STAT Analyser has done so for its use in clinical settings. Such studies have reported an ‘excellent’ (>0.8) intraclass correlation coefficient (Veldhoen, *et al.*, 2014), whilst Dascombe and others (2007) report ‘excellent’ (>0.77) intraclass correlation coefficient of the i-STAT Analyser following maximal exercise. These positive reliability assessments, and the portability of the i-STAT Analyser as required for assessment prior to, and immediately following ‘away’ soccer matches meant this assessment tool was deemed suitable for the assessment of CK.

Assessments of CK required a finger-tip blood sample (approximately 2-3 drops). A disposable lancet was used to pierce the skin, and capillary tubes were used to draw blood from the finger. A blood sample was then applied to the cartridge before being inserted into the analyser. Prior to completing the test, cartridges initiate a series of pre-set quality control diagnostics. Results were delivered in approximately 5 minutes, and assigned to player numbers to ensure anonymity.

#### 3.4.2.2.7 *Reliability of STATSports Viper and Apex GPS Units*

In order to ensure consistency in the physical load of soccer match exposure, athletes were required to wear global positioning system (GPS) units fitted with accelerometers (STATSports Apex/Viper). The GPS units reported movement variables across all axes, as well as reporting heart rate data, and have previously been shown to produce reliable GPS data (Beato, *et al.*, 2018a; Beato, *et al.*, 2018b). Due to evolution in the manufacturer’s GPS technology within the time-course of this thesis, two differing models (Apex and Viper) were used, however, to minimise error and increase reliability, studies were confined to using only one model type. Chapters 4 and 5 utilised the Viper GPS model, whilst chapters 6 and 7 utilised the Apex GPS model. Individual thresholds based on movement parameters were set according to the average of the combined variables for all full competitive matches completed. As a result, ‘match percentages’ were calculated, with the average of dynamic stress load (fatigue score calculated using player movements, steps and collisions), metres per minute, speed intensity, high metabolic load distance, high speed running distance (distance covered above 65% of an individual’s maximum speed), accelerations, decelerations, heart rate minutes above 85% of max., and heart rate exertion over all full games amounting to a match percentage of 100%. This match percentage determined the individual intensity of matches completed for all players. To ensure the intensity of each competitive soccer match remained consistent throughout the study, any participant whose match percentage for any particular game was ± 10% of their average match percentage for all completed matches were excluded from the data collection for that specific competitive match. Furthermore, participants were excluded from data collection for individual games if they failed to complete a minimum of 80% of the 80 minute match (Kinugasa & Kilding, 2009), whilst goalkeepers were excluded from the study due to the inability to control consistency in physical loading for matches.

### 3.4.3 Statistical Methods

Statistical analyses throughout this thesis were conducted using statistical software SPSS (versions 23 – 24, SPSS Inc., Chicago, IL, USA). Prior to statistical analysis, all data was investigated for normality using a Shapiro-Wilk test. Statistical significance was accepted at $p < 0.05$. The specific statistical analyses of individual studies are further detailed in the respective chapters.

#### 3.4.3.1 *Analysis of Variance*

For the majority of statistical analysis throughout this thesis, the main statistical test used was a repeated measures analysis of variance (ANOVA). Repeated measures ANOVAs are designed to determine if differences exist between two or more means of treatment conditions from a sample population. Field (2013) states that when repeated measures are used, the assumption that scores of different conditions are independent is violated because scores of repeated measures ANOVAs come from the same entity, and consequently we must assume that relationships between pairs of experimental conditions are similar – termed the assumption of sphericity. Field (2013) discusses how sphericity can be assessed using Mauchly's test which tests the null hypothesis that differences between conditions are equal, and as such, if Mauchly's test statistic is significant, the conditions of sphericity are not met and we conclude a significant difference between conditions. It is possible that when small sample sizes in studies are used, the likelihood of conditions of sphericity not being met increase due to type 1 error, however the degrees of freedom can be adjusted using the Greenhouse-Geisser correction (Field, 2013). Due to small sample sizes used within some chapters of this thesis, violation of sphericity was determined by Greenhouse-Geisser correction indicating statistical differences. In such cases, post-hoc analyses were performed to identify where the statistical differences between conditions occurred.

In cases where data was not normally distributed (chapter 5) and due to small sample sizes, a non-parametric statistical test was required. In such cases Friedman's ANOVA was used to detect differences in treatment conditions across multiple tests.

#### 3.4.3.2 *Post-Hoc Analysis*

As indicated by Field (2013), when sphericity is violated, the post-hoc Bonferroni statistical method is the most robust univariate technique, especially in terms of power and control of type 1 error, for determining where the differences lie. As such, this was the chosen post-hoc test in chapters 4 and 7. However, when statistical significance was identified in chapter 5, sample T-tests were used for post-hoc analysis.

### 3.4.3.3 *Effect Size*

Although a significant difference in measures can be identified by ANOVAs, and post-hoc assessments can indicate where the differences between measures lie, that significance does not tell us about the importance of an effect (Field, 2013). Therefore, we can measure the size of the effect termed effect size. Field (2013) describes this as a standardised measure of the magnitude of the observed effect, and can be used to compare between studies measuring differing variables. Partial eta squared ($\eta^2_\mathrm{p}$) is the proportion of variance that a variable explains, that is not explained by other variables (Field, 2013), where effect sizes are small (0.01), medium (0.006) or large (0.14) (Field, 2009). $\eta^2_\mathrm{p}$ was used to determine effect size in this thesis.

# 4 Elite Youth Soccer Matches Induce Greater Muscle Damage than Isolated Eccentric Exercise Protocols

## 4.1 Introduction

Exercise-induced muscle damage (EIMD) is the physiological alteration often associated with eccentric, high-intensity and unaccustomed exercise, resulting in delayed onset muscle soreness (DOMS) (Bieuzen *et al*., 2013; Lavender & Nosaka, 2008; Peake, *et al*., 2005; Byrne, *et al*., 2001; Clarkson, *et al*., 1992). Although the exact mechanisms of muscle damage are yet to be defined, research has demonstrated a significant reduction in performance markers (Roberts, *et al*., 2014; Elias, *et al*., 2013; Duffield, *et al*., 2010; Ingram, *et al.,* 2009; Howatson, *et al.*, 2009; Tessitore, *et al*., 2007; Dawson, *et al*., 2005), significant myoprotein deterioration (Ascensão, *et al*., 2011; Ingram, *et al.*, 2009; Howatson, *et al*., 2009; Rowsell, *et al*., 2009; Dawson, *et al*., 2005) and perceived soreness increase (Ascensão, *et al*., 2011; Ingram, *et al*., 2009; Rowsell, *et al*., 2009; Dawson, *et al*., 2005) following eccentric, unaccustomed exercise.

Owens and others (2019) suggest the mechanisms associated with muscle damage can be simplified to two stages: 1) the initial phases or primary muscle damage that occur as a consequence of mechanical work performed, and 2) secondary damage that exacerbates pre-existing muscle damage as a result of an inflammatory response. Although alternative theories suggest metabolic factors influence primary muscle damage, it would appear that mechanical factors play a more prevalent role (Proske & Morgan, 2001). Morgan and Proske (2004) suggest that during an eccentric contraction, weakening of sarcomeres occurs by non-uniform lengthening beyond their optimum length, with repetition of lengthening contractions leading to disruption of myofibril cross-bridges (Kendall & Eston, 2002), stretching myofilaments of weaker sarcomeres into a state of no overlap (Peake, *et al*., 2005). On completion of the eccentric action, the majority of myofilaments of overstretched sarcomeres return to their normal function however some fail to reconnect and become disrupted, with repeated eccentric bouts resulting in structural disruption (Peake, *et al.*, 2017a). Failure of the excitation-contraction (E-C) coupling process has also been suggested to contribute to the primary muscle damage phase (Hyldahl & Hubal, 2014), however it is generally acknowledged (Owens, *et al.*, 2019) that the initial events after eccentric contractions disrupt muscle fibre structure, resulting in membrane damage and E-C coupling dysfunction (Proske & Morgan, 2001). These initial phases have been suggested to be followed by secondary damage, characterised by a calcium ion influx and resulting inflammatory cascade (Owens, *et al.*, 2019). The mechanical stress results in the opening of stretch-activated channels, cell membrane disruption and excitation-contraction coupling dysfunction (Peake, *et al.*, 2017a). The opening of these stretch-activated channels, and disruption of cellular membranes may allow calcium ($Ca^{2+}$) to enter the cytosol stimulating the activation of calpain, phospholipase $A_2$ ($PLA_2$) and reactive oxygen species (ROS) production, leading to a 'vicious cycle' of increased cellular permeability, increased cytosolic $Ca^{2+}$ concentrations [$Ca^{2+}$], and cellular necrosis (Gissel, 2005).

Due to the vast research surrounding physiological effects of muscle damage, and the effects of recovery interventions following exercise induced muscle damage, numerous muscle damaging protocols have been implemented, ranging from isolated eccentric exercise, to repeated plyometric exercise, simulated sport specific protocols such as simulated soccer matches, and non-simulated match exposure (Leeder, *et al.*, 2012; Bieuzen, *et al.*, 2013; Hill, *et al.*, 2013), successfully inflicting a significant occurrence of muscle damage. However, despite the vast amount of research conducted, few studies have considered the application of their findings to a highly-trained, elite youth soccer population (chapter 5, Pooley, *et al.*, 2017; de Hoyo, *et al.*, 2016; de Hoyo, *et al.*, 2015; Ascensão, *et al.*, 2011). Problems arise when recommendations of recovery strategies for the attenuation of muscle damage are derived from a limited simulated body of research, without fully understanding the extent to which these protocols reflect EIMD sustained through sport-specific competition exposure such as soccer match play.

To the authors knowledge, no study to date has compared EIMD elicited from isolated eccentric exercise to that sustained following soccer matches of elite youth soccer players. It is therefore plausible that damage sustained as a result of elite soccer match play might differ in magnitude to that of simulated matches or isolated eccentric exercise and current recovery interventions assessed from studies utilising such exercise protocols are inappropriate for practice. The purpose of this study was to therefore identify to what extent muscle damage sustained by an isolated eccentric muscle damaging protocol might differ compared to that induced by competitive youth soccer matches.

## 4.2 Methodology

### 4.2.1 Participants

Ten elite youth soccer players (mean [± SD]: age 16 [1] years, stature 177.2 [4.9] cm, mass 66.2 [4.4] kg) were voluntarily recruited from a professional soccer academy in the English Premier League to participate in this study. Prior to commencing this study, participants were informed of any risks that may occur, and player and parental consent was obtained (see appendices). All procedures were conducted in accordance with the Declaration of Helsinki, and the study was ethically approved by the Faculty of Science, Engineering and Computing Ethics Committee (Kingston University London, UK).

### 4.2.2 Experimental Design

Participants were required to complete a minimum of 80% (chapter 5, Pooley, *et al.*, 2017) of three 80-minute competitive soccer matches (2x40min per match) [Match], and one eccentric exercise protocol [ECC] designed to elicit muscle damage (Saka, *et al.*, 2009). In order to assess the extent of muscle damage elicited, creatine kinase (CK), countermovement jump with arm swing (CMJA), muscle oedema and perceived muscle soreness (PMS) were measured before (pre) and immediately after (post) each competitive soccer match and the eccentric exercise protocol. Upon completion of testing, average indicators of muscle damage for all three soccer matches were calculated and compared to indicators of muscle damage obtained following ECC protocol.

### 4.2.3 Experimental Protocol

#### 4.2.3.1 *Physical Assessments*

Figure 4.1 displays a schematic diagram of the assessments time-points of muscle damage and implementation of exercise protocols. Prior to any physical exercise, participant stature and mass were recorded, immediately followed by the recording of PMS at 2.5 hours prior to exercise commencing. PMS was indicated using a 10-point visual analogue scale (VAS) from 0.5 to 5, with 0.5 increments. Immediately following PMS assessment, muscle oedema was indicated using a constant-tension tape measure to assess muscle circumference (chapter 3) using three sites of the lower body; the two sites on the lower leg were identified by 1/3 (OedemaG1) and 2/3 (OedemaG2) of the lower leg length calculated by the distance from the medial condyle of the Tibia to the Calcaneus. The site on the upper leg (OedemaQ) was identified by the midpoint of the distance from the Patella to the anterior superior iliac spine. Following these initial assessments, CK levels were assessed using fingertip whole blood samples, and analysed using the i-STAT 1 Analyser (Abbott Point of Care, Abbott Park, Illinois, USA). Two hours prior to exercise, CMJA was recorded using the Smart Speed Jump Mat (Fusion Sport), with participants completing three maximal jumps with peak jump performance recorded for analysis.

Prior to any participation in matches, a physical and technical preparation warm-up was conducted by a sport scientist and UEFA qualified coach respectively. Warm-ups remained consistent throughout the duration of the study, comprising 15 minutes physical preparation involving muscle activation, movement preparation and dynamic stretching and mobility, and 15 minutes of positional and technical football work. For consistency, the warm-ups were conducted by the same sport scientist and coach prior to all competitive matches.

#### 4.2.3.2 *Eccentric Protocol*

At 20 minutes prior to testing, participants completed a 15 minute warm-up which replicated the physical aspects used in the pre-match warm-up. Upon completion of the warm-up participants were briefed on the proceedings of the eccentric exercise test (ECC).

The eccentric exercise protocol modified a protocol utilised by Saka and co-workers (2009) which inflicted significant muscle soreness of knee extensor muscle for up to three days post-exercise. Participants completed three sets of 12 maximal voluntary repetitions of isokinetic eccentric contractions of the knee flexors and extensors at an angular velocity of 30°/sec$^{-1}$ (0.5 rad/s$^{-1}$) using a CSMi Humac Norm Dynamometer (Stoughton, MA, USA). Participants were required to complete the bilateral eccentric protocol on both legs in a randomised order, completing 3x12 repetitions of the first leg, followed by 3x12 repetitions of the second leg. The starting position of the leg was set to 0° (full extension of the knee) and worked to the point of 100° flexion. Participants were braced at their upper body and pelvis by a harness and their upper leg of the resting limb strapped to prevent any extraneous movement. Full testing of the eccentric protocol lasted approximately 15 minutes.

Following eccentric exercise, participants remained seated for 10 minutes. At 45 minutes post-exercise, muscle damage assessments were taken using the same format as previously stated.

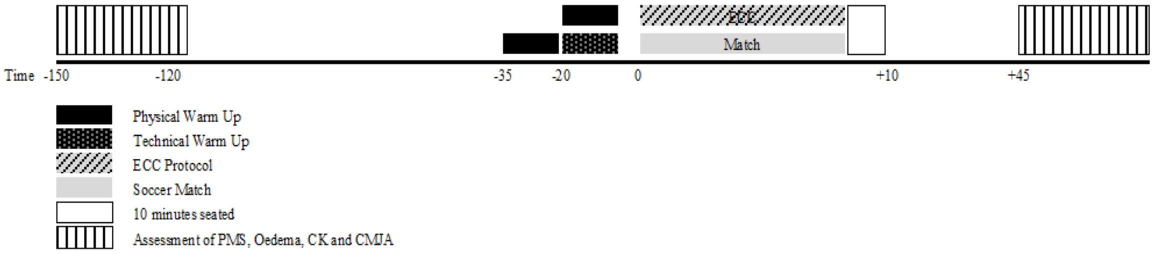


**Figure 4.1** Schematic diagram depicting the chronological process (mins) of pre-exercise muscle damage assessments, pre-exercise warm ups, ECC and Match protocols and post-match assessments of muscle damage with initiation of exercise protocols occuring on 0 minutes.
- represents minutes preceding exercise protocols. + represents minutes

#### 4.2.3.3 *Player Exclusions*

For the purpose of control, and for the monitoring of physical outcomes of competitive soccer matches, Global Positioning Systems (GPS) units fitted with accelerometers were worn by all participants during match play. The GPS units (STATSports Viper) reported movement variables across all axes, as well as reporting heart rate data. Individual thresholds based on these movement parameters were set according to the average of the combined variables for all full competitive matches completed. As a result, match percentages were calculated, with the average of dynamic stress load (fatigue score calculated using player movements, steps and collisions), metres per minute, speed intensity, high metabolic load distance, high speed running distance (distance covered above 65% of an individual's maximum speed), accelerations, decelerations, heart rate minutes above 85% of max., and heart rate exertion over all full matches amounting to a match percentage of 100%. This match percentage determined the individual intensity of matches completed for all players. To ensure the intensity of each competitive soccer match remained consistent throughout the study, only participants whose match percentage for each match was within ± 10% of their average match percentage for all completed matches were considered for data collection. Furthermore, participants were excluded from data collection for individual matches if they failed to complete a minimum of 80% of the 80 minute match, whilst goalkeepers were excluded from the study due to the inability to control consistency in physical loading for matches. Of a potential squad of 16, four players were excluded for not completing a minimum of 80% of match play and two were goal keepers. As such a total of ten players were include in the final analysis.

### 4.2.4 Statistical Analysis

Data are presented as mean ± standard deviation (SD) unless otherwise stated. Analysis of indicators of muscle damage commenced following assessment for normality in all data, verified using the Shapiro-Wilk test. The assessment markers recorded (CK, CMJA, PMS, Oedema) were compared across two conditions (Match and ECC), and two timepoints (pre and post) using a two-way within-subjects repeated measures analysis of variance (ANOVA) in statistical software SPSS 24 (SPSS Inc., Chicago, IL, USA). A Bonferroni statistical test was used for post-hoc analysis. Effect sizes were calculated using partial eta$^2$ ($\eta^2_p$) where 0.01 = small; 0.06 = medium; and 0.14 = large effect (Field, 2009). Statistical significance was accepted at $p < 0.05$.

## 4.3 Results

### 4.3.1 Eccentric Exercise Protocol

Analysis of indicators of muscle damage between pre- and post-ECC protocol revealed a significant increase in OedemaG1 ($p = 0.019$, $\eta^2_p$:0.474), OedemaG2 ($p = 0.021$, $\eta^2_p$: 0.463), OedemaQ ($p = 0.001$, $\eta^2_p$: 0.883), and PMS ($p = 0.001$, $\eta^2_p$: 0.900), and a significant reduction in CMJA performance ($p = 0.015$, $\eta^2_p$: 0.500), however no significant increase in CK ($p > 0.05$) was observed (Table 4.1, Figure 4.2).

### 4.3.2 Soccer Match

Analysis of average indicators of muscle damage between pre- and post-match measures revealed a significant increase in PMS ($p = 0.001$, $\eta^2_p$: 0.959) and CK ($p = 0.004$, $\eta^2_p$: 0.630), and a significant reduction in CMJA performance ($p = 0.001$, $\eta^2_p$: 0.730), however no significant differences between pre- and post-match assessments of OedemaG1, OedemaG2 and OedemaQ ($p > 0.05$) were identified (Table 4.1, Figure 4.2).

### 4.3.3 Eccentric Exercise Protocol v Soccer Match

Analysis of muscle damage indicators between exercise conditions revealed no significant differences across all assessments pre-exercise ($p > 0.05$), suggesting comparable baseline measures were identified. Post-exercise indicators of muscle damage revealed a significantly higher PMS ($p = 0.045$, $\eta^2_p$: 0.377) and CK ($p = 0.004$, $\eta^2_p$: 0.623), and significantly reduced CMJA ($p = 0.007$, $\eta^2_p$: 0.574) following soccer matches when compared to ECC. OedemaG2 revealed a significant increase ($p = 0.01$, $\eta^2_p$: 0539) following ECC when compared to soccer matches. No significant differences ($p > 0.05$) were identified between OedemaG1 and OedemaQ when compared between post-ECC and post-match assessments (Table 4.1; Figure 4.3).

**Table 4.1** Comparison of mean (± SD) physiological, psychological and performance indicators of muscle damage at pre- and post- eccentric protocol and soccer matches.

| | **Assessment Timepoint** | |
|---|---|---|
| | **Pre** | **Post** |
| OedemaG1 (cm) | | |
| ECC | 26.9 (1.8) | 27.1 (1.8)$^{(a)}$ |
| Match | 26.9 (1.8) | 26.9 (1.9) |
| OedemaG2 (cm) | | |
| ECC | 36.5 (2.2) | 36.7 (2.2)$^{(a)(c)}$ |
| Match | 36.2 (2.0) | 36.2 (2.0)$^{(b)}$ |
| OedemaQ (cm) | | |
| ECC | 52.7 (3.7) | 53.6 (3.6)$^{(a)}$ |
| Match | 53.0 (3.5) | 53.2 (3.6) |
| Perceived Soreness | | |
| ECC | 2.9 (0.5) | 3.8 (0.4)$^{(a)(c)}$ |
| Match | 1.5 (0.4) | 4.1 (0.4)$^{(a)(b)}$ |
| CK (ng/mL) | | |
| ECC | 4.6 (2.6) | 6.0 (4.4)$^{(c)}$ |
| Match | 3.3 (2.0) | 8.1 (5.4)$^{(a)(b)}$ |
| CMJA (cm) | | |
| ECC | 56.8 (7.5) | 54.0 (7.1)$^{(a)(c)}$ |
| Match | 55.3 (7.8) | 52.0 (7.8)$^{(a)(b)}$ |

$^{(a)}$ $p < 0.05$ = significantly different from Pre; $^{(b)}$ $p < 0.05$ = significantly different from ECC; $^{(c)}$ $p < 0.05$ = significantly different from Match. (ECC: eccentric protocol; Match: soccer match).

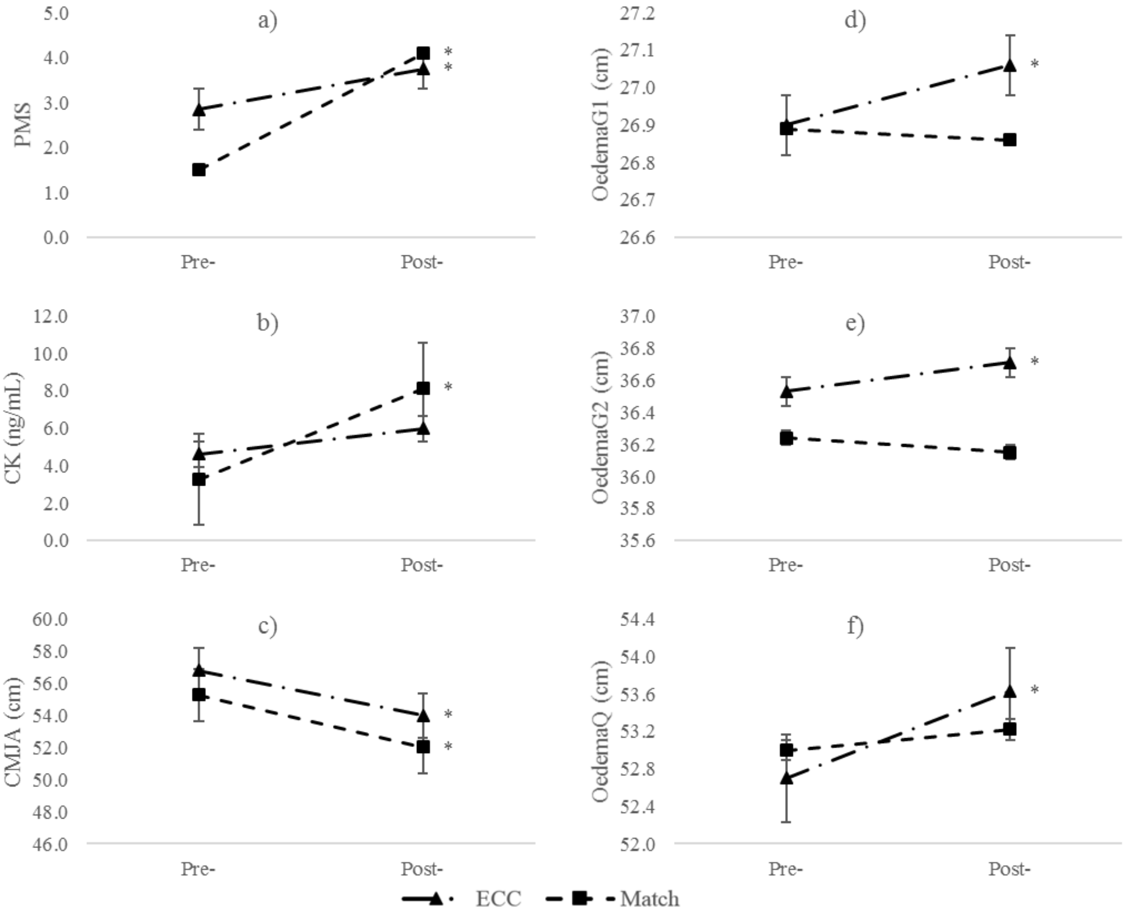


**Figure 4.2** Comparison of mean change in PMS (a), CK (b), CMJA (c), OedemaG1 (d), OedemaG2 (e) and OedemaQ (f) from pre- to post-exercise assessments for eccentric (ECC) and soccer match (Match) protocols. *$p < 0.05$, significantly different from pre-.

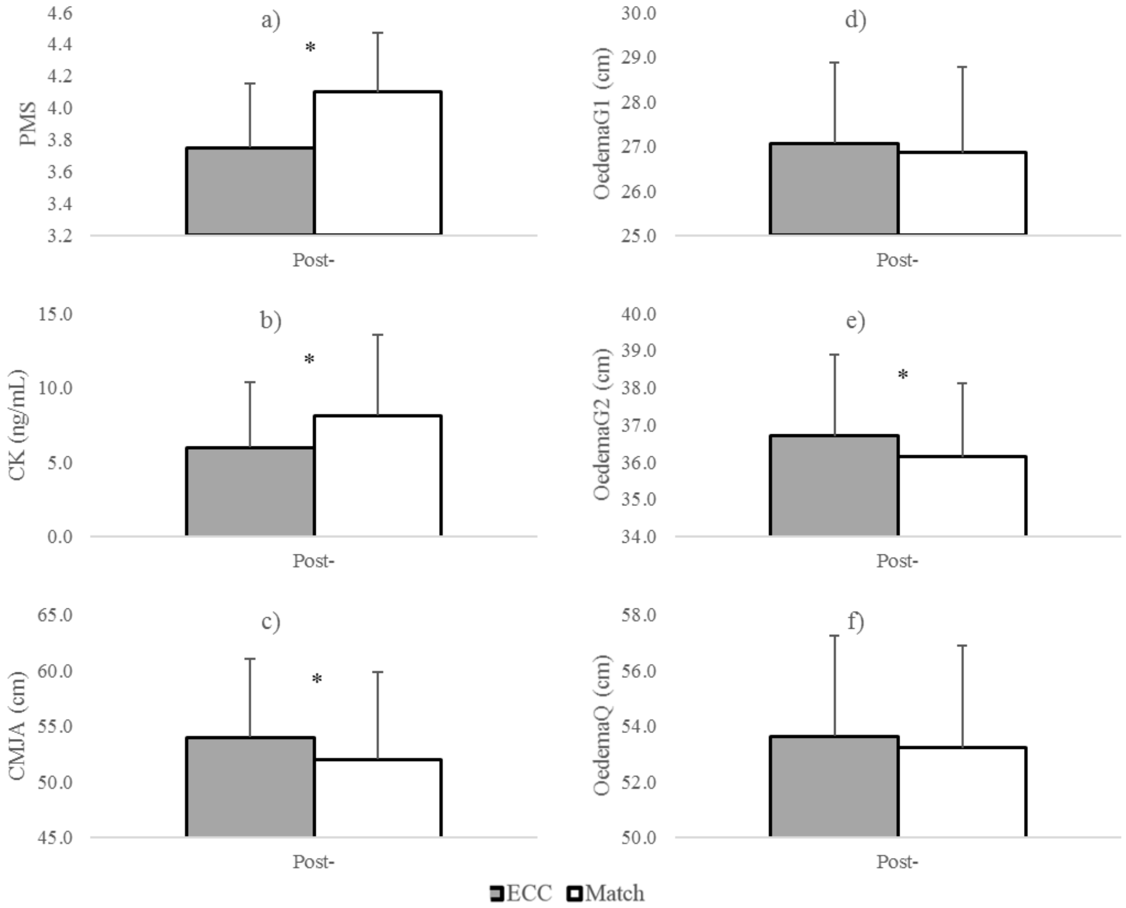


**Figure 4.3** Comparison of post-exercise indicators PMS (a), CK (b), CMJA (c), OedemaG1 (d), OedemaG2 (e) and OedemaQ (f) between eccentric (ECC) and soccer match (Match) protocols. $*p < 0.05$, significant difference between protocols.

## 4.4 Discussion

The aim of this study was to identify to what extent isolated eccentric muscle damaging exercise sufficiently replicates muscle damage sustained following elite youth soccer matches. The main findings from the present study demonstrated both ECC and elite youth soccer matches significantly induce muscle damage, as shown by the significant increase in PMS, and significant reduction in CMJA performance for both exercise protocols, and a significant increase in CK following competitive youth soccer matches when compared to pre-exercise measures (Table 4.1, Figure 4.2). In addition, this study was the first of its kind to demonstrate that, despite the ECC protocol significantly eliciting muscle damage, the damage inflicted does not sufficiently represent that sustained following elite youth soccer matches, as displayed by a significantly higher PMS and CK, and significantly reduced CMJA following youth soccer matches in comparison to ECC protocol (Figure 4.3) illustrating the importance of considering applied research for implementation of interventions to applied settings, and the need for future research to be conducted in applied settings.

When comparing the findings of the present study to previous research, the ECC and soccer match protocol must be considered separately, as no previous research has considered these protocols comparatively. In comparison to literature implementing eccentric muscle damaging protocols, the present study offers similar findings. Saka and others (2009) reported comparable findings to the present study, revealing a significant increase in CK immediately post-exercise when compared to baseline assessments following unilateral eccentric muscle contractions of knee extensors. Unfortunately, due to differences in indicators of muscle damage assessed, direct comparisons to the present study cannot be made, however, the eccentric protocol of Saka and co-workers (3x15 maximal eccentric contractions at 30°/sec$^{-1}$) successfully induced muscle damage as observed by a significant increase in CK post-exercise despite only considering unilateral assessment of knee extensors. In comparison, the present study considered eccentric bilateral assessment of both knee extensor and knee flexor muscle groups, and given the similarities in the eccentric protocols utilised, it can be estimated that the present study induced greater muscle damage. It can therefore be stated that the ECC protocol of the present study is suitable for inflicting significant muscle damage.

In contrast to findings of the present study, Magal and others (2010) reported no significant increases in CK immediately following unilateral eccentric exercise when implementing a protocol comprising eccentric contractions of knee extensors. The use of only one muscle group (quadriceps) in comparison to the inclusion of both knee flexors and extensors in the present study may result in lower muscle damage elicited, and therefore reduced detection of indicators of muscle damage post-exercise, however it must also be considered that assessment time post-exercise may impede findings in the study of Magal and others (2010). Although the exact timeframes are not stated, Magal *et al* report assessing indicators of muscle damage immediately following eccentric exercise, in comparison to the assessments taken 45 minutes post-exercise in the present study, potentially allowing greater time for the onset of muscular adaptations, and the manifestation of indicators of muscle damage post-exercise to occur.

When considering the effects of match performance on muscle damage sustained post-match, findings from the present study complement previous research (chapter 5, Pooley, *et al*., 2017; de Hoyo, *et al*., 2016; Ascensão, *et al*., 2011; Gill, *et al*., 2006) revealing significant alterations in muscle damage indicators when compared to pre-match assessments. Gill and co-workers (2006) report significant increases in CK immediately post-match following competitive rugby match-play of elite adult rugby players. However, despite similar findings to the present study, caution must be taken when considering the transferability of findings of damage caused by professional rugby competition as it is likely that the impacts sustained, and physical capabilities of elite adult rugby players, surpass that of elite youth soccer players, consequently increasing muscle damage. It is therefore important to compare findings of the present study to research assessing soccer players, and damage elicited by soccer matches. Nédélec and colleagues (2014) report a significant reduction in CMJ performance and hamstring maximal voluntary contraction (MVC), and a significant increase in CK at 24 hours post-match when assessing professional adult soccer players. Findings of reduced performance indicators and increased myoprotein assessment post-match are supported by the present study, however the athlete age and training status must again be considered when comparing findings to elite youth soccer players, as it can be expected that the physical demands of elite adult soccer matches are greater than that of youth soccer matches, therefore eliciting higher muscle damage. Research of de Hoyo and others (2016), and Pooley and colleagues (2017) (chapter 5) assessing elite youth soccer players following soccer match exposure display similarities to the present study, reporting significant reductions in CMJ and CMJA performance and increases in CK immediately post-match when compared to pre-match values, demonstrating soccer matches of elite youth soccer players significantly induce muscle damage.

The present study identified that elite youth soccer matches induce greater muscle damage than isolated eccentric exercise protocols. Due to this being the first study to demonstrate ECC protocols do not sufficiently replicate muscle damage induced by elite youth soccer matches, the current literature lacks explanations to why this might be, however it is important to consider the physical work completed throughout soccer matches in comparison to eccentric EIMD protocols. It has been suggested that throughout soccer matches, players are exposed to 1000-1500 discrete movement changes (Bloomfield *et al*., 2007), with up to 250 brief high-intensity actions (Ade, *et al.*, 2014) such as accelerations and decelerations occurring per game. In comparison the ECC protocol implemented in the present study, despite eliciting significant muscle damage, incorporated a total of 144 eccentric contractions, offering a potential explanation for the greater damage observed following elite youth soccer matches. In addition to this eccentric load from soccer matches, the volume of physical work, although not typically associated with muscle damage, may exacerbate fatigue when combined with eccentric exposure, resulting in greater perceived muscle soreness. It is also possible that, although difficult to quantify, neuromuscular fatigue inflicted by the unpredictable changes in intensity of the explosive, eccentric actions in soccer matches (Scott, *et al.,* 2016) may result in reduced performance indicators of muscle damage as represented by a significantly reduced CMJA performance when compared to predictable eccentric contractions of ECC protocol.

When comparing the eccentric actions of ECC protocols to soccer matches, it is important to consider the speed at which the contractions occur. It has been reported that eccentric actions that occur at greater angular velocities inflict significantly higher muscle damage (Owens, *et al*., 2019), and as such it is plausible to suggest that the eccentric contractions of soccer matches were of greater speed than the eccentric actions of the ECC protocol. Again, this appears to be a topic lacking in research, and it may be recommended that future studies consider the assessment of eccentric torque and angular velocities following EIMD protocols in comparison to eccentric actions of elite soccer matches. It is also important to consider that data collected following elite youth soccer matches in the present study was completed over three competitive fixtures throughout the season. It may therefore be expected that due to the repeated bout effect whereby a muscle's susceptibility to damage is reduced for subsequent exercise bouts as a result of prior exposure to eccentric, unaccustomed exercise (Owens, *et al*., 2019), muscle damage observed following elite youth soccer matches would reduce, however the present study did not display such trends. It is possible that this is due to training exposure of athletes ill-preparing them for competitive soccer match play, or that application of athletes in soccer matches was greater than that of training sessions, resulting in exposure to unaccustomed exercise. This is an area of potential future consideration, to assist in the knowledge of preparation for elite youth match play.

## 4.5 Practical Implications

In light of the findings of the present study, it may be advised that when considering the application of research to applied environments, researchers should aim to accurately recreate the physical conditions experienced within the applied setting, instead of using simulated, or replicated exercise protocols. To date, large bodies of research concerning the efficacy of recovery interventions for assisting in the muscle repair process post-exercise have used eccentric exercise protocol to inflict muscle damage (Hill, *et al*., 2013; Bieuzen, *et al*., 2013; Leeder, *et al*., 2012), however the present study would suggest that such articles do not sufficiently replicate damage elicited from applied setting, raising questions over the effectiveness of recovery interventions assessed. As a result, current practices based on findings from research using isolated eccentric exercise protocols may not produce replicable results when introduced within applied settings. It may therefore be useful for future research concerning the recovery of elite youth athletes from competition to focus on the effects of interventions implemented following match exposure, ensuring results are transferable and applicable to their intended setting. It may also be advised that caution be taken when considering recommendations of recovery strategies based on eccentric EIMD for the implementation following competition, and researchers should aim to accurately recreate competition when assessing the effects of such recovery interventions for implementation following matches.

# 5 Static Stretching Does Not Enhance Recovery in Elite Youth Soccer Players

## 5.1 Introduction

As discussed in chapter 4, current research concerning the implementation of recovery interventions following eccentrically induced muscle-damaging exercise protocols may not elicit equivalent muscle damage to that of soccer matches, and as such, previously assessed recovery interventions must be assessed in applied research. Static stretching (SS) is an intervention historically recommended as a recovery method following exercise to prevent or reduce muscle soreness (Keil, 2019; Cheung, *et al.*, 2003; Wessel & Wan, 1994), often referred to as the delayed-onset muscle soreness (DOMS). When performing static stretching, muscles are elongated, often to the point of slight discomfort where they are held for a prescribed period of time (Keil, 2019; Nédélec, *et al.*, 2013). It has been suggested that static stretching may assist in the dispersion of post-exercise muscle oedema (Delextrat, *et al.*, 2014), reducing the potential damaging effects of reactive oxygen species (ROS), neutrophils, lymphocytes and pro-inflammatory cytokines which, although act as a protective mechanism, may inadvertently cause further cell damage (Owens, *et al.*, 2019; Peake, *et al.*, 2017a; Peake, *et al.*, 2005).

In addition to the recommendations for using static stretching with non-elite participants (Cheung, *et al.*, 2003), static stretching is known to be used by elite athletes with research highlighting the application on national level basketball (Delextrat, *et al.*, 2014) and soccer (Nédélec, *et al.*, 2013) players. Furthermore, Dadebo and others (2004) reported English Premier League football clubs dedicate almost 40% of training time to flexibility training, with the most common technique being static stretching, although this study did not specify the context in which static stretching was implemented (warm-up or cool-down). This has been supported by Van Crombrugge and colleagues (2019) reporting 50% of soccer teams in their cohort comprising Premier League teams of the Belgium and English soccer systems complete static stretching, however again this study did not specify the context in which static stretching was implemented.

Applied studies investigating the effects of static stretching have explored numerous sports such as basketball (Delextrat, *et al*., 2014; Montgomery, *et al*., 2008; Calleja-González, *et al*., 2016), Australian rules football (Bahnert, *et al*., 2013) and soccer (Kinugasa & Kilding, 2009; Dawson, *et al*., 2005; Wessel and Wan, 1994). Research assessing the effects of static stretching in basketball, an intermittent sport comprising high intensity actions, has revealed varied and inconclusive findings. Montgomery and colleagues (2008) compared the effects of static stretching on physical performance markers of basketball players to pre-tournament baseline values, with static stretching having no significant effect on performance. These findings are supported by a review study of Calleja-González and others (2016) advising against the use of static stretching as a recovery intervention for improving flexibility and reducing adhesions post-competition. Limitations to the study produced by Montgomery and colleagues (2008) can be highlighted, as data was collected following competitive basketball matches on three consecutive days, suggesting an element of cumulative muscle damage may exist, questioning the transferability of these findings to elite youth soccer as this may not truly represent an expected competitive schedule.

In contrast to research demonstrating static stretching has no effect on recovery, Delextrat and co-workers (2014) report a significant reduction in PMS (females) and a significant improvement in countermovement jump (CMJ) performance (males) as a result of a static stretching intervention on state level basketball players. Again, limitations to this study exist, as the recovery intervention incorporated massage therapy and therefore findings cannot be directly compared to studies focusing solely on static stretching. Additionally, caution must be taken when assessing the transferability of findings from various sports to soccer as physical demands between sports are likely to differ, resulting in varying degrees of muscle damage sustained.

In order to truly assess the effects of static stretching on recovery of soccer players post-competition, it is important to use soccer players as participants in research studies. Dawson and others (2005) reported a significant improvement in peak power 15-hours post-exercise when assessing semi-professional male soccer players, however no differences in subjective assessments or range of movement were found. These findings may appear more applicable to elite youth soccer; however, athlete training status and participant age (24 ± 3 years) may not truly represent the elite youth population. It may be suggested that muscle damage elicited from a competitive semi-professional soccer match would differ substantially to that experienced at elite standard, therefore affecting the impact of static stretching as a recovery technique.

In contrast to the findings of Dawson *et al.*, Kinugasa and Kilding (2009) report static stretching had no significant effect on non-elite youth soccer players following three soccer matches compared to a control. Despite the use of youth participants (14 ± 1 years), athletes were considered non-elite, again questioning the applicability of findings to the elite youth population. Additional limitations to this study may also be highlighted; firstly, the primary assessment method was subjective and therefore caution must be taken when considering the effectiveness of static stretching for recovery based on these results. Secondly, participants completed three competitive soccer matches within one week, and an average of all pre-match, post-match, and post-recovery assessments for all matches were calculated and used for statistical analysis. The grouping of all data for statistical analysis may impact the ecological validity of this study as results represent cumulative muscle damage rather than the effects of static stretching on single trials. Alternative applied research supports the findings of Kinugasa and Kilding, reporting static stretching did not significantly improve muscle recovery following soccer training when compared to a control group (Sermaxhaj, *et al*., 2017), however it must be considered that the control group completed an active recovery intervention only, whilst the static stretching intervention group completed both active recovery and static stretching. As has been displayed in chapter 6, active recovery may aid in the recovery process, and as such could explain the lack of significant differences in the findings of Sermaxhaj and colleagues (2017). It should also be noted that the recovery interventions were implemented following training exposure, which may not sufficiently elicit significant muscle damage, and therefore recovery from training can be expected to occur sooner than that of soccer matches.

The use of static stretching as a recovery intervention has been extensively studied, and despite inconclusive findings, would appear to be used as a recovery technique following exercise (Keil, 2019; Sermaxhaj, *et al.,* 2017; Nédélec, *et al*., 2013; Delextrat, *et al*., 2014; Dadebo, *et al*., 2004). To date, limited research has been conducted on the effects of static stretching as a recovery intervention following competitive soccer matches for elite youth soccer players, and with a dearth of research in this area, it may be hypothesised that current practices in elite youth soccer are supported by inapplicable evidence. The variance in sports studied and participant training status and age throughout literature does not allow for current research to be applied to the elite youth population, therefore the aim of this study was to compare the effects of static stretching to a control of passive recovery following competitive soccer matches using elite youth soccer players.

Due to reduced accessibility to elite youth athletes for research studies, there is a dearth of applied evidence to support the use of recovery intervention on elite youth soccer players. In order to address these issues, this study was conducted in an elite soccer academy, and as such, assessed practices implemented within the soccer club, and their effects on indicators of muscle damage following elite youth soccer matches.

## 5.2 Methodology

### 5.2.1 Participants

Ten elite youth soccer players (mean (± SD): age 16 (1) years, stature 173.5 (6.1) cm, mass 63.3 (6.5) kg) were voluntarily recruited from a professional football academy in the English Premier League to participate in this study. All participants had been playing soccer at an elite academy for a minimum of 12 months prior to testing. All prior consent was obtained, and procedures explained as mentioned in chapter 3. All procedures were conducted in accordance with the Declaration of Helsinki, and the study was ethically approved by the Faculty of Science, Engineering and Computing Ethics Committee (Kingston University London, UK).

### 5.2.2 Experimental Design

Participants were required to complete a minimum of three 80-minute competitive soccer matches for each recovery intervention (Static stretching (SS) or passive recovery (PR)). In order to assess the extent of muscle damage elicited from matches, markers of muscle damage (CK), countermovement jump with arms (CMJA), muscle oedema and perceived muscle soreness (PMS) were measured before (pre), immediately after (post), and 48-hours after (48-hours post) each competitive soccer match.

### 5.2.3 Experimental Protocol

#### 5.2.3.1 *Physical Assessments*

Upon arrival to the match facility, participant stature and mass was recorded, immediately followed by the recording of PMS at 2.5 hours prior to exercise commencing. PMS was indicated using a 10-point visual analogue scale (VAS) from 0.5 to 5.

Immediately following subjective soreness assessment, muscle oedema was taken using a constant-tension tape measure to assess muscle circumference (Howell, *et al.*, 1993; Stanton, *et al.*, 2000), using three sites of the lower body; the two sites on the lower leg of the gastrocnemius were identified by 1/3 (OedemaG1) and 2/3 (OedemaG2) of the lower leg length calculated by the distance from medial condyle of the Tibia, to the Calcaneus Tarsal. The site on the upper leg of the quadriceps (OedemaQ) was identified by the midpoint of the distance from the Patella to the Iliac Crest. Following these initial assessments, CK levels were assessed using the i-STAT 1 Analyser (Abbott Point of Care, IL, USA) using fingertip whole blood samples according to the manufacturer's instructions. Two hours prior to exercise, CMJA were recorded using the Smart Speed Jump Mat (Fusion Sport).

#### 5.2.3.2 *Recovery Interventions*

Prior to any participation in matches, a physical and technical preparation warm-up was conducted by a sport scientist and UEFA qualified coach respectively. Warm-ups remained consistent throughout the duration of the study, comprising 15 minutes physical preparation involving muscle activation, movement preparation and dynamic stretching and mobility, and 15 minutes of positional and technical football work. For consistency, the warm-ups were conducted by the same sport scientist and coach prior to all competitive games.

Immediately following completion of the competitive games, participants were required to complete either the PR protocol which consisted of 10 minutes passive seating, or the SS protocol consisting of two 15 second stretches to the gastrocnemius, hamstrings, quadriceps, glutes, hip flexors, adductors and abductors. Upon completion of the recovery protocols participants were required to repeat the assessment of muscle damage markers in the same order as was taken pre- match. Post-match assessments were undertaken within 30 minutes of completing the match. The same assessments of muscle damage were recorded at 48 hours post-exercise. On every occasion, the assessments were carried out in the same order and by the same sport scientist. The time intervals of assessments (pre-, post- and 48 hours post-match) were consistent with those used throughout literature (Ascensão, *et al.*, 2011; Magal, *et al.*, 2010; Brown, *et al.*, 1997).

#### 5.2.3.3 *Player Exclusions*

For the purpose of control, and for the monitoring of physical outcomes of competitive soccer matches, Global Positioning Systems (GPS) were worn by all participants when competing in games. The GPS units (STATSports Viper) report movement variables across all axes, as well as reporting heart rate data. Individual thresholds based on these movement parameters are set according to the average of the combined variables for all full competitive matches completed. As a result, match percentages were calculated (see chapter 3). This match percentage determined the individual intensity of training and matches completed for all players. To ensure the intensity of each competitive football match remained consistent throughout the study, any participant whose match percentage for any particular game was ± 10% of their average match percentage for all completed matches were excluded from the data collection for that specific competitive match. Furthermore, participants were excluded from data collection for individual games if they failed to complete a minimum of 80% of the 80-minute match whilst goal keepers were excluded from the study due to the inability to control consistency in physical loading for matches. A total of 5 players (2 goal keepers and 3 outfielders) were excluded from participating in this study.

### 5.2.4 Statistical Analysis

Means and standard deviations of all anthropometric data were recorded, and all data analysed for normal distribution, with analysis of muscle damage markers only commencing following assessment of normality. The assessment markers recorded were compared across the two conditions (static stretching and passive recovery) and three time intervals (pre, post-, and 48-hours post-exercise) using a two-way within subjects repeated measures analysis of variance (ANOVA) in statistical software SPSS 23 (SPSS Inc., Chicago, IL, USA). Paired samples T-tests were used as post-hoc analysis. In the case of no homogeneity of variance, Friedman's non-parametric statistical test was conducted with a Bonferoni post-hoc adjustment. Effect sizes were calculated using partial eta$^2$ ($\eta^2_p$) where 0.01 = small; 0.06 = medium; and 0.14 = large effect (Field, 2009). Statistical significance was accepted at $p < 0.05$.

## 5.3 Results

Analysis of data within conditions across time intervals (pre-, post-, 48 hours post-match) produced significant differences ($p < 0.05$) between all time intervals for static stretching and passive recovery interventions when assessing CK, CMJA and perceived soreness as markers of muscle damage (Table 5.1). Additionally, significant differences were found between pre- and 48-hours post-exercise for passive recovery of OedemaG1 ($p = 0.024$, $\eta^2_p$: 0.450, CI: $^{-}0.054$-1.334) and OedemaQ ($p = 0.023$), whilst OedemaQ following static stretching showed significant differences at pre- and immediately post-match ($p = 0.028$).

Further analysis of results for each assessment method of muscle damage showed no significant difference ($p > 0.05$) between recovery interventions of static stretching and passive recovery for OedemaG1, OedemaQ, perceived soreness and CMJA (Table 5.1). However, analysis of CK indicated a significant difference between static stretching and passive recovery interventions 48 hours post-match ($p = 0.032$, $\eta^2_p$: 0.427, CI: 0.024-0.362), whereas analysis of OedemaG2 showed significant differences between static stretching and passive recovery immediately post-match ($p = 0.029$, $\eta^2_p$: 0.596, CI:$^{-}1.047$- $^{-}0.073$), and 48 hours ($p = 0.006$, $\eta^2_p$: 0.642, CI: $^{-}1.109$-$^{-}0.251$) following completion of competitive soccer matches.

**Table 5.1** Comparison of mean (± SD) physiological, psychological and performance markers of muscle damage at pre-, post- and 48 hours post-competitive soccer matches in recovery interventions.

| | **Assessment time point** | | |
|---|---|---|---|
| | **Pre** | **Post** | **48hrs Post** |
| OedemaG1 (cm) | | | |
| PR | 26.3 (2.5) | 26.6(2.5) | 26.9 (2.5)(a) |
| SS | 26.2 (2.6) | 26.5 (2.6) | 26.9 (2.7) |
| OedemaG2 (cm) | | | |
| PR | 35.8 (1.9) | 36.1(1.9) | 36.3 (1.9) |
| SS | 35.7 (2.1) | 35.5 (2.0)* | 35.6 (1.9)* |
| OedemaQ (cm) | | | |
| PR | 49.4 (3.9) | 50.1 (4.1) | 50.3 (4.2)(a) |
| SS | 49.2 (4.0)(b) | 50.1 (4.1)(a) | 49.9 (4.0) |
| Perceived Soreness | | | |
| PR | 1.5 (0.3)(b) | 4.0 (0.3)(a) | 3.0 (0.2)(a) (b) |
| SS | 1.5 (0.3)(b) | 4.0 (0.3)(a) | 3.0 (0.2)(a) (b) |
| CK | | | |
| PR | 2.9 (1.6)(b) | 7.6 (2.1)(a) | 5.8 (2.3)(a) (b) |
| SS | 2.8 (1.7)(b) | 7.4 (2.2)(a) | 5.5 (2.4)*(a) (b) |
| CMJA (cm) | | | |
| PR | 46.8 (6.4)(b) | 41.1 (7.4)(a) | 43.3 (7.1)(a) (b) |
| SS | 46.7 (6.4)(b) | 40.7 (8.0)(a) | 43.4 (7.2)(a) (b) |

(a) $p < 0.05$ = significantly different from Pre; (b) $p < 0.05$ = significantly different from Post;
* $p < 0.05$ = significantly different from PR

PR, passive recovery; SS, static stretching

Due to the inter-individual variability of CK and to demonstrate recovery percentage, further analysis was conducted to assess the percentage change in CK from pre-competition levels at post- and 48 hours post-exercise between conditions (Figure 5.1). Results show no significant differences ($p > 0.05$) between passive recovery and static stretching interventions for percentage change, however a significant difference ($p < 0.05$) across all time intervals was recognised for passive recovery and static stretching.

Additionally, the variability between individual measurements of muscle oedema was large, and therefore further analysis was conducted to assess the percentage change in muscle oedema at three lower leg sites from baseline to post- and 48 hours post-exercise (Figure 5.2). Results show a significant difference ($p = 0.009$, $\eta^2_p$: 0.601, CI: 0.334-1.401) between static stretching and passive recovery at 48 hours post-match when measuring OedemaG2. Furthermore, significant differences ($p = 0.027$, $\eta^2_p$: 0.440, CI: $^-$0.285-5.225) were identified across time for OedemaG1 between pre- and 48 hours post-exercise when assessing static stretching, whilst OedemaQ demonstrated significant differences between pre- and post- (SS: $p = 0.004$, $\eta^2_p$: 0.716, CI: 0.644-2.870; PR: $p = 0.006$, $\eta^2_p$: 0.714, CI: 0.400-2.065), and pre- and 48 hours post-match (SS: $p = 0.046$, $\eta^2_p$: 0.726, CI: 0.353-2.670; PR: $p = 0.008$, $\eta^2_p$: 0.647, CI: 0.471-2.196) for both static stretching and passive recovery conditions.

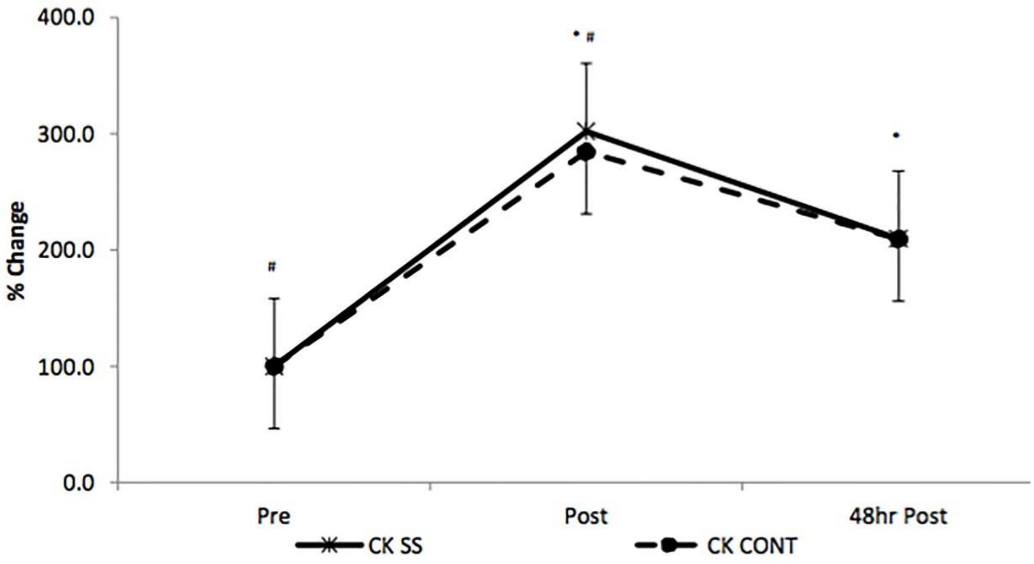


**Figure 5.1** Percentage change in CK levels between pre- (baseline), immediately post- and 48 hours post-match grouped by condition (SS = static stretching, CONT = Control).

Error bars represent standard error at respective time points.

$^{*}p < 0.05$ = significantly different from baseline values

$^{\#}$ $p < 0.05$ = significantly different from 48 hours post-exercise values

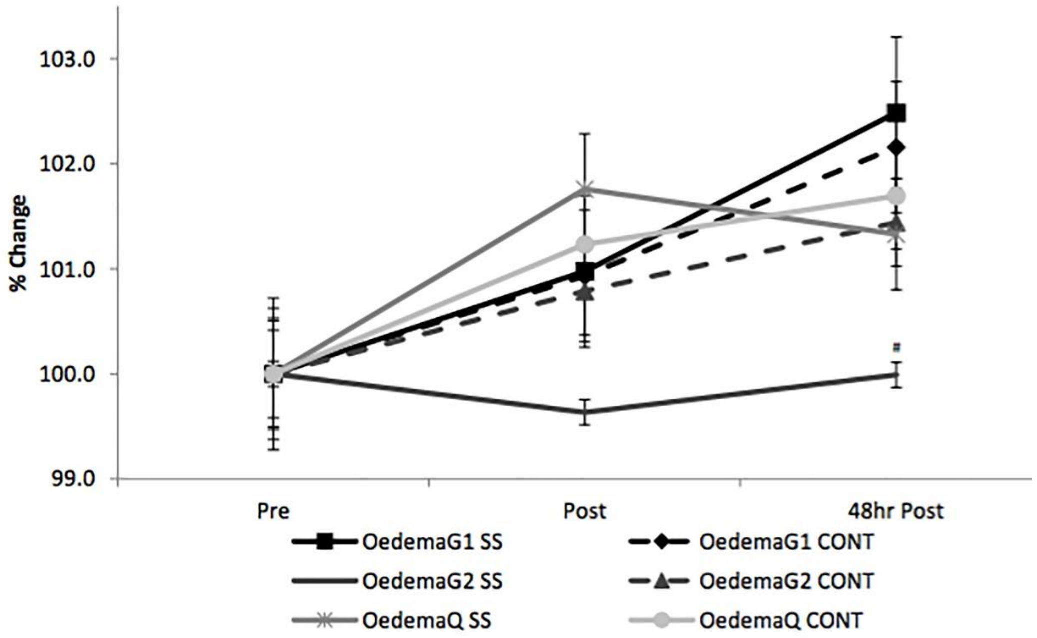


**Figure 5.2** Percentage change in oedema levels between pre- (baseline), immediately post- and 48 hours post-match grouped by condition (SS = static stretching, CONT = Control).
Error bars represent standard error at respective time points.
$^{\#}$ $p < 0.05$ = significantly different from CONT

## 5.4 Discussion

The aim of this study was firstly, to compare the effects of static stretching to a control of passive recovery, and secondly to assess the effects of elite youth soccer matches on muscle damage elicited. In support of the findings of chapter 4, the present study again demonstrated that competitive soccer matches of elite youth soccer players significantly induced muscle damage as demonstrated by the significant increase ($p < 0.05$) in perceived soreness and CK, and significantly reduced CMJA assessments immediately post-match when compared to baseline values (Table 5.1). Furthermore, and continuing from the findings of chapter 4, this study demonstrated muscle damage was evident for at least 48 hours post-match as indicated by the significant difference in perceived soreness, CK and CMJA assessments recorded at this time point ($p < 0.05$) in comparison to baseline values (Table 5.1). The findings of the present chapter, support a review study of Dupuy and others (2018), reporting that stretching provides no recovery benefit to perceived fatigue of DOMS. These findings differ from those of Dawson and others (2005), whose results showed a significant increase in PMS only, at 15 hours post- and 48 hours post-match in comparison to baseline values with no significant differences in performance measures, therefore suggesting soccer matches of semi-professional soccer players do not elicit significant muscle damage. Additionally, Kinugasa and Kilding (2009) reported no significant differences in muscle damage markers across time intervals when assessing perceived soreness and performance measures following competitive matches of non-elite youth soccer players. The findings of the current study may therefore suggest that the intensity of competitive soccer matches at elite standard is higher than non-elite and semi-professional matches and as a result elicit significant occurrences of muscle damage. As demonstrated in chapter 4, eccentric exercise protocols do not elicit muscle damage that is comparable to elite soccer matches, however it is possible that the damage elicited is similar to that of non-elite soccer matches, and research assessing recovery interventions following eccentrically induced muscle damage may be applicable to sub-elite athletes in applied settings. These findings present new information on the effects of competitive soccer matches on muscle damage of elite youth soccer players, providing applied evidence of the time to recovery of elite athletes, however it may also be of interest to coaches and practitioners to understand the physical components of elite youth soccer matches that are correlated to markers of muscle damage, and to identify recovery interventions that significantly reduce muscle damage and aid the recovery process following elite youth soccer matches.

Further examination of the data from the present study demonstrated a significant reduction in perceived soreness, CK and CMJA assessments between immediately post- and 48 hours post-match, suggesting that although competitive soccer matches elicit muscle damage post-match, at 48 hours the recovery process had begun and values were returning to baseline for both passive recovery and static stretching. That said, the report of a significant difference between pre- and 48 hours post-exercise for perceived soreness, CK and CMJA may suggest that the body is going through a process of repair and regeneration (Kendall & Eston, 2002; Figure 2.1) and a return to optimal performance is yet to be achieved, presenting new information on the recovery process of elite youth athletes in the days following competitive soccer matches, and may prove useful to coaches preparing training sessions at 48 hours post-match. It may be advised that the volume and intensity of training sessions 48 hours post-match is reduced to assist in the recovery process, allowing for an earlier return to optimal performance and an increase in training exposure in subsequent training sessions upon full recovery. In order to determine the athletes in need of reduced training volume and intensity to assist in their recovery, it may again be useful to understand the physical match variables correlated to indicators of muscle damage.

Comparisons between static stretching and passive recovery show no significant differences for OedemaG1, OedemaQ, perceives soreness and CMJA ($p > 0.05$). These results are consistent with previous research, suggesting static stretching as a recovery technique has no effect on recovery time when considering muscle oedema (Jayaraman, *et al*., 2004), perceived soreness (Dawson, *et al*., 2005; Kinugasa & Kilding, 2009; Jayaraman, *et al*., 2004) and CMJA performance (Montgomery, *et al*., 2008; Dawson, *et al*., 2005), therefore, it can be argued that in contrast to findings by Delextrat and others (2014), static stretching is an ineffective method for reducing muscle oedema for elite youth soccer players. It is plausible to suggest that the stretching of muscle fibres via static stretching assists in the process of opening stretch-activated channels aiding the calcium influx, which has been suggested to play a role in secondary muscle damage (Owens, *et al*., 2019) and as such static stretching as a recovery intervention may be ineffective. Additionally, due to the elite training status of these individuals, the body may be accustomed to managing the repair of damaged muscle fibres and the removal of myoproteins (Brancaccio, *et al*., 2007), and as a result static stretching has little effect on the repair process.

However, in contrast to Gill and colleagues (2006), CK was significantly elevated ($p = 0.032$) 48 hours post-match as a result of no recovery intervention when compared to static stretching. Although this may suggest static stretching significantly reduces CK following exercise ($p < 0.05$), it may be possible that the individual variability of CK levels at baseline may have an influence on statistical interrogation of results post-match, therefore the CK values were aligned for relative change to baseline and expressed as a percentage. As reported by Nowakowska and others (2019) it is likely that resting CK values of elite soccer players are high due to regular training and match exposure, and the physical load players are subjected to. It is therefore likely that there are positional differences in resting CK values, again due to the varying physical demands of players and their positions, providing justification of alignment of baseline values. Statistical analysis in this form showed no significant differences between static stretching and passive recovery (Figure 5.1), similar to findings of previous studies (Nédélec, *et al.*, 2013; Kinugasa & Kilding, 2009).

Analysis of OedemaG2 also showed significant differences between static stretching and passive recovery interventions immediately post- and 48 hours post-match. Again, this contrasts to the findings in current literature (Nédélec, *et al.*, 2013; Dawson *et al.*, 2005; Kinugasa & Kilding, 2009), however when assessing these results, considerations of compulsory attire of soccer players must be taken into account. The assessment of OedemaG2 was taken at the upper third of the gastrocnemius, an area potentially compressed by the wearing of supportive tape. This may provide a compression aid which could not be avoided due to the applied nature of this study. As participant use of supportive tape was not recorded, it cannot be assumed that it was used across both recovery interventions, and as such, the effects of strappings cannot be directly determined, and future research may be advised. Again, in order to reduce inter-individual variability in muscle oedema measures, manipulation of data to percentage change allowed for further statistical analysis. Results from this analysis (Figure 5.2) showed a significant difference between static stretching and passive recovery of OedemaG2 at 48 hours post-match ($p < 0.05$). Again, the potential influence of the supportive tape may have a contribution to the results, leading to the significant difference (Gill, *et al.*, 2006).

In conclusion the results of this study indicate that competitive soccer matches of elite youth soccer players significantly induce muscle damage, and muscle damage markers remain elevated at 48 hours post-match. Furthermore, the use of static stretching showed no significant reduction in muscle damage markers when compared to passive recovery following competitive soccer matches. This supports the advice provided by Calleja-

González and others (2016) who warn against the use of static stretching as a recovery intervention for reducing muscle damage post-competition.

## 5.5 Practical Implications

In light of these findings, it may be recommended that the use of static stretching following competitive soccer matches for elite youth athletes provides no definitive recovery effect, and that alternative post-match recovery interventions may be advised. In the applied settings of elite academy soccer, situations such as away fixtures provide little opportunity for alternative recovery interventions to be implemented post-match, and as such, in these situations it may be recommended that training sessions in the days following (at least 48 hours post-exercise) have a reduced load.

# 6 Using the Physical Demands of Elite Youth Soccer Matches as indicators for Post-Match Muscle Damage

## 6.1 Introduction

As demonstrated in chapters 4 and 5, elite youth soccer matches significantly induce muscle damage immediately post-match (chapter 4), which remains for up to 48 hours (chapter 5; Pooley, *et al.*, 2017), however the physical parameters of elite youth soccer matches that significantly correlate to markers of muscle damage are yet to be identified. Soccer is considered a high-intensity, intermittent sport (Thorpe & Sunderland, 2012), involving frequent and unpredictable changes in intensity ranging from standing stationary to maximal sprints (Scott, *et al.*, 2016). It has been observed that during a 90 minute soccer match players can cover between 9 and 14 km (Bradley & Ade, 2018), of which between 2 and 3 km consists of high-intensity running, and up to 0.5 km sprinting (Devrnja & Matković, 2018). It has been further estimated that players complete 1000 – 1500 discrete movement changes throughout soccer matches, with these changes occurring every 4 to 6 seconds (Bloomfield, *et al.*, 2007), and a substantial number of the explosive activities in soccer are eccentric muscle actions comprising jumps, accelerations, decelerations and changes of direction which have been suggested to be causally related to post-match muscle damage (Devrnja & Matković, 2018). Such eccentric actions likely explain the muscle damage elicited through soccer matches, causing skeletal muscle microtrauma resulting in perforations in the sarcolemma and damage to the sarcomeres (Russell, *et al.*, 2016).

Previous research has considered various methods for the assessment of muscle damage inflicted following soccer match exposure, including physical performance assessments, perceived soreness, and myoprotein assessment (de Hoyo, *et al.*, 2016: Russell, *et al.*, 2016; Scott, *et al.*, 2016; Romagnoli, *et al.*, 2015; Nédélec, *et al.*, 2014; Thorpe & Sunderland, 2012; Rampinini, *et al*., 2011) with inconsistent findings. It has been accepted that creatine kinase (CK) is a useful tool for the indirect assessment of muscle damage following exercise (Thorpe & Sunderland, 2012; Lazarim, *et al*., 2009), however it may be considered an invasive and impractical assessment method when used in an applied, elite soccer environment. Alternative assessment tools such as performance measures including jump performance (de Hoyo, *et al*., 2016: Russell, *et al*., 2016; Romagnoli, *et al*., 2015; Rampinini, *et al*., 2011) have been used throughout literature, although may be considered inappropriate methods to implement in the hours and days following elite soccer matches when recovery is to be prioritised. As such, it may be of use to coaches to understand the physical elements of soccer matches that elicit muscle damage in correlation to recognised and previously used methods.

Previous studies concerning the correlation of CK and match performance outcomes exist (de Hoyo, *et al*., 2016: Russell, *et al*., 2016; Scott, *et al*., 2016; Thorpe & Sunderland, 2012), although inconclusive findings and limitations abound when comparing the outcomes to elite youth soccer players. Scott and others (2016) compared CK measures 48 hours post-match to physical parameters recorded using a semi-automated multi-camera recognition system reporting no significant relationships, however as recognised by the authors, limitations to their applied setting and elite male population utilised restricted the use of GPS (Global Positioning System) and alternative timepoints of CK assessments. In contrast, Russell and co-workers (2016) report significant correlations between CK and physical match parameters in an elite male population for high-intensity distance, high speed running, sprints and decelerations compared to CK measured 24 hours post-match. The use of GPS and timepoints of CK measurement differ to those of Scott and others (2016), however caution must be taken when considering the application of these findings to the elite youth population as it can be assumed that the adult population assessed by Russell and others

(2016) possessed greater physical capacity than elite youth athletes, therefore potentially exacerbating the physical load of a soccer match and damage sustained. In agreement with the findings of Russell *et al*, de Hoyo and colleagues (2016) reported significant correlations between physical match performance and CK measured 48 hours post-match, however no relationship was identified in CK measured immediately post-match. Again, limitations with data only collected from one soccer match, and of the population used, only seven completed the full game, reduces the impact of their findings. De Hoyo and others (2016) also reported significant correlations between GPS parameters and CMJ performance, although no relationship between GPS and CMJ height was established.

Although the use of CK as a marker of muscle damage has been reported to be reliable and optimal (Thorpe & Sunderland, 2012; Lazarim, *et al.*, 2009), alternative indicators of muscle damage and soreness have been utilised in comparison to match physical performance (Nédélec, *et al.*, 2012), reporting findings of a significant relationship between CMJ at 24 hours post-match and the number of 'hard change of directions'. Furthermore, a significant correlation between muscle soreness at 48 hours post- and 72 hours post-match, and 5m sprint was observed.

Despite several studies comparing the physical demands of soccer matches to indirect assessments of muscle damage, it remains unclear whether physical demands of games significantly correlate to markers of muscle damage, and if so, what specific physical parameters are correlated to indirect assessments. Due to the advances in technology, it is possible for coaches and practitioners to assess live GPS metrics during match play, with physical reports available immediately post-match, therefore providing potential information of estimated muscle damage sooner than the peak manifestation of alternative indirect markers of muscle damage, allowing immediate implementation of recovery strategies and planning of future sessions. Therefore, the purpose of this study was to compare the relationships of physical match performance variables of competitive youth soccer matches to indicators of muscle damage.

## 6.2 Methodology

### 6.2.1 Participants

Nineteen elite youth soccer players (mean [± SD]: age 16 [1] years, stature 174.2 [5.8] cm, mass 64.2 [5.2] kg) from a professional football academy in the English Premier League voluntarily participated in this study. Prior to participating in the study, players were informed of any risks that may occur, and player and parental consent was obtained. All procedures were conducted in accordance with the Declaration of Helsinki, and the study was ethically approved by the Faculty of Science, Engineering and Computing Ethics Committee (Kingston University London, UK).

### 6.2.2 Experimental Design

Participants were required to complete a minimum of 80% (chapter 3) of three 80-minute competitive soccer matches (2x40min per match) with averages of physical parameters across all completed matches calculated for analysis. In order to assess the extent of muscle damage elicited from matches, indicators of muscle damage via creatine kinase (CK), countermovement jump with arm swing (CMJA) and perceived muscle soreness (PMS) were measured before (pre) and immediately after (post) each competitive soccer match. All indicators of muscle damage are commonly used markers in the assessment of muscle damage and recovery (Darani, *et al*., 2017; Gill, *et al.*, 2006; Ingram, *et al*., 2009; Ascensão, *et al*., 2011; Kinugasa & Kilding, 2009; Bahnert, *et al*., 2013).

### 6.2.3 Experimental Protocol

#### 6.2.3.1 *Physical Assessments*

Upon arrival to the match facility, player stature and mass were recorded, immediately followed by the recording of PMS at 2.5 hours prior to the match commencing. PMS was indicated on a 10-point visual analogue scale (VAS) from 0.5 to 5 with 0.5 increments. Immediately following PMS assessment CK levels were assessed using fingertip whole blood samples, and analysed using the i-STAT 1 Analyser (Abbott Point of Care, Abbott Park, Illinois, USA). Two hours prior to exercise, CMJA was recorded using the Smart Speed Jump Mat (Fusion Sport), with participants completing three maximal jumps with peak jump performance recorded for analysis (Chapter 7; Pooley, *et al.*, 2019).

#### 6.2.3.2 *Procedures*

Match performance data was collected using Global Positioning Systems (GPS) units fitted with accelerometers (STATSports Apex) worn by all participants when competing in games and recorded movement variables across all axes, as well as heart rate. The following physical variables were selected for analysis of the relationship between match performance and indicators of muscle damage (CK, perceived soreness, CMJA): accelerations, decelerations, distance covered, high speed running, explosive distance, high metabolic loading distance (HMLD), match percentage and minutes played. Of the physical parameters chosen, number of accelerations and decelerations, and high-speed running distance (m) were further explored to analyse the intensity at which these physical parameters were completed, resulting in a total of 10 physical parameters and one duration parameter, the definitions of which are detailed below.

#### 6.2.3.3 *Match Physical Variable Definitions*

Table 6.1 displays the definitions of physical parameters used to assess physical match load, based on the GPS manufacturer guidelines.

**Table 6.1** Definitions of match performance variables

| Metric | Definition |
|---|---|
| **Distance Covered** | Total distance (m) accumulated throughout the duration of the match. |
| **Accelerations zone 4-6 (AccelsZ4-6)** | Accelerations > 3m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **Accelerations zone 5-6 (AccelsZ5-6)** | Accelerations > 4m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **Decelerations zone 4-6 (DecelsZ4-6)** | Decelerations > 3m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **Decelerations zone 5-6 (DecelsZ5-6)** | Decelerations > 4m.s$^{-1}$ with minimum duration of 0.5 sec. |
| **Absolute high-speed running (HSRAb)** | Distance covered (m) above 5.5m.s$^{-1}$. |
| **Relative high-speed running (HSRRe)** | Distance covered above 65% of individual's maximum speed. |
| **Explosive distance (Ex.D)** | HMLD minus HSRRe |
| **High metabolic loading distance (HMLD)** | Distance covered performing any activity above 25.5 W/Kg |
| **Match Percentage (Match %)** | Individual average of dynamic stress load (fatigue score calculated using player movements, steps and collisions), metres per minute, speed intensity, HMLD, HSRRe, AccelsZ4-6, DecelsZ4-6, heart rate minutes above 85% of max., and heart rate exertion over all full games amounting to a match percentage of 100%. Match percentage for any individual game is then compared to their average (100%) and a percentage for that game then calculated. |
| **Minutes played (MP)** | Total time played in any one game by individual players. |

#### 6.2.3.4 *Player Exclusions*

On the occasions when players completed less than 80% of a competitive soccer match, data was not included for analysis. Players completing less than 80% of three competitive soccer matches were removed from sample collection. Goalkeepers were excluded from the study due to the inability to monitor match load. In total, seven players were excluded from participation (three goalkeepers and four outfielders).

### 6.2.4 Statistical Analysis

Data are presented as mean ± standard deviation (SD) unless otherwise stated. Analysis of relationships between match performance variable and indicators of muscle damage commenced following assessment for normality in all data, verified using the Shapiro-Wilk test. Indicators of muscle damage were analysed in delta form (de Hoyo, *et al.*, 2016; Russell, *et al.*, 2016), whereby pre-match values are deducted from post-match values. Average match data for individual participants was calculated for all physical match parameters and indicators of muscle damage in delta form. To analyse the relationship between match performance variables and indicators of muscle damage, three multiple linear regressions were completed using statistical software SPSS 24 (SPSS Inc., Chicago, IL, USA) with indicators of muscle damage set as the independent variable in each of the multiple linear regression models. Statistical significance was accepted at $p < 0.05$.

## 6.3 Results

Average match physical locomotion data, and average assessments of indirect measures of muscle damage are displayed in Table 6.2 and Table 6.3 respectively. Analysis of the linear regression for predicting the effects of physical match performance on CK revealed a significant regression ($F(11,7) = 4.627$, $p = 0.03$, $R^2 = 0.879$), demonstrating the physical parameters of soccer matches assessed were significantly related to the increases in CK post-match. Analysis of the physical parameters revealed  HSRAb ($\beta = -1.20$, $t(15) = -3.89$, $p < 0.01$), AccelsZ5-6 ($\beta = 0.91$, $t(15) = 2.46$, $p = 0.04$), DecelsZ4-6 ($\beta = 0.78$, $t(15) = 2.31$, $p = 0.05$) and DecelsZ5-6 ($\beta = -0.63$, $t(15) = -2.35$, $p = 0.05$) significant impacted on the increases in CK observed post-match (Table 6.4), however no other physical parameters significantly effect post-match CK measures ($p > 0.05$). Analysis of the linear regression model for predicting the effects of physical match performance on CMJA ($F(11,7) = 0.854$, $p > 0.05$, $R^2 = 0.57$) and perceived soreness ($F(11,7) = 1.131$, $p > 0.05$, $R^2 = 0.64$) revealed a non-significant regression (Table 6.4). Predictors of the relationship between physical match performance and CMJA or perceived soreness showed no significant relationships (Table 6.4).

**Table 6.2** Average (±SD) GPS data collected following elite youth soccer matches

| **Metric** | **Average** |
|---|---|
| **Distance Covered (m)** | 8897 (571) |
| **Accelerations zone 4-6 (n)** | 57 (7) |
| **Accelerations zone 5-6 (n)** | 11 (3) |
| **Decelerations zone 4-6 (n)** | 70 (9) |
| **Decelerations zone 5-6 (n)** | 22 (4) |
| **Absolute high-speed running (m)** | 855 (286) |
| **Relative high-speed running (m)** | 433 (91) |
| **Explosive distance (m)** | 1171 (94) |
| **High metabolic loading distance (m)** | 1612 (133) |
| **Match Percentage (%)** | 99.4 (9.1) |
| **Minutes played (min)** | 78 (2) |

**Table 6.3** Average of assessments of muscle damage taken pre-, post- and in delta form.

| | Pre- | Post- | Delta |
|---|---|---|---|
| **CK (ng/mL)** | 3.5 | 8.4 | 4.9 |
| **CMJA (cm)** | 42.4 | 36.8 | 5.6 |
| **Perceived Soreness** | 1.2 | 4.0 | 2.8 |

**Table 6.4** Relationship between physical match performance variables and CK, CMJA and perceived soreness.

| | CK (ng/mL) | | | CMJA (cm) | | | Perceived Soreness | | |
|---|---|---|---|---|---|---|---|---|---|
| | *β* | *t* | Sig. ($p < 0.05$) | *β* | *t* | Sig. ($p < 0.05$) | *β* | *t* | Sig. ($p < 0.05$) |
| **Distance (m)** | -0.17 | -0.59 | 0.58 | -0.17 | -0.31 | 0.77 | 0.31 | 0.63 | 0.55 |
| **HMLD (m)** | -0.45 | -0.44 | 0.67 | 1.61 | 0.85 | 0.42 | -1.67 | -0.96 | 0.37 |
| **HSRRe (m)** | 1.15 | 1.60 | 0.15 | -1.40 | -1.04 | 0.34 | 1.02 | 0.83 | 1.44 |
| **HSRAb (m)** | -1.20 | -3.89 | 0.01 | -0.21 | -0.36 | 0.73 | 0.28 | 0.54 | 0.61 |
| **Ex.D (m)** | 0.86 | 1.33 | 0.23 | -1.42 | -1.16 | 0.28 | 1.03 | 0.92 | 0.387 |
| **AccelsZ4-6 (no.)** | -0.30 | -0.88 | 0.41 | 0.75 | 1.18 | 0.28 | -0.15 | -0.25 | 0.81 |
| **AccelsZ5-6 (no.)** | 0.91 | 2.46 | 0.04 | -0.95 | -1.38 | 0.21 | 0.49 | 0.78 | 0.46 |
| **DecelsZ4-6 (no.)** | 0.78 | 2.31 | 0.05 | 0.36 | 0.54 | 0.59 | 0.46 | 0.79 | 0.46 |
| **DecelsZ5-6 (no.)** | -0.63 | -2.35 | 0.05 | 0.38 | 0.75 | 0.48 | 0.34 | 0.74 | 0.48 |
| **Match %** | 0.20 | 1.04 | 0.33 | -0.48 | -1.33 | 0.22 | -0.11 | -0.34 | 0.74 |
| **MP (min)** | -0.09 | -0.48 | 0.65 | 0.01 | 0.03 | 0.981 | 0.46 | 1.45 | 0.19 |

## 6.4 Discussion

The objective of the present study was to investigate whether physical match parameters significantly impact on the indicators of muscle damage assessed, and if so, whether specific physical performance variables accumulated throughout a competitive youth soccer match were significantly correlated to the post-match disturbances in indicators of muscle damage.

The main findings of the present study showed that the number of accelerations zone 5-6 (AccelsZ5-6), decelerations zone 4-6 (DecelsZ4-6), decelerations zone 5-6 (DecelsZ5-6) and distance of absolute high-speed running (HSRAb) demonstrate a positive relationship to alterations in post-match creatine kinase (CK) (Table 6.4). Previous support for the use of CK as an indicator of muscle damage following exercise (de Hoyo, *et al*., 2016; Russell, *et al*., 2016), alongside the relationship of CK taken post-match with AccelsZ5-6, DecelsZ4-6, DecelsZ5-6 and HSRAb observed in the present study, suggests that these physical parameters may be a suitable tool to use in the determination of muscle damage severity following competitive youth soccer matches, therefore impacting on the immediate recovery strategies implemented on individual players, and assisting in the management of training exposure in the days following competitive soccer matches. Interestingly, AccelsZ4-6 revealed no significant relationship to the alteration in post-match indicators of muscle damage, therefore suggesting the intensity of acceleration actions significantly contributes to the muscle damage sustained.

The findings of a significant regression between the identified physical match performance variables and indicators of muscle damage from the present study differ to those of Scott and others (2016) concerning elite soccer players, potentially due to the parameters assessed. As demonstrated by the present study, the parameters identified as significantly altering the post-match assessments of CK (AccelsZ5-6, DecelsZ4-6, DecelsZ5-6 and

HSRAb), are those considered to be eccentric and explosive in nature (Varley, *et al.*, 2017). Such actions have previously been suggested to inflict skeletal muscle microtrauma resulting in perforations in the sarcolemma and damage to the sarcomeres (Russell, *et al.*, 2016). In contrast, the physical parameters considered by Scott and co-workers (2016) comprising jogging, running, high speed running and sprinting, and excluding the assessment of accelerations and decelerations may not completely incorporate all eccentric activities of soccer matches. The observations of Davrnja and Matković (2018) indicate that explosive actions such as accelerations, decelerations and sprint-induced muscle damage may provide an explanation as to why Scott and co-workers (2016) were unable to establish a relationship between physical match performance and CK assessments, and may be supported by the findings from the present study indicating AccelsZ5-6, DecelsZ4-6, and DecelsZ5-6 significantly induce muscle damage following elite youth soccer matches. These findings support research of Young and others (2012), who suggest that accelerations and decelerations in Australian rules football matches significantly contribute to muscle damage. In support of the current findings, and the notion that markers of muscle damage are positively correlated to explosive and eccentric actions, Russell and others (2016) also report a significant correlation between CK assessments taken 24 hours post-match and high intensity distance, high speed running, sprints and decelerations, however caution must be taken when considering the application of the findings of Russell *et al* to elite youth soccer players, due to their use of elite professional adult soccer players. It can be assumed that the physical capacity of elite professional adult soccer players surpasses that of elite youth soccer players, therefore potentially exacerbating the physical load of a soccer match and resulting muscle damage sustained. It is also likely that the training exposure of elite professional adult soccer players is greater than that of elite youth soccer players, elevating baseline assessments of muscle damage, potentially altering their findings.

It may also be possible that the timepoint of CK assessment of Scott and others (2016) impacted on the non-significant relationship between soccer match performance variables and CK assessments. Coelho and others (2011) suggested assessments of CK taken closer to the termination of the exercise event are significantly higher than CK assessments taken 48 hours post-event. Although the exact mechanisms of CK clearance are not currently completely understood, this may be due to the assessments of Scott and others being taken too late, should CK clearance commence before 48 hours post-match resulting in lower assessments taken in comparison to immediately post-match, prior to CK clearance occurring. As such, it may be possible that had Scott and others (2016) been able to measure CK immediately post-match, significant relationships with physical match performance may have been established, however the applied nature of their study, and use of elite professional soccer players may prevent such desirable assessments. In support of the hypothesis that CK clearance may have commenced at 48 hours post-match, hence the insignificant correlations identified by Scott and co-workers (2016), Russell and others (2016) report a significant correlation between physical match performance and CK measured 24 hours post-match. This may suggest that the optimal range for assessment of CK exists between immediately post- and prior to 48 hours post-match, therefore inhibiting the findings of Scott *et al*. Given that the peak changes in CK would appear to occur between immediately post-match and 48 hours post-match, match physical performance data would be available to coaches and practitioners sooner than the peak manifestation of indicators of muscle damage, therefore displaying an advantage of using physical data to indirectly indicate muscle damage, and implement recovery interventions and plan appropriate training exposure in the days following games.

Despite the contrasting findings of Scott *et al* (2016), the research of Thorpe and Sunderland (2012) is supported by the present study, demonstrating a significant regression between CK and high intensity activity. However, it must be noted that the speed that defined the high-intensity activity (>15kph) of Thorpe and Sunderland (2012) differed from that of the present study (>5.5m.s$^{-1}$/19.8kph) which also demonstrated a significant relationship to elevated post-match CK values. It is possible that the use of sub-elite athletes by Thorpe and Sunderland (2012) with a reduced training exposure in comparison to the elite athletes used in the present study resulted in higher CK due to a lower tolerance to soccer matches, and as a result it is possible that the slower running speeds significantly induces muscle damage of such athletes.

## 6.5 Practical Implications

In light of the current findings, it may be advised that the physical parameters assessed in the present study significantly contribute to the muscle damage elicited during elite youth soccer matches, and more specifically the number of AccelsZ5-6, DecelsZ4-6, and DecelsZ5-6 and distance of HSRAb directly contribute to the muscle damage sustained immediately post-match. These findings may be of use to coaches and practitioners, as they provide insight to the important physical components of soccer matches that elicit muscle damage. Due to the evolution in GPS technology offering the ability to monitor athletes' GPS live in games and publishing physical match outcomes immediately post-match, the physical components identified in the present study for correlating with indicators of muscle damage will be available to coaches and practitioners sooner than the time taken for peak manifestation of muscle damage indicators, thus allowing earlier implementation of recovery strategies and planning of subsequent training sessions, ensuring optimal recovery and preparation for upcoming training and fixtures. It may also be advised that future research examines the possibility of identifying thresholds of athletes for the determination of muscle damage sustained following soccer matches in comparison to indicators of muscle damage, both on a squad and positional level to better identify muscle damage sustained and implement the most suitable recovery strategies following soccer matches ensuring optimal physical performance in subsequent training and competition.

# 7 Comparative Efficacy of Active Recovery and Cold-Water Immersion as Post-Match Recovery Interventions in Elite Youth Soccer

## 7.1 Introduction

As demonstrated in chapters 4 and 5, elite youth soccer matches significantly induce muscle damage that has been shown to remain for up to 48 hours post-match. Chapter 5 also demonstrated that the commonly used recovery intervention static stretching does not aid muscle recovery at 48 hours post-match, and as such, further research on alternative recovery interventions is necessary. To allow for the greatest opportunity for success in an elite sporting career, athletes are required to train and compete at exceptionally high physical, technical, and psychological intensities, resulting in high stress loads being placed on their bodies (Reilly & Ekblom, 2005), and as demonstrated in chapter 6, high intensity, eccentric actions often contribute to muscle damage. In order to maintain or enhance training and performance levels, athletes require sufficient time to recover from match stimulus and allow muscular adaptations to take place (Peake, *et al.*, 2017a) as indicated in chapter 5. Consequently, athletes and their coaches will seek recovery interventions that will reduce time taken to recover upon cessation of exercise exposure. As such, optimal recovery interventions following training and competition in elite sport are commonly sought (Kinugasa & Kilding, 2009), with active recovery and cold-water immersion often considered in applied elite settings.

Static stretching (SS) is a historically recommended recovery intervention, said to minimise muscle soreness following exercise via the dispersion of post-exercise muscle oedema, reducing the potential damaging effects of reactive oxygen species, neutrophils, lymphocytes and pro-inflammatory cytokines (Cheung, *et al.*, 2003). This intervention has been implemented with varying levels of success, with studies in basketball (Delextrat, *et al.*, 2014) reporting significant improvements on perceived muscle soreness (PMS) and countermovement jump performance, whilst research involving adult semi-professional soccer players suggest improvements in peak power (Dawson, *et al*., 2005). However, these positive findings focused on non-elite participants, and therefore research concerning elite youth soccer players must be considered. As demonstrated in chapter 5 and as indicated in Pooley and others (2017) static stretching following elite youth soccer matches has been found to be ineffective. As Pooley *et al.* (2017) (chapter 5) focused only on the effects of static stretching, further research on the alternative recovery interventions used in elite youth soccer must also be considered.

Within elite soccer, a number of interventions are frequently used, with some of the most prevalent being active recovery (AR) and hydrotherapy techniques such as cold-water immersion (CWI) and contrast water therapy (CWT) (Nédélec, *et al*., 2013). AR – a low intensity ‘cool down’ (Peake, *et al*., 2017b) - has been suggested to aid in the recovery process by facilitating in the removal of metabolic by-products from skeletal muscles post-exercise (Reilly & Ekblom, 2005). Low-intensity exercise completed following the cessation of intense physical activity is reported to increase cardiac output and muscle blood flow (Peake, *et al*., 2017b), accelerating the clearance of exercise-induced by-products such as blood lactate (Ortiz Jr, *et al.*, 2019; Martin, *et al*., 1998) reducing the potential pooling effects of pro-inflammatory cytokines, ultimately preventing further cell damage (Chazaud, 2016).

Difficulties arise when comparing findings between AR interventions (Ortiz Jr, *et al*., 2019; Mika, *et al*., 2016), as the aim of increasing blood flow to respiring muscles can be achieved through various modes of exercise (Darani, *et al*., 2018; Bahnert, *et al*., 2013; Tessitore, *et al*., 2007; Gill, *et al*., 2006). Tessitore *et al*. (2007) compared the effects of an AR intervention comprising 8 minutes walking, 8 minutes jogging, 4 minutes dynamic stretching to passive recovery, electrostimulation and water-based aerobic exercise following soccer training of elite young soccer players with no significant improvements in recovery markers identified as a result of any recovery intervention. However, findings of Tessitore *et al.* (2007) contradict those of Gill and others (2006), reporting significant improvements in CK following elite competitive rugby matches using a cycle ergometer AR intervention when compared to passive recovery. Limitations on the transferability of the findings of Tessitore and colleagues (2007), and Gill and co-workers (2006) to elite youth soccer players are evident. Although using elite youth soccer players within their sample population, Tessitore and others (2007) implement recovery interventions around pre-season training events potentially producing lower physical outcomes and exertion in comparison to soccer matches, therefore limiting the amount of muscle damage inflicted. In comparison, Gill and colleagues (2006) utilised elite adult rugby players within their sample population, again affecting the transferability of findings between adult and youth athletes, and rugby and soccer players. As a result, further research on the effects of AR in elite youth soccer players following competitive soccer matches is required.

Hydrotherapy techniques, including CWT, CWI, and aerobic water-based exercise involve submersion muscles in water in an attempt to accelerate muscle recovery (Qu, *et al.*, 2020). These techniques are often implemented following competition in an elite applied environment, and may also be used following training sessions in preparation for upcoming fixtures (Seco-Calvo, *et al.*, 2019). Such techniques have shown significant improvements in muscle recovery when compared to alternative recovery techniques (Higgins, *et al.*, 2017; Stephens, *et al.*, 2016; Elias, *et al.*, 2013; Vaile, *et al.*, 2011). CWI is suggested to aid in the recovery process by assisting in the removal of metabolic by-products following exercise by improving venous return via vasoconstriction as a result of exposing muscle to cold water (Stephens, *et al.*, 2016) dampening the potentially harmful effects of pro-inflammatory cytokines involved in the removal of damaged muscle tissue (Peake, *et al.*, 2005), and the possibility of lipid peroxidation of ROS and $PLA_2$ resulting in a calcium influx and potential exacerbation of muscle damage. As with AR, numerous CWI protocols have been implemented throughout research, with literature of Machado and others (2016) supporting its use suggesting protocols using water temperatures between 11 and 15°C with submersion times between 10 and 15 minutes were most effective for both immediate and delayed recovery effects. Despite the extensive research of CWI interventions showing significant improvements in muscle recovery (Machado, *et al*., 2016; Elias, *et al*., 2013; Ascensão, *et al*., 2011; Rowsell, *et al*., 2009; Ingram, *et al*., 2009), there is a dearth of evidence supporting the use of CWI following competitive matches concerning elite youth soccer players when compared to alternative recovery interventions, and therefore this area requires further research. A meta-analysis of Machado and other (2016) advocated the use of CWI as a recovery intervention, however the literature reviewed failed to consider elite soccer players, and as such, further research may be required. Ascensão and colleagues (2011) compared the effects of CWI protocol to a thermoneutral protocol following a one-off soccer match of youth soccer players, reporting a significant improvement in indicators of muscle damage as a result of CWI, however limitations to their research are evident, as the training status of athletes may not qualify as elite, and the use of a one-off soccer match may not provide sufficient data to accurately assess the effects of recovery interventions following soccer matches, and again further research is necessary.

To date, no clearly defined recovery protocol has been identified and compared across recovery modalities demonstrating its effectiveness on improving recovery following elite youth soccer matches, and as such, coaches and athletes must rely on past research from non-specific sports or incomparable participant age and training statuses when implementing recovery interventions. It has been demonstrated that the use of traditional static stretching as a recovery intervention was ineffective for the elite youth sample population (Pooley, *et al*., 2017; chapter 5), and therefore this study aimed at determining the efficacy of alternative recovery interventions when compared to static stretching following competitive soccer matches of elite youth soccer players. As with chapter 5, the present study aimed to assess the current recovery practices of an elite youth soccer academy, to determine the effects of such interventions on indicators of muscle damage.

## 7.2 Methodology

### 7.2.1 Participants

Fifteen elite youth soccer players (mean [± SD]: age 16 [1] years, stature 176.4 [5.1] cm, mass 64.9 [5.6] kg) from a professional football academy in the English Premier League voluntarily participated in this study. Prior to participating in the study, participants were informed of any risks that may occur, and player and parental consent was obtained. All procedures were conducted in accordance with the Declaration of Helsinki, and the study was ethically approved by the Faculty of Science, Engineering and Computing Ethics Committee (Kingston University London, UK).

### 7.2.2 Experimental Design

Participants were required to complete three 80-minute competitive soccer matches (2x40min per match) for each recovery intervention [static stretching (SS), cold water immersion (CWI) and active recovery (AR)], where interventions were implemented in a cross-sectional fashion upon completion of the respective soccer matches. In order to assess the extent of muscle damage elicited from matches, markers of muscle damage via muscle oedema, creatine kinase (CK), countermovement jump with arms (CMJA) and perceived muscle soreness (PMS) were measured before (pre), immediately after (post), and 48 hours after (48 hours post) each competitive soccer match. All indicators of muscle damage are commonly used markers in the assessment of muscle damage and recovery (Darani, *et al*., 2017; Gill, *et al*., 2006; Ingram, *et al*., 2009; Ascensão, *et al*., 2011; Kinugasa & Kilding, 2009; Bahnert, *et al*., 2013).

### 7.2.3 Experimental Protocol

#### 7.2.3.1 *Physical Assessments*

Upon arrival to the match facility, participant stature and mass were recorded, immediately followed by the recording of PMS at 2.5 hours prior the match commencing. PMS was indicated on a 10-point visual analogue scale (VAS) from 0.5 to 5 with 0.5 increments.

Immediately following PMS assessment, muscle oedema was taken using a constant-tension tape measure to assess muscle circumference (chapter 3) using three sites of the lower body; the two sites on the lower leg were identified by 1/3 (OedemaG1) and 2/3 (OedemaG2) of the lower leg length calculated by the distance from the medial condyle of the Tibia to the Calcaneus. The site on the upper leg (OedemaQ) was identified by the midpoint of the distance from the Patella to the anterior superior iliac spine. Following these initial assessments, CK levels were assessed using fingertip whole blood samples, and analysed using the i-STAT 1 Analyser (Abbott Point of Care, Abbott Park, Illinois, USA). Two hours prior to exercise, CMJA was recorded using the Smart Speed Jump Mat (Fusion Sport), with participants completing three maximal jumps with peak jump performance recorded for analysis.

#### 7.2.3.2 *Recovery Interventions*

Prior to any participation in matches, a physical and technical preparation warm-up was conducted by a sport scientist and UEFA qualified coach respectively. Warm-ups remained consistent throughout the duration of the study, comprising 15 minutes physical preparation involving muscle activation, movement preparation, dynamic stretching and mobility, and

15 minutes of positional and technical football work. For consistency, the warm-ups were conducted by the same sport scientist and coach prior to all competitive games.

Immediately following completion of the competitive ‘home’ fixtures, participants were randomly assigned to either the CWI [10 minutes submersion to the point of Illiac Crest in cold water set to 14 ±0.8$^o$C (Machado, *et al*., 2016)] or AR [10 minutes low-intensity exercise on a cycle ergometer at 80-100rpm ~80 Watts (Gill, *et al*., 2006)] intervention, or the static stretching protocol (chapter 5; Pooley, *et al*., 2017) performed following ‘away’ fixtures (two 15 second stretches to the gastrocnemius, hamstrings, quadriceps, glutes, hip flexors, adductors and abductors, see chapter 5). Upon completion of the recovery protocols, participants were required to repeat the assessment of muscle damage markers in the same order as taken pre-match. Post-match assessments were undertaken within 30 minutes of completing the match. The same assessments of muscle damage were recorded at 48 hours post-exercise. On every occasion, the assessments were carried out in the same order and by the same sport scientist. The time intervals of assessments (pre-, post- and 48 hours post-match) were consistent with those used throughout literature (Pooley, *et al*., 2017; Ascensão, *et al*., 2011; Magal, *et al*., 2010; Brown, *et al*., 1997).

#### 7.2.3.3 *Player Exclusions*

For the purpose of control, and for the monitoring of physical outcomes of competitive soccer matches, Global Positioning Systems (GPS) units fitted with accelerometers were worn by all participants when competing in games. The GPS units (STATSports Apex) reported movement variables across all axes, as well as reporting heart rate data. Individual thresholds based on these movement parameters were set according to the average of the combined variables for all full competitive matches completed. As a result, match percentages were calculated, with the average of dynamic stress load (fatigue score calculated using player movements, steps and collisions), metres per minute, speed intensity, high metabolic load distance, high speed running distance (distance covered above 65% of an individual's maximum speed), accelerations, decelerations, heart rate minutes above 85% of max., and heart rate exertion over all full games amounting to a match percentage of 100%. This match percentage determined the individual intensity of matches completed for all players. To ensure the intensity of each competitive football match remained consistent throughout the study, any participant whose match percentage for any particular game was ± 10% of their average match percentage for all completed matches were excluded from the data collection for that specific competitive match. Furthermore, participants were excluded from data collection for individual games if they failed to complete a minimum of 80% of the 80 minute match whilst goalkeepers were excluded from the study due to the inability to control consistency in physical loading for matches. Of a potential squad of 24, seven players were excluded for not completing a minimum of 80% of match play and two were goal keepers. As such a total of 15 players were included in the final analysis.

### 7.2.4 Statistical Analysis

Data are presented as mean ± standard deviation (SD) unless otherwise stated. Analysis of indicators of muscle damage commenced following assessment for normality in all data, verified using the Shapiro-Wilk test. To analyse differences in assessment markers (CK, CMJA, PMS) a two-way within-subjects repeated measures analysis of variance (ANOVA) was used comparing [conditions SS, CWI and AR] x time points (pre, post-, and 48-hours post-exercise) using statistical software SPSS 23 (SPSS Inc., Chicago, IL, USA). Data of assessment markers were also transformed to indicate percentage change from pre-match baseline values to indicate the percentage of recovery achieved (Pooley, *et al.*, 2017; Duffield, *et al.*, 2010; Howatson, *et al.*, 2009; Ingram, *et al.*, 2009; Goodall & Howatson, 2008; Montgomery, *et al.*, 2008; Gill, *et al.*, 2006). A Bonferroni statistical test was used for post-hoc analysis. Effect sizes were calculated using partial eta$^2$ ($\eta^2_p$) where 0.01 = small; 0.06 = medium; and 0.14 = large effect (Field, 2009) and 95% bootstrap confidence intervals were used to detect differences in trends in the data, where negative/positive values indicate negative/positive relationships respectively, and intervals crossing zero indicate no effect (Field, 2009). Statistical significance was accepted at $p < 0.05$.

## 7.3 Results

All analysis of all pre-match markers of muscle damage between static stretching, AR and CWI revealed no significant differences suggesting that for all three conditions, participant's pre-match physical conditions were similar allowing an accurate comparison of post-match and 48 hours post-match data.

Analysis of data within conditions across time intervals (pre-, post-, 48 hours post-) revealed a significant increase in PMS immediately following (post-) competitive soccer matches when compared to pre-match measures ($p < 0.001$, $\eta^2_p$: 0.961, 95% CI 2.373 to 2.767) and was significant for all conditions (SS: $p < 0.001$, $\eta^2_p$: 0.981, 95% CI 2.558 to 3.162, AR: $p < 0.001$, $\eta^2_p$: 0.960, 95% CI 2.156 to 2.944, CWI: $p < 0.001$, $\eta^2_p$: 0.961, 95% CI -1.950 to 2.650; Table 7.1). PMS collected at 48 hours post-match was significantly elevated in comparison to pre-match values for all recovery interventions (SS: $p < 0001$, $\eta^2_p$: 0.956, 95% CI 1.450 to 1.970, AR: $p = 0.003$, $\eta^2_p$: 0.643, 95% CI 0.328 to 1.172, CWI: $p = 0.013$, $\eta^2_p$: 0.514, 95% CI 0.122 to 0.798), with all three interventions significantly reduced from post-match values (SS: $p < 0.001$, $\eta^2_p$: 0.956, 95% CI 0.965 to 1.335, AR: $p < 0.001$, $\eta^2_p$: 0.967, 95% CI 1.550 to 2.050, CWI: $p < 0.001$, $\eta^2_p$: 0.923, 95% CI 1.438 to 2.242). CMJA (cm) performance was significantly reduced immediately post-match when compared to pre-match values for all recovery interventions (SS: $p < 0.001$, $\eta^2_p$: 0.847, 95% CI 4.019 to 7.803, AR: $p < 0.001$, $\eta^2_p$: 0.896, 95% CI 2.811 to 4.757, CWI: $p < 0.001$, $\eta^2_p$: 782, 95% CI 2.431 to 5.649). At 48 hours post-match, CMJA following static stretching intervention remained significantly reduced, ($p < 0.001$, $\eta^2_p$: 0.784, 95% CI 1.944 to 4.496) however no significant differences were observed between pre- and 48 hour post-match CMJA performance following AR and CWI interventions (Table 7.1).

A significant increase in CK (ng/mL) was observed between pre- and post-match measure within all conditions (SS: $p < 0.001$, $\eta^2_p$: 0.944, 95% CI 3.782 to 5.478, AR: $p < 0.001$, $\eta^2_p$: 0.918, 95% CI 3.502 to 5.538, CWI: $p = 0.001$, $\eta^2_p$: 0.734, 95% CI 2.276 to 6.064), with a significant reduction in CK observed between post- and 48 hour post-match measures taken following all recovery interventions (SS: $p < 0.001$, $\eta^2_p$: 0.814, 95% CI 1.280 to 2.700, AR: $p < 0.001$, $\eta^2_p$: 0.857, 95% CI 2.594 to 4.906, CWI, $p < 0.001$, $\eta^2_p$: 0.829, 95% CI 2.770 to 5.650), however no significant difference between pre- and 48 hour post-match CK was observed following CWI intervention.  No significant differences in muscle oedema were identified between any time intervals, for all recovery interventions.

**Table 7.1** Comparison of mean (± SD) physiological, psychological and performance markers of muscle damage at pre-, post- and 48 hours post-competitive soccer matches in recovery interventions (n=15).

| | **Assessment time point** | | |
|---|---|---|---|
| | **Pre** | **Post** | **48hrs Post** |
| PMS (0.5-5) | | | |
| SS | 1.2 (0.2) (b) | 4.1 (0.3) (a) | 3.0 (0.3) (a) (b) |
| AR | 1.6 (0.5) (b) | 4.1 (0.2) (a) | 2.3 (0.3) (a) (b)* |
| CWI | 1.7 (0.5) (b) | 4.0 (0.5) (a) | 2.1 (0.4) (a) (b)* |
| CK (ng/mL) | | | |
| SS | 3.0 (1.6) (b) | 7.6 (2.1) (a) | 5.7 (2.2) (a) (b) |
| AR | 3.0 (1.0) (b) | 7.5 (1.3) (a) | 3.8 (1.0) (a) (b)* |
| CWI | 3.2 (1.2) (b) | 7.4 (2.9) (a) | 3.2 (1.2) (b)* |
| CMJA (cm) | | | |
| SS | 46.6 (6.3) (b) | 40.6 (8.1) (a) | 43.0 (6.9) (a) (b) |
| AR | 44.8 (5.0) (b) | 41.1 (4.1) (a) | 44.4 (4.1) (b) |
| CWI | 44.2 (5.3) (b) | 40.2 (4.7) (a) | 44.4 (5.2) (b) |
| OedemaG1(cm) | | | |
| SS | 26.0 (2.7) | 26.2 (2.4) | 26.7 (2.5) |
| AR | 25.2 (1.5) | 25.7 (1.4) | 25.6 (1.5) |
| CWI | 25.0 (1.7) | 25.6 (1.5) | 25.9 (2.0) |
| OedemaG2 (cm) | | | |
| SS | 35.5 (2.3) | 35.2 (1.8) | 35.4 (1.6) |
| AR | 34.7 (1.9) | 35.2 (1.9) | 34.9 (2.0) |
| CWI | 34.6 (2.0) | 35.0 (1.7) | 34.3 (1.8) |
| OedemaQ (cm) | | | |
| SS | 49.3 (4.2) | 50.3 (4.0) | 49.5 (3.8) |
| AR | 49.7 (2.6) | 50.3 (2.7) | 50.0 (2.8) |
| CWI | 49.3 (2.9) | 50.3 (3.0) | 49.6 (2.7) |

(a) $p < 0.05$ = significantly different from Pre; (b) $p < 0.05$ = significantly different from Post;
* $p < 0.05$ = significantly different from SS, ▪ $p < 0.05$ = significantly different from AR

SS, static stretching; AR, active recovery; CWI, cold water immersion

Analysis of data between conditions across time intervals produced no significant differences ($p > 0.05$) between any interventions pre-match, and immediately post-match when assessing CK (ng/mL), CMJA (cm), muscle oedema (cm) and PMS, however at 48 hours post-match, PMS significantly reduced following AR ($p = 0.003$, $\eta^2_p$: 0.644, 95% CI 0.281 to 0.999) and CWI ($p < 0.001$, $\eta^2_p$: 0.766, 95% CI 0.478 to 1.162) when compared to SS. Assessments of CK at 48 hours post-match significantly reduced following AR ($p = 0.020$, $\eta^2_p$: 0.468, 95% CI 0.334 to 3.066), and CWI ($p = 0.031$, $\eta^2_p$: 0.430, 95% CI 0.255 to 4.205) when compared to static stretching.

In order to determine the effects of recovery interventions on the percentage change of CK, CMJA and PMS, further statistical analysis was conducted. This analysis, consistent with previous research (Pooley, *et al.*, 2017; Duffield, *et al.*, 2010; Howatson, *et al.*, 2009; Ingram, *et al.*, 2009; Goodall & Howatson, 2008; Montgomery, *et al.*, 2008; Gill, *et al.*, 2006), also eliminated the potential inter-individual variability of CK. Results showed a significant reduction in CK following AR ($p = 0.014$, $\eta^2_p$: 0.507, 95% CI 20.106% to 135.634%), and CWI ($p < 0.001$, $\eta^2_p$: 0.794, 95% CI 67.572% to 151.769%) when compared to SS at 48 hours post-match (Figure 7.1). A significant reduction in CK values following CWI recovery intervention at 48 hours post-match was also reported ($p = 0.049$, $\eta^2_p$: 0.366, 95% CI 0.217% to 62.383%) when compared to AR. Percentage change from pre-competition levels at immediately post- and 48 hours post-match between conditions were also analysed for CMJA and PMS, with results showing significant differences were identified between static stretching and AR ($p = 0.016$, $\eta^2_p$: 0.494, 95% CI 1.530% to 11.390%), and SS and CWI ($p = 0.001$, $\eta^2_p$: 0.693, 95% CI 3.811% to 11.509%) at 48 hours post-competition (Figure 7.2) when assessing CMJA, whilst significant differences at 48 hours post-match between static stretching and AR ($p = 0.005$, $\eta^2_p$: 0.605, 95% CI 33.284%

to 137.016%), and static stretching and CWI ($p = 0.001$, $\eta^2_p$: 0.725, 95% CI 57.452% to 157.208%) were identified when assessing PMS (Figure 7.3).

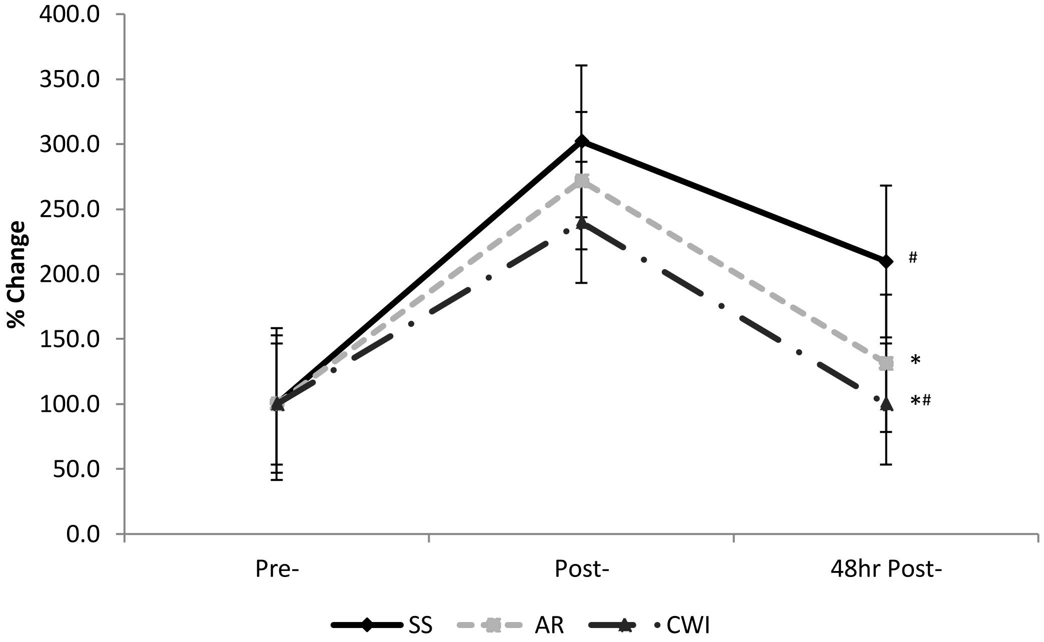


**Figure 7.1** Percentage change in CK (ng/Ml) levels between pre-exercise, immediately post-exercise and 48 hours post-exercise, grouped by condition (SS, static stretching; AR, active recovery; CWI, cold water immersion).

Error bars represent SE at respective time points.
*$p < 0.05$, significantly different from SS.
$^{\#}p < 0.05$, significantly different from AR (n=15).

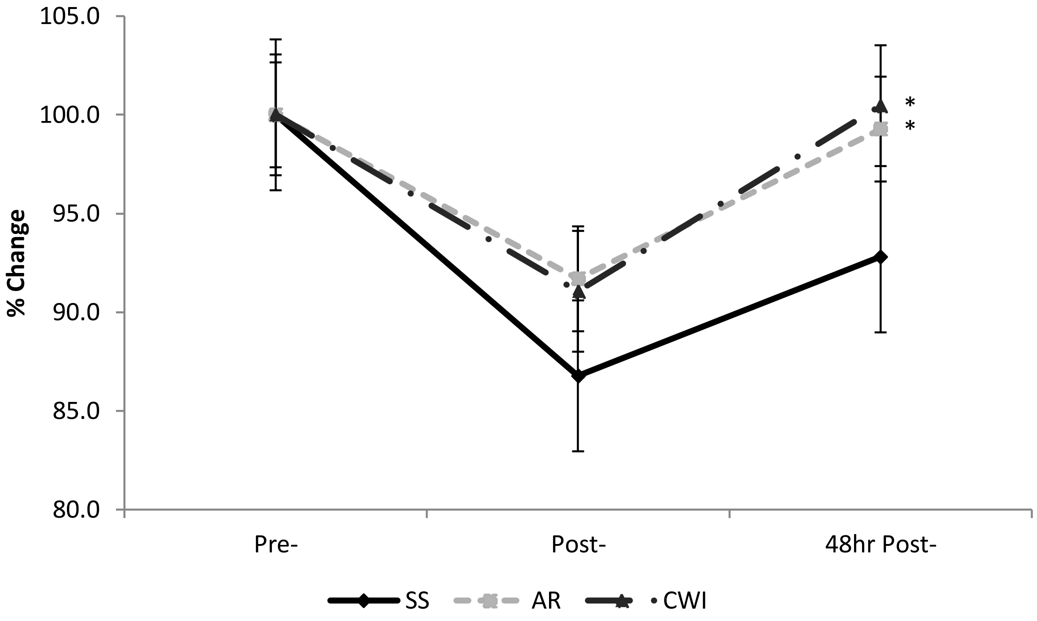


**Figure 7.2** Percentage change in CMJA (cm) performance between pre-exercise, immediately post-exercise and 48 hours post-exercise, grouped by condition (SS, static stretching; AR, active recovery; CWI, cold water immersion).

Error bars represent SE at respective time points.
*$p < 0.05$, significantly different from SS (n=15).

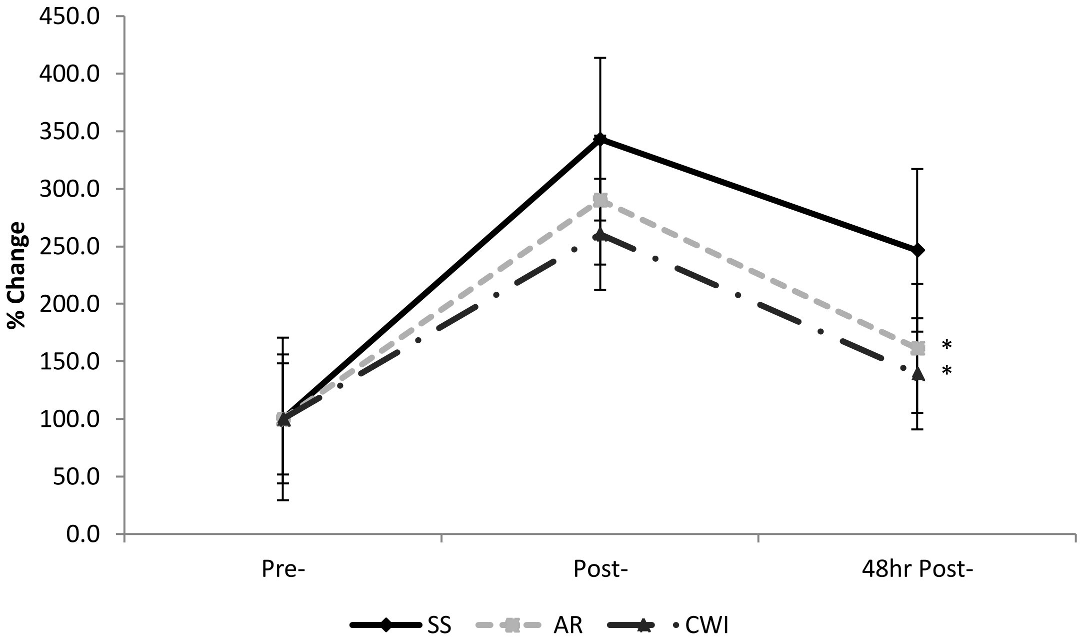


**Figure 7.3** Percentage change in PMS between pre-exercise, immediately post-exercise and 48 hours post-exercise, grouped by condition (SS, static stretching; AR, active recovery; CWI, cold water immersion).

Error bars represent SE at respective time points.
*$p < 0.05$, significantly different from SS (n=15).

## 7.4 Discussion

The aim of the present study was to assess the effects of active recovery (AR) and cold water immersion (CWI) on muscle damage and muscle recovery interventions following elite youth soccer matches. The main findings from the present study showed CWI and AR significantly attenuate markers of muscle damage over a 48 hour period in comparison to conventional static stretching, as demonstrated by a significant reduction in perceived soreness and CK, and a significant increase in CMJA when compared to the static stretching protocol at 48 hours post-match. Immediately post-match, no significant differences between recovery intervention groups were identified for all markers of muscle damage (Table 7.1), suggesting the exposure to soccer matches elicited a consistent level of muscle damage allowing for an accurate comparison of the effects of recovery interventions at 48 hours post-match. All indicators of muscle damage (CK, PMS, CMJA) revealed a significant increase immediately post-match when compared to pre-match values, demonstrating that competitive youth soccer matches significantly induce muscle damage, which further supports the findings of previous research (Pooley, *et al*., 2017; chapter 5), and highlights a demand for the use of effective recovery interventions to assist in returning athletes to their pre-competition status.

When considering the effects of static stretching and AR on the attenuation of CK, markers of CK taken at 48 hours post-match were significantly reduced in comparison to post-match values, however were also significantly elevated when compared to pre-match values. This would suggest that full recovery had not occurred at 48 hours post-match and supports the findings of chapter 5, although when analysing the effects of CWI on CK, data collected 48 hours post-match showed a significant reduction in comparison to that collected immediately post-match, with no significant difference between pre-match and 48 hours post-match with CK values returning to baseline. These findings contradict those of

Ascensão *et al*. (2011) who, although presenting significant differences in CK values following CWI and thermoneutral recovery interventions, found CK remained elevated at 24-, and 48-hours post-match in comparison to pre-match data, therefore not returning to baseline. The differences in findings may be attributed to the use of a one-off soccer match by Ascensão and co-workers for sample collection – potentially exposing participants to physical outcomes of soccer matches that are outside an average range, therefore affecting the validity of their findings. Additionally, the participant training status of Ascensão and others (2011) may not replicate the elite standard of those assessed in the present study, and as has been previously suggested muscle damage sustained, and therefore recovery time, may be reduced due to the use of elite participants (Owens, *et al*., 2019). The present study showed no such limitations and so adds to the existing evidence for support of CWI as a recovery intervention following elite youth soccer matches. Further research comparing the changes in CK levels following recovery interventions can be compared to the present study. Gill and others (2006) report a recovery percentage of CK at 36 hours post-match at ~60% following an AR intervention, with the recovery percentage improving to 88% at 84 hours post-match. Although time intervals used by Gill and others differ from those used in the present study, the recovery trends are complementary, with both interventions having a positive effect on CK following exposure to game situations at 48 hours post-match in the present study and 36 and 84 hours post-match in the study of Gill and others (2006). Furthermore, Goodall and Howatson (2008) report a significant reduction in CK at 48 hours post-eccentric exercise when compared to values taken at 24 hours post-exercise following CWI, again supporting the findings of the present study. It has been suggested that exposure to CWI following exercise may elicit a reduction in capillary permeability (Broatch, 2015) due to the vasoconstrictive effect of CWI, as a result attenuating the efflux of CK from damaged muscle fibres. This theory may provide an explanation for the reduction in CK at

48 hours post-match following CWI intervention, and the return of CK to baseline measure seen in the present study. Although capillary permeability was not measured in the present study, this may be an area for further investigation in future studies.

A comparison of CMJA performance across time intervals showed a significant increase in jump height at 48 hours post-match when compared to values recorded immediately post-match in all three conditions suggesting muscle recovery had occurred, and supporting the findings of chapter 5, however when comparing values recorded at 48 hours post-match to pre-match measures following the static stretching intervention, jump heights remained significantly lower. In comparison AR and CWI interventions showed no significant differences between pre-match and 48 hour post-match jump heights suggesting that performance indicators of muscle damage have returned to baseline, and participants had fully recovered (Table 7.1; Figure 7.2). These results support the findings of Elias and others (2013) who reported a return to baseline in performance markers (CMJA and sprint time) at 48 hours post-match following a CWI intervention (14 min submersion at 12˚C) when monitoring male professional Australian soccer players. Additionally, Ascensão and colleagues (2011) reported a significant reduction in CMJA performance at 24 hours post-match when compared to baseline values, however no significant differences between baseline and CMJA performance at 48 hours post-match were found, suggesting the AR intervention assisted in returning performance markers to baseline values, and supporting the findings of the present study. As described by Peake and co-workers (2017a), one potential mechanism that may explain muscle damage is the ‘popping sarcomere theory’, whereby muscle damage is caused following eccentric muscle contractions, where sarcomeres are overstretched beyond the point of filament overlap and is likely to result in a reduction in force production, as seen in the present study with reduced CMJA heights immediately post-match. It may be possible that the use of static stretching as a recovery mechanism exacerbates muscle damage by stretching further sarcomeres beyond the point of filament overlap. Peake and others (2017a) also suggested that stretching of sarcomeres may open stretch-activated channels allowing calcium to enter the cytosol through these open channels of the sarcolemma which may in turn stimulate Calpain enzymes to degrade contractile proteins, providing further reductions in force production. The stimulation of Calpain enzymes and degradation of contractile proteins was not measured in the present study, however, may be an area for consideration in future studies.

When considering PMS, measurements of all three recovery interventions at 48 hours post-match demonstrated a significant improvement from values recorded immediately post-match, however all remained significantly elevated compared to pre-match data. This would suggest that although perceived soreness had improved, the recovery process was yet to be completed. These findings support those of chapter 5 (Pooley, *et al.*, 2017), demonstrating PMS remained elevated at 48 hours post-match when comparing static stretching to passive recovery (Table 5.1). The findings of the present study suggest that even with the addition of AR or CWI interventions, perceived soreness remains elevated. These findings may be of benefit to coaches to understand that although some physiological and performance markers of muscle damage suggest muscle recovery has occurred, athletes perceptions suggest otherwise and therefore may demonstrate a reduced exertion resulting in reduced performance in sessions in the days immediately following competitive soccer matches, potentially as a protective mechanism for the prevention of injury or further damage. These findings should be taken into consideration by coaches, alongside the technical and tactical aims of training sessions, and in order to cater for these athletes still in a state of recovery, a reduced training volume and intensity may be advised for sessions 48 hours post-match.

When comparing the effects of recovery interventions on PMS at 48 hours post-match, AR and CWI demonstrated significantly lower values than static stretching, suggesting these interventions provide a superior recovery benefit, however no differences between AR and CWI were identified which may imply both interventions provide similar levels of recovery effects. It has been suggested that the use of CWI as a recovery intervention may assist in reducing perceived soreness (Broatch, 2015) due to reduced firing rate of pain sensory receptors in the skin after cooling and therefore reducing the pain sensation. Furthermore, Broatch (2015) suggests CWI induced vasoconstriction may limit perceived pain via a reduction in inflammation and the osmotic pressure of exudate, therefore decreasing pressure exerted on pain signalling nociceptors. The assessments of muscle oedema in the present study demonstrated a lack of significant differences between all time intervals, and as seen in chapter 5, may be attributed to the use of compressive tapes of athletes during soccer matches which was not altered due to the applied nature of the study. It may also be possible that the assessment method of muscle oedema was not sensitive enough to detect changes of swelling across time intervals, and therefore future research may be required to assess the suggestions of Broatch (2015) that reduced inflammation may result in a reduction in pain post-exercise, using alternative methods of muscle oedema assessment.

Analysis of the effects of static stretching, AR and CWI on CMJA revealed no significant differences between any recovery interventions, suggesting AR and CWI provided no greater recovery benefit than static stretching (Table 7.1). As such, analysis was undertaken using a recovery percentage, aligning baseline values. When analysing data in the form of percentage change, pre-match values were set to 100%. The purpose of this was to remove the inter-individual variance of markers of muscle damage, whilst determining the recovery percentage at 48 hours post-match in comparison to baseline figures to establish intervention effectiveness. PMS percentage change revealed a significant reduction for AR and CWI when compared to static stretching at 48 hours post-match. Furthermore, a significant improvement between 48 hour post-match and immediate post-match data was identified for all recovery conditions, with no significant differences identified between pre-match and 48 hour post-match PMS for AR and CWI suggesting an improved recovery effect in comparison to SS. The significant reduction in PMS following CWI when compared to static stretching may be supported by the findings of Elias *et al.* (2013) who reported CWI interventions the most effective for reducing perceived muscle soreness following a practice match of professional male Australian football players when compared to CWT. As aforementioned, these findings may be attributed to CWI dampening the neural signalling, reducing the firing rate of pain sensory receptors in the skin after cooling, and a reduced nerve conduction velocity resulting in decreased sensation of pain (Broatch, 2015).

Analysis of CK data in the form of percentage change at 48 hours post-match also shows a significant reduction in AR and CWI values when compared to static stretching, suggesting both interventions provide a significant recovery benefit. Again these findings support those of previous research – Ascensão and colleagues (2011) report a significant improvement in CK at 48 hours post-match following CWI when compared to a thermoneutral intervention, suggesting the vasoconstriction effects of the CWI protocol assisted in the removal of waste products and inflammatory cytokines (Broatch, *et al.*, 2018) following soccer match exposure, reducing the potential for the exacerbation of muscle damage due to lipid peroxidation of healthy cell membranes (Close, et al., 2005) caused by phagocytosis (Kendall & Eston, 2002) and the production of ROS (Close, et al., 2005). Similar findings were made by Gill and others (2006), reporting a significant improvement in CK as a result of AR when compared to a passive recovery intervention, however this was found at 84 hours post-match. The AR intervention utilised in the present study was based on that of Gill *et al.* (2006) and with such positive findings would suggest a similar outcome would have been witnessed by Gill and others had measures been taken at 48 hours post-match. Interestingly, the present study found a significant reduction in CK following CWI when compared to AR, suggesting CWI may be a superior recovery intervention for elite youth soccer players, and expanding on the findings of chapter 5.

A significant improvement in 48 hour post-match CMJA performance in the form of percentage change was identified when comparing AR and CWI to static stretching. The findings of this study support previous research conducted by Gill and co-workers (2006) stating a significantly improved recovery effect when comparing AR to passive recovery assessments of elite adult rugby players. However, the significant improvement in markers of muscle damage following the AR intervention have been conflicted by findings of more recent research on elite youth soccer players produced by Tessitore and co-workers (2007), with no significant differences between recovery interventions being identified. The differences in these results may be due to the AR intervention that was implemented, as that used by Tessitore and others (2007) consisted of 8 minutes walking, 8 minutes jogging and 4 minutes of dynamic movements – a protocol that, although longer in duration, may have been lower in intensity, resulting in a lower systemic blood flow than the AR intervention of the present study, potentially limiting the removal of blood toxins in skeletal muscles and allowing for possible further damage to be caused by pro-inflammatory cytokines (Chazaud, 2016), although further research to confirm these suggestions and compare the effectiveness of both AR interventions may be required. Positive findings for the CWI intervention may be attributed to reduced fluctuations of limb blood flow as demonstrated by Fiscus and others (2005). Peiffer and colleagues (2009) report a reduction in femoral venous diameter following cold water immersion post-exercise (20 minutes submersion at 14.3˚C) which remain for up to 45min post-intervention at which point, sample collection was terminated. This may suggest CWI interventions provide an enhanced recovery benefit due to a continued reduced muscle temperature and femoral venous diameter in the time following intervention termination, potentially providing an enhanced recovery when compared to alternative interventions.

In conclusion, the results of this study showed CWI and AR significantly attenuate markers of muscle damage over a 48 hour period in comparison to conventional static stretching. The findings of the present study support previous research (Elias, *et al*., 2013; Ascensão, *et al*., 2011; Ingram, *et al*., 2009; Gill, *et al*., 2006), suggesting AR and CWI significantly improve indicators of muscle damage following competitive sporting matches, whilst providing new evidence to support the use of AR and CWI as recovery interventions for elite youth soccer players following competitive soccer matches. The findings of 48 hour post-match values of CK and CMJA returning to pre-performance measures following the CWI intervention may indicate this is a superior recovery intervention over AR, however this would require further investigation. Owing to the mode of AR protocol used in the present study, the practicality of a soccer team completing a CWI intervention may be advantageous, along with the superior significant findings when deciding the intervention of choice.

## 7.5 Practical Implications

In light of the current findings, it may be advised that CWI and AR interventions provide an enhanced recovery effect when compared to static stretching. Additionally, there would appear to be limited differences in recovery benefit between CWI and AR, suggesting use of either protocol would provide similar and significant improvements in muscle recovery, however it is feasible to predict that a combination of both AR and CWI may provide superior recovery benefits than either alone, although the order of intervention use must be considered. The findings of this study would suggest that, in the absence of CWI (in cases such as away fixtures), the use of AR would provide a significant recovery benefit similar to that of CWI, however again mode of intervention must be considered for practicality, as the AR protocol used in the present study requires cycle ergometers. Should similar recovery results be found using little or no equipment, AR on away fixtures may be advised for improving markers of muscle damage at 48 hours post-match. It must also be considered that PMS remains elevated at 48 hours post-match, regardless of the effectiveness of recovery intervention, potentially resulting in reduced physical exertion of athletes in subsequent training sessions or possible injury should coaches chose to ignore perceived soreness and plan sessions based on physiological markers of muscle damage. In applied settings such as elite academy soccer it may be recommended that recovery interventions are sought after all competitive soccer matches (home and away) to assist in reducing muscle damage at 48 hours post-match.

# 8 General Discussion

In elite youth soccer, where the emphasis is on the development of young athletes for successful careers in the adult game, coaches will aim to maximise training opportunities to optimise the learning possibilities of players within elite academies. As a result, it is imperative that players are given the opportunity to recover, and it is therefore important for practitioners to understand the physiological effects of elite youth soccer matches on muscle damage and muscle recovery and utilise effective recovery interventions.

The aim of this thesis was to examine the effects of elite youth soccer matches by establishing the extent of elicited muscle damage which remained for up to 48 hours; the physical components of soccer matches (high-intensity accelerations, decelerations, and absolute high speed running) that contribute to muscle damage (as indicated by elevated post-match creatine kinase); and the physiological and performance effects of recovery interventions (static stretching, active recovery and cold-water immersion) on accelerating the recovery process following muscle damage induced by elite youth soccer matches (of which static stretching was ineffective). The investigations were conducted within an elite soccer academy of a professional English soccer team competing at the highest domestic and European level, with the objective of providing practical knowledge to the applied industry for the assessment of muscle damage and improvement in recovery interventions, hence improving and optimising training and performance.

## 8.1 Overall Discussion

The assessment of recovery interventions has gained considerable academic attention, however due to the accessibility of elite sporting environments often being restricted to researchers, much of the research to date has been conducted on non-elite participants using eccentric exercise protocols. Despite the use of such protocols successfully inducing muscle damage, and subsequent recovery interventions being implemented to assess their effectiveness, it was considered likely that soccer matches of elite youth soccer players would induce greater muscle damage than eccentric exercise protocols. Until now, and to the best of the authors knowledge, no research has compared muscle damage elicited via isolated eccentric muscle damaging exercise to that of elite youth soccer matches (chapter 4).

The findings of chapter 4 revealed that despite eliciting significant muscle damage, the eccentric exercise protocol assessed induced significantly less muscle damage than elite youth soccer matches. These findings, although unique to their environment, may provide evidence that current practices of applied environments that are implemented based on laboratory-style research findings may not be providing the expected outcomes as reported in research articles. It is therefore advised that where possible, club and institution practices are assessed and evaluated in their intended environment prior to being implemented. This study, and studies of this type cross the boundary of efficacy and effectiveness research, whereby the ability to maintain complete control and consistency was ensured throughout the assessment of the eccentric protocol, however the unpredictable nature of soccer matches resulted in inevitable ‘noise’, and whilst every effort to ensure control was maintained, as with many applied studies and such is the nature of effectiveness research, some ‘noise’ was unavoidable. This demonstrates a possible drawback to chapter 4, however as discussed throughout this thesis, the dearth of applied research in this subject area creates a demand for such studies to inform athletes, coaches and practitioners. The interpretation of findings from chapter 4 must be considered with caution, as although assessment markers indicated significant muscle damage due to both exercise modalities, samples were taken immediately (45 minutes) post-exercise, and it may be argued that at such a time-point, the manifestation of muscle damage is yet to occur, and impairment of assessment measures is due to fatigue.

The terms muscle damage and fatigue appear to be used interchangeable throughout research, particularly in cases referring to physiological assessments immediately post-exercise, and is something researchers and practitioners should consider when interpreting research articles and conducting studies.

Chapter 4 demonstrated both the eccentric exercise protocol and elite youth soccer matches significantly induce muscle damage immediately post-exercise, however in identifying soccer matches induced significantly greater muscle damage and considering the argument that assessments taken immediately post-exercise may be indicative of fatigue, chapter 5 assessed the effects of soccer matches on muscle damage up to 48 hours post-match. In doing so, chapter 5 also assessed the effects of static stretching on muscle recovery – a post-match recovery intervention regularly implemented within the professional soccer club involved in this thesis, and throughout academy soccer. The findings of chapter 5 support previous research advising against the use of static stretching as a recovery intervention, however do so in a relatively unexplored setting – following elite youth soccer match exposure. Given the continued use of static stretching in elite youth soccer academies despite literature advising against its use, the applied nature of chapter 5 may be of interest to coaches and practitioners when considering its implementation in future, however as with chapter 4, the findings of this thesis are unique to their environment, and may not be exactly replicated in other elite youth soccer academies or across other sports.

Chapter 5 also indicated elite youth soccer players are still in recovery at 48 hours post-match, providing coaches and practitioners a new understanding of the physiological adaptations of these athletes following match exposure. This may prove crucial in the planning and implementation of training sessions in the days following soccer matches, and continued exposure to high intensity actions and high volume sessions may be counter-productive for athletes still recovering, potentially exacerbating muscle damage or resulting in muscle injury. This may be of particular importance to coaches in periods of high fixture congestion when the recovery time between soccer matches is significantly reduced, as allowing greater recovery time and optimising recovery techniques is likely to reduce injury risk and allow for a greater physical performance in subsequent games.

Upon discovering that elite youth soccer matches significantly induce muscle damage remaining for up to 48 hours post-match, it was considered important to ascertain the physical parameters of soccer matches that significantly contribute to muscle damage immediately post-match. Chapter 6 assessed the correlation of physical soccer match parameters collected via global positioning systems (GPS) to indicators of muscle damage, revealing the number of high intensity accelerations and decelerations and absolute high speed running significantly contribute to post-match rises in creatine kinase. These findings may allow practitioners to determine which athletes have completed a significant physical performance and are therefore likely to suffer significant muscle damage. This may prove useful to practitioners, as GPS data can be fed to coaches in-game (live) and immediately upon its conclusion, allowing the implementation of optimal recovery interventions to players likely to experience muscle damage based on their physical match outputs in an attempt to minimise damage sustained and accelerate the recovery process. Given the nature of these physical performance variables, it is possible to suggest that the initial damaging mechanism appears to be mechanical (Owens, *et al.*, 2019; Varley, *et al.*, 2017). Research has demonstrated that following an initial bout of eccentric activity, a physical adaptation occurs referred to as the repeated bout effect, providing a protective mechanism against future eccentric exposure to limit the muscle damage experienced (Owens, *et al.*, 2019; Peake, *et al.*, 2017a). Despite this and given the samples in this thesis were collected over numerous soccer matches throughout soccer seasons, the significant increases in muscle damage observed would suggest there was little repeated bout effect for these athletes. This may be due to the participant age and training status, as it has been suggested that young athletes are less susceptible to muscle damage, whilst highly trained athletes experience a reduced repeated bout effect in comparison to non-trained athletes (Chen, *et al*., 2014). It is therefore possible that a sample population of highly trained, young participants would demonstrate a limited repeated bout effect, and therefore experience significant muscle damage post-match. This may suggest that the repeated bout effect has a limiting capability in providing a protective mechanism, however this is an area for possible future research. Alternatively, in addition to the mechanical muscle damage elicited as a result of eccentric and explosive muscle contractions such as accelerations, decelerations and high speed running, the volume of work completed, potentially accounted for by total distance may contribute to a metabolic mechanism of muscle damage as demonstrated by research assessing long distance cyclists (Tee, *et al*., 2007). However, a lack of significant correlation between total distance and muscle damage markers suggests further research may be required, and alternative muscle damage assessments such as muscle biopsies may be required to assess muscle glycogen concentrations, which have been shown to be depleted following soccer match exposure (Krustrup, *et al*., 2006), reducing ATP and therefore ATPase of calcium within muscle cells resulting in an inflammatory cascade of metabolic events that lead to muscle fibre degradation (Tee, *et al*., 2007). The correlations established in chapter 6 may also be of use to practitioners in future research studies as indicators of muscle damage. This would provide an alternative method for use as monitoring tools by coaches and practitioners for the early indication of muscle damage and subsequent recovery time, providing less invasive or counter-productive assessments when promoting recovery from elite soccer matches.

Chapters 4 to 6 of this thesis established that elite youth soccer matches significantly induce muscle damage greater than that of eccentric exercise protocols which remains for up to 48 hours post-match. Additionally, it was established that high intensity accelerations and decelerations, and absolute high-speed running contribute to muscle damage sustained post-match, and static stretching as a recovery intervention proved ineffective for reducing muscle damage and accelerating muscle recovery. Given these findings, it was necessary to determine if two commonly used recovery interventions (active recovery and cold-water immersion) assist in the recovery process and reduce muscle damage observed at 48 hours post-match. Chapter 7 assessed the effects of AR and CWI as recovery interventions following elite youth soccer match exposure, and in doing so, examine the protocols implemented within the elite youth soccer academy (as described in chapter 7). This study revealed CWI and AR significantly improve muscle recovery as indicated by improvements in muscle damage markers at 48 hours post-match, however, no significant differences were identified between AR and CWI, suggesting these interventions would provide similar and significant recovery benefits. It must be noted that, whilst this study compared the effects of static stretching, AR and CWI, the static stretching recovery intervention was implemented upon completion of away soccer matches only, whilst AR and CWI recovery interventions were implemented following home soccer matches only. Although the muscle damage inflicted from competitive matches did not differ significantly between conditions therefore allowing for comparisons between recovery interventions, this study did not account for the potential effects of uncontrollable factors, such as the travel to and from soccer matches which may consist of long inactive periods of sitting, allowing a pooling of an inflammatory response, potentially exacerbating muscle damage, and may be an area for future research.

The positive findings for the use of AR as a recovery intervention may be attributed to AR facilitating the removal of metabolic by-products post-exercise (Reilly & Ekblom, 2005) due to increased cardiac output and muscle blood flow (Peake, *et al.*, 2017b). This increase in muscle blood flow may assist in the clearance of pro-inflammatory cytokines preventing further cell damage (Chazaud, 2016) producing positive recovery benefits. Additionally, this study demonstrated that CWI is an effective muscle recovery intervention for the attenuation of muscle damage following elite youth soccer matches. The exposing of skeletal muscle to cold water has been suggested to assist venous return via increased vasoconstriction (Stephens, *et al*., 2017) and hydrostatic pressure, dampening the potentially harmful effects of pro-inflammatory cytokines involved in the removal of damaged muscle tissue (Peake, *et al*., 2005). This mechanism may be similar in principle to that of AR by accelerating the clearance of metabolic by-products, however CWI may also reduce perceived soreness via dampening of neural signalling (Broatch, 2015) and reducing inflammation and therefore osmotic pressure of exudate on pain signalling nociceptors (Broatch, 2015). Finally, CWI has been suggested to reduce capillary permeability (Broatch, 2015), potentially reducing the calcium influx and unwanted leakage of ROS and neutrophils to healthy cells (Close, *et al*., 2005), preventing further muscle damage. As previously mentioned, the implementation of AR and CWI following home games and static stretching following away games may highlight a limitation to this study, however in doing so provided a greater sample population to be included in statistical analysis, as the use of AR and CWI at away soccer matches was not possible. It must also be considered that the control of nutritional intake was limited due to the athletes departing the training centre post-match, however every attempt was made to ensure sufficient nutritional content was provided for both home and away soccer matches in an attempt to maximise the control of the study. Finally, the findings of this study are specific to their environment and may not be exactly replicated in other soccer academies and across different sports, and it is advised that the recovery interventions that practitioners intend to implement are assessed in their environment prior to implementation. Factors such as training programmes and strength and conditioning provision differ significantly across soccer clubs, and may be a contributing factor to the muscle damage and repair process.

## 8.2 Practical Applications and Impact

The findings of this thesis have impacted upon the professional soccer academy at which this research was conducted. Firstly, the identification of elite youth soccer players experiencing muscle damage that remained for up to 48 hours post-match, resulted in the alteration of the training programme to ensure recovery was aided. Previous training sessions 48 hours post-match may have contained actions of a high intensity and explosive nature, and although volume was always minimised, this exposure may have exacerbated muscle damage and hindered muscle recovery. As such, recommendations were passed to coaching staff to reduce the explosive actions of training sessions in the days following games, with these sessions adopting a highly technical focus, allowing an enhanced recovery process. In order to ensure soccer players maintained the same exposure to coaching staff and continued development throughout the training week, additional sessions were included once full recovery had been realised (later within the training week), maximising coach contact time. In alternative applied soccer settings, this may represent a suitable adaptation to the training programme to ensure recovery whilst maintaining relationships with coaching staff. It is possible that the recommendation of minimising coach contact to improve recovery without offering suggestions to increase coaching opportunities could impact negatively on the relationship with coaching staff, hindering the ability to implement new or improved practices. This thesis also identified that, the exposure to duration of soccer matches did not correlate to muscle damage elicited, rather the amount of exposure to high-intensity eccentric components, and as such it is advised that players should be considered on an individual physical level when planning subsequent training sessions, as opposed to time played. For many elite soccer teams it is common practice that ‘starters’ of soccer matches complete recovery sessions 48 hours post-match, with little consideration for substitutes given they complete a reduced number of match minutes, however it is recommended that is reconsidered and instead the amount of physical work completed – specifically high intensity accelerations and decelerations, and absolute high speed running – is utilised to determine who completes recovery sessions 48 hours post-match. That being said, a reduced duration of exercise is likely to reduce total load exposure and therefore limit potential metabolic muscle damage and is an area in need of further research.

The indicators of muscle damage used within this thesis (CMJA performance, perceived soreness and creatine kinase) have been adopted throughout the soccer club following their use and given the findings of this research. The assessment of jump performance and perceived soreness has continued to be implemented giving continuity to the monitoring assessments utilised within the soccer club, whilst creatine kinase assessments have been introduced at both First Team and academy level with great success. Although these measures now form part of the monitoring test battery, decisions are very rarely made based on their outcomes alone, and is something that should be considered if implemented within alternative soccer clubs or elite sporting environments. As has been shown throughout this thesis, muscle damage and muscle recovery are multi-factorial and these assessments, even when used collectively, may not give practitioners the full picture of the recovery process. Having said that, in some cases these assessments have been used in isolation to support decisions on player participation. When considering the transferability of these assessments to new environments, practitioners and researchers must consider numerous factors that may impact on their suitability. Firstly, the ability to maintain control and reproducibility of assessments in an applied sporting environment can prove difficult. Furthermore, some assessments used (e.g. CMJA) require complete participant cooperation. This may be difficult to ensure when considering the influence of coach and peer interference, and in some cases, assessments taken post-match may be influenced by athlete performance levels or team results. Consideration must also be taken when implementing subjective assessment markers, as athletes may feel their response determines squad selection and their resulting participation level. In order to avoid these limitations, it is essential athletes are able to trust the researchers and practitioners implementing the assessments, and education given to ensure their understanding of the protocols undertaken.

This thesis supported previous research in identifying static stretching as a recovery intervention provided no recovery benefit, and as such it is recommended its use is avoided. These findings have impacted the recovery protocol within the soccer academy of which this thesis was based, as static stretching as a recovery intervention has since been removed and alternative techniques sought. Given the academic support against static stretching, it may be advised soccer clubs avoid its use for assisting in muscle recovery. It should be noted that although it has been advised against the use of static stretching to aid muscle recovery, this thesis did not consider the assessment of range of motion or mobility, and therefore it has not been recommended to abandon static stretching completely as it may be useful in these situations. Furthermore, the identification of the effectiveness of active recovery and cold-water immersion has impacted on the recovery interventions implemented within the soccer academy, with CWI now implemented where possible post-match. In situations where CWI is not possible, active recovery is instead implemented due to there being no significant differences identified between protocols within this thesis. As has been suggested within this thesis, it is advised that prior to determining the recovery interventions being implemented, practitioners should consider their assessment within the environment of their intended use to determine their suitability.

Despite the findings of this research, it must be considered that all recovery interventions implemented were assessments of club protocols at the time the research was conducted. As such, alternative protocols may reveal differing results, and so the positive findings for active recovery and cold-water immersion may not be optimal. As aforementioned, one issue with identifying the optimal recovery protocols for interventions is the variety of protocols used throughout literature, and the many muscle damage assessments used, making it difficult to compare between studies for optimal muscle recovery. This is further hindered by the numerous sporting modalities and participant training and competition level, again making comparisons difficult, and should be considered by practitioners and researchers when interpreting the findings of this thesis.

## 8.3 Reflective Considerations

Whilst the completion of this thesis is very rewarding and leaves a feeling of fulfilment, the process itself was often very stressful, with little enjoyment and no view of the end goal. The combination of a full-time role in elite sport and completing a PhD was far more challenging than ever imaginable, and future researchers considering such a combination should understand the extent to which the roles combined will dominate both personal and professional time. It is also recommended that the research project and full-time role occupy topics of enjoyment and interest, in an industry that is both challenging and rewarding.

The balance between efficacy and effectiveness research was an area of discussion throughout this thesis. On one hand efficacy research was essential to ensure meaningful findings were maintained throughout, but on the other hand effectiveness research in an elite applied environment is often lacking due to limited researcher accessibility, and with such research come unavoidable ‘noise’ and a reduction in control. Both aspects of research are essential to developing the industry knowledge and application of findings at the highest level, and is something all researchers should consider before undertaking a research project.

# 9 Recommendations for Future Research

In attempting to answer questions around muscle damage and muscle recovery in elite youth soccer, this thesis uncovered numerous areas for further investigation. One area that remains somewhat unexplored is the assessment of metabolic muscle damage following soccer match exposure. As discussed in this thesis it would appear that the initial mechanisms of muscle damage can be attributed to mechanical stress as displayed by significant correlations of markers of muscle damage to physical match performance variables of a high intensity, eccentric nature (chapter 6). However, a dearth of research surrounding metabolic stress exists, and given soccer consists of a large proportion of low intensity physical exertion as well as high intensity bursts it is plausible that metabolic stress contributes to muscle damage elicited. In support of this, previous research has demonstrated that an increase in muscle damage following long distance cycling (Tee, *et al.*, 2007) which consists of minimal eccentric actions and may suggest metabolic stress contributes to damage.

In order to assess metabolic stress post-match in soccer players, assessment markers beyond those used in this thesis must be considered. Previous research has assessed muscle cell glycogen depletion indicating reduced concentrations results in a decrease in ATP and therefore inhibited calcium removal via ATPase (Tee, *et al.*, 2007) increasing cytosol calcium concentrations. Increased cytosol calcium concentrations may lead to a loss in calcium homeostasis, activation of $PLA_2$, ROS production and calpain stimulation (Owens, *et al.*, 2019; Howatson & van Someren, 2008; Gissel, 2005) increasing muscle cell permeability, resulting in a ‘vicious cycle’ of calcium influx potentially culminating in cell necrosis and death. Research on metabolic stress and assessment of muscle cell glycogen depletion in soccer exist (Krustrup, *et al.*, 2011; Krustrup, *et al.*, 2006), however a great deal more is required to fully understand its place in soccer and soccer specific muscle damage.

This knowledge may aid the understanding of muscle damage mechanisms, allowing for further development of techniques to minimise future damage and optimise recovery techniques.

Although this thesis assessed commonly used markers of muscle damage, it by no means exhausted the available assessment markers which may be of interest to assess in future studies to provide a greater understanding of the muscle damage and repair process. Assessments of antibodies such as Immunoglobulin (Ig) A, IgG and IgM (Thorpe & Sunderland, 2012) have received some attention in relation to elite soccer and the effects of exercise on their expression (Freitas, *et al*., 2016; Morgans, *et al*., 2015; Owen, *et al*., 2014; Mortatti, *et al*., 2012; Thorpe & Sunderland, 2012), and may be important to consider with regards to the immune response to muscle damaging exercise. Additionally, calcium plays a vital role in the function of muscle and the muscle damaging process yet is often overlooked when considering muscle damaging assessments. Its measurement in the hours and days following muscle damaging exercise and elite soccer matches may be of interest for future research and may provide greater knowledge in the damage and repair process. The ability to reduce calcium influx post-exercise may drastically alter the recovery process of elite sport, and may impact on the implementation of optimal recovery techniques.

This thesis displayed that some markers of muscle damage can return to baseline values at 48 hours post-match which may indicate a readiness to compete at optimal levels, however it may also be of importance for future research to consider the assessment of alternative indirect markers such as performance measures. The loss of strength and power appears to be a consequence of muscle damage as displayed by a reduced CMJA in the present thesis (chapter 4 & 7), however assessments of true strength measures such as isometric mid-thigh pull, maximal adductor squeeze and abductor pull (Norris, *et al*., 2018; Wollin, *et al*., 2018) are gaining attention and may provide an insight in to the return to baseline of muscle function post-soccer match. Assessments of these measures pre- and post-match will provide an insight into the effects of soccer matches on muscle strength measures, whilst measurements in the days following soccer matches will assist in the understanding of the time taken to return to optimal strength performance, and the resulting optimal soccer performance and may aid the development of session planning and training load periodisation.

Research assessing muscle fibre composition has elucidated type II and type IIb fibres may be more susceptible to muscle damage than type I fibres due to being preferentially recruited in eccentric contractions (Owens, *et al.*, 2019; Douglas, *et al.*, 2017; Magal, *et al.*, 2010), and as such athletes with a higher type II and type IIb composition may experience greater muscle damage than those of a higher type I composition. To date, there is a dearth of research on elite athletes and their fibre type in relation to muscle damage, likely due to the reluctance of athlete participation and limited availability of researchers to collect muscle biopsies. Development of the understanding of muscle fibre type damage may be of great use to practitioners and could significantly change the applied recovery practice. In elite soccer, athletes are seldom considered on an individual basis where recovery from soccer matches is concerned, and sessions in the two days following soccer matches often focus on recovery techniques and low intensity training. However, it is possible that type II dominant athletes require a greater recovery period before returning to optimal performance, and the development of personalised training plans based on their needs may prove beneficial. Conversely, type I dominant athletes may require less recovery time, potentially meaning they are able to complete high intensity sessions sooner post-match, which may assist in preparation for upcoming fixtures and provide greater learning opportunities for young athletes. As yet, this knowledge is lacking, and may be of interest to future researchers.

In order to prevent or reduce the severity of muscle damage, research has assessed the effects of the repeat bout effect (RBE) and as discussed within this thesis, it may be possible that the RBE has a limited capacity, beyond which it provides no further protective mechanism against exercise induced muscle damage. Further understanding on the RBE, and a potential limit of a protective effect may provide knowledge to the applied industry, from which optimal training preparations can be made limiting future muscle damage and maximising training and learning opportunities. If applied to competition, it may assist practitioners in providing training recommendations in the build up to busy fixture periods to aid in minimising muscle damage and maximising performance capabilities.

Although this thesis has focused predominantly on muscle damage from soccer matches, the RBE is not restricted to eccentric actions from match settings. Instead, the majority of an elite soccer schedule consists of training and gym exposure, yet despite this there is a dearth of research on the effects of strength and conditioning programmes on the RBE providing a protective mechanism from muscle damage sustained via elite youth soccer matches. Future research should seek to determine the effects of a strength and conditioning intervention on reducing muscle damage of soccer matches, and whether such programmes consisting mainly of eccentric actions aid the RBE in comparison to concentric only, and mixed (eccentric and concentric) programmes.

Acute muscle damage and muscle recovery following elite youth soccer matches formed the focus of this thesis, yet it may be of great interest to assess the longitudinal muscle damage, and in particular, accumulation of muscle damage throughout a competitive soccer season. Findings from such research may assist coaches and practitioners in the periodisation of a training programme, allowing for a reduced training load at periods of chronically high muscle damage, and greater training and coaching opportunities to maximise learning at periods of chronically low muscle damage. Additionally, the longitudinal assessment of muscle recovery techniques may be an area of future interest. This thesis has demonstrated successful acute implementation of interventions following elite youth soccer matches, however there is a dearth of research on the longitudinal effects of recovery interventions, and whether a similar benefit is observed following regular implementation of interventions, or whether adaptations to interventions occur and a nullified effect occurs limiting the recovery benefits. This may also aid the development of research concerning the potential benefit of removing recovery interventions at suitable periods within a season to allow a natural recovery process to take place and development of a protective effect to occur, so that an additional recovery benefit is observed when effective recovery interventions are later re-introduced. This may apply to elite youth soccer at times such as pre-season where competition is yet to begin, meaning skeletal muscle can adapt to eccentric exercise exposure, potentially enhancing a RBE, whereas the use of recovery interventions may hinder this natural muscular adaptation.

Chapter 6 of this thesis discussed the possible positional differences in match load, and the subsequent differing physical variables that correlate to muscle damage. Due to differing playing styles, players of different positions are exposed to large variances in physical parameters, and as such the muscle damage they experience may correlate to different physical parameters. This is a recommended area of future research which may assist in the evolution of training programmes. Understand the muscle damaging physical demands by position, will allow coaches and practitioners to adapt training sessions to expose these physical variables, better preparing athletes for soccer matches, potentially reducing muscle damage elicited. Chapter 6 also discusses the development of thresholds for variables correlating to muscle damage to identify physical capacities of players beyond which they fail to perform at their optimal level. Research in this area will assist coaches in real-time decision making, optimising team physical performance, and aid practitioners in the development of physical training programmes for enhancing physical match conditioning.

## 9.1 Concluding Statement

The research of this thesis has provided a unique insight into the assessment of muscle damage and muscle recovery in an applied elite soccer environment. It has been established that soccer matches of elite youth players significantly induce muscle damage of a greater magnitude than isolated eccentric exercise protocols. It has also been identified that the muscle damage sustained from elite youth soccer matches remains elevated at 48 hours post-match in the absence of effective recovery interventions. Static stretching as a recovery intervention has been shown to provide no recovery benefit following elite youth soccer matches leading to recommendations against its use for the attenuation of muscle damage. Alternative recovery interventions active recovery and cold-water immersion have been identified to provide a significant recovery benefit at 48 hours post-match following soccer matches of elite youth soccer players, and are therefore recommended for their use in applied elite settings to minimise muscle damage and accelerate the muscle recovery process.

This thesis has identified that specific physical demands (high intensity accelerations and decelerations, and absolute high speed running) of elite youth soccer matches significantly correlate to indicators of muscle damage, emphasising the importance of recording and monitoring GPS data of elite youth soccer players, and providing a new indirect tool to assess and predict muscle damage with feedback from matches available prior to the peak manifestation of alternative indicators of muscle damage, providing coaches and practitioners with information required to implement optimal recovery interventions immediately post-match.

# 10 References

Ade, J.D., Harley, J.A., and Bradley, P.S. (2014) Physiological Response, Time–Motion Characteristics, and Reproducibility of Various Speed-Endurance Drills in Elite Youth Soccer Players: Small-Sided Games Versus Generic Running, *International Journal of Sports Physiology and Performance*, 9, 471 -479.

Akenhead, R. (2014) *Examining the physical and physiological demands of elite football.* Doctoral Thesis. Northumbria University, Newcastle.

Anderson, J.C. (2005) Stretching Before and After Exercise: Effect on Muscle Soreness and Injury Risk, *Journal of Athletic Training,* 40(3), 218-220.

Archer, J. D. (1982) Elevated CK-MB levels in Marathon runners, *Journal of the American Medical Association*, 247(17), 2368-2369.

Armstrong, R.B. (1990) Initial events in exercise-induced muscular injury, *Medicine and Science in Sports and Exercise*, 22, 429-35.

Arnaston, A., Andersen, T.E., Holme, I., Engebretsen, L., and Bahr, R. (2008) Prevention of hamstring strains in elite soccer: an intervention study, *Scandinavian Journal of Medicine and Science in Sport*, 18, 40-48.

Ascensão, A., Leite, M., Rebelo, A.N., Magalhäes, S., and Magalhäes, J. (2011) Effects of cold water immersion on the recovery of physical performance and muscle damage following a one-off soccer match, *Journal of Sports Sciences*, 29(3), 217-25.

Bahnert, A., Norton, K., and Lock, P. (2013) Association between post-game recovery protocols, physical and perceived recovery, and performance in elite Australian Football League players, *Journal of Science and Medicine in Sport*, 16(2), 151-6.

Baird, M.F., Graham, S.M., Baker, J.S., and Bickerstaff, G.F. (2012) Creatine-kinase- and exercise-related muscle damage implications for muscle performance and recovery, *Journal of Nutrition and Metabolism*.

Baumert, P., Lake, M.J., Stewart, C.E., Drust, B., and Erskine, R.M. (2016) Genetic variation and exercise-induced muscle damage: implications for athletic performance, injury and ageing, *European Journal of Applied Physiology*, 116, 1595-1625.

Beato, M., Devereux, G., and Stiff, A. (2018a) Validity and reliability of global positioning system units (STATSports Viper) for measuring distance and peak speed in sports, *Journal of Strength and Conditioning Research*, 32(10), 2831–2837.

Beato, M., Coratella, G., Stiff, A., and Iacono, A.D. (2018b) The Validity and Between-Unit Variability of GNSS Units (STATSports Apex 10 and 18 Hz) for Measuring Distance and Peak Speed in Team Sports, *Frontiers in Physiology*, 9, 1288.

Best, T.M., Fiebig, R., Corr, D.T., Brickson, S., and Ji, L. (1999) Free radical activity, antioxidant enzyme, and glutathione changes with muscle stretch injury in rabbits, *Journal of Applied Physiology*, 87(1), 74-82.

Bieuzen, F., Bleakley, C.M., and Costello, J.T. (2013) Contrast Water Therapy and Exercise Induced Muscle Damage: A Systematic Review and Meta-Analysis, *PLoS ONE*, 8(4).

Bloomfield, J., Polman, R., and O’Donoghue, P. (2007) Physical demands of different positions in FA Premier League soccer, *Journal of Sports Science and Medicine*, 6, 63-70.

Bowen, L., Gross, A.S., Gimpel, M., and Li, F. (2017) Accumulated workloads and the acute:chronic workload ratio relate to injury risk in elite youth football players, *British Journal of Sports Medicine*, 51, 452–459.

Bradley, P.S., and Ade, J.D. (2018) Are Current Physical Match Performance Metrics in Elite Soccer Fit for Purpose or Is the Adoption of an Integrated Approach Needed? *International Journal of Sports Physiology and Performance*, 13(5), 656-664.

Brancaccio, P., Lippi, G., and Maffulli, N. (2010) Biochemical markers of muscular damage, *Clinical Chemistry and Laboratory Medicine*, 48(6), 757-67.

Broatch, J.R. (2015) *The Influence of Cold-Water Immersion on the Adaptive Response to High-Intensity Interval Training in Human Skeletal Muscle*. Doctoral Thesis. Victoria University. Melbourne.

Broatch, J.R., Petersen, A., and Bishop, D.J. (2018) The Influence of Post-Exercise Cold-Water Immersion on Adaptive Responses to Exercise: A Review of the Literature, *Sports Medicine*, 48, 1369–1387.

Brown, S.J., Child, R.B., Day, S.H., and Donnelly, A.E. (1997) Exercise-induced skeletal muscle damage and adaptation following repeated bouts of eccentric muscle contractions, *Journal of Sports Sciences*, 15(2), 215-222.

Buroker, K.C., and Schwane, J.A. (1989) Does post-exercise static stretching alleviate delayed muscle soreness? *The Physician and Sports Medicine*, 17(6), 65-83.

Butterfield, T.A. (2010) Eccentric exercise in vivo: strain-induced muscle damage and adaptation in a stable system, *Exercise and Sports Sciences Review*, 38(2), 51-60.

Byrne, C., Eston, R.G., and Edwards, R.H.T. (2001) Characteristics of isometric and dynamic strength loss following eccentric exercise-induced muscle damage, *Scandinavian Journal of Medicine and Science in sports,* 11, 134-140.

Calleja-González, J., Terrados, N., Mielgo-Ayuso. J., Delextrat, A., Jukic, I., Vaquera, A., Torres, L., Schelling, A., Stojanovic, M., and Ostojic, S.M. (2016) Evidence-based post-exercise recovery strategies in basketball, *The Physician and Sportsmedicine*, 44(1), 74-8.

Chazaud, B. (2016) Inflammation during skeletal muscle regeneration and tissue remodeling: application to exercise-induced muscle damage management, *Immunology and Cell Biology*, 94, 140-145.

Chen, T.C., Chen, H.L., Liu, Y.C., and Nosaka, K. (2014) Eccentric exercise-induced muscle damage of pre-adolescent and adolescent boys in comparison to young men, *European Journal of Applied Physiology,* 114, 1183–1195.

Cheung, K., Hume, P.A., and Maxwell, L. (2003) Delayed Onset Muscle Soreness: Treatment Strategies and Performance Factors, *Sports Medicine*, 33(2), 145-164.

Clarkson, P.M., Nosaka, K., and Braun, B. (1992) Muscle function after exercise-induced muscle damage and rapid adaptation, *Medicine and Science in Sports and Exercise*, 24(5), 512-520.

Clarkson, P.M., Kearns, A.K., Rouzier, P., Rubin, R., and Thompson, P.D. (2006) Serum creatine kinase levels and renal function measures in exertional muscle damage, *Medicine and Science in Sports and Exercise*, 38(4), 623-7.

Cleak, M.J., and Eston, R.G. (1992) Delayed onset muscle soreness: Mechanisms and management, *Journal of Sports Sciences*, 10, 325-341.

Close, G.L., Ashton, T., Cable, T., Doran, D., and MacLaren, D.P.M. (2004) Eccentric exercise, isokinetic muscle torque and delayed onset muscle soreness: the role of reactive oxygen species, *European Journal of Applied Physiology*, 91, 615–621.

Close, G.L., Ashton, T., McArdle, A., and MacLaren, D.P.M. (2005) The emerging role of free radicals in delayed onset muscle soreness and contraction-induced muscle injury, *Comparative Biochemistry and Physiology*, 257-266.

Cochrane, D.J. (2004) Alternating hot and cold water immersion for athlete recovery: a review, *Physical Therapy in Sport*, 5, 26-32.

Coelho, D.B., Morandi, R.F., de Melo, M.A.A., and Silami-Garcia, E. (2011) Creatine kinase kinetics in professional soccer players during a competitive season, *Brazilian Journal of Kinanthropometry and Human Performance*, 13(3), 189-194.

Coffey, V., Leveritt, M., and Gill, N. (2004) Effect of recovery modality on 4-hour repeated treadmill running performance and changes in physiological variables, *Journal of Science and Medicine in Sport*, 7(1), 1-10.

Dadebo, B., White, J., and George, K.P. (2004) A survey of flexibility training protocols and hamstring strains in professional football clubs in England, *British Journal of Sports Medicine*, 38, 388–394.

Darani, M.S.M., Abedi, B., and Fatolahi, H. (2018) The Effect of Active and Passive Recovery on Creatine Kinase and C-reactive Protein after an Exercise Session in Football Players, *International Archives of Health Sciences,* 5, 1-5.

Dascombe, B.J., Reaburn, P.R.J., Sirotic, A.C., and Coutts, A.J. (2007) The reliability of the i-STAT clinical portable analyser, *Journal of Science and Medicine in Sport*, 10, 135—140.

Dawson, B., Cow, S., Modra, S., Bishop, D., and Stewart, G. (2005) Effects of immediate post-game recovery procedures on muscle soreness, power and flexibility levels over the next 48 hours, *Journal of Science and Medicine in Sport*, 8(2), 210-221.

de Hoyo, M., de la Torre, A., Pradas, F., Sañudo, B., Carrasco, L., Mateo-Cortes, J., Domínguez-Cobo, S., Fernandes, O., and Gonzalo-Skok, O. (2015) Effects of Eccentric Overload Bout on Change of Direction and Performance in Soccer Players, *International Journal of Sports Medicine,* 36(4), 308-14.

de Hoyo, M., Cohen, D.D., Sañudo, B., Carrasco, L., Álvarez-Mesa, A., del Ojo, J.J., Domínguez-Cobo, S., Mañas, V., and Otero-Esquina, C. (2016) Influence of football match time–motion parameters on recovery time course of muscle damage and jump ability, *Journal of Sports Sciences*, 34(14), 1363-1370.

de Vries, H.A. (1966) Quantitative electromyographic investigation of the spasm theory of muscle pain, *American Journal of Physical Medicine*, 45, 119-35.

Delextrat, A., Hippocrate, A., Leddington-Wright, S., and Clarke, N.D. (2014) Including stretches to a massage routine improves recovery from official matches in basketball players, *Journal of Strength and Conditioning Research*, 28(3), 716–727.

Devrnja, A., and Matković, B.R. (2008) The effects of a soccer match on muscle damage indicators, *Kinesiology*, 50(1), 112-123.

Douglas, J., Pearson, S., Ross, A., and McGuigan, M. (2017) Eccentric Exercise: Physiological Characteristics and Acute Responses, *Sports Medicine*, 47(4), 663-675.

Duffield, R., Cannon, J., and King, M. (2010) The effects of compression garments on recovery of muscle performance following high-intensity sprint and plyometric exercise, *Journal of Science and Medicine in Sport*, 13, 136–140.

Dupuy, O., Douzi, W., Theurot, D., Bosquet, L. and Dugué, B. (2018) An Evidence-Based Approach for Choosing Post-exercise Recovery Techniques to Reduce Markers of Muscle Damage, Soreness, Fatigue, and Inflammation: A Systematic Review With Meta-Analysis, *Frontiers in Physiology*, 9, 403.

Elias, G.P., Wyckelsma, V.L., Varley, M.C., McKenna, M.J., and Aughey, R.J. (2013) Effectiveness of Water Immersion on Postmatch Recovery in Elite Professional Footballers, *International Journal of Sports Physiology and Performance*, 7, 243-253.

Field, A. (2009) *Discovering Statistics using SPSS*. 3rd Edition. London: SAGE Publications.

Field, A. (2013) *Discovering Statistics using IBM SPSS Statistics*. 4th Edition. London: SAGE Publications.

Fiscus, K.A., Kaminski, T.W., and Powers, M.E. (2005) Changes in lower-leg blood flow during warm-, cold-, and contrast-water therapy, *Archives of Physical Medicine and Rehabilitation,* 86, 1404-10.

Freitas, C.G., Aoki, M.S., Arruda, A.F.S., Franciscon, C., and Moreira, A. (2016) Monitoring Salivary Immunoglobulin A Responses to Official and Simulated Matches In Elite Young Soccer Players, *Journal of Human Kinetics*, 53, 107-115.

Gill, N. D., Beaven, C. M., and Cook, C. (2006) Effectiveness of post-match recovery strategies in rugby players, B*ritish Journal of Sports Medicine,* 40, 260-263.

Gissel, H. (2005) The role of Ca2+ in muscle cell damage, *Annals of the New York Academy of Science*, 1066, 166-80.

Goodall, S., and Howatson, G. (2008) The effects of multiple cold water immersions on indices of muscle damage, *Journal of Sports Science and Medicine*, 7, 235-241.

Gunst, J.J, Langlois, M.R., Delanghe, J.R, De Buyzere, M.L., and Leroux-Roels, G.G. (1998) Serum creatine kinase activity is not a reliable marker for muscle damage in conditions associated with low extracellular glutathione concentration, *Clinical Chemistry*, 44(5), 939-943.

Habets, B., Staal, J.B., Tijssen, M., and van Cingel, R. (2018) Intrarater reliability of the Humac NORM isokinetic dynamometer for strength measurements of the knee and shoulder muscles, *BMC Research Notes*, 11(1), 15.

Herbert, R.D., and Gabriel, M. (2002) Effects of stretching before and after exercising on muscle soreness and risk of injury: systematic review, *British Medical Journal*, 468.

Herbert, R.D., de Noronha, M., and Kamper, S.J. (2011) Stretching before or after exercise does not reduce Stretching to prevent delayed-onset muscle soreness, *British Journal of sports Medicine*, 45, 1249-1250.

Hidalgo, C., Gonzalez, M.E., and Garcia, A.M. (1986) Calcium transport in transverse tubules isolated from rabbit skeletal muscle, *Biochimica et Biophysica Acta*, 854, 279–286.

Higgins, T.R., Greene, D.A., and Baker, M.K. (2017) Effects of cold water immersion and contrast water therapy for recovery from team sport: a systematic review and meta-analysis, *Journal of Strength and Conditioning Research*, 31(5), 1443-1460.

Hill, J., Howatson, G., van Someren, K., Leeder, J., and Pedlar, C. (2013) Compression garments and recovery from exercise-induced muscle damage: a meta-analysis, *British Journal of Sports Medicine*, 48(18), 1340-1346.

Hody, S., Rogister, B., Leprince, P., Wang, F., and Croisier, J-L. (2013) Muscle fatigue experienced during maximal eccentric exercise is predictive of the plasma creatine kinase (CK) response, *Scandinavian Journal of Medicine and Science in Sports*, 23, 501–507.

Hopkins, W.G. (2000) Measures of Reliability in Sports Medicine and Science, *Sports Medicine*, 30(1), 1-15.

Howatson, G., and van Someren, K.A. (2008) The Prevention and Treatment of Exercise-Induced Muscle Damage, *Sports Medicine*, 38(6), 483-503.

Howatson, G., Goodall, S., and van Someren, K.A. (2009) The influence of cold water immersions on adaptation following a single bout of damaging exercise, *European Journal of Applied Physiology*, 105, 615–621.

Howell, J.N., Chleboun, G., and Conatser, R. (1993) Muscle stiffness, strength loss, swelling and soreness following exercise-induced injury in humans, *Journal of Physiology*, 464, 183-196.

Hyldahl, R.D., and Hubal, M.J. (2014) Lengthening our perspective: morphological, cellular, and molecular responses to eccentric exercise, *Muscle and Nerve*, 49(2), 155-70.

Hyldahl, R.D., Chen, T.C., and Nosaka, K. (2017) Mechanisms and mediators of the skeletal muscle repeated bout effect, *Exercise and Sport Sciences Reviews*, 45(1), 24–33.

Ingram, J., Dawson, B., Goodman, C., Wallman, K., and Beilby, J. (2009) Effect of water immersion methods on post-exercise recovery from simulated team sport exercise, *Journal of Science and Medicine in Sport*, 12, 417-421.

Jayaraman, R.C., Reid, R.W., Foley, J.M., Prior, B.M., Dudley, G.A., Weingand, K.W., and Meyer, R.A. (2004) MRI evaluation of topical heat and static stretching as therapeutic modalities for the treatment of eccentric exercise-induced muscle damage, *European Journal of Applied Physiology,* 93, 30-38.

Keil, M. (2019) Have we been stretching the truth? The effects of stretching post exercise and delayed onset muscle soreness, *Creative Components*, Iowa State University.

Kendall, B., and Eston, R. (2002) Exercise-Induced Muscle Damage and the Potential Protective Role of Estrogen, *Sports Medicine,* 32(2), 103-123.

Kinugasa, T., and Kilding, A.E. (2009) A Comparison of Post-Match Recovery Strategies in Youth Soccer Players, *Journal of Strength and Conditioning Research*, 23(5), 1402-7.

Krustrup, P., Mohr, M., Steensberg, A., Bencke., J., Kjaer, M., Bangsbo, J. (2006) Muscle and blood metabolites during a soccer game: implications for sprint performance, *Medicine and Science in Sports and Exercise*, 38(6), 1165-1174.

Krustrup, P., Ørtenblad, N., Nielsen, J., Nybo, L., Gunnarsson, T.P., Iaia, F.M., Madsen, K., Stephens, F., Greenhaff, P., and Bangsbo, J. (2011) Maximal voluntary contraction force, SR function and glycogen resynthesis during the first 72 h after a high-level competitive soccer game, *European Journal of Applied Physiology*, 111, 2987–2995.

LaStayo, P.C., Reich, T.E., Urquhart, M., Hoppeler, H., and Lindstedt, S.L. (1999) Chronic eccentric exercise: improvements in muscle strength can occur with little demand for oxygen, American Journal of Physiology, 276(2), 611-615.

Lavender, A.P., and Nosaka, K. (2008) Changes in markers of muscle damage of middle-aged and young men following eccentric exercise of the elbow flexors, Journal of Science and Medicine in Sport, 11, 124-131.

Lazarim, F.L., Antunes-Neto, J.M.F., da Silva, F.O.C., Nunes, L.A.S., Bassini-Cameronc, A., Cameronc, L-C., Alves, A.A., Brenzikofer, R., and de Macedo, D.V. (2009) The upper values of plasma creatine kinase of professional soccer players during the Brazilian National Championship, *Journal of Science and Medicine in Sport*, 12(1), 85-90.

Lee, J., and Clarkson, P. M. (2003) Plasma creatine kinase activity and glutathione after eccentric exercise, *Medicine and Science in Sports and Exercise*, 35, 930–936.

Leeder, J., Gissane, C., van Someren, K., Gregson, W., and Howatson, G. (2012) Cold water immersion and recovery from strenuous exercise: a meta-analysis, *British Journal of Sports Medicine*, 46(4), 233-240.

Lloyd, R.S., Oliver, J.L., Hughes, M.G. and Williams, C.A. (2009) Reliability and validity of field-based measures of leg stiffness and reactive strength index in youths, *Journal of Sports Sciences*, 27(14), 1565-1573.

Lund, H., Vestergaard-Poulsen, P., Kanstrup, I.L., and Sejrsen, P. (1998) The effect of passive stretching on delayed onset muscle soreness, and other detrimental effects following eccentric exercise, *Scandinavian Journal of Medicine and Science in Sports*, 8, 216-221.

Machado, A.F., Ferreira, P.H., Micheletti, J.K., de Almeida, A.C., Lemes, I.R., Vanderlei, F.M., Junior, J.N., and Pastre, C.M. (2016) Can Water Temperature and Immersion Time Influence the Effect of Cold Water Immersion on Muscle Soreness? A Systematic Review and Meta-Analysis, *Sports Medicine*, 46, 503–514.

Magal, M., Dumke, C.L., Urbiztondo, Z.G., Cavill, M.J., Triplett, N.T., Quindry, J.C., McBride, J.M., and Epstein, Y. (2010) Relationship between serum creatine kinase activity following exercise-induced muscle damage and muscle fibre composition, *Journal of Sports Sciences*.

Martin, N.A., Zoeller, R.F., Robertson, R.J., and Lephart, S.M. (1998) The Comparative Effects of Sports Massage, Active Recovery, and Rest in Promoting Blood Lactate Clearance After Supramaximal Leg Exercise, *Journal of Athletic Training*, 33, 30-35.

McCord, J. (1995) Superoxide radical: controversies, contradictions, and paradoxes, *Society for Experimental Biology and Medicine*, 209, 112-7.

McHugh, M.P., Connoly, D.A.J., Eston, R.G., and Gleim, G.W. (1999) Exercise-Induced Muscle Damage and Potential Mechanisms for the Repeated Bout Effect, *Sports Medicine*, 27(3), 157-170.

McKinnon, N.B., Graham, M.T., and Tiidus, P.M. (2012) Effect of creatine supplementation on muscle damage and repair following eccentrically-induced damage to the elbow flexor muscles, *Journal of Sports Science and Medicine*, 11, 653-659.

Mika, A., Oleksy, Ł., Kielnar, R., Wodka-Natkaniec, E., Twardowska, M., Kamiński, K., and Małek, Z. (2016) Comparison of Two Different Modes of Active Recovery on Muscles Performance after Fatiguing Exercise in Mountain Canoeist and Football Players, *PLoS ONE*, 11(10), e0164216.

Mohr, M., Draganidis, D., Chatzinikolaou, A., Barbero-Álvarez, J.C., Castagna, C., Douroudos, I., Avloniti, A., Margeli, A., Papassotiriou, I., Flouris, A.D., Jamurtas, A.Z., Krustrup, P., and Fatouros, I.G. (2017) Muscle damage, inflammatory, immune and performance responses to three football games in 1 week in competitive male players, *European Journal of Applied Physiology*, 116(1), 179-93.

Monedero, J., and Donne, B. (2000) Effect of recovery interventions on lactate removal and subsequent performance, *International Journal of Sports Medicine*, 21(8), 593-7.

Montgomery, P.G., Pyne, D.B., Hopkins, W.G., Dorman, J.C., Cook, K., and Minahan, C.L. (2008) The effect of recovery strategies on physical performance and cumulative fatigue in competitive basketball, *Journal of Sports Sciences*, 26(11), 1135-45.

Morgan, D.L., and Proske, U. (2004) Popping sarcomere hypothesis explains stretch induced muscle damage, *Australian Physiological and Pharmacological Society*, 34, 19.23.

Morgans, R., Owen, A., Doran, D., Drust, B., and Morton, J.P. (2015) Prematch Salivary Secretory Immunoglobulin A in Soccer Players From the 2014 World Cup Qualifying Campaign, *International Journal of Sports Physiology and Performance*, 10, 401-403.

Mortatti, A.L., Moreira, A., Aoki, M.S., Crewther, B.T., Castagna, C., de Arruda, A.F.S., and Filho, J.M. (2012) Effect of competition on salivary cortisol, immunoglobulin A, and upper respiratory tract infections in elite young soccer players, *Journal of Strength and Conditioning Research*, 26(5), 1396–1401.

Morton, J.P., Atkinson, G., MacLaren, D.P.M., Cable, N.T., Gilbert, G., Broome, C., McArdle, A., and Drust, B. (2005) Reliability of maximal muscle force and voluntary activation as markers of exercise-induced muscle damage, *European Journal of Applied Physiology*, 94, 541–548.

Nédélec, M., McCall, A., Carling, C., Legall, F., Berthoin, S., and Dupont, G. (2012) Recovery in Soccer: Part I - Post-Match Fatigue and Time Course of Recovery, *Sports Medicine*, 42(12), 997-1015.

Nédélec, M., McCall, A., Carling, C., Legall, F., Berthoin, S., and Dupont, G. (2013) Recovery in Soccer: Part II-Recovery Strategies, *Sports Medicine*, 43(1), 9-22.

Nédélec, M., McCall, A., Carling, C., Legall, F., Berthoin, S., and Dupont, G. (2014) The influence of soccer playing actions on the recovery kinetics after a soccer match, *Journal of Strength and Conditioning Research*, 28(6), 1517-1523.

Niess, A.M., Dickhuth, H., Northoff, H., and Fehrenbach, E. (1999) Free radicals and oxidative stress in exercise-immunological aspects, *Exercise Immunology Review*, 5, 22-56.

Nikolaidis, M.G., Jamurtas, A.Z., Paschalis, V., Fatouros, I.G., Koutedakis1, Y., and Kouretas, D. (2008) The Effect of Muscle-Damaging Exercise on Blood and Skeletal Muscle Oxidative Stress: Magnitude and Time-Course Considerations, *Sports Medicine*, 38(7), 579-606.

Norris, D., Joyce, D., Siegler, J., Clock, J., and Lovell, R. (2018) Recovery of Force-Time Characteristics After Australian Rules Football Matches: Examining the Utility of the Isometric Midthigh Pull, *International Journal of Sports Physiology and Performance*, 14(6), 765–770.

Nosaka, K., Newton, M., and Sacco, P. (2002) Delayed-onset muscle soreness does not reflect the magnitude of eccentric exercise-induced muscle damage, *Scandinavian Journal of Medicine and Science in Sports*, 12, 337-346.

Nowakowska, A., Kostrzewa-Nowak, D., Buryta, R., and Nowak, R. (2019) Blood Biomarkers of Recovery Efficiency in Soccer Players, *International Journal of Environmental Research and Public Health*, 16(18).

Nuzzo, J.L., Anning, J.H., and Scharfenberg, J.M. (2011) The reliability of three devices used for measuring vertical jump height, *Journal of Strength and Conditioning Research*, 25(9), 2580–2590.

Ortiz Jr, R.O., Sinclair Elder, A.J., Elder, C.L., and Dawes, J.J. (2019) A systematic review on the effectiveness of active recovery interventions on athletic performance of professional-, collegiate-, and competitive-level adult athletes, *Journal of Strength and Conditioning Research*, 33(8), 2275-2287.

Owen, A.L., Wong, D.P., Dunlop, G., Groussard, C., Kebsi, W., Dellal, A., Morgans, R., and Zouhal, H. (2014) High-intensity training and salivary immunoglobulin A responses in professional top-level soccer players: Effect of training intensity, *Journal of Strength and Conditioning Research*, 30(9), 2460-9.

Owens, D.J., Twist, C., Cobley, J.N., Howatson, G., and Close, G.L. (2019) Exercise-induced muscle damage: What is it, what causes it and what are the nutritional solutions? *European Journal of Sport Science*, 19(1), 71-85.

Paulsen, G., Mikkelsen, U.R., Raastad, T., and Peake, J.M. (2012) Leucocytes, cytokines and satellite cells: what role do they play in muscle damage and regeneration following eccentric exercise? *Exercise Immunology Review*, 18, 42-97.

Peak, J., Nosaka, K., and Suzuki, K. (2005) Characterization of inflammatory responses to eccentric exercise in humans, *Exercise Immunology Review*, 11, 64-85.

Peake, J.M., Neubauer, O., Della Gatta, P.A., and Nosaka, K. (2017a) Muscle damage and inflammation during recovery from exercise, *Journal of Applied Physiology*, 122, 559 –570.

Peake, J.M., Roberts, L.A., Figueiredo, V.C., Egner, I., Krog, S., Aas, S.N., Suzuki, K., Markworth, J.F., Coombes, J.S., Cameron-Smith, D., and Raastad, T. (2017b) The effects of cold water immersion and active recovery on inflammation and cell stress responses in human skeletal muscle after resistance exercise, *Journal of Physiology*, 595(3), 695-711.

Peñailillo, L., Blazevich, A.K., and Nosaka, K. (2017) Factors contributing to lower metabolic demand of eccentric compared with concentric cycling, *Journal of Applied Physiology*, 123, 884 –893.

Perrey S, Betik A, Candau R, Rouillon JD, Hughson RL. (2001) Comparison of oxygen uptake kinetics during concentric and eccentric cycle exercise, *Journal of Applied Physiology*, 91(5), 2135-42.

Peiffer, J.J., Abbis, C.R., Nosaka, K., Peake, J.M., and Laursen, P.B. (2009) Effect of cold water immersion after exercise in the heat on muscle function, body temperatures, and vessel diameter, *Journal of Science and Medicine in Sport*, 12, 91-96.

Pooley, S., Spendiff, O., Allen, M., and Moir, H.J. (2017) Static stretching does not enhance recovery in elite youth soccer players, *BMJ Open Sport & Exercise Medicine*, 3:e000202.

Pooley, S., Spendiff, O., Allen, M., and Moir, H.J. (2019) Comparative efficacy of active recovery and cold water immersion as post-match recovery interventions in elite youth soccer, *Journal of Sports Sciences*, 1-9.

Powers, S.K., Radak, Z., and Ji, L.L. (2016) Exercise-induced oxidative stress: past, present and future, *Journal of Physiology,* 598(18), 5081–5092.

Proske, U., and Morgan, D.L. (2001) Muscle damage from eccentric exercise: mechanism, mechanical signs, adaptation and clinical applications, *Journal of Physiology*, 537(2), 333-345.

Qu, C., Wu, Z., Xu, M., Qin, F., Dong, Y., Wang, Z., and Zhao, J. (2020) Cryotherapy Models and Timing-Sequence Recovery of Exercise-Induced Muscle Damage in Middle- and Long-Distance Runners, *Journal of Athletic Training*, 55(4).

Rampinini, E., Bosio, A., Ferraresi, I., Petruolo, A., Morelli, A., and Sassi, A. (2011) Match-Related Fatigue in Soccer Players, *Medicine and Science in Sports and Exercise,* 43(11), 2161–2170.

Redden, J., Stokes, K., and Williams, S. (2018) Establishing the Reliability and Limits of Meaningful Change of Lower Limb Strength and Power Measures during Seated Leg Press in Elite Soccer Players, *Journal of Sports Science and Medicine*, 17, 539-546.

Redden, R. (2019) *Assessing Lower Limb Strength, Power and Asymmetry in Elite Soccer Players using the Keiser Air420 Seated Leg Press*. Doctoral Thesis. University of Bath, Bath.

Reilly, T., and Ekblom, B. (2005) The use of recovery methods post-exercise, *Journal of Sports*, 23(6), 619-627.

Richter, A., Räpple, S., Kurz, G., and Schwameder, H. (2012) Countermovement jump in performance diagnostics: Use of the correct jumping technique, *European Journal of Sport Science*, 12(3), 231-237.

Roberts, L.A., Nosaka, K., Coombes, J.S., and Peake, J.M. (2014) Cold water immersion enhances recovery of submaximal muscle function after resistance exercise, *American Journal of Physiology*, 307(8), 998-1008.

Romagnoli, M., Sanchis-Gomar, F., Alis, R., Risso-Ballester, J., Bosio, A., Graziani, R.L., and Rampinini, E. (2015) Changes in muscle damage, inflammation, and fatigue-related parameters in young elite soccer players after a match, *The Journal of sports medicine and physical fitness*, 56(10), 1198-1205.

Rowsell, G.J., Coutts, A.J., Reaburn, P., and Hill-Haas, S. (2009) Effects of cold-water immersion on physical performance between successive matches in high-performance junior male soccer players, *Journal of Sports Sciences*, 27(6), 565-573.

Russell, M., Sparkes, W., Northeast, J., Cook, C.J., Bracken, R.M., and Kilduff, L.P. (2016) Relationships between match activities and peak power output and Creatine Kinase responses to professional reserve team soccer match-play, *Human Movement Science*, 45, 96-101.

Sachdev, S., and Davies, K.J. (2008) Production, detection, and adaptive responses to free radicals in exercise, *Free Radical Biology & Medicine*, 44(2), 215-23.

Saka, T., Akova, B., Yazici, Z., Sekir, U., Gür, H., and Ozarda, Y. (2009) Difference in the magnitude of muscle damage between elbow flexors and knee extensors eccentric exercises, *Journal of Sports Science & Medicine*, 8(1), 107-15.

Scott, A., Malone, J.J., Morgans, R., Burgess, D., Gregson, W., Morton, J.P., and Drust, B. (2016) The relationship between physical match performance and 48-h post-game creatine kinase concentrations in English Premier League soccer player, *International Journal of Sports Science and Coaching*, 11(6), 846–852.

Seco-Calvo, J., Mielgo-Ayuso, J., Calvo-Lobo, C., and Córdova, A. (2019) Cold water immersion as a strategy for muscle recovery in professional basketball players during the competitive season: A retrospective cohort study, *Journal of Sport Rehabilitation*, 1-29. DOI: 10.1123/jsr.2018-0301.

Sermaxhaj, S., Popovic, S., Bjelica, D., Gardasevic, J., And Arifi, F. (2017) Effect of recuperation with static stretching in isokinetic force of young football players, *Journal of Physical Education and Sport*, 17(3), 1948-1953.

Sjøgaard, G., Adams, R.P., and Saltin, B. (1985) Water and ion shifts in skeletal muscle of humans with intense dynamic knee extension, *American Journal of Physiology*, 248(2), 190-196.

Smith, L.L., Brunetz, M.H., Chenier, T.C., McCammon, M.R., Houmard, J.A., Franklin, M.E., and Israel, R.G. (1993) The effects of static and ballistic stretching on delayed onset muscle soreness and creatine kinase, *Research Quarterly for Exercise and Sport*, 64(1), 103-7.

Spierer, D.K., Goldsmith, R., Baran, D.A., Hryniewicz, K., and Katz, S.D. (2004) Effects of active vs. passive recovery on work performed during serial supramaximal exercise tests, *International Journal of Sports Medicine,* 25(2), 109-114.

Stanton, A.W., Badger, C., and Sitzia, J. (2000) Non-invasive assessment of the lymphedematous limb, *Lymphology,* 33(3), 122-135.

Steinbacker, P., and Eckl, P. (2015) Impact of Oxidative Stress on Exercising Skeletal Muscle, *Biomolecules*, 5, 356-377.

Stephens, J.M., Halson, S., Miller, J., Slater, G.J., and Askew, C.D. (2016) Cold Water Immersion for Athletic Recovery: One Size Does Not Fit All, *International Journal of Sports Physiology and Performance,* 12(1), 2-9.

Tee, J.C., Bosch, A.N., and Lambert, N.I. (2007) Metabolic consequences of exercise-induced muscle damage, *Sports Medicine*, 37(10), 827-836.

Tessitore, A., Meeusen, R., Cortis, C., and Capranica, L. (2007) Effects of different recovery interventions on anaerobic performances following pre-season soccer training, *Journal of Strength and Conditioning Research*, 21(3), 745.

Thorpe, R., and Sunderland, C. (2012) Muscle damage, endocrine, and immune marker response to a soccer match, *Journal of Strength and Conditioning Research*, 26(10), 2783–2790.

Totsuka, M., Nakaji, S., Suzuki, K., Sugawara, K., and Sato, K. (2002) Break point of serum creatine kinase release after endurance exercise, *Journal of Applied Physiology*, 93, 1280–1286.

Toumi, H., F'guyer, S., and Best, M. (2006) The role of neutrophils in injury and repair following muscle stretch, *Journal of Anatomy*, 208(4), 459-470.

Tsatalas, T., Giakas, G., Spyropoulos, G., Paschalis, V., Nikolaidis, M.G., Tsaopoulos, D.E., Theodorou, A.A., Jamurtas, A.Z., and Koutedakis, Y. (2010) The effects of muscle damage on walking biomechanics are speed-dependent, *European Journal of Applied Physiology*, 110(5), 977-88.

Tsatalas, T., Giakas, G., Spyropoulos, G., Sideris, V., Lazaridis, S., Kotzamanidis, C., and Koutedakis, Y. (2013) The effects of eccentric exercise-induced muscle damage on running kinematics at different speeds, *Journal of Sports Sciences*, 31(3), 288-298.

Van Crombrugge, G., Duvivier, B.M.F.M., Van Crombrugge, K., Bellemans, J., and Peers, K. (2019) Hamstring injury prevention in Belgian and English elite football teams, *Acta Orthopaedica Belgica*, 85, 373-380.

Vaile, J., O'Hagan, C., Stefanovic, B., Walker, M., Gill, N., and Askew, C.D. (2011) Effect of cold water immersion on repeated cycling performance and limb blood flow, *British Journal of Sports Medicine*, 45(10), 825-829.

Varley, I., Lewin, R., Needham, R., Thorpe, R.T., and Burbeary, R. (2017) Association Between Match Activity Variables, Measures of Fatigue and Neuromuscular Performance Capacity Following Elite Competitive Soccer Matches, *Journal of Human Kinetics*, 60, 93-99.

Veldhoen, E.S., de Vooght, K.M.K., Slieker, M.G., Versluys, A.B., Turner, N.McB. (2014) Analysis of bloodgas, electrolytes and glucose from intraosseous samples using an i-STAT® point-of-care analyser, *Resuscitation*, 85(3), 359-63.

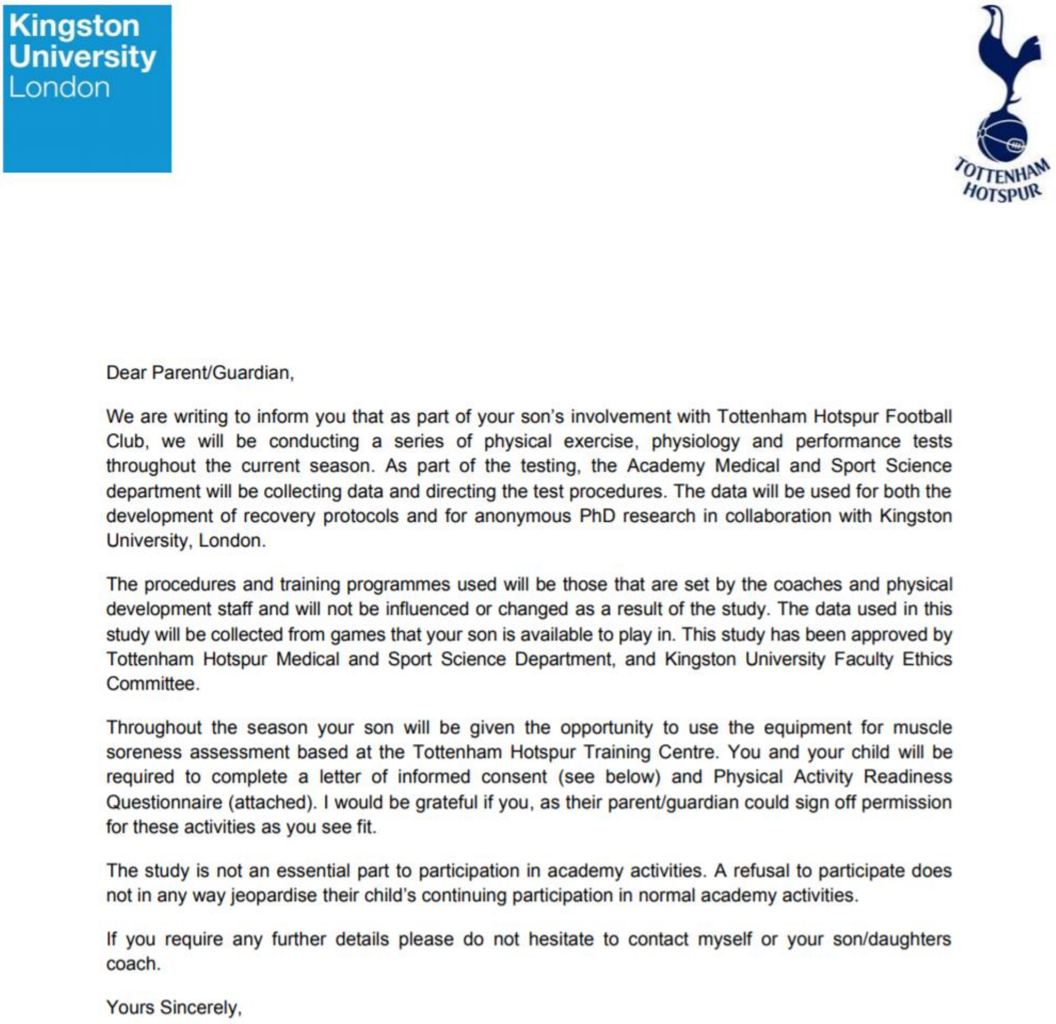
Wessel, J., and Wan, A. (1994) Effect of stretching on the intensity of delayed-onset muscle soreness, *Clinical Journal of Sports Medicine*, 4(2), 83-7.

Young, W.B., Hepner, J., and Robbins, D.W. (2012) Movement demands in Australian rules football as indicators of muscle damage, *Journal of Strength and Conditioning Research*, 26, 492-496.

Wollin, M., Pizzari, T., Spagnolo, K., Welvaert, M., and Thorborg, K. (2017) The effects of football match congestion in an international tournament on hip adductor squeeze strength and pain in elite youth players, *Journal of Sports Sciences*, 36(10), 1167-1172.

# 11 Appendices

## 11.1 Parental Information Sheet - signature and contact details redacted



Dear Parent/Guardian,

We are writing to inform you that as part of your son's involvement with Tottenham Hotspur Football Club, we will be conducting a series of physical exercise, physiology and performance tests throughout the current season. As part of the testing, the Academy Medical and Sport Science department will be collecting data and directing the test procedures. The data will be used for both the development of recovery protocols and for anonymous PhD research in collaboration with Kingston University, London.

The procedures and training programmes used will be those that are set by the coaches and physical development staff and will not be influenced or changed as a result of the study. The data used in this study will be collected from games that your son is available to play in. This study has been approved by Tottenham Hotspur Medical and Sport Science Department, and Kingston University Faculty Ethics Committee.

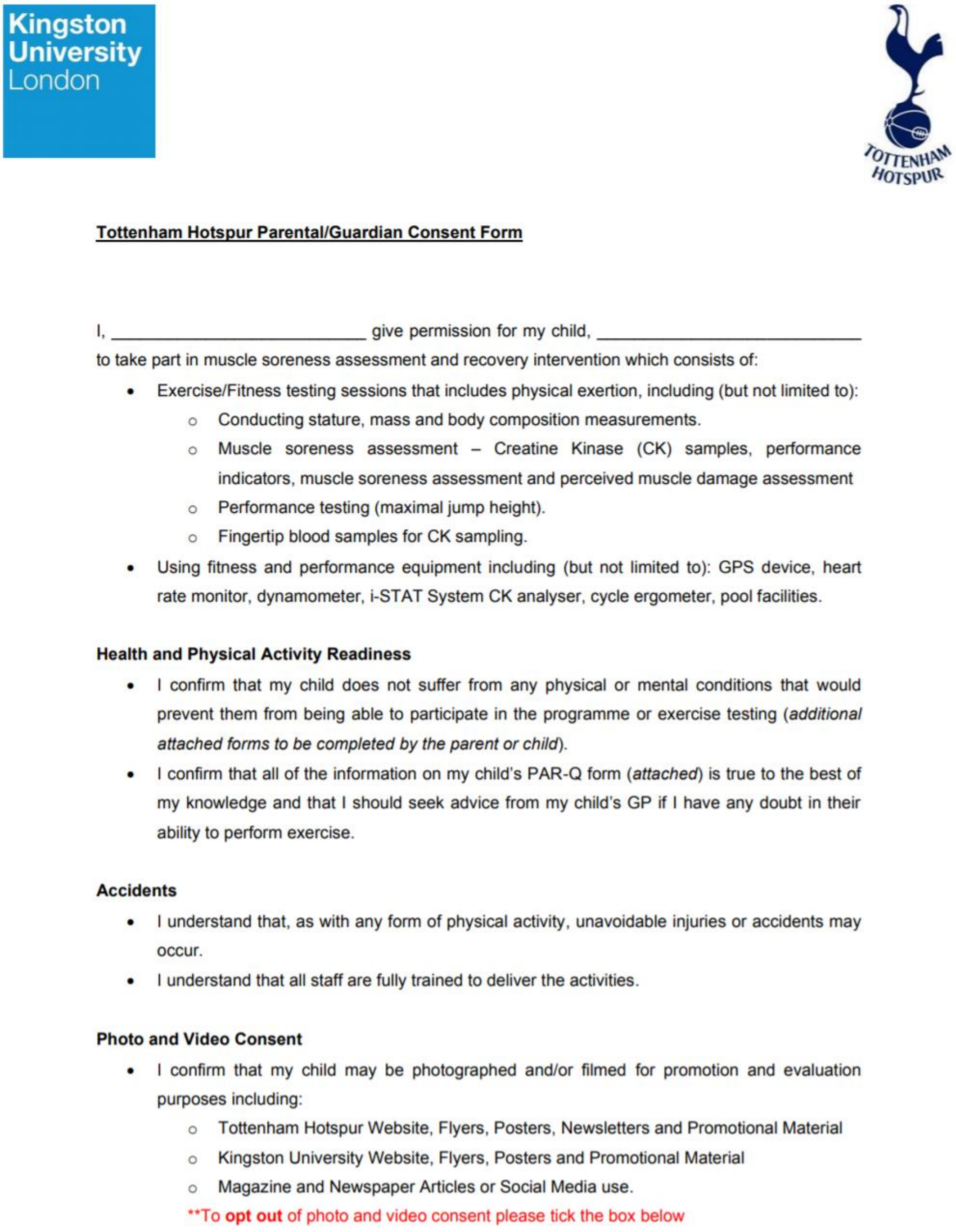
Throughout the season your son will be given the opportunity to use the equipment for muscle soreness assessment based at the Tottenham Hotspur Training Centre. You and your child will be required to complete a letter of informed consent (see below) and Physical Activity Readiness Questionnaire (attached). I would be grateful if you, as their parent/guardian could sign off permission for these activities as you see fit.

The study is not an essential part to participation in academy activities. A refusal to participate does not in any way jeopardise their child's continuing participation in normal academy activities.

If you require any further details please do not hesitate to contact myself or your son/daughters coach.

Yours Sincerely,

## 11.2 Parental Informed Consent Sheet



Kingston University London

TOTTENHAM HOTSPUR

**Tottenham Hotspur Parental/Guardian Consent Form**

I, ___________________________ give permission for my child, ____________________________ to take part in muscle soreness assessment and recovery intervention which consists of:

- Exercise/Fitness testing sessions that includes physical exertion, including (but not limited to):
    - Conducting stature, mass and body composition measurements.
    - Muscle soreness assessment – Creatine Kinase (CK) samples, performance indicators, muscle soreness assessment and perceived muscle damage assessment
    - Performance testing (maximal jump height).
    - Fingertip blood samples for CK sampling.
- Using fitness and performance equipment including (but not limited to): GPS device, heart rate monitor, dynamometer, i-STAT System CK analyser, cycle ergometer, pool facilities.

**Health and Physical Activity Readiness**

- I confirm that my child does not suffer from any physical or mental conditions that would prevent them from being able to participate in the programme or exercise testing (*additional attached forms to be completed by the parent or child*).
- I confirm that all of the information on my child's PAR-Q form (*attached*) is true to the best of my knowledge and that I should seek advice from my child's GP if I have any doubt in their ability to perform exercise.

**Accidents**

- I understand that, as with any form of physical activity, unavoidable injuries or accidents may occur.
- I understand that all staff are fully trained to deliver the activities.

**Photo and Video Consent**

- I confirm that my child may be photographed and/or filmed for promotion and evaluation purposes including:
    - Tottenham Hotspur Website, Flyers, Posters, Newsletters and Promotional Material
    - Kingston University Website, Flyers, Posters and Promotional Material
    - Magazine and Newspaper Articles or Social Media use.

  **To **opt out** of photo and video consent please tick the box below

**Agreement**

I confirm that I have read and understood the above agreement and that I have had the opportunity to ask any questions and discuss the programme details. I hereby confirm that my child is participating in

- Tottenham Hotspur muscle damage assessment and recovery strategy intervention testing.

Signed________________________________ (Parent/Legal Guardian)

Name: __________________________________

Name of child: __________________________________

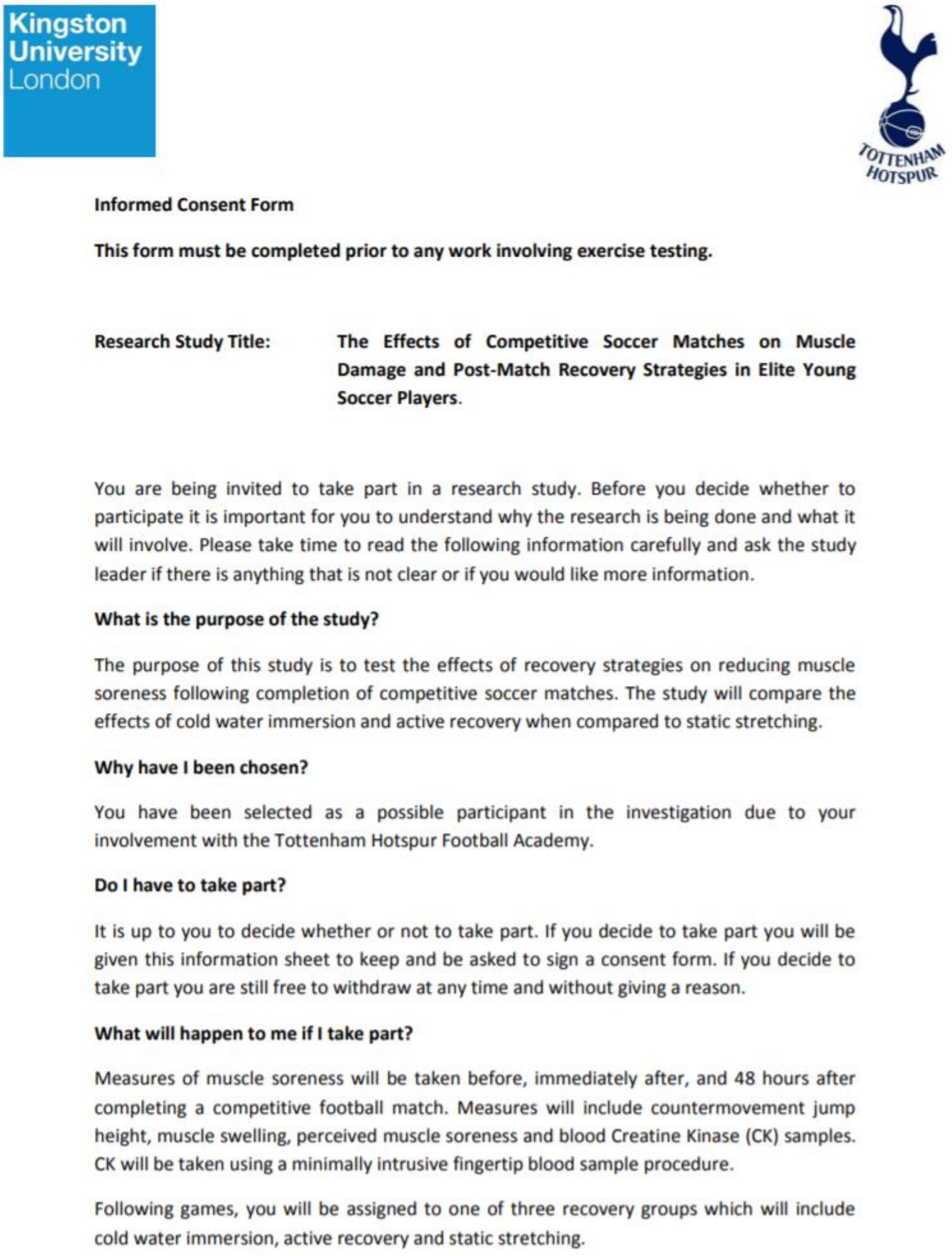
Emergency Contact details:

________________________________________________________________________________
________________________________________________________________________________
____________________________________________________________________________

Date__________________________________

**To **opt out** of photographic & video consent please tick this box ☐

## 11.3 Participant Information Sheet - contact details redacted



Kingston University London

TOTTENHAM HOTSPUR

**Informed Consent Form**

**This form must be completed prior to any work involving exercise testing.**

**Research Study Title:** **The Effects of Competitive Soccer Matches on Muscle Damage and Post-Match Recovery Strategies in Elite Young Soccer Players**.

You are being invited to take part in a research study. Before you decide whether to participate it is important for you to understand why the research is being done and what it will involve. Please take time to read the following information carefully and ask the study leader if there is anything that is not clear or if you would like more information.

**What is the purpose of the study?**

The purpose of this study is to test the effects of recovery strategies on reducing muscle soreness following completion of competitive soccer matches. The study will compare the effects of cold water immersion and active recovery when compared to static stretching.

**Why have I been chosen?**

You have been selected as a possible participant in the investigation due to your involvement with the Tottenham Hotspur Football Academy.

**Do I have to take part?**

It is up to you to decide whether or not to take part. If you decide to take part you will be given this information sheet to keep and be asked to sign a consent form. If you decide to take part you are still free to withdraw at any time and without giving a reason.

**What will happen to me if I take part?**

Measures of muscle soreness will be taken before, immediately after, and 48 hours after completing a competitive football match. Measures will include countermovement jump height, muscle swelling, perceived muscle soreness and blood Creatine Kinase (CK) samples. CK will be taken using a minimally intrusive fingertip blood sample procedure.

Following games, you will be assigned to one of three recovery groups which will include cold water immersion, active recovery and static stretching.

**What are the possible disadvantages and risks of taking part?**

All exercise involves a slight risk of injury, however the individual programmes you are given are designed to reduce this risk significantly. The discomfort experienced with such testing is only that typically experienced during normal exercise.

The potential risks to participants are very small and are acceptable in the light of the probable benefits for participating in the study to help determine whether the recovery strategies that are implemented for injury rehabilitation are an effective way to decrease muscle soreness and improve recovery time.

This is a simple procedure and causes only minor discomfort and will be performed by a trained individual following good clinical practice but may include discomfort at the site of puncture, possible bruising around the puncture site and uncommonly, infection or faintness from the procedure. Good practice however, minimises these risks.

**What are the possible benefits of taking part?**

By completing this test, we hope to determine the extent of muscle soreness you suffer as a result of football matches. We also hope to establish the most suitable method of recovery for you to improve the time you need to recover following football matches. This will allows us to set individual programmes to assist in the recovery process that is most suited to you.

**What if something goes wrong?**

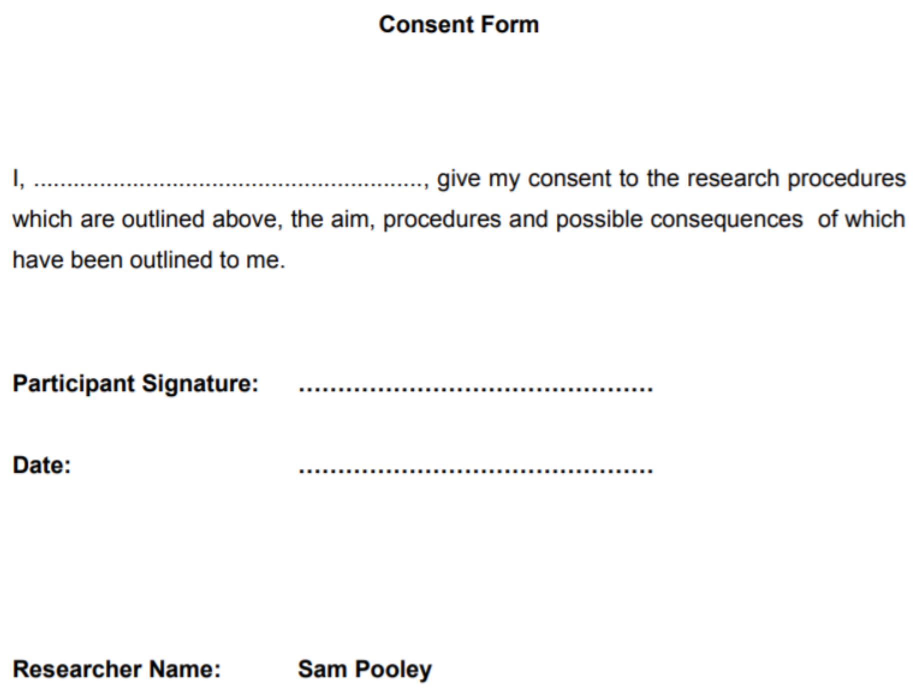
If you are harmed by taking part in this research project, there are no special compensation arrangements. If you are harmed due to someone's negligence, then you may have grounds for legal action but you may have to pay for it.

**Will my taking part in this study be kept confidential?**

All information about you that is collected during the course of the research will be kept strictly confidential.

If you wish to find out any more about this study, feel free to contact:

## 11.4 Participant Informed Consent Sheet - signature redacted



### Consent Form

I, ............................................................, give my consent to the research procedures which are outlined above, the aim, procedures and possible consequences of which have been outlined to me.

**Participant Signature:** ..............................................

**Date:** ..............................................

**Researcher Name:** **Sam Pooley**