THE EFFECT OF DIETARY SUPPLEMENTATION ON FATIGUE AND RECOVERY AFTER RESISTANCE EXERCISE IN FEMALES

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ABSTRACT
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Resistance exercise has been a popular form of muscle strength development for sport participants. This type of exercise activates a wide variety of physiological mechanisms involved with the exercising of muscle. The aim of this thesis was to investigate responses to dietary supplementation on muscular strength and biochemical indices to resistance exercise in female subjects. Firstly, to determine the reliability of the isometric test designed, and the number of trials required to obtain reproducible measurements of maximum voluntary isometric force and rate of force development. Secondly, to establish heavy resistance exercise volume and intensity to produce a fatigue effect of a 40% reduction in measured force variables. Thirdly, to determine the optimal recovery period required to overcome the effect of fatigue responses to heavy resistance exercise after ingesting carbohydrate supplement (CHO). Finally, to determine the effect of creatine supplementation (Cr) on fatigue and recovery responses after resistance exercise in female subjects.

Study one (1.A): The objective of study one was to quantify for female subjects, maximum voluntary isometric contractions (MVC) and rate of force development (RFD) and to evaluate the repeatability (between-days) of measurements. The data showed a small systemic bias between days for both, right and left leg and showed good reliability between days for MVC (range 5.4% to 11.5%), (9.55% to 36.3%) and (5.8% to 11.4%) for both legs, right leg and left leg, respectively. The LOA for RFD showed good reliability between days for all conditions (range 0.1% to 7.4%). It was concluded that the average of 3 trials between days is satisfactory for the repeatability of MVC and RFD.

Study one (1.B) This second part of study one was to determine if there was a fatiguing effect of the testing protocol and also to establish the fatigue effect of the heavy resistance exercise. The same subjects were used as in study 1A, but with the fatigue effects of an exercise trial between sessions 2 and 3. Subjects performed three sets of six different resistance exercises involving the lower body at an intensity corresponding to 60% of 1-RM (8-10 repetitions). The LOA for MVC was 0.6%, 13.7%, and 6.7%, for both legs, right leg and left leg respectively, and for RFD was 0.3%, 4.4%, and 5.3% for both legs. It was concluded that using both legs for studying the MVC was more reliable than using one leg for within-day and between-days force measurements.

Study 2: The objective of study two was to establish the heavy resistance exercise volume and intensity to produce a fatigue effect of a 40% reduction in measured force variables and to establish the fatigue and recovery responses over a 48 hour period. Subjects were familiarised with the same testing procedures as in the pilot study 1B. All subjects performed three sets of six different exercises (lying leg curls, dumbbell lunges, barbell squats, leg extensions, straight leg deadlift, leg presses) at an intensity corresponding to 70% of 1-RM (8-10 repetitions). Measurements were obtained after 2h, 24h and 48h recovery for MVC and RFD. A significant main effect was found for time on MVC and RFD for both legs and the dominant leg (P < 0.001) across recovery time, but there was no significant difference for MVC at 48h for both legs and 24h, 48h for the dominant leg, and no significant difference for RFD between pre-exercise and 24h and 48h for the dominant
leg. The fatigue protocol reduced measured force variables by 23.7% and 34.2%, and recovery from fatigue had been achieved after 48 hours.

**Study 3:** The objective was to quantify the effect of carbohydrate supplementation on muscular strength after resistance exercise in females. Carbohydrate (CHO) supplementation and placebo trials were randomised and conducted at the same time of day (9:00 am), on two separate occasions with one week between sessions. For the CHO trial, participants ingested a carbohydrate solution (0.5g CHO per kg/BM). The resistance exercise protocol described in study 2 was employed in this experiment. Instead of using 70% of the 1-RM, a work load in this study corresponding to 80% 1-RM was used. A significant (P<0.05) overall main effect for condition and time on MVC and RFD was found, but there was a non-significant interaction between condition and time. The data showed that there was a faster recovery in the CHO condition with a suggestion of super-compensation. The resistance exercise for the lower body resulted in a significant decrease (P<0.05) in MVC immediately after resistance exercise, and this occurred similarly in both CHO and placebo trials.

**Study 4:** The objective was to quantity the effect of creatine (Cr) supplementation on muscular strength and biochemical responses to resistance exercise in female subjects. The methodological studies described in the pilot and main studies were use to create the protocols to reliably assess MVC and RFD. Subjects undertook a resistance exercise session at an intensity corresponding to 80% of 1-RM. They were required to consume 20g of creatine monohydrate or placebo in a double-blind experimental design for 5 days before being-tested. Blood samples were taken before each session of tests, and analyzed for blood biochemical variables which included: creatine kinase (CK), growth hormone (GH), Myoglobin (MYO). A significant effect of Cr was found on MVC and RFD recovery (P<0.01). Body mass was not significantly different between sessions (P= 0.14) but there was a slight increase (1.0 kg) following Cr supplementation compared to other conditions. The CK and MYO, data revealed no significant main effect on time and conditions (P>0.05). Indicating that the fatigue protocol did not induce muscle damage the GH data showed a significant mean effect of time and conditions (P<0.05), conforming an hormonal response to exercise. It was concluded that oral creatine supplementation enhances recovery following a resistance exercise challenge with a suggestion of a super-compensation at 48 hours.

In summary, the procedure of resistance exercise was used in the four experimental studies and nutritional supplementation (CHO and Cr) significantly reduced the decline in maximal peak force and enhanced recovery following resistance exercise. It was concluded that the recovery from heavy resistance exercise in female appears to be aided by dietary supplementation producing an increase in the recovery of both maximal voluntary contraction force and rate of force development.
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For making the long days seem shorter, the dark days light, and the mundane tasks exciting, my mother and my father, I thank you both for being there and helping me in my times of greatest need. Also, I extend my thanks and gratitude to everyone who has helped me throughout the preparation of this work.

Finally, I am eternally grateful to the Libya Arab Jamahiriya who provided me with much needed funds to maintain my research efforts to undertake my Ph.D programme in the UK.
DEDICATION

This thesis is dedicated to my father and mother and who taught me what hard work and dedication can accomplish in life, and my brothers and sisters for the everlasting pray, love, support, and encouragement they provide me with.
DECLARATION

I declare that the work in this thesis is entirely my own. This project was supervised by two members of academic staff (Professor Adrian Lees my Director of Studies and Professor Don MacLaren). No portion of the work referred to in this thesis has been submitted in support of an application for any another degree or qualification of this or any other university or institute of learning.
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<td>1-RM</td>
<td>One repetition maximum</td>
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<td>Rate of force development</td>
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CHAPTER 1

INTRODUCTION
1.1. INTRODUCTION

Human skeletal muscle is capable of generating immense force and power output when properly activated (Astrand and Rodahl, 1986). Muscular strength is one of the major factors influencing the performance of sports activity. An adequately functioning musculoskeletal system is a key factor for functional capacity and good quality of life (McBride et al., 2004) and an enhanced musculoskeletal fitness is often associated with an improvement in health status (Kay et al., 2000; Wojtys et al., 1996). Therefore, success in many sports is closely related to the athlete's ability to develop muscular strength.

There are several ways to measure muscle strength. Currently the most common methods of measurements are isotonic, isometric, and isokinetic. In the past, muscle strength assessments were done in the isometric condition as movement changes muscle length and function, and hence affects muscle strength. Best and Taylor (1973) and Winter (1979) reported that the isometric condition is theoretically simple and experimentally well controllable. It allows a rather easy measurement of muscular effort and therefore most of the information currently available on human strength is described by the outcomes of isometric testing. Many studies have used isometric tests as a common form of muscular strength assessment in the laboratory. The main reason for the isometric mode to have been the standard for strength assessment was that the force developed in a concentric contraction decreases as a function of movement speed, and so the maximum active force production occurs during an isometric contraction (Wilson et al., 1993). Measurement of isometric strength appears to yield a reasonable estimate of the maximal possible effort for most slow body movements. The peak torque obtained from a maximal voluntary contraction (MVC) during an isometric test is a common
variable used to quantify strength (Andrews and Bohannon, 2000). Strength may also be quantified by examining the rate of force development (RFD) which is typically quantified as the greatest slope of the force-time curve obtained from a maximum effort isometric contraction (Wilson et al., 1993).

During exercise, the magnitude and mechanisms of human skeletal muscle fatigue vary widely and depend to a large extent on the individual, the type of muscle, and the exercise stimulus or task. The exercise prescription of the specific program design reflecting these targeted program goals includes variables such as option of exercises, order of exercise, amount of rest used between sets and exercises, number of repetitions and sets used for each exercise, and the intensity of each exercise (Kraemer, 2002; Kraemer and Ratamess, 2005).

The consequence of muscle fatigue is that there will be some limitation on physical performance, regardless of individual circumstances. Muscle fatigue is defined as a decline in the maximal voluntary force produced by a muscle following a period of exercise, coupled to a time-dependent reduction in the force-generating capacity of the muscle and an inability to maintain adequate force during voluntary contraction (Hainaut and Duchateau, 1999; Gandevia, 1992). Fatigue may also be accompanied by slowing of relaxation, reduced muscle shortening velocity, and recruitment of additional motor units in an attempt to maintain force output. Muscle fatigue may occur due to factors related to the muscle itself or to extra-muscular factors which influence how the muscle contracts. The concept of muscle fatigue is thought to have two main components these being central fatigue and peripheral fatigue. For example, a reduction of maximal voluntary contraction (MVC) of the quadriceps muscles has been found after 2-h cycling
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performed at a constant power output (Leveritt et al., 2000). This reduction resulted from changes in central and peripheral mechanisms such as decrease of neural input and alterations of the M-wave and isometric muscular twitch.

A common well-known problem studying human muscles is the difficulty in controlling factors such as the individual’s nutritional status (Mujika and Padilla, 1997). Creatine and carbohydrate are essential nutrients that are found in balanced diet. Many athletes nowadays consume nutritional ergogenic aids such as carbohydrate and creatine in an attempt to increase muscle mass and force output (Vandenberghe et al., 1997). Balsom et al., (1994) and Volek and Kraemer, (1996) reported that creatine has become one of the most popular nutritional supplements of the past decade. Many, but not all, scientific research studies to evaluate creatine as an ergogenic aid have found that creatine ingestion improved repetitive, short-term, strenuous exercise performance, thereby leading to an increase in muscle strength and girth. Creatine is an osmotic agent in skeletal muscle and increases water retention in cells also leading to an increase in muscle size through stimulation or protein synthesis (Volek and Kraemer, 1996).

Similarly, carbohydrate supplementation is usually associated with improvement of exercise performance and capacity and results in rapid repletion of muscle glycogen (Ivy et al., 1988). Further, a low carbohydrate intake can be detrimental to performance. Some evidence suggests that high-carbohydrate diets optimise muscle and liver glycogen stores (Bergstrom et al., 1967; Nilsson and Hultman, 1973) and have been shown to optimise performance during prolonged, moderate intensity exercise, intermittent exercise (Hargeaves, et al., 1984) and also in high-intensity exercise of short duration (Maughan and Poole, 1981; Pizza, et al., 1995).
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A true gender difference exists between males and females in muscular strength, as well as metabolic and hormonal profiles (Ditor and Hicks, 2000). Comparison between males and females has shown important metabolic and hormonal differences in response to strength exercise (Nissen, 2001). The endocrine system secretes anabolic hormones, e.g. growth hormone, which has an influence on resistance training-induced adaptations in skeletal muscle (Sheffield-Moore and Urban, 2004). Also growth factors are produced locally in worked muscles. These functional and physiologic adaptations are similar in nature among men and women at all ages. However, sex and age differences may exist in the absolute magnitude of adaptation to resistance training (Deschenes and Kraemer, 2002).

Young women are capable of longer-duration contractions than young men when performing sustained sub-maximal isometric contractions to failure at low-to-moderate intensities (Hicks et al., 2001). This sex difference is observed for several muscle groups, including the adductor pollicis (Ditor and Hicks, 2000), elbow flexors (Hunter and Enoka, 2001; Hunter et al., 2002), the extrinsic finger flexors (Petrofsky et al., 1975; West et al., 1995), the back extensors (Clark et al., 2003), and the knee extensors (Maughan et al., 1986). While men are stronger than women, the latter are able to sustain a contraction for a longer duration before failure of the task (Hunter Enoka, 2001). These results are consistent with the hypothesis that men, who are usually stronger than women and sustain greater absolute forces when the contraction is performed at a relative intensity, experience increased intramuscular pressures, greater blood flow occlusion, increased accumulation of metabolites and impairment of oxygen delivery to the muscle (Mitchell, 1980; Sadamoto et al., 1983), and an earlier onset of task failure during a sustained contraction. Consistent with these results is that the sex difference in muscle
fatigue for maximal contractions is eliminated when blood flow to the muscle is occluded (Russ and Kent-Braun, 2003).

Although muscular strength is an important factor in achieving optimum sports performance in female athletes, the majority of studies on the effects of resistance exercise using indices pertinent to muscular strength along with biochemical and hormonal responses have generally been carried out in male populations.
1.2 Aim of the thesis

The aim of this thesis is to explore the fatigue and recovery responses to resistance exercise and the effects of nutritional supplements in females.

1.2.1 Objectives of the thesis

The aim of this thesis was achieved by following objectives:

1. To establish the reliability of measurements of maximum voluntary isometric force (MVC) and rate of force development (RFD) in females.
2. To establish the intensity of exercise required to produce a pre-determined reduction in MVC in females.
3. To characterise the fatigue and recovery responses to heavy resistance exercise in females in terms of isometric force variables
4. To examine the effects of carbohydrate supplementation on the neuromuscular responses to heavy resistance exercise in females
5. To examine the effect of creatine supplementation on the neuromuscular and biochemical responses to heavy resistance exercise in females.

These objectives were achieved with reference to the studies reported within the thesis. Objectives 1 and 2 are addressed in the first study, whereas objectives 3, 4 and 5 are addressed in studies 2, 3 and 4 respectively.
1.3 Operational definitions

**Central fatigue:** Fatigue affected by those factors influencing the central nervous system control of voluntary contraction.

**Endurance:** The capacity to continue a physical performance over a period of time.

**Fatigue:** A loss of strength to continue a given level of physical performance.

**Isometric:** A condition in which the length of the muscle-tendon complex remains constant.

**Isometric contraction:** A muscle contraction characterized by rising tension production but no change in muscle-tendon complex length.

**Muscle:** A bundle of fibres, able to contract or be lengthened. In this context, striated (skeletal) muscle that moves body segments about each other under voluntary control.

**Muscle contraction:** The result of contractions of motor units distributed through a muscle so that the muscle length is shortened.

**Muscle strength:** The ability of a muscle to generate and transmit tension in the direction of its fibres.

**Maximal voluntary contraction:** Maximal voluntary contraction (MVC) means the muscle has contracted to the best of its ability.
**Torque:** The product of force and the perpendicular length of the lever-arm at which it acts.

**One Repetition Maximum (1-RM):** The maximum resistance with which a person can execute one repetition of an exercise movement. See repetition.

**Peripheral fatigue:** Fatigue affected by the factors within the muscle.

**Resistance:** The force that a muscle is required to work against.

**Repetition:** Performing the same activity more than once.

**Strength:** The amount of muscular force that can be exerted.

**Set:** A group of repetitions of an exercise movement done consecutively, without rest, until a given number, or momentary exhaustion, is reached.
CHAPTER 2

REVIEW OF THE LITERATURE
Chapter 2 - Review of the literature

The Review of the Literature is divided into three sections. The first section introduces the physiological responses and adaptations to individual strength and endurance training sessions. The second section outlines the findings of concurrent training research including the effects of prior bouts of endurance or strength training on subsequent muscle force generating capacity and recovery dynamics. Finally, the third section presents possible mechanisms for compromised responses and adaptations with concurrent training. Furthermore, due to the vast volume of literature in relation to muscle strength and endurance training responses and adaptations, the Review of the Literature could only cover those areas that are deemed most relevant to the concept of training.

2.1. Muscle strength

2.1.1. Physiological Muscle strength

Muscular strength is important in sport as well as in daily activities. The need for muscular strength runs across a spectrum of people from elite athletes attempting to optimize sports performance to frail elderly trying to perform activities of daily living. An adequately functioning musculoskeletal system (musculoskeletal fitness) is a key factor for functional capacity and good quality of life (McBride, et al., 2004) and an enhanced musculoskeletal fitness is often associated with an improvement in health status (Kay et al., 2001, Hunter et al., 2001). Furthermore, if muscle strength is not maintained, musculoskeletal fitness is then compromised which can significantly affect physical health and well-being (Kay et al., 2001).

Strength is generally defined as the capacity to produce force or torque generated during maximal isometric contraction (Atha, 1981). As early as 1903 Fick, at the Croonian
Lectures, reported that the strength of a muscle depends upon the number of fibres in what is known as the physiological cross-section area. In a muscle with parallel or nearly parallel fibres which have the same direction as the tendon, this corresponds to the anatomical cross-section, but in uni-pinnate and bi-pinnate muscles the physiological cross-section may be nearly at right angles to the anatomical cross-section as shown in Figure 2.1. Within the muscle fiber, strength is developed by filament contraction in the longitudinal direction. The filament tensions combines to give a resultant tension of the muscle. Its magnitude depends mostly on the number of muscle fibers involved, i.e. on the cross-sectional thickness of the muscle. Maximal isometric stress in human skeletal muscle is reported in the range of 16 - 61 N/cm; Enoka, (1988) uses 30 N/cm as a typical value.

Figure 2.1. A, fusiform; B, unipinnate; C, bipinnate, the physiological cross-section (Enoka, 1988).

Elevated muscle tension has been linked to an increased incidence of injury and accelerated depletion of muscular fuel stores. Athletes can reduce muscle tension by maintaining flexibility. Stretching disengages the cross bridges, which allows blood to flow to this area, removing accumulated metabolic by-products from heightened muscle tension (Atha, 1981).
2.1.2. Gender and muscle function

In terms of muscle contractile characteristics and the ability to produce force, muscle is identical in both males and females. The difference that exists in strength levels are primarily a function of total muscle mass. Only 24 percent of the typical female body is muscle mass, whereas the male is 40 percent muscle mass. Strength of the female lower body is similar to that of men relative to body mass and lean body mass. Men are stronger in the upper extremities due to their greater development of muscle mass in that area. Because of this and the fact that a female typically uses the muscle mass in her lower body to a much greater degree then she uses the muscle mass of her upper body, the female is seldom as strong in absolute measurements as the male (Margareta et al., 2005; Barry and Gallagher, 2003; Lambert et al., 2002).

The literature on muscle fatigue suggests that women generally have longer endurance times than men, especially at low-to-moderate forces (Kahn et al., 1986; West et al., 1995; Zijdewind and Kernell, 1994). For example, the endurance time of women was longer than that of men when performing an isometric contraction at 20% of maximum with the knee extensor muscles but not at 50 or 80% of maximum. Similarly, women were able to perform a greater number of repetitions with the elbow flexor muscles when lifting loads that were 50, 60, and 70% of maximum but not with loads that were 80 or 90% of maximum (Maughan et al., 1986).

Resistance exercise programs for women do not need to be drastically different from those for men, except for a few physiological issues (e.g. joint laxity, menstrual cycle). Nevertheless, understanding differences in how women generally respond and adapt to exercise can facilitate design of an individualized and optimal exercise program for women. Fulco, (1999) reported that research on resistance exercise is mostly studied on
men. However some studies investigate muscular strength and function for both genders (Margareta et al., 2005; Barry and Gallagher, 2003), were performed: hand-grip strength; abdominal strength; arm/shoulder strength; quadriceps muscle strength; and a functional test of leg muscle endurance. It is well known that there are differences in muscular strength and function between healthy men and women.

Generally, males have a higher capacity for anaerobic metabolism and their muscles generate a higher maximum power output than females. There are also important differences between female and male muscles during prolonged intense activity leading to fatigue, where female muscles have been found to be more fatigue resistant and to recover faster than male muscles (Fulco et al., 1999; Lindle et al., 1997). Conversely, other studies did not observe any change in muscle function in response either to increased estrogen levels (Greeves et al., 1999) or to fluctuations during the menstrual cycle (Janse et al., 2001). There is still a dearth of consistent results for female subjects. Another confounding point of many studies is the conditioning status of the subjects: trained or untrained individuals. The exercise response in trained individuals is often several times different than that of untrained individuals. This complicates extrapolation of results from one group to the other.

2.2. Strength Training

Muscle strength can be defined as the maximum force generation capacity (Macaluso and De Vito, 2004). The neural factors regulate muscle force generation. Increased levels of muscle activation and consequent increase in muscular force are achieved by increases in the firing rate of each motor unit, changes in the model of motor part activation and the recruitment of more motor units (Komi, 1978; Häkkinen, 1994; Drew et al., 2002;
Kamen and Ratamess, 2005). Regular contact to heavy resistance exercise will result in increases in maximal muscular strength and changes in both neuromuscular function and muscle morphology (Tesch, 1988; Haff et al., 1997; Aagaard, 2002; Fry, 2003).

However it has been well known that systematic resistance training, especially among untrained healthy subjects, has a powerful effect in promoting increases in size and strength of skeletal muscle. This is true both in men and women. Although women have lower absolute strength than men, the relative increases in strength following a training programme are similar between genders, at least in the beginning of resistance training (Häkkinen and Pakarinen, 1993; Stroud et al., 1994; Häkkinen et al., 2000).

2.2.1. Neural adaptations to resistance training

Neuromuscular performance depends not only on the quantity and quality of the involved muscles, but also by the ability of the nervous system to appropriately activate the muscles. Adaptive changes in the nervous system in response to training are referred to as neural adaptation (Moritani and DeVries, 1979; Sale, 1991; Moritani et al., 1995). Resistance training may cause adaptive changes within the nervous system that allow a trainee to more fully activate prime movers in specific movements (Sale, 1991).

Adaptations of the neuromuscular system to resistance training are focused on the development and maintenance of the neuromuscular unit needed for force production. Resistance training induces adaptations are mediated by supraspinal mechanisms, which include increased excitation (Aagaard et al., 2002; Gandevia, 2001) and changes in the organization of the motor cortex (Barry and Gallagher, 2003). This can influence the manner in which trained muscles are recruited by the Central Nervous System (CNS)
during related functional tasks (Carroll et al., 2001). Nervous system adaptation to resistance training may also include descending neural tracts and spinal cord circuitry.

Resistance training-induced changes in synaptic efficacy within the moto-neuron pool (Semmler et al., 1999) and neural pathways at the spinal cord may help the way in which muscles are co-ordinated during related movement tasks (Carroll et al., 2001). Nervous system adaptation to resistance training may also include the motor end plate connections between moto-neurons and muscle fibres (Carroll et al., 2001). Increased activity of the myo-neural synapse results in morphological changes of the neuromuscular junction which are associated with functional alterations in neuromuscular transmission that enhance neuromuscular transmission (Deschenes et al., 2002). These adaptations can enhance the activation of muscles and are likely to be expressed whenever the moto-neuron pool of the trained muscle is activated (Carroll et al., 2001; Barry and Gallagher, 2003).

Early increases in muscle strength due to resistance training are thought to result from neural adaptations and improvements in coordination while later strength increases arise from increased muscle hypertrophy (e.g. Komi and Viitasalo, 1977; Sale, 1991; Staron et al., 1994). During the first few weeks of resistance training there is an increase in maximal muscle force output that cannot be accounted for by muscle hypertrophy (Griffin and Cafarelli, 2005). Increases in muscular strength due to resistance training may be produced by increased neural drive resulting increases in motor unit release rate to agonist muscles (Schillings et al., 2003; Aagaard et al., 2002) and maybe also increases in the recruitment of additional motor units (Barry and Gallagher, 2003).
Furthermore, cross-sectional studies suggest that years of resistance training may be associated with increased maximal firing rates (Griffin and Cafarelli, 2005). Also synchronization among motor unit firing rate and frequency of firing may increase during resistance training (Enoka, 1997; Van Cutsem et al., 1998; Griffin and Cafarelli, 2005; Kamen, 2005). Neural adaptations to resistance training include reductions in the level of coactivation of the antagonist muscles (Sahin, 1992; Häkkinen et al., 1998, 2000) and changes in synergistic muscle activation (Rutherford, et al., 1986; Rate et al., 2003), which could contribute to maximal force generation.

Resistance training may cause adaptive changes within the nervous system that allow a trainee to better organize the activation of all relevant muscles, thereby effecting a greater net force in the intended direction of movement (Sale, 1991). While resistance training leads to strength increases by increasing the force-generating capacity of individual muscles, it is likely that neural adaptations also comprise changes in the neural activation of muscles, with modifications occurring in both intramuscular and inter-muscular coordination (Rutherford, et al., 1986; Enoka and Stuart, 1992; Häkkinen et al., 1998, 2000). Some of the adaptations associated with resistance training may be regarded as motor learning, i.e. learning to produce the specific patterns of muscle recruitment that are associated with optimal performance of movement task (Carroll et al., 2001).

2.2.2. Rest Intervals

Rest interval is the pause between exercise sets that allows muscles to partially recover before beginning the next set, the rest interval between strength training sessions depends on the conditioning level and recovery ability of the individual, the training phase, and
the energy source used in training. Well-conditioned athletes always recover faster, especially as training progresses toward the competitive phase, when they are supposed to reach their highest physical potential (Fox et al., 1974). Sale (1991) suggested that one minute of rest be provided between trials. Caldwell et al., (1974) and Chaffin (1975) recommended a rest interval of two minutes between trials if a large number of trials (e.g., 10). In addition Diane et al. (2006) used 2- to 3-minute rest interval between each block of contractions, but rest intervals may be as short as 30-s if only a few trials are performed. Collectively, the available literature suggests that a one-minute rest period should be sufficient to allow adequate recovery between trials. It should be noted however, that these recommendations are derived from testing experience as opposed to experimental validation.

2.2.3. Number of Repetitions
Edwards et al. (1977) used three maximal voluntary contractions in testing the quadriceps since the first contraction was usually “tentative”, while the second and third maximal contractions were usually similar to one another (coefficient of variation = 2.8%). Zeh et al. (1986) reported that the mean of three trials was highly correlated with the first score of the three and concluded that one repetition provides “a reasonably good indicator of the subject’s strength in that position”. They also noted that use of two repetitions increased the precision of the measurement. The advantage of using few test repetitions is decreased injury risk, especially for testing that stresses the lumbar spine (Zeh et al., 1986). In addition, fewer repetitions will minimize the confounding effects of fatigue on the strength data. However, their regression analysis did not address potential systematic bias in the use of only one or two trials. While there is no consensus in the literature, three test repetitions are likely to be sufficient to elicit a maximal value.
2.3. Muscular Strength Testing

2.3.1. Isometric strength

The term isometric in physics refers to static, because there is no change in movement. Isometric resistance refers to a muscular action during which no change in the length of the muscle-tendon complex takes place. This type of resistance training is normally performed against an immovable object such as a wall, a barbell, or a weight machine loaded beyond an individual's maximal concentric strength (Fleck et al., 1997). The static condition is theoretically simple and experimentally well controllable. It allows for a rather easy measurement of muscular effort. Apart from maximum isometric strength and isometric force–time curve characteristics, such as the rate of force development, are important capacities of the neuromuscular system for developing maximal force rapidly, and are related to athletic performance (Katartzi et al., 2005; Papadopoulos, 1997). These force–time parameters are the starting force at 100 ms, the peak force relative to body mass, and the rate at which isometric force can be developed (rate of force development RFD); (Katartzi et al., 2005; Papadopoulos et al., 2006; Papadopoulos and Salonikidis, 2000). In isolated muscle preparations, contractile RFD is obtained from the slope of the force-time curve (_force_/time), whereas, for intact joint actions, RFD is calculated as the slope of the joint moment-time curve (_moment_/time).

The time period for which the rate of change in force is determined has varied from an interval of 5 ms (Wilson et al., 1993) though to 60 ms (Christ et al., 1994) with most researchers tending to use a value towards the lower end of this range as it produces significantly higher values for RFD (Wilson and Murphy, 1996). The RFD parameter has important functional significance in fast and forceful muscle contraction. Measurement of isometric strength appears to yield a reasonable estimate of the maximal possible
effort for most slow body link movements. Force-velocity curves reported by Best and Taylor (1973) and by Winter (1979) indicate that the largest tension or force is indeed developed at zero velocity of muscle shortening or lengthening, which is the isometric case. However, the strength developed in faster motions, especially when concentric and ballistic, is not similar to that under static conditions.

2.3.2. Isometric testing

The isometric technique requires the individual to push or pull maximally against a recording device without movement taking place. Isometric testing is also called static testing. The primary advantage of isometric strength testing is that with the proper equipment, it is relatively quick and easy to perform which lends itself to testing of large groups of subjects (Fry et al., 1991). Varieties of devices have been used to measure isometric strength. These include cable tensiometers, strain gauges, and isokinetic dynamometers (with speed set to zero). In addition, with the exception of isokinetic devices, testing equipment is relatively inexpensive. Further, computer interfacing with isometric recording devices allows for the calculation of additional variables besides strength, such as the rate of force development (Haff et al., 1997). Isometric strength maximal isometric force of the leg extensor muscles was measured in a sitting position (knee and hip angle 90 degrees) (Katartzi et al. 2005) and testing as a highly reliable as assessed by reliability coefficients correlations between 0.85 and 0.99 (Abernethy et al., 1995). Testing at multiple joint angles allows for determination of strength throughout the range of motion.

Sale, (1991) suggested that isometric contractions of five seconds duration are long enough to allow for peak force development. As maximal force can only be maintained
for less than 3 seconds, Caldwell et al., (1974) recommended that contraction duration of
four seconds with a one second transition period from rest to maximal force should be
used. They also suggested that a four-second effort ensures that a three-second plateau
will occur and that the mean force over this three-second period be recorded.
Collectively, the available literature indicates that a contraction period with a one-
second-transition period and a four to five second plateau should be adequate to achieve
a maximal isometric contraction.

The disadvantage of isometric testing is that the strength values recorded are specific to
the point(s) in the range of motion at which the isometric contraction occurred, and
strength scores at one position may be poorly correlated with strength scores at other
positions (Bigland and Lippold, 1945; Drew et al., 2002). It has been questioned whether
static strength measures provide strength data that are specific to activities of interest
(Murphy, 1995; Zeh, 1986) and there are conflicting results in the literature as to whether
isometric testing is predictive of dynamic performance (Wilson and Murphy, 1991).
However, conflicting results regarding static versus dynamic relationships may be a
reflection of the joint angle used during isometric testing (Murphy et al., 1995).

2.3.3. One repetition maximum test (1-RM)
The gold standard for muscular strength testing is the 1-RM. Fry and Kramer, (1991)
suggest the following protocol for 1-RM testing. The test procedure begins with a warm
up of 5-10 repetitions at 40% to 60% of the subjects estimated maximum. After a brief
rest period, the load is increased to 60% to 80% of the estimated maximum for 3-5
repetitions. At this point a small increase in weight is added to the load and a 1-RM lift is
attempted. The goal is to determine the 1–RM in 3 to 5 trials. The subject should be
allowed rest at least 3-5 minutes before each 1-RM attempt. Therefore, Fry and Kramer, (1991) emphasize that ongoing encouragement and communication with the subject during this testing is crucial to obtain the best performance.

In addition to improved muscular strength, guidelines recommend beginner and intermediate exercisers train for 8 to 12 repetitions, at 60 to 70 percent of their maximum capacity for one lift (American Fitness, 2002). They should progress at a 2 to 10 percent increase depending on the muscle group, when one or two repetitions, more than the desired repetition range can be performed on two consecutive training sessions.

**2.3.4 Measurement of maximal voluntary muscle strength (MVC) and contractile rate of force development (RFD)**

Strength measured with a dynamometer can be quantified in different ways. Peak torque obtained from a maximal voluntary contraction (MVC) during an isometric test is a common measure of strength. MVC is typically defined as the maximal muscle force that a highly motivated subject is able to produce voluntarily under particular contractile conditions. The reductions in MVC force associated with muscle fatigue persist over the entire timescale of the progression of the degenerative and regenerative process i.e. until the muscle returns to its pre-fatigue condition (Warren et al., 1999). The assessment of skeletal muscle function as fatigable or a marker of damage necessitates reliable measures of maximal force. Routine measurements of maximal muscle force may include many potential sources of error of which the most important may be the possible lack of central drive to the muscle fatigue (Merton, 1954; Rutherford et al., 1986). Therefore, reliability should be considered as the amount of measurement error that has been deemed acceptable for the practical use of a measurement tool. Logically it is
reliability that should be first tested in a new measurement tool, since it will never be valid if it is not adequately consistent in whatever value it indicates from repeated measurements.

Although a number of researchers have examined the reliability of the twitch interpolation technique for the assessment of maximal quadriceps muscle force and voluntary activation (Behm et al., 2001; Oskeui et al., 2003; Todd et al., 2004), to the author's knowledge none have tested the reliability of these variables (using the twitch interpolation technique) across a timescale of several days (Morton et al., 2005). Such temporal evaluations of reliability estimates are of particular importance given that the reliability of any assessment should always be established with respect to its intended use or 'analytical goal' (Atkinson and Nevill 1998).

Strength can also be quantified by examining the rate of force development (RFD) (Aagaard et al., 2002). Explosive muscle strength is the rate of rise in contractile force at the onset of contraction, exerted within the early phase of rising muscle force (Schmidtbleicher and Haralambie, 1981; Sleivert and Wenger, 1994). In isolated muscle preparations, RFD is obtained from the slope of the force time curve (force/time), whereas, for intact joint actions, RFD is calculated as the slope of the joint moment-time curve (moment/time). The RFD parameter has important functional significance in fast and forceful muscle contraction (Figure 2.2). For example, fast movements such as sprint running, karate, or boxing typically involve contraction times of 0.50–2.50 ms. In contrast, it typically takes a longer time to reach maximum force in most human muscles, i.e., 300 ms for the elbow flexors (Sukop and Nelson, 1974) and knee extensors (Thorstensson et al., 1976). During fast limb movements, therefore, the short contraction time may not allow maximal muscle force to be reached. As a result, any
increase in contractile RFD becomes highly important as it allows reaching a higher level of muscle force in the early phase of muscle contraction. In addition to RFD, another important strength parameter is the total contractile impulse that can be produced within a given contraction time (Baker et al., 1994). In accordance with classic mechanical physics, the angular impulse, defined as the time-integrated moment of force, is identical to the momentum reached during limb movement.

**Figure 2.2** Rate of force development (RFD) for explosive muscle action. Also shown in maximum force (MVC). (Baker et al., 1994).
2.4. Physiology of Muscle fatigue

2.4.1. Muscle fatigue
Physiologists have several definitions of fatigue, all of which describe progressive loss of force-producing capacity and ultimately the inability to perform a desired level of physical work. Piper et al. (1987) and Meyerowitz (1979) described fatigue as a multi-component sensation with behavioural, affective, sensory, and cognitive components. They also designed a simple measurement tool for assessing fatigue that combined multiple fatigue-associated elements into an overall fatigue score. It is divided into peripheral and central components, a division based on whether a loss of capacity to generate a maximum force is found to originate in the muscle tissue or in the central nervous system, respectively. During a sustained maximal voluntary contraction (MVC), healthy subjects develop both peripheral and central fatigue (Kent-Braun, 1999; Schillings et al., 2003).

Muscle fatigue is thought to have both central and peripheral origins. By convention, central fatigue is confined to the breakdown in any process proximal to the neuromuscular junction and may comprise branch point failure in the motor unit, failure of axonal propagation, some non-specific inhibition at the level of the anterior horn, or a reduction of central drive from the supraspinal structures. Transmission failure at the neuromuscular junction, the sarcolemma or perhaps the T-tubular system are possible peripheral sources of fatigue, whilst other peripheral sites may be localized within the contractile apparatus, as the muscle contraction requires energy. This later is supplied via metabolic energy conversion in the mitochondria, cells located within the muscle. They release energy from the breakdown of adenosine triphosphate (ATP) to adenosine
diphosphate (ADP), which in turn is restored to ATP using energy derived from nutrients (Lynn and Taylor, 1971; Bagshaw and Trentham, 1974).

Central Fatigue: The central component to fatigue is generally described in terms of a reduction in the neural drive or nerve-based motor command to working muscles those results in a decline in the force output. (Gandevia 2001; Kay et al., 2000; Vandewalle et al., 1991) It has been suggested that the reduced neural drive during exercise may be a protective mechanism to prevent organ failure if the work was continued at the same intensity. (Bigland-Ritchie and Woods, 1986; Noakes, 2000) The exact mechanisms of central fatigue are unknown although there has been a great deal of interest in the role of serotonergic pathways (Davis, 1995; Newsholme and Blomstrand, 1995). Peripheral Fatigue during physical work is considered an inability for the body to supply sufficient energy to the contracting muscles to meet the increased energy demand or a failure of energy producing capacity of skeletal muscle. This is the most common case of physical fatigue-affecting a national average of 72% of adults in the work force in 2002 (Aagaard et al., 2002). This causes contractile dysfunction that is manifested in the eventual reduction or lack of ability of a single muscle or local group of muscles to do work. The insufficiency of energy, i.e. sub-optimal aerobic metabolism, generally results in the accumulation of lactic acid and other acidic anaerobic metabolic by-products in the muscle, causing the stereotypical burning sensation of local muscle fatigue.

The fundamental difference between the peripheral and central theories of fatigue is that the peripheral model of fatigue assumes failure at one or more sites in the chain that initiates muscle contraction. Peripheral regulation is therefore dependent on the localised metabolic chemical conditions of the local muscle affected, whereas the central model of
fatigue is an integrated mechanism that works to preserve the integrity of the system by initiating fatigue through muscle derecruitment, based on collective feedback from the periphery, before cellular or organ failure occurs. In summary, the conscious sensation of fatigue does not arise directly from the action of metabolites in the periphery but rather from the regulatory centres in the subconscious parts of the brain, the function of which is to ensure homoeostasis during all forms of exercise.

2.4.2. Metabolic characteristics of fatigue

'Metabolic fatigue' is a common term for the reduction in contractile force due to the direct or indirect effects of the reduction of substrates or accumulation of metabolites within the muscle fibre. This can occur through a simple lack of energy to fuel contraction, or interference with the ability of (Ca^{2+}) to stimulate actin and myosin to contract.

Karl and Kroemer, (1999) reported that this metabolic process requires sufficient supply of the muscle tissue with arterial blood. Blood brings the needed energy carriers and oxygen and it removes metabolic by-products, particularly lactic acid and potassium as well as heat, carbon dioxide and water liberated during metabolism. Sufficient blood supply and its unimpeded flow through the muscle's capillary bed into the venules and veins are crucial because they determine the ability of the metabolic processes and hence of the contractile efforts of the muscle to continue (Astrand and Rodahl, 1986; Kahn and Monod, 1989). A strongly contracting muscle generates strong pressure inside itself, as can be felt by touching a tightened biceps or calf muscle. By this pressure, the muscle compresses its own blood vessels thus shutting off its own circulation. Therefore, a maximal contraction can be maintained for only a few seconds.
Haylock (1979) and Piper (1987) repeated that muscular effort was made possible because chemically stored energy is released in the mitochondria of cells in muscle tissue. To keep this metabolic process going, blood flow through the tissues is necessary. If the flow is insufficient, muscle fatigue occurs.

Regardless of the mechanism, the damage appears to result in a pronounced weakness, the recovery of which, at least in unconditioned individuals, may take several days or even weeks (Newham et al., 1983; Clarkson and Tremblay, 1988). During high-intensity activity, this non-metabolic component would be expected to be progressive with the duration of the activity and, in fact, may represent a major aspect of the fatigue observed.
2.4.3 Resistance exercise-induced muscular fatigue

A heavy resistance exercise procedure performed with progressive overload leads to acute responses observed as a temporary decrease in maximal force production and electromyographic activity of the loaded muscles associated with increases in blood lactate concentrations (e.g. Tesch et al., 1983, Häkkinen et al., 1988). Therefore, the magnitude of neuromuscular responses can be measured as important indicators of training effects of different heavy resistance exercises. The performance of muscle gradually declines when muscles are used repeatedly at near their maximum force. This muscle fatigue is reflected in reduced force production, reduced shortening velocity and a slower time-course of contraction and relaxation (Allen, 2004).

Fatigue may be caused by diminished efferent neural direct to activate muscles from the central nervous system (i.e. central fatigue) which inhibits exercise activity before any severe damage to muscles and organs occurs. Fatigue may also be caused by factors within the muscle cells (i.e. peripheral fatigue) (St Clair Gibson et al., 2001; Westerblad et al., 2000). Phosphocreatine (PCr) depletion, intramuscular acidosis and carbohydrate depletion are all potential causes of the fatigue during resistance exercise (Lambert and Flynn, 2002).

Metabolic acidosis during the resistance exercise is caused by an increased reliance on non-mitochondrial ATP turnover. Lactate production is necessary for muscle to produce cytosolic NAD+ to support continued ATP regeneration from glycolysis. However, accumulation of lactate within skeletal muscle or blood directly contributes to intracellular acidosis and is therefore good indirect indicators of increased proton release and decreased cellular pH (Robergs et al., 2004, Lindinger et al., 2005).
2.4.4 Recovery from muscle fatigue

Recovery from fatigue has been found to be complex, with both fast and slower components. The faster component is due to reversal of the metabolic changes which caused periphery fatigue in the first place; for example, wash-out of lactic acid and restoration of the PCR store which take up the excess of phosphate ions. These processes are relatively fast and are completed in minutes. There remains a second component of fatigue, which recovers much more slowly, taking several hours or even days for muscles to regain their normal capacity. Experiments on isolated muscles suggest that this delayed recovery is caused by reduced Ca\(^{2+}\) release (Westerblad et al., 2000). The action potential is normal and the resistance session is normally loaded with Ca\(^{2+}\) but the coupling between the action potential and Ca\(^{2+}\) release is damaged. One suggestion is that some Ca\(^{2+}\) activated process might damage the proteins involved in Ca\(^{2+}\) release. With too much training the slow phase of recovery is never completed and performance can start to decline. Some athletes respond to this decline by training even more and a vicious overtraining cycle develops.

Various studies have also provided evidence of a failure at the level of the sarcoplasmic reticulum, for the sarcoplasmic reticulum to be implicated in fatigue, the coupling signal from the T-tubule, designed to elicit elevations in Ca\(2+\) consistent with maximal activation under non-fatigued conditions, must result in an inappropriate response from the sarcoplasmic reticulum (Gollnick et al., 1991; Allen et al., 1995).

This enzyme, as with the other major enzymes involved in excitation and contraction, is capable of hydrolysing ATP for the production of the energy necessary to pump the cytosolic Ca\(^{2+}\) against a concentration gradient, into the lumen of the sarcoplasmic
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reticulum, where it is stored or used for release through the Ca$^{2+}$ channel of the sarcoplasmic reticulum. The energy supplying systems, oxidative phosphorylation, glycolysis and high-energy phosphate transfer, must be precisely geared to regenerate ATP at a rate necessary to prevent any substantial depletion of ATP, which exists only in low concentration in the muscle (Sahlin, 1992).

Pascoe et al. (1990) reported that recovery of muscle glycogen stores underpins the recovery of endurance capacity for moderate to high intensity exercise. Low intensity exercise can be pursued when muscle glycogen stores have not been restored fully, although the duration and intensity will be limited by the inadequacy of the carbohydrate stores.

Furthermore, glycogen granules are physically associated with a number of proteins (including glycogen phosphorylase, phosphorylase kinase, glycogen synthase, glycogenin and phosphatases) that are involved in the metabolism of glycogen itself and other substrates such as glucose. This information implies that glycogen is not only a substrate for exercise metabolism, but may also have an important role in metabolic regulation (Marchand et al., 2002).

Reduction of muscle glycogen during exercise activates glycogen synthase (Nielsen and Richter, 2003), and this activation is greater when muscle glycogen is lower (Zachwieja et al., 1991), resulting in a faster rate of glycogen resynthesis in the early post-exercise period. The link between glycogen and glycogen synthase may be mediated by protein phosphatase 1, which is targeted to the glycogen molecule. The regulatory getting subunit of protein phosphatase 1 is essential for the exercise-induced activation of glycogen synthase in skeletal muscle (Aschenbach et al., 2001). As glycogen is
associated with the sarcoplasmic reticulum (Marchand et al., 2002), it is possible that glycogen has a key role in excitation-contraction coupling. It has been shown that low muscle glycogen following fatiguing contractions is associated with reduced sarcoplasmic reticulum Ca2\textsuperscript{+} release (Chin and Allen, 1997) and hence reduced force development.

2.4.5. Effect of fatigue and recovery during strength exercise

The type of fatigue-resistance exercise utilised by athletes for their training purposes leads to acute decreases not only in maximal peak force but also in the explosive force production capability (Hakkinen et al., 1988). The differences in magnitude of the decreases in neuromuscular performance and the various mechanisms leading to muscle fatigue and recovery may also be related in part to the type of fatiguing load (Hakkinen et al., 1988). For example, neuromuscular fatigue and recovery in the period up to 48 hours post-exercise has been investigated following maximum strength loading and explosive strength loading force (Linnamo et al., 1998). The authors attributed the specific motor unit recruitment patterns which may occur during explosive training. And suggested that fatigue after heavy exercise loading is of both central and peripheral origin, supporting the finding by Hakkinen (1992, 1994), whereas fatigue after explosive loading seems to be primarily central.

It is well known that impairment of performance resulting from muscle fatigue differs according to the types of contraction involved, the muscular groups tested, and the exercise duration and intensity. Depending on these variables, strength loss with fatigue can originate from several sites from the motor cortex through to contractile elements. A great number of studies have examined the event of fatigue after heavy resistance
exercise. However, when comparing men and women, besides lower absolute forces, women have been shown to have lower rates of maximal force production (Komi and Karlsson 1978; Ryushi et al., 1988). Also women have been shown to demonstrate less fatigue than men in heavy resistance exercise (Hakkinen, 1994). It would be of interest therefore to examine whether the differences in fatigue and recovery period would remain for resistance exercise.

With resistance exercise there is an immediate increase in epinephrine and nor-epinephrine (Kraemer and Ratamess, 2005). These hormones increase blood glucose and are important for increasing force production, muscle contraction rate, and energy production (i.e., the synthesis of ATP-the energy currency of cells). These hormones actually begin to rise prior to the resistance training aerobics (Kraemer and Ratamess, 2005). This is a preventative response of the body preparing for the challenging exercise to follow. Interestingly, the elevated blood glucose levels do not typically lead to an increase in insulin unless protein/carbohydrate supplementation precedes the workout (Kraemer and Ratamess, 2005). The increased uptake of blood glucose by the skeletal muscle is occurring due to the increase in function of the cell's glucose transporters, which increase glucose uptake and thus glucose metabolism in the muscle cell. Thus, regular resistance exercise training has been shown to increase 'insulin sensitivity', meaning the body can intake and use glucose more effectively (Pollock et al., 2001).
2.4.6. The Menstrual Cycle

One striking difference between male and female athletes can be attributed to the female hormonal cycle during exercise performance with the female reproductive hormone concentrations fluctuate significantly (Sarwar et al., 1996). There are four hormonal markers of the menstrual cycle and these are oestrogen, progesterone, follicle stimulating hormone (FSH), and luteinising hormone (LH). These hormones vary with the cycle and the fluctuations in female steroid hormones affect the autonomic nervous system and metabolic functions (Florini et al., 1996). Oestrogen concentration measurements are important in identifying the late-follicular oestrogen peak. Measurement of both oestrogen and progesterone is the only method that can identify between the three distinct phases: (i) early-follicular phase (low oestrogen and progesterone); (ii) late-follicular phase (high oestrogen and low progesterone); and (iii) mid-luteal phase (high oestrogen and progesterone) [see Table 2.1].

Table 2.1 Menstrual cycle phase terminology with corresponding days of the menstrual cycle, where possible accompanied by an indication of corresponding hormone concentrations of oestrogen and progesterone (Xanne and Janse, 2003).

Therefore certain physiological parameters and athletic performance could be influenced by the phase of the menstrual cycle. However, the influence of the menstrual cycle phase on exercise performance, particularly muscle strength, is unclear.
Interestingly, while some research has shown a significant difference in exercise-induced hormone changes during different phases of the menstrual cycle (Jurkowski et al., 1978; Hornum et al., 1997), others have demonstrated no significant phase effect. (Bonen, et al., 1983; Kanaley et al., 1992). However, these studies used very different exercise protocols, and the training status of the participants ranged from highly trained (Kanaley et al., 1992) to untrained, (Hornum et al., 1997) which could influence the results. To the authors' knowledge only one study has attempted to compare the short-term anabolic hormone responses between an endurance and resistance session, in the same group of pre-menopausal women. Consitt et al. (2001) reported that 40 minutes of cycling (75% of maximal heart rate) was capable of increasing serum levels of testosterone and oestradiol, compared with a resting session. Although increases were observed in these hormones after a resistance-exercise session including three sets of eight exercises at 10-RM (RM), they did not reach statistical significance when compared with the resting session.
2.4.6.1. Muscle strength during phases of the menstrual cycle.

Sarwar et al. (1996) tested skeletal muscle strength, relaxation rate and fatigability of the quadriceps during the menstrual cycle. They found no changes in these parameters for women taking oral contraceptives, whereas in women not taking oral contraceptives, the quadriceps were stronger, more fatigable and had a longer relaxation time at mid-cycle (day 12–18). Phillips et al. (1996) reported a higher adductor pollicis strength during the follicular phase than during the luteal phase, with a rapid decrease in strength around ovulation. They suggested that oestrogen has a strengthening action on skeletal muscle, although the underlying mechanism is not clear. Greeves et al. (1999), however, reported the highest quadriceps strength during the mid-luteal phase and found a positive relationship between strength and progesterone concentration. They showed that females undergoing in-vitro fertilization, in which there are those supra-physiological levels of oestrogen, did not cause any changes in strength of the first dorsal interosseus muscle. So the findings that suggest an effect of oestrogen on strength can be questioned.

Several other studies have found no changes in skeletal muscle strength over the menstrual cycle (Lebrun et al., 1993; Gür, 1997). Elliott (2003) and others (Janse de Jonge et al., 2001) found that menstrual cycle phase had no affect on skeletal muscle contractile characteristics in humans. The main problem in the measurement of maximum voluntary strength is ensuring that the contraction truly reflects the maximum force-generating capacity of the muscle. Even well-motivated subjects may not always reach full neural activation of their muscles (Rutherford et al., 1986). The extent of neural activation can be evaluated by applying a superimposed electrical stimulus to the muscle during the performance of a maximal voluntary contraction (MVC). When
comparing strength over a period of time, such as in menstrual cycle research, it is especially important to ensure maximal neural activation during each test.

A further problem encountered in research on the influence of the menstrual cycle on physical performance is the timing of the testing. It is difficult to predict the exact phases of the menstrual cycle and the concurrent reproductive hormone concentrations. Counting days from the onset of bleeding and basal body temperature charting can be used to estimate the different phases of the menstrual cycle. These methods, however, only provide predictions, and serum hormone level measurements of at least oestrogen and progesterone are necessary to confirm the menstrual cycle phase.

Janse et al. (2001) measured muscle function during three phases of the menstrual cycle with significantly different concentrations of circulating female reproductive hormones (Table 2.2). The results showed similar isometric quadriceps strength at menses (571 ± 114N), and decline the late follicular (551 ± 114N) and luteal (570 ± 109N) phases. No correlations were found between any of the strength and fatigue parameters and the serum concentrations of oestrogen, progesterone, LH and FSH.

Table 2.2 Serum hormone concentrations of oestrogen, progesterone, follicle stimulating hormone (FSH) and luteinising hormone (LH) throughout the menstrual cycle (Janse et al., 2000).
Muscle function measures show no correlations with any of the hormone concentrations. The studies suggest that the fluctuation in female reproductive hormones throughout the menstrual cycle does not significantly affect muscle strength, fatigability and contractile properties.

2.4.7. Nutritional considerations and fatigue

Energy balance is an important nutritional consideration for strength athletes, particularly women. Differences between men and women in substrate use during moderately intense exercise may be related to the reproductive hormones oestrogen and progesterone. Elevated concentrations of oestrogen and progesterone that occur during the luteal (Lut) phase of the menstrual cycle have been associated with enhanced glycogen storage (Hackney et al., 1994; Nicklas et al., 1989) and fat utilization during rest and exercise (Hackney et al., 1994). Although, this change in substrate metabolism may be beneficial during prolonged low-intensity exercise (Gollnick, 1988), higher intensity endurance exercise in the absence of glucose feedings may be adversely affected (Davis, et al., 1995). Furthermore, the effects of menstrual cycle phase on the potential positive effect of CHO supplementation on endurance performance during prolonged exercise are unknown.

Stephen et al. (2000) reported that the performance-enhancing effects of CHO supplementation during prolonged exercise at 70% $\dot{V}O_2$max are not influenced by menstrual cycle phase. Furthermore, menstrual cycle phase did not alter endurance performance or any hormonal and metabolic factors that could influence endurance performance.
Furthermore, when women were matched for lean body mass with men and compared, consuming a high carbohydrate diet resulted in absolute and relative increases in muscle glycogen that were much lower than for men, and only significant for women if total energy intake was substantially greater than habitual intake Tarnopolsky et al. (2001). Over time, the increased energy required to obtain adequate glycogen synthesis in women would lead to an energy imbalance. A number of studies showed that endurance exercises performed previously to strength exercises may have prejudicial effects on these, such as impairment of acute performance on specific tests. A possible explanation for such is that an endurance exercise session could promote acute metabolic changes on a subsequent strength training session (Leveritt et al., 2000).

On the other hand, a large body of scientific literature over the last decade has documented the physiological and performance effects of creatine supplementation. Short term creatine supplementation improves anaerobic performance, and long term creatine supplementation consistently augments strength and lean body mass gains with resistance training. Although the majority of studies have been in men, a significant amount of research indicates women are also responsive to creatine supplementation Volek and Rawson, (2004). Short term creatine supplementation enhanced high intensity exercise performance in women, (Eckerson et al., 2004; Tarnopolsky and MacLennan, 2000).

Earnest et al. (1995) have reported that short-term creatine intake increases fat-free mass in strength trained athletes. Accordingly, some (Bessman, and Savabi, 1990; Ingwall, 1976) but not all (Fry and Morales, 1980) in vitro findings indicate creatine stimulates the biosynthesis of muscle myosin. Given these earlier in vivo and in vitro findings, it is reasonable to consider that oral creatine supplementation may produce an anabolic action
in humans and thus is likely to enhance the effects of resistance training on muscle mass and strength. In summary, the nutritional needs of women engaged in strength training are important and are generally similar to those recommended for men.

Women in particular should ensure enough energy is consumed to optimise adaptations to training and improve general health Volek, (2004). There are gender differences in exercise metabolism that may give good reason for specific nutritional recommendations for women. These include less importance on carbohydrate intake after exercise. In addition, generous consumption of a healthy diet from a variety of sources is encouraged to support a positive energy balance, hormonal balance, and optimal health. Like men, women respond to the favourable effects of creatine supplementation during resistance training.

2.5. Blood parameters and hormonal responses to resistance exercise.

Resistance exercise acts as a powerful stimulus leading to the acute increases in serum concentrations of several hormones such as growth hormone, testosterone, oestradiol and cortisol. The nature of this stimulation varies according to the direction of the acute programme variables i.e. intensity (load) of exercise, number of sets and repetitions per set, length of rest periods between sets, and muscle mass involved (Häkkinen and Pakarinen, 1993) as well as subject characteristics of age, gender, health, and nutritional and training status (Sheffield-Moore and Urban, 2004). The acute decreases in maximal isometric force were quite similar in degree after “hypertrophic” resistance exercise performed with a 10-RM protocol for ten sets (Häkkinen, 1994) as compared to the
respective acute decreases after the "neural" high loading 1RM protocol of twenty sets (Häkkinen and Pakarinen, 1993).

In contrast to the 1-RM protocol, hypertrophic heavy resistance exercise is known to induce the greatest acute hormone responses when performed by multiple sets per exercise (e.g. 3-5 sets) with short rest periods (e.g. 60-120 sec.) between the sets and with a moderately high number of (e.g. 8-12 RM) repetitions per set (Häkkinen and Pakarinen, 1993). Therefore, the data of these previous studies suggest that the training mode seems to have an important influence on the magnitude and/or duration of acute hormonal responses. A previous study by Häkkinen et al., (2001) also suggests that acute hormone responses might have a relationship with gains in muscle mass or strength during resistance training. Thus, exercise-induced stimulation of the hormonal system may be a cause for the adaptation processes in skeletal muscle cells leading to increases in the contractile proteins. It is well known that the secretion of anabolic steroids results from resistance training, yet it is not clear why women show similar responses to training (compared to men) in the absence of increased testosterone levels (Taylor et al., 2000).

Taylor (2000) examined the differences in growth hormone (GH) response to acute bouts of resistance exercise in weight-trained and non-weight-trained women. Growth hormone is responsible for increasing protein synthesis and for mediating the release of insulin-like growth factor (IGF-1), which is another potential anabolic factor (Taylor et al., 2000). They hypothesized that women with weight training experience would have a greater GH response to the exercise stimulus than the non-weight-trained women (Taylor et al., 2000). These findings established that both weight-trained and non-weight-trained women have an acute rise in GH levels following resistance exercise, although, the
weight-trained women were able to continue exercise at the increased GH levels for a longer period of time.

Table 2.3 Main functional responses to exercise and glandular tissues and hormones involved in acute adaptation in men and women (from Robergs and Roberts, 1997).

FSH; follicle stimulating hormone; LH; lutenizing hormone; ACTH; drenocorticotropin

2.5.1 Creatine Kinase

Creatine Kinase (CK) is found predominantly in muscle and is released into the circulation during muscular lesions. Consequently, serum CK activity has been employed as a marker in exercise physiology and sports medicine for the detection of muscle injury and overload (Clarkson and Tremblay, 1988; Houmard et al., 1990; Schneider et al., 1995; Totsuka et al., 1996). However, some studies on CK release have not clearly demonstrated its value as a marker for these states (Nakaji et al., 1992; Newham et al., 1987).

Numerous studies have evaluated changes in CK activity after exercise and found that it differs markedly according to exercise conditions. For example, following isometric
muscle contraction, peak serum CK activity is observed relatively early i.e. 24-48 h after exercise (Clarkson et al., 1985; Graves et al., 1987; Kirwan et al., 1986), whereas it is seen 3-7 days after exercise following eccentric muscle contraction (Newham et al., 1987; Nosaka and Clarkson, 1992; Smith et al., 1994), and a biphasic pattern is observed after weight training (Tokuda, 1985).

These studies used short-duration, high-intensity workloads, which damaged muscle tissue and induced CK release, so their relevance to the actual process of CK release during and after strength exercise is not clear. There is a clear social trend toward increased physical activity, and regular physical activities with low or moderate intensity have been recommended for improving general health. However, non-athletes often experience fatigue and injury from daily exercise. This may be due to the lack of an objective marker for overtraining. Serum CK activity may act as a marker for fatigue or overwork in non-athletes, and it is therefore important to examine the effects of daily repeated aerobic exercise on serum CK activity in such subjects during and after non-injurious endurance exercise.

It is also possible that the CK response to exercise depends on the individual's physical characteristics or training background (Hortobagyi et al., 1989; Nakaji et al., 1992; Norton et al., 1985). Newham et al., (1986) found high and low responders after a stepping exercise.
2.5.2.1 Changes in CK activity

Serum concentrations of CK are increased after muscle is damaged by physical or biochemical injury. This has been reported after strenuous physical activity, (Schneider, et al., 1995) pain, crush injury, myositis, and muscular dystrophy. Other enzyme markers for skeletal muscle injury such as aldolase, enolase, aspartate aminotransferase, and lactate dehydrogenase isoenzyme 5 are not as specific as CK, Laurence, 2000; Wu and Perryman, (1992). The best studied of the muscle proteins that efflux into the blood following exercise damage is creatine kinase (CK). Because CK is found almost exclusively in muscle tissue, serum or plasma, it is considered an indicator of muscle damage (Armstrong, 2000). The role of CK is the hydrolysis of creatine phosphate within the muscle cell. Following muscle damage, changes in cell membrane permeability results in a release of CK into the blood (Nosaka et al., 1992).

There is large inter-subject variability in serum CK response after eccentric exercise, questioning the reliability of CK as an indicator of muscle damage. In one study, 10 males performed 24 maximal eccentric contractions. Over 72 hours post-exercise, peak CK response ranged from 236 U \cdot L^{-1} \text{ to } 2524 U \cdot L^{-1} \text{ (Nosaka and Clarkson, 1992). This is exaggerated further with significant differences observed between genders (Ebbeling and Clarkson, 1989). It appears that androgenic steroids increase CK efflux while, oestrogens inhibit CK release, resulting in women exhibiting lower CK responses after exercise (Amelink and Bär, 1986). Nosaka and Clarkson, (1992) hypothesised that larger post-exercise increase in plasma CK activity would be produced when a larger amount of muscle was damaged by eccentric exercise. In comparing eccentric work done on one arm followed by the other five weeks later to eccentric work on both arms on the same day, it appeared that CK activity did not reflect total muscle damage.
Nosaka and Clarkson (1996) found that peak CK correlated significantly with peak changes in isometric strength, range of motion, and circumference. These results contradicted the earlier experiment, but still contained massive inter-subject variations. At present there is no clear explanation for CK variability. Finally, it should be noted that when measuring CK in plasma or serum, total CK concentration represents both the efflux and clearance of the enzyme. Thus, interpretation of peak changes in CK should be made with caution (Ebbeling and Clarkson, 1989).

2.5.3 Growth hormone

Interactions of GH with other hormones have been noted. Therefore, it is often difficult to distinguish effects due to GH versus its influence on other hormones such as the somatomedins, testosterone, insulin-like growth factor-1 (IGF-1) and insulin-like growth factor-2 (IGF-2) (Florini, 1996; Kraemer, 1988). However, GH is considered an anabolic hormone since experimental evidence supports its associations with muscle hypertrophy, increased protein and RNA content in muscle, and increased activities of RNA polymerases, and increased ribosomes (Florini, 1987). Approximately 30 years ago, it was first reported that GH administration stimulated amino acid uptake in rat diaphragm muscle (Kostyo, 1968). Follow-up studies indicated RNA synthesis increased in muscles following GH administration in hypophysectomized rats, although the effect is delayed 18-24 hr. If GH administration is continued there is increased DNA content in the muscles by 7 days (Kostyo and Reagan, 1976). As is the case with testosterone, intensity of exercise may play a role on increased serum GH response that is typically seen following acute resistance exercise.
Takano et al. (2005) reported that three sets of low-intensity resistance exercise (20% 1 RM) performed under an occlusive stimulus (1.3 times greater than systolic blood pressure) was capable of elevating GH concentrations to near 100 times that of baseline values, but exercise at the same intensity without occlusion did not result in similar GH responses. Interestingly, these studies reported that ISC plus low-intensity exercise conditions resulted in GH responses similar to those reported during resistance exercise at much higher intensities (75% 1 RM) without occlusion (Kraemer et al., 1999).

Kraemer et al. (1991) found increased serum GH in both male and female subjects following a high volume, 10-RM protocol with 1-min rest between sets in comparison to a high-intensity protocol involving loads of 5-RM and 3-min rest between sets. The GH values observed during high-volume exercise were more than double those reported for the high-intensity protocol. Blood samples taken pre-, mid-, immediate post-, and 5, 15, 30, and 60 min after the high-volume protocol continued to show significant differences from pre-exercise levels as well as from the high-intensity protocol.

There are minimal data regarding effects of long-term resistance training on GH secretion (Kraemer, 1988). McMillan et al. (1993) found a 100% higher GH response immediately following resistance exercise in untrained versus trained subjects although both demonstrated larger responses than non-exercised controls. They felt the higher serum free fatty acids seen in the trained subjects following the exercise session would suggest a GH sensitivity response with training. Other studies have suggested that higher GH leads to increased free fatty acid mobilization, which may be related to the inhibition of glucose by GH (Yarasheski, 1994).
Chandler et al. (1994) examined serum GH responses following the resistance exercise protocol and consumption of different macronutrient beverages immediately and 2 hr post-resistance exercise. Although all groups showed an elevated serum GH response immediately post-exercise, GH responses were elevated 6 hr post-exercise for both the carbohydrate-protein beverage and the carbohydrate beverages compared to protein and water. These results were interpreted as a more favorable environment for protein synthesis with ingestion of carbohydrate-protein beverage. Thus, post-exercise nutrient consumption has the potential to modify the GH response to resistance exercise.

2.5.3.1 Acute growth hormone response to resistance exercise

The release of GH is sensitive to many physiological stimuli, including exercise (Godfrey et al., 2003). Multiple-set protocols have elicited greater GH responses than single-set protocols (Craig and Kang, 1990; Gotshalk et al., 1997). Moderate- to high-intensity, high-volume programmes using short rest periods have shown the greatest acute GH response compared with conventional strength or power training using high loads, low repetitions and long rest intervals in men (Kraemer et al., 1990, 1991, 1993; Häkkinen and Pakarinen, 1993; Bosco et al., 2000; Williams et al., 2003; Goto et al., 2003; Smilious et al., 2003). The magnitude appears dependent upon exercise selection and subsequent amount of muscle mass recruited (Kraemer et al., 1992), muscle actions used (i.e. greater response during concentric than eccentric muscle actions) (Durand et al., 2003), intensity (VanHelder et al., 1984; Pyka et al., 1992), volume (Gotshalk et al., 1997), rest intervals between sets (Kraemer et al., 1990, 1991) and training status (e.g. greater acute elevations based on individual strength and the magnitude of total work performed) (Rubin et al., 2005).
Acute resistance exercise can increase GH release in men and women of all age groups (e.g. Kraemer et al., 1993, 1999; Nicklas et al., 1995, Marcell et al., 1999, Bosco et al., 2000, Nindl et al., 2000; Takarada et al., 2000; Hymer et al., 2001). The serum GH concentration peaks at or slightly after the termination of resistance exercise and returns to baseline levels by approximately within 90 minutes post-exercise (Wilder et al., 2002). GH is an anabolic hormone, and therefore, heavy resistance exercise-induced increases in secretion of GH may be important for the process of training-induced muscle hypertrophy (Kraemer et al., 1987). However, the inter-individual GH response to acute resistance exercise is highly variable. Dependent on the protocol employed, the average peak GH concentration attained during acute resistance exercise in young men and women ranges between 5–25 µg/L. Similarly, the average peak GH concentration attained during acute aerobic exercise is also between 5–25 µg/L (Wideman et al., 2002).

Resistance exercise programmes that elicit the greatest GH response also elicit the greatest demand on anaerobic glycolysis and lactate formation as well as acute cortisol response (Roemmich and Rogol, 1997, Takarada et al., 2000, Kraemer and Ratamess, 2005). The isoforms of GH that are measurable in the circulation may be altered by muscle afferent stimulation (McCall et al., 1996). It may be possible that the nervous system has an important role in regulating GH secretion during resistance exercise and this regulatory mechanism may be sensitive to specific muscle actions used during resistance training (Kraemer et al., 2001). With progressive overload, motor unit recruitment increases (Sale, 1991). The anterior pituitary is innervated by nerve fibres from the central nervous system, i.e. the motor cortex (Ju, 1999). Therefore, hormonal responses to exercise may be triggered by the central motor command to working muscles (Galbo et al., 1987; Kjaer et al., 1987) and the responses are further modulated
by muscle afferent-pituitary axis feedback such as from cholinergic pathways and proprio and metaboreceptors in muscles (Few and Davis, 1980; Kjaer et al., 1989, 1992; Thompson et al., 1993; McCall et al., 1996; Giustina and Veldhuis, 1998; Gosselink et al., 1998).

The research suggests that women should be encouraged to engage in resistance exercise, since there may be an influential effect of growth hormone response in women attempting to develop strength and power. However, further investigations are necessary to fully describe the relationship between hormonal responses and resistance-training in women.

2.5.1 Myoglobin

Myoglobin and haemoglobin are haemeproteins whose physiological importance is principally related to their ability to bind molecular oxygen. Myoglobin is a monomeric haeme protein found mainly in muscle tissue where it serves as an intracellular storage site for oxygen. During periods of oxygen deprivation oxy-myoglobin releases its bound oxygen, which is then used for metabolic purposes.

"Each myoglobin molecule contains one haeme prosthetic group inserted into a hydrophobic cleft in the protein. Each heme residue contains one central coordinately bound iron atom that is normally in the Fe²⁺, or ferrous, oxidation state. The oxygen carried by hemepeptides is bound directly to the ferrous iron atom of the heme prosthetic group. Oxidation of the iron to the Fe³⁺, ferric, oxidation state renders the molecule incapable of normal oxygen binding" (Michael, 2006). Hydrophobic interactions between the tetrapyrrole ring and hydrophobic amino acid R groups on the interior of the
cleft in the protein strongly stabilize the heme protein conjugate. In addition a nitrogen atom from a histidine R group located above the plane of the haeme ring is coordinated with the iron atom further stabilizing the interaction between the heme and the protein. In oxymyoglobin the remaining bonding site on the iron atom (the 6th coordinate position) is occupied by the oxygen, whose binding is stabilized by a second histidine residue Michael, (2006).

2.6 Dietary supplementation

2.6.1. Creatine intake

Creatine is produced naturally by the body and helps to improve muscles' performance during exercise. Foods such as meat and fish provide much of the body's creatine and the rest is made in the body by the liver, kidneys and pancreas. It is stored in the muscles as phosphocreatine contributing to the body's energy stores used during intense exercise.

Increasing the muscle stores of phosphocreatine by taking a creatine supplement theoretically improves the ability to maintain power output during intensive exercise. Taking creatine supplements can increase muscle stores of phosphocreatine by roughly 20% on average. However, the exact increase can vary depending on the individual—ranging between 10% and 40% (Hultman et al., 1996).

Evidence suggests that creatine supplementation is probably more useful for those sports whose activities require a good anaerobic performance (Volek, 1996). Such activities include weight lifting, sprinting, football and rugby. There have been many studies to examine its effect in this area and over half of these have shown quite positive outcomes.
However, for sports requiring mainly aerobic performance there is less evidence that creatine supplementation is helpful. So, for the athletes' individuals - such as runners, cyclists and long distance swimmers - the case is not so strong. Nevertheless, a few studies have shown some improvement in performance. For example, Prevost et al. (1997) found that creatine supplements delayed the onset of muscle fatigue in endurance athletes by boosting their lactate thresholds. Studies have shown that creatine supplementation does increase body weight and also has an effect on body composition (Kreider, 1996; Clark, 1997).

In particular, creatine supplement increases muscle mass and this effect has been found in both male and female athletes. Weight increases of up to 4 kg have been reported after a period of six weeks with creatine supplementation. It is thought that this weight gain occurs because increases in the concentration of creatine in the muscles has the effect of drawing water into the muscle cells, thus increasing cell volume. This increase in volume acts as an anabolic signal which helps to reduce protein breakdown and improves the body's usage of protein. The end result is an increase in lean body tissue. Creatine supplementation in humans elevates total creatine (TC) and phosphocreatine (PC) concentration in the muscle (Willoughby and Rosene, 2001). This is usually associated with enhanced power and strength performance due to an increase in muscle mass (Vandenberghe et al., 1997). Because of these effects, creatine has been used as a therapeutic agent in neuromuscular disease and in muscular dystrophies (Tarnopolsky and Martin, 1999).

2.6.1.1 Creatine in human muscle metabolism

The weight of scientific evidence, together with subjective reports from athletes, points to an important role for creatine supplementation; In broad terms, exercise can be
considered as "short-term, high-intensity" and "prolonged, sub-maximal." For many years, athletes involved in prolonged exercise have been aware of the benefits of carbohydrate loading, but until recently there has been little in the way of dietary supplementation that has been shown scientifically to aid high-intensity exercise performance.

Most of the total creatine is restricted in skeletal muscle, with about 65% in a phosphorylated form as phosphocreatine (Casey, 1996). Phosphocreatine assumes a pivotal role in the energetics of muscle contraction during high-intensity (maximal) exercise. Muscle contraction and relaxation are fuelled exclusively by free energy liberated from the dephosphorylation of ATP, and thus muscle function depends critically on ATP availability. The ATP concentration of skeletal muscle amounts to about 24 mmol/kg dry mass (Harris et al., 1992), but ATP use during maximal, short-term, voluntary exercise (Gaitanos et al., 1993; Bogdanis et al., 1995) is such that the store of skeletal muscle ATP would be exhausted within 1–2 s of the onset of contraction without a means of resynthesizing ATP at an equally rapid rate. During such exercise, resynthesis of ATP is achieved mostly by the anaerobic degradation of phosphocreatine and glycogen (Hultman et al., 1991) resulting in an increasing rate of ATP production close to 15 mmol ⋅ kg dry mass⁻¹ ⋅ s⁻¹ during the first 6 s of maximal exercise (Gaitanos et al., 1993). In this manner the concentration of skeletal muscle ATP can be maintained to some degree during both a single bout (Boobis et al., 1987; Nevill et al., 1989) and repeated bouts (Gaitanos et al., 1993; McCartney et al., 1986) of short-term maximal exercise.
Therefore, these findings raise the question of just how important is the availability of phosphocreatine and glycogen to ATP resynthesis, and thus to performance, during high-intensity exercise. It is well documented that in mixed muscle the glycogen concentration is still relatively high at the end of this type of exercise (McCartney et al., 1986).

2.6.1.2 Creatine supplement and resistance Exercise

The early studies on Cr supplementation in the 1990s in humans focused on exercise performance, which served as a basis for subsequent scientific research and applications. As mentioned earlier, supplementation increases intramuscular t-Cr content. The increase in Cr in young healthy males has been shown to anaerobic exercise performance by increasing power output (Earnest et al., 1995), muscular strength and work (Vandenberghe et al., 1997; Volek et al., 1999), and muscle fibre size (Volek et al., 1999). Studies have also been performed on young healthy females, middle-aged males (30–60 years) of age, and the elderly (60 years) of age. Both females (Vandenberghe et al., 1997) and middle-aged males (Smith et al., 1998) benefited from Cr supplementation, but the elderly did not show an exercise performance enhancement (Rawson et al., 1999; Rawson and Clarkson, 2001). The lack of an effect in the elderly may be explained by changes in transporters density related with aging and decreased Cr uptake. The American College of Sports Medicine recently had a discussion group on the physiological and health effects of Cr supplementation (Terjung et al., 2000). Performance has been enhanced in swimming, all-out cycling, sprinting, repeated jumping, and resistance training (Juhn and Tarnopolsky, 1998). The greatest improvements in performance have been found in series, high-power output exercises and the final exercise bouts of a series, those performance that are repetitive in nature.
and those of high-energy output, which would stress the PCr system, would likely benefit from Cr supplementation (Terjung et al., 2000).

Although the majority of studies have been in men, a significant amount of research indicates women are also responsive to creatine supplementation (Volek and Rawson, 2004). Short-term creatine supplementation enhanced high intensity exercise performance in women (Eckerson et al., 2004; Tarnopolsky and MacLennan, 2000) but in contrast to the situation in men, suggesting other mechanisms are responsible for the long term effects creatine on lean body mass (Volek and Rawson 2004). Studies have examined strength and body composition responses during a resistance training program indicate women respond favourably to creatine supplementation (Brenner et al., 2000; Vandenberghe et al., 1997).

**2.6.2 Carbohydrate intake**

Carbohydrate (CHO) is a broad term used to describe sugars involved in nutrition. CHO or sugar is one of the body's primary sources of energy for life in general, as well as during exercise. In our diet, CHO takes two forms, simple CHO (sugars) and complex CHO (starches). Following digestion, CHO enters the blood stream in the form of glucose where it circulates until it is stored in the muscle or the liver as glycogen first has to be transported by the blood, and taken up by the muscle before it can be oxidised (Jeukendrup, 1997). Gluconeogenic substrates such as lactate, glycerol and amino acids can be converted to glucose in the liver and so indirectly serve as an energy substrate. Some simple CHO, sugars like glucose and fructose, are found naturally in many fruits. Other simple CHO are sucrose (common table sugar) and lactose (milk sugar). Starches,
which are found in grains and vegetables, are "complex" because they are simple CHO related together into long molecules (Kreider, 2003).

The total amount of muscle glycogen of an 80 kg man is about 400 grams in which each gram of CHO stored also retains about 3 g of water, further decreasing the efficiency of CHO as an energy source. Liver glycogen represents about 80-100 g. The total amount of plasma substrates (glucose and lactate) is about 20 g. Expressed in terms of energy; the body CHO stores represent approximately 8000 kJ. In comparison with this, fat stores are large and theoretically they could provide energy for days whereas the glycogen stores can become depleted within 60-90 min.

CHO are important to athletes because they can produce energy at a faster rate for exercise if they use CHO instead of fat. Basically, CHO is a more efficient fuel than fat. Unfortunately, CHO in the muscles and the liver has limited storage possible, whereas the body's fat supplies are general, CHO is also necessary for efficiently burning fat. While the oxygen energy system is designed for endurance, an insufficient supply of the optimal fuel, CHO, can limit performance (Costill et al., 1992; Zachwieja et al., 1993). Muscular fatigue is associated with severe muscle glycogen store depletion (Akermark et al., 1996). Research has found that by consuming a high CHO diet (60-95% or 7-10g CHO/kg/d) a few days prior to exercise/competition while reducing exercise intensity causes muscle glycogen stores to become super-compensated (Ward, 1996; Goforth et al., 1997). Elevated muscle glycogen stores do not influence muscle power, strength or performance during short-term exhaustive exercise (anaerobic modes) but do prolong performance time during exercise (aerobic modes) at high intensities (Goforth et al., 1997; Ward, 1996; Akermark et al., 1996; Walberg-Rankin, 1995). Consuming a meal
high in easy to digest CHO's 3-4 hrs before exercise improves endurance running and cycling capacity (Williams and Nicholas, 1998).

2.6.2.1 Metabolic responses to carbohydrate supplement

The majority of well-controlled studies show that, compared with men, women rely more on fat and less on carbohydrate (CHO) oxidation during aerobic exercise performed at the same relative strength. The differences in the metabolic response to exercise could have implications for nutritional recommendations and physical performance for male and female athletes (Tarnopolsky, 2000, 2003). In this respect, the metabolic and performance responses to the ingestion of CHO during exercise have been investigated over the last 25 years, although the amount to which sex directly influences these responses has not been methodically studied.

CHO supplementation during exercise increases endurance exercise capacity and performance in females (Bailey et al., 2000; Campbell et al., 2001). This effect is largely qualified to increased CHO oxidation and protection of euglycemia during exercise, particularly as exercise duration increases and endogenous CHO stores become low (Coggan and Coyle, 1989; Coyle et al., 1986). In addition, in men (Bosch et al., 1994; Jecukendrup et al., 1999; McConell et al., 1994) and women (Campbell et al., 2001), CHO ingestion during exercise reduces and replaces hepatic glucose production, which is indicative of liver glycogen careful. It is generally accepted that CHO feeding does not affect the rate of muscle glycogen utilization during exercise (cycling) in males (Bosch et al., 1994; Febbraio et al., 2000; McConell et al., 1994). In contrast, it has been reported that CHO ingestion during cycling exercise could reduce muscle glycogen consumption during exercise in females (Campbell et al., 2001).
In one study, muscle glycogen stores were unchanged in women in response to a 4-day high-CHO diet (HCD; ~75% CHO) compared with a moderate-CHO diet (MD; ~57% CHO), whereas the men increased muscle glycogen content by ~40% after the high-CHO regimen (Tarnopolsky, et al., 1995). Furthermore, only men increased cycle time to exhaustion (80–85%VO2 max, after 1 h at 70–75% VO2 max) in response to the dietary CHO-loading regimen. Therefore, women may not be able to super-compensate muscle glycogen stores before exercise and increase exercise performance in response to increases in dietary CHO.

The female athletes engaged in the above studies were tested during the follicular phase (FP) of the menstrual cycle when circulating reproductive hormones are low (Horton, et al., 1998; Tarnopolsky, et al., 1999; 2000). Hackney et al. (1994) found a 13% higher resting muscle glycogen content in the luteal phase (LP) compared with the FP of the menstrual cycle when they controlled for diet and exercise in the 36 h before muscle glycogen sampling. In contrast, Nicklas et al., (1989) studied moderately trained women and reported no significant difference in muscle glycogen content between phases of the menstrual cycle after a glycogen-depleting exercise bout and 3 days of a controlled diet (~56% CHO) with no exercise. However, a significantly greater amount of muscle glycogen repletion occurred during the 3 days in the LP vs. the FP [379 ± 20 vs. 313 ± 25 mmol/kg dry muscle]. These studies suggest that glycogen synthesis may be increased during the LP of the menstrual cycle. Therefore, the testing of women during the LP of the menstrual cycle may show that female athletes are able to super-compensate muscle glycogen stores and improve exercise performance.
It has been recommended that athletes ingest CHO following exercise in order to enhance glycogen resynthesis, promote an anabolic hormonal environment, enhance PRO synthesis, and/or lessen the immuno-suppressive effects of intense exercise (Campbell, et al., 2007; Conley and Stone, 1996). The type of CHO ingested is an important consideration because the glycemic index of a CHO may enhance glycogen storage and/or anabolic responses to exercise by promoting a greater glucose and insulin response (Burke and Hargreaves, 1993; Burke et al., 1996). These recommendations are based on findings that ingestion of CHO following exercise increases insulin levels promoting glycogen restoration (Roy, Tarnopolsky, 1997; Zawadzki et al., 1992). Additionally, increasing insulin levels following exercise optimizes an anabolic hormonal environment and can serve as a potent stimulator of PRO synthesis pathways (Tarnopolsky et al., 1999; 1995).
2.7 Summary.
In summary, several authors (Baker et al., 1982; Hunter and Enoka, 2001) have studied the metabolic and physiological responses difference between males and females and have shown important metabolic and hormonal differences in response to resistance exercise. Even though muscular strength is an important characteristic in achieving optimum sports performance, the majority of previous studies on the effects of resistance exercise on indices related to muscular strength along with biochemical and hormonal responses have been carried out in male populations. Therefore, there is a need to examine the characteristics of fatigue and recovery responses to heavy resistance exercise in females in terms of isometric force and blood analysis variables. There is also a need to examine the effect of dietary supplementation on the neuromuscular and biochemical responses to heavy resistance exercise.
CHAPTER 3

THE RELIABILITY OF ISOMETRIC FORCE ON FEMALES
3.1. The between-day reliability of maximal voluntary isometric contraction and rate of force development measures

3.1.1 Introduction

Reliability refers to the precision of values of a test, assay or other measurements in repeated trials on the same individuals. Greater reliability implies better precision of single measurements and better tracking of changes in measurements in research or practical settings. In sports science, a frequently assessed variable is the maximal voluntary isometric contraction (MVC) which is typically defined as the maximal muscle force that a highly motivated subject is able to produce voluntarily under particular contractile conditions. Another measure employed is the rate of force development (RFD). Although some researchers have examined the reliability of the technique for the assessment of MVC, few studies have been undertaken using women (Constance et al., 1994).

Several studies have established a high reliability of maximal force measurements and intra-individually reproducible values in men for the voluntary activation of the single quadriceps during isometric knee extension (Morton et al., 2005). The MVC and RFD have been shown to be reliable (Viitasalo et al., 1980; Bemben et al., 1992). For example, Bemben (1992) reported correlations of 0.92 to 0.98 between test scores administered on successive days for a variety of muscle groups. Maximal isometric strength tests generally have high test-re-test reliability. Viitasalo et al. (1980) reported a high correlation between repeated maximal isometric trials which were conducted with a 5-min rest interval. Similar reliability values were achieved by Bemben et al. (1992) for subjects of differing ages, and greater reliability was achieved when several practice
tests were provided prior to data collection and data collected over a 2-day period and the values averaged.

As rehabilitation training programs aim at enhancement of functional performance, it is also important to know what development in values indicates a good physical performance and therefore a better training status. Several scientific studies have dealt with reliability issues of muscle function variables during knee extension (McDonnell et al., 1987; Feiring et al., 1990; Oldham and Howe, 1995; Behm et al., 1996; Norregaard et al., 1997; Pincivero et al., 1997; Oeffinger et al., 2001; Dauty and Rochcongar, 2001; Larsson et al., 2003; Horemans et al., 2004; Todd et al., 2004; Symons et al., 2004; Mathur et al., 2005; Morton et al., 2005). Several of them have established a high reliability of maximal voluntary force measurements Intra-Class Correlation coefficients (ICC > 0.8) for men in the single quadriceps during isometric (Oldham and Howe, 1995; Behm et al., 1996; Horemans et al., 2004; Symons et al., 2004; Morton et al., 2005) and isokinetic knee extension (Feiring et al., 1990; Pincivero et al., 1997; Dauty and Rochcongar, 2001; Larsson et al., 2003; Symons et al., 2004; Todd et al., 2004).

One study presented data on woman, for isometric function of six muscle groups (Constance et al., 1994). Repeated measures of maximal force, maximal rate of force increase, total impulse, time to maximal force, average rate to maximal force, and time between 90% maximal force and maximal force were obtained on the finger flexors, thumb extensors, forearm flexors, forearm extensors, dorsiflexors and plantar flexors. Repeated measures ANOVA yielded significant day effects for the finger flexors maximal rate, dorsiflexors (maximal force, average rate to maximal force), and plantar flexors, and significant trial effects for the forearm extensors and plantar flexors.
(maximal force, average rate to maximal force). Intra-class correlation coefficients (ICC) varied among muscle groups and parameters of muscle function, ranging from ICC = 0.30 to 0.94. Estimates of the component variances also varied among muscle groups and parameters of muscle function. Coefficients of variation ranged from 7.7% to 44.1%.

Morten et al. (2001) determined reliability measurements of maximal voluntary contractions (MVC) in females during isometric back flexion, back extension, shoulder elevation, shoulder abduction and handgrip. All tests showed reasonable reliability at group level judged by 95% confidence intervals, although the variation was wide according to the calculated limits of agreement at the individual level. If this variation is not taken into consideration the tests can be of limited use at the individual level. Finally, this study clearly showed the correlation coefficients as a single number to estimate reliability.

The few studies reviewed on the reliability of female strength measures suggests that reliability, while good, is also variable in females. In particular, no study exists that has examined the reliability of isometric quadriceps strength with single and both legs together for females. Therefore, the aim of this study was to establish the between-day reliability of the testing protocol for maximum force isometric contraction and rate of force development with similar procedure for both legs and each leg separately when using an isometric chair with female subjects.
3.1.2 Methods

3.1.2.1. Subjects and study design

Subjects. Eight healthy females volunteered to participate in the study. All subjects were medically screened (Appendix 1) to determine their health and exercise habits prior to exercising lower body muscles. Each individual gave written informed consent to participate in the study, which was approved by the Ethics Committee of Liverpool John Moores University. The mean (±SD) age, height and body mass of the subjects were 25±2 years, 1.78 ± 0.04 m and, 58.3 ±3.11 kg, respectively.

Experimental design. The knee extensor muscle strength of each leg was obtained using a strain gauge dynamometer attached to an adapted Lido Dynamometer chair (Loredan, USA, see figure 3.1). The subject was seated with the trunk vertical with 90° flexion at the hip and knee. To prevent extraneous body movements, Velcro straps were applied tightly across the thorax and distal thigh. The subject was seated in the adjustable chair and strapped at the waist and chest to maintain muscle length and prevent substantial use of the hip extensors. Quadriceps muscle force was measured from the ankle where the attachment was connected to a strain gauge (previously calibrated with known weights) by a metal rod. A separate device was used for each leg. The subject was instructed to exert maximal force as rapidly as possible and maintain that force for 6 s while verbal encouragement was given. Force data from the strain gauge were amplified and collected on-line by computer via a 12 bit analogue-to-digital converter (figure 3.1). Data were recorded for MVC, and smoothed used a Hanning 3-point filter. The RFD was calculated as the first differential of the smoothed force data. Data were collected on 3 successive days at the same time of day (Figure 3.2).
Figure 3.1 Isometric strength testing chair and measurement equipment.

Figure 3.2 The experimental design for study one (A).
3.1.2.2. Measured variables

3.1.2.2.1 Body mass (BM)

Body mass of each subject was measured to the nearest 0.1 kg using calibrated balance scales (Cranlea and Company, Bournvile, Birmingham, UK), in light clothing, and without jacket, and footwear. The subject stood on the scales with their feet together, arms hanging loosely by their sides and head facing forward. Body mass was measured in the first session of each experiment.

3.1.2.2.2 Body height (BH)

The height of each subject was measured without shoes using a calibrated stadiometer (Cranlea and Company, Bournvile, Birmingham, UK). Measurements were recorded to the nearest 0.5 cm. The subject's head was held straight looking forward with their line of vision parallel to the floor.

3.1.2.3 Determination of one repetition maximum (1-RM)

Two familiarisation sessions were designed to habituate subjects with the testing procedures and laboratory environment. Subject's 1-RM was required for the exercise used for the exercise component of the protocol, used in later studies. The main aims of these sessions were to familiarise subjects with resistance exercise using weight-training equipment. Subjects reported to the weight training room in their appropriate sporting attire. During the familiarisation sessions, it was ensured that all subjects performed the correct technique for all exercises prior to taking part in the main testing trials. Following familiarisation, subjects were asked to report to the laboratory for an additional session designed to determine 1-RM.
Chapter 3- The between-day reliability

The leg 1-RM muscular strength tests were used to evaluate each subject’s maximal level of strength for each of the six exercises which was used in later studies in order to determine the loads (percentage of 1-RM) used. The 1-RM has been defined as the weight that can be successfully lifted no more than once, through a specific range of movement (Sale, 1991).

During the leg-press the subjects lowered the weight to a flexed knee angle of 90° (Hoeger et al., 1990), measured manually using a goniometer. The subjects were given two standardized sub-maximal warm-up trials with descending (10 and 5) repetitions with progressively heavier loads before performing a series of one-repetition trials with progressively heavier weights to determine their 1-RM (Humphries et al., 1999; Sale, 1991). The weight was increased in increments until the subject could not complete the lift (Murphy and Wilson, 1997). The heaviest weight lifted across the trials was used for estimating 80% of each subject’s 1-RM in each exercise (Feigenbaum and Pollock, 1997; Hoeger et al., 1990) and corresponding 10-12-repetition for each strength-exercise (Figure 3.3).
Prior to commencing the test, the subjects warmed up on a cycle ergometer for five-minutes at a workload of 1 watt per kilogram (kg) of gross-body-mass and performed light stretches of the major leg muscle groups (quadriceps, hamstrings, and calves). Isometric assessments are generally performed to quantify various force-time characteristics such as the MVC and RFD. Subsequent to each strength test session, the subjects completed an identical progression of MVC (right and left leg separately then together) at 90° of knee flexion on a custom-built testing chair. Ninety degrees of knee flexion was chosen due to its association with force production during the down stroke of the isometric chair pedal action (Cavanagh and Sanderson, 1986). The subjects were given a series of progressive warm-up trials before performing the three MVCs with a 1–2 min rest period between each contraction (Bennet and Stauber, 1989). The subjects performed the contractions in response to a verbal command and held the contractions for a period of 6 seconds.
3.1.3. Statistical analysis

In order to evaluate the between-day repeatability, the averages of the first and second day, and second and third day. The CV was calculated to represent intra-subject variation between the two testing sessions. A one-way ANOVA with repeated measures was used with Tukey's post hoc test of honestly significant differences to assess the systematic bias between tests with statistical significance set at $P<0.05$. The limits of agreement (LOA) were determined (Bland and Altman, 1986, 1999). The data for all subjects were expressed as mean ± standard deviations, unless otherwise stated.

3.1.4. Results

The mean (±SD) values of MVC and RFD for both legs together and each leg separately Figure 3.4. The one-way ANOVA with repeated measures revealed a significant trial effect across days determined between day1 vs. day2 and day2 vs. day3. The LOA comparisons were made for each variable MVC and RFD and for both legs and each leg.

**Figure 3.4** Mean of three trials for MVC and RFD (units= N) of both legs, right and left leg (D=days).
3.1.4.1. Reliability of MVC for both legs together and right and left leg separately.

The day-to-day measurements of MVC are presented in Figure 3.4. The three days data for MVC were approximately similar. There was a small systemic bias between days for both legs, right leg and left leg. The one-way ANOVA with repeated measures established that there was significant main effect when the data was compared across days for both legs and left leg ($F_{2,14} = 5.77$, $P <0.05$), and for the right leg ($F_{2,14} = 9.12$, $P <0.01$). The reliability of the measured variables had wide-ranging levels of consistency, with CV’s across days ranging from 5.4% to 28.8% (Table 3.1). The 95% limits of agreement showed good reliability between-days, The variables displayed a relatively low-degree of systematic bias on the both legs, as the vast majority of the observed variance was due to random error (Table 3.2).

Table 3.1 Between-days limits of agreement for MVC (N).

<table>
<thead>
<tr>
<th>DAYS</th>
<th>Mean (1)</th>
<th>Mean (2)</th>
<th>Grand mean</th>
<th>Mean diff (Bias)</th>
<th>SD diff *1.96 (error)</th>
<th>Mean CV as %</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Both legs</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1 vs. D2</td>
<td>249.2</td>
<td>224.5</td>
<td>236.8</td>
<td>24.7</td>
<td>12.9</td>
<td>5.4</td>
</tr>
<tr>
<td>D2 vs. D3</td>
<td>224.5</td>
<td>226.8</td>
<td>225.7</td>
<td>-2.3</td>
<td>14.5</td>
<td>6.4</td>
</tr>
<tr>
<td><strong>Right leg</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1 vs. D2</td>
<td>304.7</td>
<td>266.0</td>
<td>285.4</td>
<td>38.7</td>
<td>27.0</td>
<td>9.5</td>
</tr>
<tr>
<td>D2 vs. D3</td>
<td>266.0</td>
<td>267.4</td>
<td>266.7</td>
<td>-1.4</td>
<td>76.8</td>
<td>28.8</td>
</tr>
<tr>
<td><strong>Left leg</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1 vs. D2</td>
<td>287.3</td>
<td>253.8</td>
<td>270.6</td>
<td>33.5</td>
<td>46.4</td>
<td>17.1</td>
</tr>
<tr>
<td>D2 vs. D</td>
<td>253.8</td>
<td>264.7</td>
<td>259.3</td>
<td>-10.9</td>
<td>15.1</td>
<td>5.8</td>
</tr>
</tbody>
</table>

[Grand mean= (1+2)/2, Mean diff= (1-2), %CV= SD diff/Grand mean *100], 1.96 = the 95th percentile.
3.1.4.2 Reliability of RFD for both legs together and right and left leg separately.

The day-to-day measurements of RFD are presented in Figure 3.5 which were very similar across days. The one-way ANOVA with repeated measures established that there was significant main effect when the data was compared across days for just the both legs condition ($F_{2, 14} = 19.97, P < 0.01$), and a significant interaction ($P \leq 0.01$) for day*time, but no significant difference across days for both left and right leg ($F_{2, 14} = 3.48, P > 0.05$). The reliability of the measured variables had wide-ranging levels of consistency, with CV's across days ranging from 0.1% to 7.4% (Table 3.2). The 95% limits of agreement showed good reliability between-days, The variables displayed a relatively low-degree of systematic bias on the both legs, as the vast majority of the observed variance was due to random error (Table 3.2).

Table 3.2 Between-days limits of agreement for RFD (N/s).

<table>
<thead>
<tr>
<th>DAYS</th>
<th>Mean (1)</th>
<th>Mean (2)</th>
<th>Grand mean</th>
<th>Mean diff (Bias)</th>
<th>SD diff *1.96 (error)</th>
<th>Mean CV %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Both legs</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1 vs. D2</td>
<td>786.1</td>
<td>709.8</td>
<td>747.9</td>
<td>18.5</td>
<td>24.1</td>
<td>3.2</td>
</tr>
<tr>
<td>D2 vs. D3</td>
<td>709.8</td>
<td>720.3</td>
<td>715.1</td>
<td>19.1</td>
<td>11.4</td>
<td>1.6</td>
</tr>
<tr>
<td>Right leg</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1 vs. D2</td>
<td>856.3</td>
<td>784.6</td>
<td>820.5</td>
<td>71.7</td>
<td>17.4</td>
<td>2.1</td>
</tr>
<tr>
<td>D2 vs. D3</td>
<td>784.6</td>
<td>811.8</td>
<td>798.2</td>
<td>-27.2</td>
<td>0.8</td>
<td>0.1</td>
</tr>
<tr>
<td>Left leg</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1 vs. D2</td>
<td>852.2</td>
<td>807.8</td>
<td>829.9</td>
<td>44.5</td>
<td>61.6</td>
<td>7.4</td>
</tr>
<tr>
<td>D2 vs. D3</td>
<td>807.8</td>
<td>807.8</td>
<td>807.8</td>
<td>0.01</td>
<td>0.04</td>
<td>4.9</td>
</tr>
</tbody>
</table>

[Grand mean= $(1+2)/2$, Mean diff= $(1-2)$, %CV= SD diff/Grand mean *100], 1.96 = the 95th percentile.
3.1.5 Discussion and conclusion

The aim of this study was to assess the reliability of MVC and RFD measurements for knee extension using an isometric chair system in females across three days for each leg separately and for both legs together. It was found that the mean between-days LOA for MVC for both legs was less than the right leg or left leg separately, and that the left leg was less than the right leg. Although previous studies (e.g. Tillman et al., 2004) have used either the dominant leg or each leg separately for MVC measurements, the MVC data for both legs were more reliable than MVC measured separately for each leg and for the RFD measurements, the typical error in all three conditions (both legs, right leg and left leg) were similar and so there was no difference in their reliability when the three days data were compared.

The reliability of this measure was rather good and ranged approximately (CV = 2.4%, 1.1%, 6.2%) for both, right and left leg evaluating the reliability of MVC using the LOA method indicates that a small amount of systematic bias (-2.3, -1.4, -10.9N) was present between the day 2 and day 3 for all conditions (Table 3.1), and so it is recommended that the MVC measurements are made using both legs instead of using each leg independently. However, for the RFD measurements, the typical error in all three conditions (both legs, right leg and left leg) were similar and so there was no difference in their reliability when the three days data were compared.
time separates testing sessions, the reliability of isometric strength is compromised (Kollmitzer, et al., 1999). The findings do not support this previous observation as excellent reliability in assessing MVC and RFD was observed even when testing sessions were separated by a relatively long-time period as in the between-days reliability data (Table 3.1 and 3.2).

However the systematic bias was significant when compared data between days for both legs and left leg ($P < 0.05$) and was either right leg ($P < 0.01$). The LOA measure of absolute reliability and represent the range within which a participant's difference score ($D_1, D_2, D_3$) would occur most of the time (Hopkins, 2000). Therefore, assuming that the errors are normally distributed, 95% of the participant's difference scores should lie within plus or minus two standard deviations of the mean of the difference scores (Portney and Watkins, 2000). Like CV the limits of agreement should be presented as a ratio because of the heteroscedastic data (Atkinson and Nevill, 1998). Data from the current study revealed LOA values for both leg, right and left leg ranging from 12% to 15%, 27% to 76% and 15% to 46.4. Consequently, any difference between the two tests of the previously mentioned isometric measures should differ by no more than the corresponding percentages above and below the mean bias. The percentages above and below the mean bias for all measures observed were again larger than desired.

**In conclusion**, the results of the present study demonstrated that the reliability of measurement presented the relative and absolute reliability of different methods and subsequent variables commonly used to asses the isometric strength muscle of humans. Using to measures both legs together is acceptable and that the method of muscle strength evaluation using an isometric chair is a reliable method for testing female
muscle strength. In this pilot study it was found that using both legs to measure isometric contraction force was more reliable for between-days measurements than using one leg.
3.2. The within-day reliability of MVC and RFD measurements and the effect of fatigue induced by resistance exercise.

3.2.1 Introduction

Fatigue is a complex, multi-factorial phenomenon and has been associated with impairments at a number of sites, ranging from central activation to myofilament interaction (Fitts, 1994; Stephenson, 1998). Further complication arises from the fact that fatigue is task specific, in that for a given task one particular site or mechanism may be more or less responsible for the decline in muscle performance (Enoka and Stuart, 1992). Fatigue after repeated heavy resistance loading results in momentary decreases both in voluntary activation and force production, but their magnitudes may be specific to the type of exercise loading (Komi and Viitasalo, 1977; Hakkinen, 1994).

Some researches suggest that a combination of resistance machines and free weights are recommended for beginners, while advanced exercisers should emphasize using free weights (Brill et al., 1998). It has been recommended that during weight training, a) large muscle groups should be challenged before smaller muscle groups, b) multiple-joint exercises should precede single-joint exercises, c) higher intensity exercises should be done before lower intensity, and (d) the training should be started with slow to moderate velocities. Moderate velocities are recommended for intermediate exercisers, while advanced exercisers can use a continuum of slow to fast velocities (Kraemer, 2002). Therefore a fatigue indicating protocol should follow these general recommendations.

Consequently, the first pilot study suggested that MVC and RFD measurements for knee extension using an isometric chair system in females could be used to quantify the improvement following resistance exercise from one day to another during studies.
Therefore, the purpose of this study was to establish exercise volume and intensity to produce fatigue effect defined as 40% reduction in measured force variables, and as a consequent of the protocol used, to assess the within-day reliability of muscle strength measures in females.

3.2.2 Methods

Eight female subjects, as used in the pilot study 1, took part in this study. The subjects were required to attend the laboratory in the same day. The within-day repeatability of the measurements for MVC and RFD was calculated by comparing the test results from session 1 and 2, where the time interval between sessions was one hour and the subjects were allowed on three sessions to rest. Session 2 was followed by the exercise protocol. Measurement for session 3 was conducted within 30-min of the completion of the exercise protocol. The fatigue effect of the resistance exercise was established by comparing between sessions 2 and 3. The apparatus and measurement protocol designed in first study one were used with both legs, right and left leg separately. The subjects were instructed to exert their maximal force as rapidly as possible and maintain that force for 6 s. Verbal encouragement was given to each subject during exercise. On each testing session subjects were required to perform three maximal isometric contractions. The time interval between maximal efforts was 6 s.

Figure 3.5 The experimental design for study one (B)
For the resistance exercise, subjects performed three sets of six different weight exercises (leg curls, dumbell lunges, barbell squats, knee extensions, squat exercise, leg presses) at an intensity corresponding to 60% of 1RM (8-10 repetitions). The resistance exercises used was designed to produce fatigue in the quadriceps. A one-minute rest period was allowed between exercises and a 3-min rest period allowed between sets. The duration of the session was approximately 45 min.

3.2.3 Familiarization
Subjects underwent extensive familiarization prior to participating in the study. During the resistance exercise session, the subjects were introduced to and familiarised with the performance of the MVC. Approximate loads for the 1-RM were determined during the familiarization session (Section 3.1.2.3).

3.2.4 Statistical analysis
In order to evaluate within-day repeatability, 95% Limits of Agreement was determined between the first and the second sessions (Bland and Altman, 1999). The CV was calculated to represent intra-subject variation between the two testing sessions. The mean values of MVC and RFD in session two and three was compared using a t-test with a level of significance of P<0.05. Data are presented as means ± SD, unless otherwise stated.
3.2.5 Results

The within-day repeatability of measurements and the effect of fatigue was evaluated using the data in Table 3.4.

![Figure 3.6 Mean of three sessions for MVC (N, N/s) per session of both legs, right and left leg. (S= Session)](image)

3.2.5.1 Reliability of MVC for both legs together, right and left leg.

The variability in measurements of MVC across sessions is presented in Figure 3.6. The data for MVC were approximately similar when compared between sessions 1 and 2. The reliability of the measured variables had wide-ranging levels of consistency, with CV's between sessions for both legs condition (0.6%), and other two conditions were 6.7% to 13.7% for right and left leg (Table 3.3). There was little bias between sessions for both legs, right leg and left leg which ranged from (1.3 to 32.8). The 95% Limits of Agreement showed good within-day reliability for both legs (Table 3.3).
Table 3.3 Mean value, bias and random error for MVC.

<table>
<thead>
<tr>
<th></th>
<th>S1</th>
<th>S2</th>
<th>Grand mean</th>
<th>Mean diff (Bias)</th>
<th>SD diff 1.96 (Error)</th>
<th>% CV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Both Legs</td>
<td>322.3</td>
<td>321.01</td>
<td>321.7</td>
<td>1.3</td>
<td>1.8</td>
<td>0.6</td>
</tr>
<tr>
<td>Right Leg</td>
<td>349.5</td>
<td>316.7</td>
<td>333.1</td>
<td>32.8</td>
<td>45.5</td>
<td>13.7</td>
</tr>
<tr>
<td>Left Leg</td>
<td>326.2</td>
<td>310.8</td>
<td>318.5</td>
<td>15.5</td>
<td>21.4</td>
<td>6.7</td>
</tr>
</tbody>
</table>

[Grand mean = (1+2)/2, Mean diff = (1-2), %CV = SD diff/Grand mean * 100].

3.2.5.2 Reliability of RFD for both legs together and right leg, left leg.

The variability in measurements of RFD across sessions is presented in Figure 3.6. The RFD data were approximately similar when compared between sessions 1 and 2. The reliability of the measured variables had wide-ranging levels of consistency, with CV's between sessions for both legs condition (3.0%), and other two conditions were 4.4% to 5.3% for right and left leg (Table 3.4). There was little bias between sessions for both legs, right leg and left leg which ranged from (1.3 to 32.8). The 95% Limits of Agreement showed good within-day reliability for both legs (Table 3.4).

Table 3.4 Mean value bias and random error of RFD.

<table>
<thead>
<tr>
<th></th>
<th>S1</th>
<th>S2</th>
<th>Grand mean</th>
<th>Mean diff (Bias)</th>
<th>SD diff 1.96 (Error)</th>
<th>%CV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Both Legs</td>
<td>1057.0</td>
<td>1034.7</td>
<td>1045.8</td>
<td>22.3</td>
<td>31.0</td>
<td>3.0</td>
</tr>
<tr>
<td>Right Leg</td>
<td>1601.5</td>
<td>1551.7</td>
<td>1576.6</td>
<td>49.8</td>
<td>69.0</td>
<td>4.4</td>
</tr>
<tr>
<td>Left Leg</td>
<td>1315.3</td>
<td>1266.3</td>
<td>1290.8</td>
<td>49.0</td>
<td>67.9</td>
<td>5.3</td>
</tr>
</tbody>
</table>
3.2.5.3 T-Test between session 2 and 3.

Analysis using a t-test on the data in Table 3.5 showed that fatigue induced by exercise decreases the MVC measured in both legs \( [t_7 = 10.36, P < 0.001] \), the right leg \( [t_7 = 11.95, P < 0.001] \) and the left leg \( [t_7 = 7.21, P < 0.001] \) leg separately. Data analysis revealed that there was a significant effect of fatigue induced by exercise on RFD in both legs \( [t_7 = 14.94, P < 0.001] \), in the right leg \( [t_7 = 5.76, P < 0.001] \) and in the left leg \( [t_7 = 7.21, P < 0.001] \).

Table 3.5 Means and standard deviations for Session 2 and 3 for MVC (N) and RFD (N/s) of both legs and right and left legs.

<table>
<thead>
<tr>
<th>TRIAL</th>
<th>(Mean of both leg) MVC</th>
<th>RFD</th>
<th>(Mean of right leg) MVC</th>
<th>RFD</th>
<th>(Mean of left leg) MVC</th>
<th>RFD</th>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Session 2</td>
<td>329.7±47.5</td>
<td>1133.6±189.3</td>
<td>344.5± 74.3</td>
<td>1525.3±389.8</td>
<td>328.0±65.3</td>
<td>1297.7±321.9</td>
</tr>
<tr>
<td>1</td>
<td>333.3±61.9</td>
<td>1122.2±157.3</td>
<td>335.4±80.7</td>
<td>1622.7±357.9</td>
<td>324.7±60.0</td>
<td>1234.8±308.5</td>
</tr>
<tr>
<td>2</td>
<td>302.9±60.7</td>
<td>982.9±212.1</td>
<td>300.2±55.7</td>
<td>1507.2±354.6</td>
<td>327.3±58.8</td>
<td>1254.0±362.7</td>
</tr>
<tr>
<td>3</td>
<td></td>
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<td>211.9±62.8</td>
<td>687.7±239.1</td>
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</table>
3.2.6 Discussion and conclusion

The aim of the second pilot study was to assess the effect of fatiguing resistance exercise and to establish the within-day reliability of MVC and RFD in females. There was no change found from session 1 to session 2 in MVC and RFD within a one-hour period. The reliability of within-day measurements was assessed using the 95% LOA. It was therefore concluded that there was no 'learning effect' on the measured variables or fatigue produced by the test protocol itself. The data therefore indicated that well familiarised subjects are competent at producing their perceived maximal force during a within-day protocol.

The reduction in force following the exercise session was due to some form of peripheral fatigue. These findings are in agreement with that of Bigland-Ritchie et al. (1986) who reported a reduction in force following exercise. The change between the second and third session is in agreement with reports of the effect of a high-intensity strength training session in men. Häkkinen (1992) reported a significantly greater reduction in maximal strength along with a significant reduction in RFD.

While the relative reliability of MVC was rather good for both, right and left leg, evaluating the reliability of MVC using the LOA method indicated that a small amount of systematic bias was present between two sessions for all conditions (Table 3.1). However, for the RFD measurements the typical error in all three conditions (both legs, right leg and left leg) were similar and there was no difference in their reliability, when the two session's data were compared. Therefore it is recommended that the MVC and RFD measurements are made using both legs instead of using each leg independently.
These findings indicated that the level of fatigue in MVC following the resistance exercise program was around 25.4%, 34.1% and 27% for both legs, right and left leg respectively. Although data on resistance exercise session showed significant reduction in RFD for both legs, right and left leg of 18.7%, 43.8% and 39.7%, respectively. The force reduction achieved by the exercise was around 25-30% at an intensity corresponding to 60% of 1-RM (8-10 repetitions) which did not reach the required 40% reduction as described on male subjects by Fell (2002). Therefore, the intensity will need to be increased for future study.

In conclusion, the findings of the present study showed good within-day reliability. Future studies will need to use a more intense exercise programme. Although all previous studies have used one leg or the dominant leg (Tillman et al., 2004) for measuring MVC or RFD, in these pilot studies it was found that using both legs for studying the MVC was more reliable within same day and between-days contractions force measurements than using one leg.
CHAPTER 4

NEUROMUSCULAR FATIGUE AND RECOVERY FROM RESISTANCE EXERCISE IN FEMALES
4.1. Introduction

Fatigue is one of the major factors limiting human performance during sporting activity, in everyday life and in any occupation that involves physical effort. A great deal is known about the metabolic changes associated with muscle failure during high intensity exercise, but the sensations of fatigue do not necessarily relate well to the more obvious physiological changes. Muscle fatigue occurs when physical tasks require high-power, short-duration repetitive contractions, or when there is low power, sustained or repetitive contractions (Faulkner and Brooks, 1997). Muscle fatigue can be defined as the reduction in maximum force-generating capability of the muscle. During exercise, the magnitude and mechanisms of human skeletal muscle fatigue vary widely and depend to a large extent on the individual, the type of muscle, and the exercise or task.

Fatigue is caused by a combination of different physiological mechanisms occurring at both the central and peripheral levels (Noakes, 2000) affecting afferent neuromuscular pathways, observed as proprioceptive deficiency (Skinner et al., 1986; Lattanzio and Petrella 1998) and efferent neuromuscular pathways, for example, a delay in muscle response (Nyland et al., 1994; Wojtys et al., 1996). Thus, muscular fatigue leads to a decline in work performance. A decreased ability to maintain balance in single-limb position (Johnston et al., 1998; Yaggie and McGregor 2002; Ageberg et al., 2003) after fatiguing exercise (i.e., higher values after exercise) has been reported in uninjured subjects, and it has been suggested that individuals are therefore at increased risk of injury when fatigued (Yaggie and McGregor 2002; Johnston et al., 1998; Lundin et al., 1993).
To enhance development and decrease risk of injury of muscular strength with heavy-resistance exercise, the best possible conditions for recovery from the individual exercise training sessions are necessary. Therefore, recovery involves the coordinated functioning of several physiological processes that are heavily influenced by the availability and actions of specific individual characteristics. However, the understanding of these complex interactions is incomplete, especially related to heavy-resistance exercise in which several other aspects of programme design (e.g., strength and duration, rest periods) and individual characteristics (e.g., age, gender, training status) also contribute to the exercise-induced hormonal responses (Kraemer, 1988).

Although recent studies have shown that women have greater muscular endurance than men (Hunter and Enoka 2001; Clark et al., 2003), there is a lack of information on sex differences in fatigue patterns considering the amount of information documenting strength differences (Clarke, 1986). The mechanisms for these differences are largely unknown, but there are two widely proposed hypotheses; 1) differences in muscle mass, and 2) differences in activation model (Clark et al., 2003). Recent research (Hunter and Enoka 2001; Clark et al., 2003) appears to indicate that the first hypothesis is most likely related to the sex differences in muscular fatigue due to similar findings that differences in endurance times during a fatiguing task were not related to differences in the neuromuscular recruitment strategy when both men and women were assessed (Albert et al., 2006).

It is well known that impairment of performance resulting from muscle fatigue differs according to the types of contraction involved, the muscular groups tested, and the exercise duration and intensity. Depending on these variables, strength loss with fatigue
can originate from several sites from the motor cortex through to contractile elements (Strojnik and Komi, 2000).

Neuromuscular fatigue also appears to develop differently, depending on the muscular action modes. Defined as the decline in muscle performance during and after repetitive contractions, fatigue has been shown to be task dependent (Nicolas et al., 2005). However, most studies dealing with the comparison of the different contraction modes on neuromuscular fatigue have used quite similar fatiguing procedures that produced different torque decrements (Kay, et al., 2000; Perry-Rana et al., 2002; Nicolas et al., 2005). When comparing men and women, besides lower absolute forces, women have been shown to have lower rates of maximal force production (Komi and Karlsson 1978; Ryushi et al. 1988). Also women have been shown to demonstrate less fatigue than men in heavy resistance exercise (Hakkinen, 1994). It would be of interest therefore to examine whether the differences in fatigue would be the same after resistance exercise in females. Accordingly, the present study was designed to examine the effects of maximal strength (heavy resistance) loading on neuromuscular fatigue and recovery in female during period of 48 hours post-exercise.
4.2 Methods

Ten trained female volunteers (18-30 years) participated in this experiment. The mean (±SD) age, height and body mass of the subjects were 25± 2 years, 1.78 ± 0.04 m and, 58.3 ±3.13 kg, respectively.

4.2.1 Resistance exercise protocol

All subjects were familiarised with the testing procedures. Subjects performed a resistance exercise session as described in section 3.2.2, which consisted of 45 minutes of weight training including 3 sets of 10-12 repetitions of six exercises (lying leg curls, dumbbell lunges, seated calf raises, leg extensions, straight leg deadlift, leg presses) but at an intensity corresponding to 70% of 1-RM. This was greater than used in the pilot study in order to obtain a greater fatigue effect. A 1-min rest period was allowed between exercises and a 3-min rest period between sets. The isometric force measurements were taken before and after exercise, and after 2, 24 and 48 hours of recovery (Figure 4.1).

Figure 4.1 The experimental design for study two.
4.2.2 Measurement of MVC and RFD

Subjects were required given a standard warm-up and then asked to perform three maximal isometric contractions each as each rapidly as possible and maintain that force for 6s for both legs together. Both legs were used as recommended from the results of pilot studies. Verbal encouragement was given to each subject during each trial and visual feedback of their performance was provided during and after each trial via the projection of the computer display onto a large screen placed in front of the subject. The time interval between each maximal effort was 60 s. Data on MVC and RFD were computed as described in pilot study one (in section 3.1.2.4).

4.2.3 Statistical analysis

The average of the three contractions was calculated for each variable and compared across each time point. Descriptive statistics (mean and ±SD) were calculated for all measured variables. The significance of differences between five times points in same group for each condition was assessed by a one-way analysis of variance (ANOVA) with repeated measures for within-subject. Post-hoc comparisons using the Bonferroni method were applied to determine pair-wise differences. Statistical significance was set at P<0.05.
4.3 Results

The mean and standard deviations of the averages of the three contractions for MVC and RFD are presented in Table 4.1 and figures 4.2 and 4.3. The one-way ANOVA with repeated measures revealed that when MVC and RFD were compared across all time, a significant trial effect was observed (P<0.05), which reflects the changes in MVC and RFD after 2, 24 and 48 hours.

4.3.1 Maximum voluntary isometric force (MVC) for both legs and dominant leg

Statistical analysis of data showed that there was a significant main effect of time on MVC for both legs (F 4, 36 = 17.25; P=0.001). Post-hoc analysis revealed a significant difference between pre-exercise and post-exercise (P=0.001), pre-exercise and 2h, 24h (P=0.002) but no significant difference for 48h (P=1.00).

There was a significant main effect of time on MVC (F 4, 36 = 18.92; P<0.001) for the dominant leg. Pairwise comparisons showed a significant difference between pre-exercise and post-exercise at 2h (P=0.001), but no significant difference between pre-exercise and after 24h and 48h (P=1.00).

Figure 4.2 The mean ±SD of two trials (n=10) for MVC (N) pre and post-exercise and into recovery time for both legs and the dominant leg.
4.3.2 Rate of force development (RFD) for dominant and both legs

Statistical analysis of data showed that there was a significant main effect of time on RFD for both legs ($F_{4,36} = 26.21; P < 0.001$). Post-hoc analysis revealed a significant difference between pre-exercise and post-exercise ($P = 0.001$) and at 2h, but no significant difference between pre-exercise and 24h and 48h ($P = 1.00$).

There was a significant main effect of time on RFD ($F_{4,36} = 11.44; P < 0.001$) for the dominant leg. Pair-wise comparisons showed a significant difference between pre-exercise and post-exercise ($P = 0.000$) but no significant difference between pre-exercise and 2h, 24h and 48h ($P = 1.00$).

![Figure 4.3](image.png)

**Figure 4.3** The mean ±SD of two trials (n=10) for RFD (N/s) pre and post-exercise and into recovery time for both legs and the dominant leg.
4.4 Discussion and conclusion

It has already been established from the pilot studies that the average of three contractions was sufficient to obtain repeatable MVC and RFD data between test sessions. It was also established that repeatable measurements could be made over successive days and when using the average of three contractions at each time point without any concomitant effect on the measured variable. Therefore this allows an experimental protocol which has both within-days and between-days reliability requirements.

The aim of the present study was to examine the characteristics of fatigue and recovery responses to heavy resistance exercise in females in terms of isometric force variables. Specifically, the effects of resistance exercise on MVC and RFD at each time interval of 2, 24, and 48 hours post-exercise were investigated. The results showed a significant effect of fatigue induced by resistance exercise for the both legs condition on MVC measured pre-exercise and post-exercise and at 24 hours but no significant difference at 48 hours of recovery. Similar results were found for the dominant leg except recovery appeared to have been achieved by 24 hours.

For the RFD, recovery appeared to have been achieved by 24h in the both legs conditions. The non-significant difference at 2h for the dominant leg condition is probably due to the large standard deviation of the data (Table 4.2), as the mean value is only a little greater than the both legs condition.

The fatigue recovery of the both legs compared to the dominant leg condition is probably due to a weaker-non-dominant leg. If that were the case, than the use of either the both legs condition or the weaker leg condition would be more likely to show difference in the
further studies. However, in order to retain consistency, comparison with other studies and to avoid any learning effects it is recommended that the both legs and the dominant leg protocol be retained.

The subjects were familiarized with the performance of maximal isometric contraction one week prior to the testing sessions and there appear to have been no learning effect as the data at 24 and 48 hours did not differ significantly from the pre-exercise level. The familiarization of the subjects was deemed sufficient in order to remove any learning effect. The reduction in force following the exercise session was due to some form of peripheral fatigue. The findings indicated that the level of fatigue in MVC and RFD percentage drop following the resistance exercise program was around 23.7% and 25.2% for MVC in the dominant leg and both legs respectively, and 32.9% and 34.2% in RFD, at an intensity corresponding to 70% of 1-RM (10-12 repetitions) which did not reach the required 40% reduction as described on male subjects by Fell (2002).

In conclusion the results suggest that fatigue under both leg conditions caused by muscular work depends on the amount of effort exerted. Contraction at 70% 1-RM was a significant contributor to the loss of muscle strength after resistance exercise, observed clearly in the females. In this study it was found that using both legs at 70% of 1-RM was still not enough for the desired reduction, so further studies should increase the intensity of 1-RM.
CHAPTER 5

CARBOHYDRATE INGESTION AND RESISTANCE EXERCISE IN FEMALES
5.1 Introduction

It is well documented that carbohydrate supplementation improves prolonged aerobic exercise performance, and numerous studies have shown that carbohydrate ingestion delays fatigue during such exercise (Coggan and coyle, 1988; Costill et al., 1992; Zachwieja et al., 1993). Mechanisms suggested to explain the beneficial effects of carbohydrate ingestion include maintenance of blood glucose concentration, promotion of high rates of carbohydrate oxidation, and muscle glycogen sparing (Bailey et al., 2000; Campbell et al., 2001). In addition, carbohydrate supplementation prior to endurance exercise decreases markers of protein and membrane breakdown (Tarnopolsky, 1999; 2000). In spite of the wealth of documented evidence for the beneficial effects of ingested carbohydrate on prolonged exercise, evidence is sparse in relation to resistance exercise.

It is generally accepted that resistance exercise performed on a regular basis elicits sport-related and health-related benefits in men and women (Tarnopolsky, 2001, 2003). Therefore, resistance exercise training is recommended by the American College of Sport Medicine. Available literature suggests that nutrition has a major effect on the physiological and biochemical responses to resistance exercise in men and women and plays a pivotal role in exercise performance (Angus et al., 2000; Bailey et al., 2000; Campbell et al., 2001). In addition, the nutritional needs of women engaged in resistance exercise training are, to a great extent, similar to those for men (Tarnopolsky, 2002). It is important therefore to recognise that sound nutritional strategies, as an adjunct to the training process, facilitate strength development and enhance exercise performance. Strength athletes and habitual resistance exercise performers usually ingest various mixtures of water, carbohydrate and electrolytes (Jeukendrup, 2004). The alleged
benefits include improved performance and or reduced physiological stress induced by resistance exercise.

Earlier studies indicated that carbohydrate drinks consumed before and during exercise are beneficial when exercise intensity is low and the duration is long (Friedlander et al., 1998; Horton et al., 1998; Jarvis et al., 1999). However the value of carbohydrate on delaying fatigue and enhancing performance in high intensity exercise of shorter duration such as resistance exercise is equivocal. Furthermore, reports on the effect of carbohydrate ingestion before and during resistance exercise on metabolic and neuromuscular responses are limited and produced conflicting results (Braun and Horton 2001; Friedlander et al., 1998; Horton et al., 1998; Tarnopolsky et al., 2001). Understanding the physiological basis for, and the beneficial effects of, carbohydrate ingestion before and during resistance exercise, and elucidation of biological control mechanism(s) that affects the strength indices is therefore of scientific significance.

Although the effect of sex-related differences on metabolic responses and exercise performance has received considerable interest in the last 20 years (Jeukendrup, 2004), meagre information is available on the effects of nutritional ergogenic aids such as carbohydrate on muscular of strength in females. Information on the influence of sex on strength-related indices response to resistance exercise is needed. The previous study indicated that the level of fatigue in MVC and RFD as indicated by the percentage drop following the resistance exercise program was around 25.2% for MVC and 34.2% in RFD in both legs. Since carbohydrate ingestion is understood to be beneficial in helping performance of other activities it would be of benefit in reducing fatigue following resistance training. In addition, we wished to promote fatigue to a greater level than the previous study and so the aim of this study was to investigate the effect of ingestion of
carbohydrate on MVC and RFD during recovery after resistance exercise by use an intensity increased to 80% of 1-RM in females.

5.2 Methods

5.2.1 Subjects and study design

This study was approved by the Ethics Committee of the Liverpool John Moores University. After the experimental protocol and testing procedures were fully explained, eight moderately trained female subjects gave their written consent before participating in the study (Appendix A) having read and understood the details of the experiments (Appendix B). Criteria for inclusion were a recent training history of at least 12 months of resistance exercise training. This was ascertained using a questionnaire (Appendix C). The mean (±SD) age, height and body mass of the subjects were 26.1 ± 2.7 years, 1.74 ± 0.05 m and, 64.3 ± 3.21 kg, respectively.

5.2.2 Design and experimental protocol

One week prior to the main experiment, the 1-RM for six resistance exercises for the lower body part was ascertained using the procedures described in section 3.1.2.3. Thereafter, all subjects were asked to report to the laboratory to undertake two further trials to examine the effects of carbohydrate supplementation or placebo on muscular strength indices. These trials were randomised and conducted at the same time of day (9:00 am), on two separate occasions with one week intervening. Subjects were either supplemented with carbohydrate (CHO) solution or carbohydrate-free placebo solution (CF) prior to the performance of a resistance exercise session encompassing different muscle groups of the lower body part. Prior to the main experiments, all subjects were
familiarized with the laboratory environment and equipment, testing procedures, and exercise protocol.

5.2.3 Protocol for the administration of the solutions

When a subject reported to the laboratory for testing, body weight was determined using the procedures described in section 3.1.2.2. The total volumes of test drink ingested were the same for each subject participated in the experimental trials and the apportioned volumes of drink ingested pre and post resistance exercise were equivalent in both trials.

For the CHO trial, participants ingested a 500ml stock of a carbohydrate solution (0.5g CHO per kg/BM) was prepared for each subject and divided into two equal portions. The first portion was given to each subject at 15-20 min before exercise, while the second portion was administered immediately after the completion of a resistance exercise session. In the placebo trial, a 500 ml stock of a carbohydrate-free solution as the placebo (CF) was prepared for each subject and divided into two equal portions and administered at the same points of time as in the carbohydrate trial. The carbohydrate and carbohydrate-free solutions were indistinguishable in appearance and taste. This was ascertained through a small pilot study using 3 subjects who participated in the main experiments.

5.2.4 Resistance exercise protocol

The resistance exercise protocol described in section 4.2.1., was employed in this experiment. However, instead of using 70% of the 1-RM, a work load corresponding to 80% 1-RM was utilised, as the result found in last study that the level of fatigue in MVC and RFD percentage drop following the 30-minute of resistance exercise program was
around 23.7% and 25.2% for MVC in the dominant leg and both legs respectively, and 32.9% and 34.2% in RFD, therefore the difference was still found to be not enough for the desired 40% reduction, so in the this study the an intensity was increased to 80% of 1-RM.

5.2.5 Measurement of MVC and RFD
Using the procedures described in section 4.2.2., the MVC and RFD were measured 20-min following the oral administration of the test solution, and immediately following the completion of the resistance exercise session. Further measurements of MVC and RFD by using both leg and were ascertained at 2 h, 24 h and 48 h into recovery after exercise. The average of three contractions of MVC at each time point was calculated and utilised in the statistical analysis. Similarly the average of RFD was calculated and employed in the statistical analyses of the data.

5.2.6 Statistical analyses
Descriptive statistics were used for the calculation of the means, ±SD. Data presented in the text are means ±SD and data presented in tables and figures are means ±SE unless otherwise stated. In order to determine the effects of carbohydrate ingestion on MVC and RED, data were analysed using a two-way ANOVA with repeated measures for treatment (carbohydrate and control) and time (pre-exercise, immediately after exercise, 2h, 24h and 48h into recovery). When a significant F ratio was found, differences in mean values between specific times points were determined by student’s paired t-test. Significance was accepted with alpha level of P < 0.05. A computerised statistical package (SPSS version 14.0) was used for all data analyses.
5.3 Results

The mean and standard deviations of MVC and RFD before and after resistance exercise and into recovery are presented in and graphically illustrated in figures 5.1 and 5.2.

5.3.1. Maximum Voluntary Contraction (MVC) for both legs together.

Statistical analysis of data showed that there was a significant main effect of treatment on MVC ($F_{1.7} = 18.99; P = 0.003$), time ($F_{4.28} = 38.48; P = 0.00$), and non-significant interaction between treatment and time ($F_{4.28} = 2.72; P = 0.08$). Post-hoc analysis revealed pre-exercise MVC mean values were similar when comparing carbohydrate (CHO) and placebo (CF) trials.

There was a non significant difference within pre and post-exercise for both conditions ($P > 0.05$), but a significant within 2h ($P = 0.025$), 24h ($P = 0.029$) and at 48h ($P < 0.008$). Resistance exercise resulted in a significant decrease ($P<0.05$) in MVC immediately after exercise and this occurred similarly in both CHO and CF conditions. The relative decrease in force immediately after resistance exercise session did not recover within the first 2h after exercise for both conditions and a deficit in force production was still evident after 24 h even. Even on the other hand, MVC did fully recover after 48 h and there was evidence a faster recovery and super-compensation in the CHO trial.
Figure 5.1 The mean and ±SD of MVC pre- and post-exercise and into recovery over CHO and CF trials (* = p< 0.05 between CHO and CF conditions).

5.3.2 Rate of Force Development (RFD)

Statistical analysis of data showed that there was a significant main effect of treatment on RFD ($F_{1,7} = 20.73$, $P= 0.003$) and time ($F_{4,28} = 45.19$, $P= 0.00$) and a significant interaction between treatment and time ($F_{4,28} = 10.69$, $P= 0.001$). Post-hoc analysis revealed pre-exercise RFD mean values were similar when comparing CHO and CF trials ($P< 0.05$).

There was non significant difference within pre and post-exercise ($P > 0.05$), but a significant within 2h recovery ($P = 0.034$), at 24h ($P= 0.003$) and at 48h recovery ($P< 0.001$) for both conditions. The strong resistance exercise for the lower body part resulted in a significant decrease ($P<0.05$) in RFD immediately after resistance exercise and this occurred similarly in both CHO and CF trials. The relative decrease in RFD immediately after the resistance exercise session did not recovery within the first 2h after exercise for both conditions and a deficit in rate of force production was still evident.
after 24h. On the other hand, RFD did fully recover after 48h and there was evidence of super-compensation for the CHO trial as indicated by the significant interaction.

Figure 5.2 The mean and ±SD of RFD pre- and post-exercise and into recovery over CHO and CF trials (* = \( p < 0.05 \) between CHO and CF conditions).
5.4 Discussion and conclusion

The results from this study demonstrate that when relatively high amounts of CHO are fed before and after exercise, there was a beneficial contribution of CHO during the recovery period when compared with the ingestion of a placebo. Specifically, when CHO was ingested both before and after exercise, MVC and RFD performance increased over the recovery times compared with the ingestion of a placebo. The beneficial effects CHO ingestion on MVC and RFD when undertaking resistance exercise lasted for at least 48 hours of recovery post-exercise.

Training or competition can only be continued when there is an adequate amount of carbohydrate available to fuel muscles. Fatigue is closely associated with depletion of the limited stores of carbohydrate in the muscle and in the liver. Therefore, it is not surprising that strategies have been developed to ensure that not only are the carbohydrate stores well stocked before exercise but that they are also restored as soon as possible after 2 hours of resistance exercise. Consuming carbohydrate immediately after exercise increases the rate of muscle glycogen resynthesis and also results in greater capacity during the subsequent recovery period. Conley et al., (2000) this may decrease recovery time following resistance exercise and enable an increase in training volume which may enhance physiological adaptations. Also, carbohydrate ingestion during or immediately after resistance exercise has been shown to increase post-exercise plasma insulin which may lead to increased protein synthesis. Additionally, insulin has been reported to be a potent stimulator of protein synthesis (Rasmussen et al., 2000; Tipton et al., 2007; Biolo et al., 1999). Some research has shown that ingesting CHO following exercise promotes a more rapid resynthesis of muscle glycogen (Blom et al., 1987; Burke
et al., 1996) and that ingesting CHO following exercise increases muscle glycogen replacement (Roy and Tarnopolsky, 1998; Tarnopolsky, et al., 1997).

During high intensity resistance exercise, recovery periods play an important role in limiting fatigue (Ratel et al., 2002). There is evidence to suggest that the provision of fluids and carbohydrate during intermittent exercise may improve physiological performance and also help to delay fatigue caused by dehydration (Welsh et al., 2002). Indeed, Williams (2004) found that the type of carbohydrate ingested in recovery has an influence on endurance capacity the following day. The findings in this study indicated that the level of fatigue for MVC and RFD after resistance exercise and after consuming CHO was reduced around 30.3% and 22.4% respectively, and 30.9% and 24.1% for placebo session. Some studies have shown that CHO supplementation may also be beneficial during intermittent exercise of shorter duration (Coombes and Hamilton, 2000). Athletes engaged in duration events or events that involve higher intensity exercise or multi-events are likely to deplete short muscle glycogen stores during the event, and so would benefit from CHO supplementation. CHO supplementation would provide glucose which can then be converted to glycogen for use in ATP production during the high intensity exercise. It should also be noted that during sporting events less than 30 minutes of continuous exercise, fluid replacement is a higher priority than replacing CHO in the muscle or liver (Pearce, 1996).

Although, the decrease in force was not enough for the desired 40% reduction, this was greater than the findings from the previous study without using CHO supplementation. This may represent a limit of fatigue for this type of exercise and gender as the findings indicated that the level of fatigue in MVC percentage drop following the resistance
exercise program was around 40% in maximum force by used male participants (Fell, 2002).

It must be noted that the effect of a CHO supplement on females is purely an observation, and more precise methods are needed to describe a mechanism for macronutrient feeding. Future research on the CHO supplementation should include measurements of blood glucose and some blood hormones to be more certain whether the exercise session did challenge circulating endogenous glucose, and that the supplement effectively countered that effect in females.

*In conclusion*, the aim of the present study was to gain a better understanding and provide more information on the response of women to CHO augmentation. On the basis of the findings, the results suggest that for trained female participants, the presence of CHO supplement results in quicker recovery after resistance exercise than placebo. Thus, the intake of carbohydrates after resistance exercise will improve the effect of exercise with females. This nutritional strategy may be critical for female athletes who need to engage in multiple events or training sessions during the course of a day.
CHAPTER 6

THE EFFECT OF CREATINE SUPPLEMENTATION ON FATIGUE AND RECOVERY AFTER RESISTANCE EXERCISE IN FEMALES
6.1 Introduction

Most of the research in the areas of sports nutrition, muscle metabolism and exercise physiology has been conducted predominantly using men. The physiological responses to exercise are similar between men and women. Strength training for healthy women is authorized by the American College of Sports Medicine and has many beneficial effects on health (Asikainen et al., 2002; Pollock et al., 2001) while also enhancing performance in other activities, (Kraemer et al., 2001) Women engaging in strength training range from young high school athletes, (Faigenbaum, 2000) to post-menopausal women, (Asikainen et al., 2002) to those competing in strength training sports such as weight lifting, power lifting, and body-building. Nutrition has a major influence on the magnitude of adaptation to training. Therefore nutritional recommendations for men and women strength athletes share many common elements (Volek, 2003; 2004).

In the last few years many athletes and persons engaged in recreational sports activities have used creatine supplementation. Recent research has also suggested that there may be a number of potential therapeutic uses of creatine. Over 500 research studies have evaluated the effects of creatine supplementation on muscle physiology and/or exercise capacity in healthy, trained, and various diseased populations (MacLaren, 2000).

Many studies show that creatine supplementation has positive effects on performance of short-duration exercises, (Harris et al., 1993; Birch et al., 1994; Balsom et al., 1995; Maughan 1995; Volek et al., 1997) as well as repeated isokinetic or isometric contractions of the quadriceps muscle, jumping, or high-intensity cycling exercises.

Although the majority of studies have been in men, a significant amount of research indicates women are also responsive to creatine supplementation. Short term creatine
supplementation has been shown to enhance high intensity exercise performance in
women (Eckerson et al., 2004; Tarnopolsky and MacLennan, 2000). Studies that have
examined strength and body composition responses during a resistance training program
indicate women respond positively to creatine supplementation (Brenner et al., 2000;
Vandenberghe et al., 1997).

Phosphocreatine stores play a key role for ATP resynthesis during muscle contraction
and recovery. The changes in performance following creatine supplementation is
dependent on the characteristics of the exercise, and it has been suggested that human
skeletal muscles have a higher limit for total creatine concentration. In contrast with
sedentary subjects, athletes and well-trained subjects who have high initial total creatine
concentrations in skeletal muscle, show only a slight improvement in exercise performance
(Bigard, 1998).

Dietary creatine supplementation (20–30 g/day for 4–6 days) has been reported to
increase muscle creatine concentration by as much as 50% and enhance muscle
performance during intermittent high-intensity exercise bouts (Earnest et al., 1995;
Greenhaff et al., 1993; Harris et al., 1992). The performance enhancing effect of creatine
may result from increased muscle creatine availability that sustains the initially rapid rate
of PCr resynthesis further into recovery and increases available PCr during later exercise
bouts (Balsom et al., 1995; Greenhaff et al., 1994; Harris et al., 1992). Taken together,
the results of most studies published to date suggest that only performances of repetitive
high-intensity exercise bouts are completely affected by creatine supplementation.
During this type of exercise, the expected increase in total creatine contributes to the fast
resynthesis of phosphocreatine during recovery.
Chapter 6- the effect of creatine supplementation

The majority of these studies have used a normal dosing regimen for the duration of their investigations. A typical creatine supplementation regimen involves a "loading phase" (0.3 g/kg) for 5 days to maximize muscle creatine stores, followed by a maintenance phase of (2 g/day) (Harris et al., 1992; Hultman et al., 1996; Kreider et al., 1998; Wilder et al., 2002). There is only one investigation that has shown the ergogenic benefits of a low-dose creatine supplementation (Burke et al., 2000). However, in that study, a dose 0.1 g/kg was used during 28 days of supplementation. Although this was a longer supplementation period than typically seen during short-term studies, it is the first study to show efficacy with a low-dose and more prolonged supplement regimen.

Nutritional needs of women engaged in strength training are important and are generally similar to those recommended for men (Volek, 2003; 2004). Women in particular should ensure adequate energy is consumed to optimise adaptations to training and improve general health. In addition, generous consumption of healthy fats from a variety of sources is encouraged to support a positive energy balance, hormonal balance, and optimal health. The biochemical events that occur with intense exercise are multiple and complex and involve metabolism, generation of reactive oxygen species, and disruption of cell membranes. The disruption to the cell membrane results in leakage of cytosolic proteins into the circulation, such as creatine kinase (McBride et al. 1998), and myoglobin (Volek et al., 1999). Like men, women respond to the favourable effects of creatine supplementation during resistance training. Therefore, the purpose of this investigation was to examine the effects of creatine supplementation on fatigue and recovery after resistance exercise in females. Such data are needed to provide direct evidence regarding the efficacy of creatine supplementation on the performance of typical resistance exercises used by females.
6.2 Methods

6.2.1 Subjects and General

The methodological studies described in the pilot and main studies were use to create the protocols to reliably assess MVC and RFD and were given to subjects on an information sheet. Ten subjects were medically screened to determine their health and exercise habits prior to exercising lower body muscles. Each individual gave written informed consent to participate in the study, which has been approved by the University Research Ethics Committee of Liverpool John Moores University, and asked to complete the diet and physical activity sheet. The subjects attended the laboratory on four different testing occasions. An initial unit was used for the purpose of familiarisation of the details of test (Atkinson and Reilly, 1996). A total of 24 hours was allowed between familiarisation and the further testing sessions. On each testing session the subjects were required to perform three maximal isometric force contractions from which MVC and RFD were evaluated. Venous blood samples were taken except during the 2nd control session which was to minimise discomfort to subjects. After the first test, subjects undertook a resistance exercise session for the lower body at an intensity corresponding to 80% of one repetition maximum (1-RM). Tests were also obtained at 2h, 24h and 48h of recovery (Figure 6.1). The participants were also tested with no supplementation (a placebo) as a control condition.

Subjects then were required to consumed either 20 g of creatine monohydrate for 5 days or a placebo before being tested again. Blood samples were taken before and after the exercise session, and analysed for creatine kinase (CK), growth hormone (GH), myoglobin (MYO), haemoglobin (Hb) and hematocrit (Hct). It was hypothesised that
Figure 6.1 The double blind cross-over experimental design

<table>
<thead>
<tr>
<th>Familiarization</th>
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<tbody>
<tr>
<td><strong>(Session 1)</strong></td>
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<tr>
<td>Control session (Test with no supplementation)</td>
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<thead>
<tr>
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<th>Day 2</th>
<th>Day 3</th>
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<td>Blood, MVC, RFD</td>
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<th>after</th>
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<th>+24h</th>
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<td><strong>(Session 2)</strong></td>
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<table>
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<tr>
<th>Days 1-5 20 g/day creatine or placebo</th>
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<td><strong>(Session 3)</strong></td>
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<tr>
<th>Day 1</th>
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<tr>
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<th>Two weeks rest</th>
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<td><strong>(Session 4)</strong></td>
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<th>Days 1-5 20 g/day creatine or placebo</th>
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<tr>
<th>Day 6</th>
<th>Day 7</th>
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<tbody>
<tr>
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<td>Blood, MVC, RFD</td>
<td>Blood, MVC, RFD</td>
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<table>
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<tr>
<th>Exercise</th>
<th>0</th>
<th>after</th>
<th>+2h</th>
<th>+24h</th>
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Chapter 6- the effect of creatine supplementation

creatine loading reduces the fatigue effects of exercise and improve the recovery responses in both neuromuscular and biochemical variables.

6.2.2 Height and Body-Mass

Each subject’s height and BM was measured as described for Study 1 and 2.

6.2.3 Protocol for the administration of the solutions

Subjects were randomised in a double-blind cross-over experimental design; placebo group (n= 10) and creatine-supplemented group (n= 10). The supplements were provided to the participants in identical, unmarked, sealed containers. Following session 1, subjects were providing with 20 doses (4 * 5 g doses per day) of creatine monohydrate or placebo. Subjects in placebo group consumed the same amount of all ingredients except creatine which were indistinguishable in appearance and taste from the creatine. The creatine component in the placebo was replicated by cellulose powder and methylcellulose. Compliance during the 5 days of supplementation was 100%. The subjects were then instructed to take the next five doses with 240 ml of water on an empty stomach for four times a day and one dose was required on the first and second day of testing. Prior to beginning the study, subjects were weighed before and after the three-day period of testing. The seven-day food records were subsequently photocopied via written dietary diary sheet and returned to subjects and were encouraged to eat similar diets, during all testing period before the heavy resistance session and recovery period.
6.2.4 Resistance exercise protocol

The resistance exercise protocol described in section 4.2.1. was employed in this experiment. However, instead of using 70% of the 1-RM, a work load corresponding to 80% 1-RM was utilised.

6.2.5 Measurement of MVC and RFD

Using the procedures described in section 4.2.2., the MVC and RFD were measured before and immediately following the completion of the resistance exercise session. Further measurements of MVC and RFD were ascertained at 2 h, 24 h and 48 h into recovery after exercise. The average of three contractions of MVC by using both legs at each time point was calculated and utilised in the statistical analysis. Similarly the average of RFD was calculated and employed in the statistical analyses of the data.

6.2.6 Blood sampling

Approximately 10 ml of venous blood was sampled from the antecubital fosse vein via catheterisation prior to the resistance exercise session (baseline), and 2h, 24h, 48 h. The blood was immediately placed into an ethylediniaminetetra-acetic acid (EDTA) tube, and gently inverted and rolled several times. Blood was then transferred into Eppendorf tubes and centrifuged at 3000 rpm for 15 min at 4 °C. Plasma was removed and aliquot into labelled tubes and stored at -80 °C for subsequent analysis of CK, GH and MYO.

6.2.6.1 Blood analysis

Analyses of frozen samples from all five sessions were performed at the same time using the same batch of reagents for CK, GH and MYO to minimise differential analyses effects.
6.2.6.2 Plasma Creatine Kinase

Aliquots of the plasma from the whole blood samples were left to return to room temperature before being analysed in duplicate by spectrophotometry using a Kodak Ektachem DTSC Module (Kodak Co, New York, and U.S.A). Ten micro-litres (10 ml) of plasma was deposit on a Kodak Ektachem DT CKMB slide (Johnson and Johnson Clinical Diagnostic Inc., New York, U.S.A) and inserted into the module for the determination of creatine kinase (CK). The module was calibrated prior to use using known references.

6.2.6.3 Growth hormone and Myoglobin

Blood sampling was conducted via a heparin lock catheter placed in a forearm vein at the start of the experimental day. Subsequently, blood samples were taken before, after exercise and 2 h, 24 h and 48 h recovery period. Blood drawn from the forearm vein was placed into Vacutainers prepared with EDTA and chilled to preserve the integrity of the samples. All samples were centrifuged at 3,500 rpm for 15 min at -10°C. Upon separation, the plasma was aliquotted to microcentrifuge tubes and frozen at -80°C until analysis.
6.3. Statistical analyses

Subject characteristics are reported as means ± SD. In the order to evaluate the fatigue and recovery characteristics in response to the four loading conditions all data were analysed using a two-way ANOVA (Condition x Time) with repeated measures on both MVC and RFD. Data are means and standard errors of means (± SE). The significance of differences between five times in same group for each condition was assessed by a one-way analysis of variance (ANOVA) with repeated measures as the within-subject, following post-hoc comparisons using the Bonferroni method were applied to determine pair-wise differences. The effect of creatine supplementation was investigated using the average of the three contractions calculated for each variable and compared across each time point. Statistical significance was accepted at P < 0.05. The statistical analyses were performed with the SPSS software package version 14.0 for Windows (SPSS Inc., Chicago, Ill., USA).
6.4. Results

The mean and standard deviations of MVC and RFD before and after resistance exercise and into recovery are and graphically illustrated in figures 6.2 and 6.3.

![Figure 6.2 Mean ±SD of three trials (N=10) for MVC (N) for all conditions across five times points.](image)

6.4.1 Maximum voluntary isometric force (MVC)

Statistical analysis of data showed that there was a significant main effect of condition on MVC \( [F_{3, 21} = 25.33, P = 0.001] \), and time \( (F_{4, 28} = 156.36, P = 0.001) \) with a significant interaction between condition and time \( (F_{12, 84} =3.52, P= 0.02) \). With regard to condition, post-hoc analysis revealed a significant difference between Cr supplement sessions with all other conditions \( (P < 0.05) \). There was no significant difference between the placebo condition and the control condition.

With regard the time, post-hoc analysis revealed a significant difference between pre-exercise and post and 2h of recovery but no significant difference at 24h and 48h of recovery although a deficit was still evident at 24h. With regard to the interaction term,
the Cr condition started a faster recovery from 2h to 48h than the placebo and control conditions, leading to a significant interaction and a super-compensation. The 80% resistance exercise for the lower body after Cr supplementation resulted in a significant decrease in MVC value of 123.2 N in the Cr supplement condition and 120 N in the placebo condition, immediately after resistance exercise.

![Figure 6.3 Main ±SD of three trials (n=10) for RFD (N/s) for all conditions across five times points.](image)

### 6.4.2 Rate of force development (RFD) across conditions and times.

Statistical analysis of data showed that there was a significant main effect of condition on RFD \(F_{3,21} = 13.36, P = 0.001\), and time \(F_{4,28} = 65.41, P <0.001\) with a significant interaction between condition and time \(F_{4,84} =4.87, P= 0.004\). With regard to condition, post-hoc analysis revealed a significant difference between Cr supplement sessions with all other conditions \(P < 0.05\). There was no significant difference between the placebo condition and the control condition.
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With regard to the time, post-hoc analysis revealed a significant difference between pre-exercise and post, 2h and 24h of recovery but no significant difference at 48h of recovery on RFD. With regard to the interaction term the Cr condition started a faster recovery from 2h to 48h than the placebo and control conditions, leading to a significant interaction and a super-compensation. The 80% resistance exercise for the lower body after Cr supplementation resulted in a significant decrease for RFD of 348.4 N/s in the Cr supplement condition and 366.9 N/s for the placebo condition, immediately after resistance exercise.

6.4.3 Body mass

After 5 days of Cr supplementation, the body mass of the Cr group increased from 60.1 kg to 61.1 kg, whereas the control and placebo group increased from 60.1 kg to 60.4 kg. There were no significant change between conditions ($F_{2,14}=3.67$, $P=0.14$).

| Table 6.1 Resting values (mean ± SD) of haematological variables over the time intervals of pre-, post-, 2, 24 and 48 hours recovery (n= 10) for control 1 (Con), supplement (Cr) and placebo (P) conditions. |
|---|---|---|---|---|---|
| Variables | Pre-exercise | Post-exercise | +2h | + 24h | + 48h |
| CK (IU/l) | Con | 69.3± 25.8 | 78.1± 24.3 | 84.9± 37.1 | 94.0± 42.6 | 81.0± 37.2 |
| | Cr | 85.8± 29.8 | 89.4± 24.8 | 91.6± 28.4 | 116.1± 38.6 | 100.8± 41.9 |
| | P | 87.1± 42.8 | 102.1± 42.1 | 97.6± 39.8 | 102.0± 39.5 | 97.5± 46.7 |
| GH (ng/ml) | Con | 0.58± 0.76 | 2.55± 0.99* | 0.18± 0.14* | 0.70± 0.88* | 0.48± 0.56* |
| | Cr | 0.62± 0.59 | 1.73± 0.57* | 0.28± 0.32* | 0.75± 0.84* | 0.31± 0.33* |
| | P | 0.56± 0.69 | 1.91± 0.79* | 0.25± 0.33* | 1.03± 0.96* | 0.80± 0.70* |
| MYO (mcg/l) | Con | 15.1± 4.41 | 19.06± 6.31 | 22.4± 11.94 | 16.3± 4.41 | 17.4± 5.94 |
| | Cr | 17.1± 4.70 | 17.94± 6.19 | 20.1± 8.69 | 18.3± 4.89 | 16.3± 4.38 |
| | P | 16.2± 6.60 | 18.6± 7.01 | 23.1± 6.43 | 21.0± 8.68 | 19.7± 6.35 |

CK creatine kinase (IU/l); GH growth hormone (ng/ml); MYO myoglobin (mcg/l).

*Implicates significant increase compared to pre-exercise, ($P < 0.05$)

+denotes significant decrease compared to post-exercise ($P < 0.05$)
6.4.3 Blood biochemistry

6.4.3.3 Creatine kinase (CK)

Changes in CK over the 3 days testing period are shown in table 6.1 and figure 6.4. Statistical analysis of data showed that there was no significant main effect of time (F 4, 28 = 1.41, P = 0.28), although the CK peaked at 24 h of recovery and no significant effect for condition (F 2, 14 = 0.23, P = 0.76) nor for interaction between condition and time (F 8, 56 = 1.63, P = 0.22).

Figure 6.4 The mean ±SD values CK (IU/1) of all conditions; control 1, Cr supplement and placebo group cross five times points.
6.4.3.5 Myoglobin (MYO)

Changes in MYO over the 3 days testing period are shown in table 6.1 and figure 6.8. Statistical analysis of data showed that there was no significant main effect of time (F_{4, 28} = 1.08, P = 0.38), nor for conditions (F_{2, 14} = 1.41, P = 0.28), nor for interaction between condition and time (F_{8, 56} = 1.51, P = 0.24).

![Figure 6.6](image.png)

Figure 6.6 The mean ± SD values MYO (mcg/l) of all conditions; control 1, Cr supplement and placebo group across five times points.
6.4.3.4 Growth hormone (GH)

Changes in growth hormone (GH) over the 3 days testing period are shown in table 6.1 and figure 6.5. Statistical analysis of data showed that there was a significant main effect of time on GH ($F_{4,28} = 45.10, P = 0.001$) and condition ($F_{2,14} = 14.40, P = 0.03$), but no interaction between condition and time ($F_{8,56} = 1.57, P = 0.22$). With regard to the time, post-hoc analysis revealed a significant difference between post-exercise, and 2h, 24h and 48h of recovery on GH. With regard to condition, Cr produced lower levels of GH from post-exercise to 48h than the placebo and control conditions.

Figure 6.5 The mean ± SD values GH (ng/ml) of all conditions; control 1, Cr supplement and placebo (free Cr) group across five time points.
6.5 DISCUSSION

6.5.1 The effect of creatine supplementation on MVC and RFD
This study identified that 5 days of ingesting Cr improved performance for MVC and RFD after 48 hours of recovery, but did not affect body weight and the hormonal responses except growth hormone. The significant change in isometric contraction muscle strength was higher during recovery from resistance exercise in the creatine supplementation session compared to placebo and control sessions, which also showed a beneficial effect from creatine supplementation on indirect markers of muscle damage. These results support the work by Santos et al. (2003). In the present study, the results of the average of recovery bouts indicate that PCr resynthesis might be enhanced during the latter part of the recovery from high-intensity isometric exercise in agreement with the results of Greenhaff et al. (1994) during recovery from ischemic exercise.

With regards to changes in MVC and RFD, large differences were evident between the conditions and time period and were significant. At forty-eight hours of recovery there was no significant difference when compared with the pre-exercise session and was sufficient to allow full recovery. In contrast to the previous strength measurements discussed, creatine supplementation had a significant time effect on MVC and RFD for isometric contractions post-exercise. Recovery of mean MVC and RFD was complete after 24 hours with evidence of compensation due to the resistance exercise session. This agrees with Zatisorsky (1995), Branch (2003) and Nissen and Sharp (2003) who have concluded that creatine supplementation has positive effects on strength. While it is understood that 80% of each participants strength (1-RM) should illicit the same proportion of damage, and hence, similar decreases from the reduction of 146.6 N and 398.4 N/s in the Cr supplement condition and 120 N and 366.9 N/s in the placebo condition, it was evident that the 1-RM for the creatine-supplemented participants was
greater compared to the 1-RM of participants consuming the placebo over both the pre-exercise and the recovery period. It is generally accepted that creatine supplements increase phosphocreatine in muscle and thus improves recovery (by increasing the rate of phosphocreatine resynthesis in muscle) and increases the intensity and the length of muscular contraction during short term, high intensity work.

6.5.2 The effect of creatine supplementation on body mass

The present study examined the effects of Cr supplementation on body mass. A non significant mass gain of 1.0 kg was observed in the Cr session following supplementation. There was no gain in mass in the placebo or control conditions. This increase in body mass was indicative of the muscle 'loading' the excess Cr ingested through supplementation. Hultman et al., (1996) found that the decline in urinary volume following Cr supplementation closely resembled the increase in body mass suggesting that the gain in mass was due to water retention. Although urine samples were not taken in this study to verify that the supplementation Cr was retained within the body, the observed increase in body mass indirectly suggests that the Cr supplementation was effective. This assumption is consistent with this study that has found a mean increase in body mass of 1.0 kg following supplementation with 20g of Cr/day for 5-7 day (Balsom et al., 1995; Earnest et al., 1995; Greenhaff et al., 1994; Plisk, and Kreider, 1999; Volek et al., 1997). It is therefore reasonable to assume that the Cr supplementation period was effective in increasing the Cr stores in the muscle. Three studies (Thompson et al., 1996; Hamilton-Ward et al 1997; Terrillion et al., 1997) involving female subjects reported no significant increase in body mass following creatine supplementation. Thus, there may be an operational gender effect. Overall, it would appear that short-term creatine
supplementation may contribute to increased total body mass, although much of the increase in body mass may be attributed to water retention.

6.5.3. Creatine kinase (CK)

Plasma CK activity was not significantly different across the times of recovery nor between conditions, where the difference was small and did not reach the assigned level of significance. The CK data showed the changes in MVC after each loading condition was not significantly difference across the recovery time on all sessions due to high variability. These results are comparable to Rawson et al. (2001) who demonstrated no significant changes in plasma CK levels post-exercise following 5 days of creatine supplementation. Indeed the lower CK levels during recovery post-exercise may also be critical in minimising protein degradation and thus, muscle damage.

The mechanical and biochemical stress responses to exercise are complex and involve generation of reactive oxygen species and disruption to the contractile apparatus and cell membranes that ultimately contribute to fatigue by adversely impacting the metabolic and functional integrity of muscle. Serum CK concentrations were not significantly different during recovery points of the study during Cr supplementation. It would appear that Cr supplementation has no effect on muscle membrane integrity. During ingestion of the placebo, there was no change in the total CK response to exercise at 2 h, 24 h and 48 h post-exercise. Plasma CK continued to increase above pre-exercise values to 24 h post-exercise but started to decrease at 48 h of recovery. Although there are proportional changes in all conditions during recovery points, it is generally accepted that the creatine kinase concentration is not altered during exercise and does not interact significantly with Cr metabolism in the circulation. Another possible explanation for the lack of an
increase in CK could be relatively small amount of Cr supplementation or maximal exercise session versus a sub-maximal resistance exercise session.

6.5.4 Myoglobin (MYO)

Myoglobin was not significantly different across times of recovery nor for conditions, where the difference was small and did not reach the assigned level of significance. These results are similar to these study findings in older (Gotshalk et al., 2002) and younger (Volek et al., 1997; 1999) men and women. In the normal condition myoglobin should start rise within 1-3 hours of muscle damage and highest value by about 8-12 hours before returning to normal by about 24 h after muscle damage. In the results of this study myoglobin started to fall back by 24 and 48 hours recovery for all conditions and was greater in the Cr condition. Myoglobin was increased after post-exercise but was greater with the Cr condition. Much research has shown the beneficial effects of creatine supplementation on muscle force and power production over longer-term supplementation. Analyses included in this study do not easily explain performance differences observed between conditions, so it is possible that some other factor may be responsible. In general, myoglobin does not seem to be limiting during high intensity exercise (Volek et al., 2004). Although there are proportional changes in all conditions during recovery points, it is generally accepted that the myoglobin concentration is not altered during exercise and does not interact significantly with Cr supplementation metabolism. It is important to note that in this study, blood samples were taken as subjects reached a certain fatigue level, regardless of total exercise time. Establishing a similar intermittent high intensity protocol that enabled muscle samples to be taken after the same number of intervals in all conditions may give more insight into time-dependent changes in metabolites and myoglobin function.
6.5.5 Growth hormone (GH)

Growth hormone was significantly different across times of recovery and significant between conditions. Most studies have sampled pre-exercise, at some time intervals during exercise and at varying intervals post-exercise for up to several hours. Regardless of the sampling intervals or the resistance protocol, the pattern of GH release is similar. In all cases, the GH concentration peaks at or slightly after resistance exercise session and returns to baseline levels by 2 hours post-exercise. The data in this study was largely in agreement.

Concentrations of serum GH were higher in the control and placebo session post-exercise, than in the Cr group for all time points. Although the GH secretion needs to be considered when interpreting resting measures, resting GH concentrations have been shown to change significantly during the resistance exercise session and over the recovery period (Kraemer et al., 1999). The data showed that the level of GH was depressed at 2 hours of recovery but gradually increased to baseline level after 48 h. The decrease in plasma GH concentration was much greater in Cr session than the control and placebo sessions. Kraemer, et al. (1998) demonstrated a reduction in post-exercise concentrations of growth hormone even after 3 days of intense exercise.
6.6 Conclusion

In conclusion, this study investigated the effects of creatine supplementation (Cr) on maximal muscle strength following fatigue and recovery, and hormonal responses after high intensity (80% 1-RM) resistance exercise with females. The present study suggests that Cr supplement enabled subjects to develop more MVC and RFD, and there was evidence of Cr induced responses to the recovery period but did not reduce the effect of acute fatigue. This effect is not easily interpreted with regard to blood variables, as there was no change in CK and myoglobin across the recovery time but there was a change on GH levels that indicated the effect of Cr ingestion on muscle damage. Because hydration status and muscular activity after exercise were not different between groups, the blunted CK and MYO response was likely due to inactivation of CK activity before entering circulation. These data have clearly shown that creatine supplementation increases the effect of resistance exercise on maximal muscle strength following fatigue and the capacity to perform high-intensity exercise in females. These data suggest that oral creatine supplementation does not reduce muscle damage but enhances recovery following a resistance exercise challenge. These partially explain the increases in strength and improvements in exercise performance following oral creatine ingestion and has strong support as a nutritional strategy for females.
CHAPTER 7

GENERAL DISCUSSION
7.1 Introduction

Resistance exercise is widely used, not only in elite sports, but also in recreational and health related exercise as well as occupationally related training. Isometric tests have been used extensively to assess neuromuscular function although this test modality can be obtain information on ability of muscle. The rate of force development (RFD) has been quantified using isometric testing protocols (Bemben et al., 1992; Hakkinen, 1994; Linnamo et al., 1988). Resistance exercise programmes are part of most male athletic and health training programmes today, since regular exercise and physical conditioning may reduce fatigue accrued early in sports events. Strength of the female lower body is similar to that of men relative to body mass and lean body mass. Extensive investigations have been undertaken on the effects of exercise on physiological and biochemical profiles. Little attention, however, has been devoted to examining the effect of dietary supplementation on blood parameters and hormonal response to resistance exercise with female individuals.

Fatigue is a multi-dimensional and complex phenomenon that can originate from a large array of sources ranging from metabolic factors such as the accumulation of metabolites, impairment of neuromuscular and muscle damage (MacLaren et al., 1989; Pyne, 1994). In addition, no previous reports have compared blood parameters in different conditions over the fatigue and recovery period in response to resistance exercise with females. To effectively address these issues, this research has been divided into four parts: (1A) to establish the between-day reliability of the testing protocol, and (1B) to determine the degree of fatigue induced by resistance exercise and the within-day reliability of the testing protocol; (2) to examine the effects of heavy resistance exercise on fatigue and recovery during period of 48 hours post-exercise in females (3) to investigate the effect
of ingestion of carbohydrate on MVC during recovery after resistance exercise in females, and finally (4) to examine the effects of creatine supplementation on hormonal responses during fatigue and recovery after heavy resistance exercise in females.

7.2 Synopsis of findings

In chapter 3 the measurement and the reliability of MVC and RFD by using an isometric chair system in females across three days for each leg separately and for both legs together was assessed, it was found that the mean MVC for both legs was less than the right leg or left leg separately, and that the left leg was less than the right leg. Although previous studies (e.g. Tillman et al., 2004) have used either the dominant leg or each leg separately for MVC measurements, the MVC data for both legs were more reliable than MVC measured separately for each leg and so it is recommended that the MVC measurements are made using both legs instead of using each leg independently. However, for the RFD measurements the typical error in all three conditions (both legs, right leg and left leg) were similar and there was no difference in their reliability, when the three days data were compared. The between-days reliability reported in this study ranged from very good to moderate for isometric knee extension when using both legs to measure isometric contraction force than using each leg separately.

The reliability of within-day measurements was assessed in part two of this chapter. No change was found from session 1 to session 2 in MVC and RFD within a one-hour period, therefore it was concluded that there was no 'learning effect' on the measured variables or fatigue produced by the test protocol itself. The data therefore indicated that well familiarised subjects are competent at producing their perceived maximal force during a within-day protocol. The reduction in force following the exercise session was
due to some form of peripheral fatigue. These findings indicated that the level of fatigue in MVC following resistance exercise showed a greater reduction in MVC and RFD for both legs than the right and left leg separately. The force reduction achieved by the exercise was around 25-30% which did not reach the required 40% reduction. Therefore, in this chapter it was found that using both legs for studying the MVC was more reliable than using one leg. Furthermore, this allows for an experimental protocol which has both within-day and between-day reliability requirements.

In chapter 4 the effects of resistance exercise on MVC and RFD at each time interval of 2, 24, and 48 hours post-exercise were investigated. The results showed a significant effect of fatigue induced by resistance exercise for the both legs condition on MVC measured pre-exercise and post-exercise and at 24 hours but no significant difference at 48 hours of recovery. Similar results were found for the dominant leg except recovery appeared to have been achieved by 24 hours. Further, the RFD recovery appeared to have been achieved by 24 hours in the both legs conditions. The fatigue recovery of the both legs compared to the dominant leg condition is probably due to a weaker-non-dominant leg. The findings indicated that the level of fatigue in MVC and RFD percentage drop following the resistance exercise program was still found to be not enough for the desired 40% reduction, so further studies would need to use an intensity increased to 80% of 1-RM.

In chapter 5 the results demonstrated that when CHO was ingested both before and after exercise, performance was increased during recovery compared with the ingestion of a placebo. The results showed a significant effect of conditions and time on MVC measured pre-exercise, post-exercise, 24h and 48h. Pre-exercise MVC and RFD mean
values were similar when comparing carbohydrate (CHO) and carbohydrate free (CF) trials. However, recovery was achieved more rapidly in the CHO conditions and had fully recovery after 48h, while the CF condition required longer. As a result, the mean value of MVC and RFD after during recovery from exercise in the CHO trial was greater than that observed in the CF trial.

During high intensity resistance exercise, recovery periods play an important role in limiting fatigue and may improve physiological performance and also help to delay fatigue caused by dehydration. Therefore, it is not surprising that strategies have been developed to ensure that not only are the carbohydrate stores well stocked before exercise but that they are also restored as soon as possible 2 hours after a resistance exercise session. The restoration of muscle glycogen after depletion by exercise is a central component of the recovery process. To maximize the rate of muscle glycogen storage during short-term recovery, it is important to consume a carbohydrate supplement as soon after exercise as possible. In contrast, Mitchell et al. (1997) found that after a 2 days low CHO diet, resistance exercise performance was not affected. The authors concluded that although muscle glycogen level was low, there was adequate CHO to fuel the activity. Consuming carbohydrate immediately after exercise is known to increase the rate of muscle glycogen resynthesis and results in greater capacity during subsequent recovery periods.

The findings support Williams (2004) who found that the type of carbohydrate in the recovery diet also has an influence on endurance capacity the following day. It must be noted that the effect of a CHO supplement on females is purely an observation, and more precise methods are needed to describe a mechanism between macronutrient feeding and
performance. Therefore, in trained female individuals using the both legs condition, the presence of a CHO supplement resulted in greater recovery time adaptations after resistance exercise than a placebo supplementation.

In chapter 6 the immediate effect of creatine supplementation after high intensity (80% 1-RM) resistance exercise with female subjects showed Cr supplementation significantly improved muscle force recovery compared to a placebo. Thus, the current study demonstrated that creatine supplementation was able to reduce muscle damage caused by a resistance exercise session and may have also aided muscle recovery for female participants. Isometric contraction muscle strength was significantly higher during the recovery period from resistance exercise-induced muscle damage in the creatine supplemented trial compared to placebo trail. These results support the work by Santos et al. (2003), who also showed beneficial effect from creatine supplementation on indirect markers of muscle damage, suggesting that creatine supplementation is not only an effective strategy in maintaining muscle integrity during and after intense prolonged exercise, but it may also be successful at protecting muscle fibres from more damaging exercises as used in the present study.

The last study also examined the effects of Cr supplementation on some anthropometric and blood variables. One of these, the body mass, showed a non-significant mass gain of 1.0 kg in the Cr session following supplementation. Yet there was no significant gain in mass in the other conditions. This increase in body mass was indicative of the muscle 'loading' the excess Cr ingested through supplementation. The observed increase in body mass indirectly suggests that the Cr supplementation was effective. A similar assumption however, could not be made with respect to the female subjects that were supplemented
with dietary Cr, as it was not significant. This is similar to the findings of Bermon et al. (1998) who failed to observe a gain in mass in females Overall, it would appear that short-term creatine supplementation may contribute to increased total body mass, although much of the increase in body mass may be attributed to water retention. The effect of creatine supplementation on blood variables was also established. Plasma CK activity did not change significantly across times of recovery and feeding conditions. The differences were small and did not reach the assigned level of significance (P = 0.05) although did peak at 24h. These results are comparable to Rawson et al. (2001) who demonstrated no significant changes in plasma CK levels post-exercise following 5 days of creatine supplementation. At the start of exercise, the speed of this reaction will be close to maximum, and therefore to increase the substrate (phosphocreatine) concentration further will not affect the speed of this reaction and consequently maximum force output and short-term of resistance exercise performance. However, creatine supplementation improved performance and recovery from fatigue in the supplemented group than placebo. As performance was improved, it seems possible that even 20 g creatine per day for 5 days may raise the muscle creatine content, thus providing a mechanism for the improvement. Therefore, the mechanism of improvement in the recovery period in the present study may have been that creatine concentration was maintained above the K - value for the creatine kinase reaction, thereby increasing the rate of phosphocreatine resynthesis (Greenhaff et al., 1994). The improvements in the test may additionally have been caused by improved buffering through the increase in muscle creatine. Adenosine triphosphate resynthesis from ADP and phosphocreatine consumes a hydrogen ion (H+) in the process. An increase in phosphocreatine turnover rate through greater creatine content in the muscle will therefore consume more H+ and
improve muscle buffering capacity. However, in the present study, the changes in CK were similar with or without supplementation during recovery period time.

Growth hormone was significantly different across times of recovery and all conditions, following resistance exercise-induced muscle fatigue. Most studies have sampled pre-exercise, during exercise and at varying intervals post-exercise for up to several hours. Regardless of the sampling intervals or the resistance protocol, the pattern of GH release is similar. In all cases, the GH concentration peaks by 45 minutes post-exercise. The GH data showed the mean percentage changes in MVC and RFD after each loading conditions. Concentrations of serum GH were higher in the control and placebo sessions, post-exercise, than in the Cr session. Although the GH needs to be considered when interpreting resting measures, resting GH concentrations have not been shown to change significantly over the recovery period.

It has been suggested that creatine has an indirect anabolic effect. In this comparative cross sectional study, (Schedel et al., 2000) a significantly higher growth hormone level was observed after acute creatine loading (20g). The peak plasma values of growth hormone were generally obtained immediate after resistance exercise session on control and placebo conditions higher than creatine condition. In this study, venous blood was sampled before, immediately after and 2 h, 24 h and 48 hours after the training session, but no measures were done after 27 hours creatine administration, when the potential indirect anabolic effect of creatine is higher. Therefore, this mechanism is not clear at present and further research is warranted.

The last variable measured was myoglobin. It was not significantly different for MVC across times of recovery and between conditions. These results are similar to the findings
in older (Gotshalk et al. 2002) and younger (Volek et al. 1997, 1999) men and women. There was no significant difference between conditions, but the difference was greater in Cr condition although did not reach the assigned level of significance (P = 0.5). In the normal condition, myoglobin should start to rise within 1-3 hours of muscle damage and highest value by about 8-12 hours and fall back to normal by about one day after muscle damage occurred. In this study myoglobin started to fall back by 24 and 48 hours recovery with all conditions but it was greater in the Cr condition and when compared across conditions.

In summary, the aim of this thesis was to examine the effect of dietary supplementation on fatigue and recovery in athletically trained females following short-term and high intensity resistance exercise. The result of this thesis clearly demonstrate that short-term resistance exercise for the development of strength produces fatigue but the recovery is quicker when nutritional supplements are used by females. Ingestion of CHO allowed full recovery after 48 hours, quicker than the placebo but more precise methods are needed to describe a mechanism explaining this performance. Creatine supplement also improved muscle force recovery compared to a placebo. Blood variables were established and although plasma CK and Myoglobin did not change significantly across times of recovery, growth hormones did change significantly across the times of recovery following an expected pattern.
7.3 Conclusion

It is concluded that the recovery from heavy resistance exercise in women appears to be added by dietary supplementation producing an increase in the recovery of both maximal voluntary contraction force and rate of force development. The same procedure of resistance exercise was used in the four experimental studies and nutritional supplementation significantly reduced the decline in maximal peak force following resistance exercise. The benefit of CHO feeding is that it is immediate, while using Cr as a supplement requires a prolonged period of ingestion and is associated with a small gain in body mass. Following short-term resistance exercise with a high intensity of 80% of 1-RM, a statistically significant difference in favour of the experimental groups was detected.
7.4 Limitations

The following are potential limitations of this study:

1. The results of this study may only be generalized to females of similar age and training status as defined by the subject sample employed her.

2. Although exercise sessions were given, the subjects’ lack of familiarity with exercise training may have influenced their effort during the isometric testing, one repetition maximum testing, and eccentric resistance exercise.

3. The timing of blood draws was pre-, post-exercise, 2 hr, 24 hr and 48 hours post-exercise on day 5. Therefore, the response of serum CK and MYO to the Cr supplement is limited to these time points. More frequent sampling was not possible due to technician availability during Cr supplementation, and subjects’ work schedules.

4. A dietary baseline period was limited to five days and may have affected baseline blood and urine measurements.

5. No biochemical assessments of nutritional status were performed on the subjects prior to the start of the experimental period.
7.5 Recommendation for future work

From conducting series of experimental studies in the present thesis and from reviewing the literature, the researcher recommends the following directions for future work in the area of dietary supplement with resistance exercise on females subjects.

- Establishing the biomechanical responses to CHO supplementation after resistance exercise would provide more complete data to support study 3, in which it was not possible to collect this data.

- Examination of the effects of a low and high carbohydrate status in terms of muscle glycogen level, before and after resistance exercise as this is thought to increase the rate of muscle glycogen resynthesis and results in greater capacity during subsequent recovery periods. This may lead to a substantial increase in performance and recovery after high CHO loading over 24h of recovery.

- It would be important to investigate the effect of using CHO together with Cr supplement on fatigue and recovery responses in female subjects after resistance exercise.
CHAPTER 8

REFERENCES
References:


APPENDICES
SUBJECT INFORMATION SHEET

Title:

Effects of muscular strength and biochemical responses to resistance exercise in females.

Investigator:

Majda Taher Touba

Supervisors:

- Professor. Adrian Lees (Director of Studies)
- Professor. Don MacLaren (Supervisor)

Department:

School of Sport and Exercise Science Liverpool John Moores University
Aims of the study:

The overall aim of the research programme is to determine the effects of dietary supplementation on muscular strength fatigue and recovery and biochemical responses to resistance exercise in females.

To achieve this aim experiments will be designed to fulfil the following objectives:

1. To establish the reliability of measurements of maximum voluntary isometric force and rate of force development in females.
2. To establish the intensity of exercise required to produce a fatigue in MVC in females.
3. To characterise the fatigue and recovery responses to heavy resistance exercise in females in terms of isometric force and blood variables.
4. To examine the effects of carbohydrate supplementation on the neuromuscular and biochemical responses to heavy resistance exercise in females.
5. To examine the effect of creatine supplementation on the neuromuscular and biochemical responses to heavy resistance exercise in females.

Pre-test procedure:

- The experimental protocol and testing procedure will be fully explained.
- The correct technique of all exercise will be explained and shown by researcher before the subjects practice the correct technique.
- Habituation and familiarizations sessions will be undertaken; these are orientation and practice sessions.
• Tests will establish you one repetition maximum (1-RM) for six exercises including lower body part.

• Do not engage in vigorous exercise and physical activity for 24 hours before tests.

• Warm-up and stretching for five minutes will be carried out on upper and lower body parts.

Experimental protocol:

Muscular strength measurements:

Muscular strength indices will be measured using the isometric chair Data on voluntary maximal peak force, rate of force development and rate of force decline will be generated. All measurements will be ascertained before and after a session of resistance exercise for lower body extremity at an intensity corresponding to 70% of 1 repetition maximum (1RM). Measurement will also be obtained 2h, 24h and 48h after the completion of the exercise protocol in order to assess fatigue and recovery characteristics, the possible effects of dietary manipulations on muscular strength.

Resistance exercise session:

All subjects will perform three sets of six different exercises (lying leg curls, dumbbell lunges, Seated Calf Raises, leg extensions, straight leg dead lift, leg presses) involving lower body part at an intensity corresponding to 70% 1RM (8-10
repetition). One-minute rest period will be allowed between exercises and 3 min rest period will be allowed between sets.

Anthropometrics measurement (age, weight and height) will be obtained before exercise; 2 ml of finger blood will be obtained before, immediately after exercise and recovery.
Liverpool John Moores University Strength Muscle Laboratory
Pre-test Questionnaire
Physical Activity Readiness Questionnaire (PAR-Q)

This questionnaire is designed to help you. For your health and safety, please answer the following questions and inform the experimenter of any factors that might affect your performance in the test. It is important to ensure that you are in a fit and healthy state to complete the exercise test, because testing involves strenuous activity. If there are any questions you don’t understand you should ask the experimenter for clarification.

All information you provide will be treated with the strictest confidence

I am interested in participating in the "strength training programme and test"

Name
Age
Date of Birth
Address
Telephone No.
Marital Status
Liverpool John Moores University  
Health and Human Science  
Pre-Test Health History Questionnaire

ANY INFORMATION CONTAINED HEREIN WILL BE TREATED AS CONFIDENTIAL

Please tick ✓ appropriate box (Yes or No)

Has your Doctor ever said you have?

<table>
<thead>
<tr>
<th></th>
<th>YES</th>
<th>NO</th>
</tr>
</thead>
<tbody>
<tr>
<td>High blood pressure / any heart trouble</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Disease of arteries</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Varicose veins</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lung disease</td>
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<tr>
<td>Asthma</td>
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<tr>
<td>Liver disease</td>
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<tr>
<td>Kidney disease</td>
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<tr>
<td>Diabetes</td>
<td></td>
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<tr>
<td>Epilepsy</td>
<td></td>
<td></td>
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<tr>
<td>Any blood clotting disorders</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Any other medical problems:

____________________________________________________________________________

____________________________________________________________________________

____________________________________________________________________________
Do you currently smoke?
Yes □ No □

If yes, how much per day? ________________

Have you ever given up smoking? ________________

For how long did you smoke? ________________

How many caffeinated beverages do you consume per day?
What type? Coffee (cups) □ Tea (cups) □ Other □

How would you describe your state of well being at this time?
(Please tick one).

Excellent □
Very good □
Good □
Poor □
APPENDIX 2

FORM OF CONSENT TO TAKE PART AS SUBJECT IN RESEARCH PROJECT
Project title:

The effect of dietary supplementation on muscular strength and biochemical responses to resistance exercise in females.

I, ................................................. agree to take part in the above named (Subject full name) Project/Procedure, the details of which have been fully explained to me

Signed ..................................... Date..............................................

I, ................................................. certify that the details of this project/Procedure (Investigator’s full name) Have been fully explained and described in writing to the subject name above and have been understand by him/ her.

Singed .....................................Date..............................................

I, ................................................. certify that the details of this project/Procedure Have been fully explained and described in writing to the subject name above and have been understand by him/ her.

Singed .....................................Date..............................................

NB: The witness must be an independent third party.
APPENDIX 3
Participant personal details (female)

Name: 
Address: 
Date: 

Date of birth: 
Contact number: 

Have you ever had? 

Yes 
No 

Knee joint injury 

☑️ ☐ how long ago? 

Quadriiceps muscle injury 

☑️ ☐ how long ago? 

Hamstring injury 

☑️ ☐ how long ago? 

Other 

☑️ ☐ how long ago? 

Have you ever suffered from? 

Yes 
No 

Arterial hypertension? 

☑️ ☐ 

Chronic fatigue syndrome? 

☑️ ☐ 

Loss of sleep, sleep deprivation, insomnia? 

☑️ ☐ 

Did you practice any sport(s)? 

Yes 
No 

Which one(s)? 

-------------------------------

How many times: 

A day ----------- a week ----------- (including competitive events)?

At which time(s) of day? -------------------------------

Do you avoid practising sport or physical activity at which period(s) of your menstrual cycle? 

During your period? 

☑️ ☐ 

Just after your period? 

☑️ ☐ 

Just before your period? 

☑️ ☐ 

Other? 

☑️ ☐ 

Do you prefer practising sport or physical activity at which period(s) of your menstrual cycle? 

During your period? 

☑️ ☐ 

Just after your period? 

☑️ ☐ 

Just before your period? 

☑️ ☐ 

Other? 

☑️ ☐ 

Are you taking any contraceptive or hormonal pills? 

Yes 
No 

If yes, which one(s)? 

☑️ ☐ 

☑️ ☐ 

An appointment will be made for you to undertake the testing protocol at a suitable time for you.
APPENDIX 4
EXERCISE TESTING FORMS
Name:

Date of first measures:
Date of second measures:
Date of third measures:
Date of fourth measures:

<table>
<thead>
<tr>
<th>Blood Variable</th>
<th>First session</th>
<th>After 2 hours</th>
<th>After 24 hours</th>
<th>After 48 hours</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Pre-1 Exercise</td>
<td>Pre 2 Exercise</td>
<td>post-Exercise</td>
<td></td>
</tr>
<tr>
<td>Haemoglobin</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Haematocrit</td>
<td></td>
<td></td>
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<tr>
<td>Creatine Kinas</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Growth hormones</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Myoglobin</td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>
Body Weight and Anthropometric Data

Subject: ____________
Age: ____________ lb
Date: ____________

Height: _______ in.
_________ cm _______ m
Weight: _______ kg.

<table>
<thead>
<tr>
<th>Day/Pre-test</th>
<th>Weight (Kg)</th>
<th>Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
<td></td>
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<tr>
<td>2</td>
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<tr>
<td>12</td>
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</tr>
</tbody>
</table>
APPENDIX 6
Isometric Contraction Exercise Protocol

Subject: __________________  Age: _______  Weight: __________
Date: _________________  Time: _______

<table>
<thead>
<tr>
<th>Set</th>
<th>Repetitions</th>
<th>Time Begin</th>
<th>Time End</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
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<td>3</td>
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<tr>
<td>12</td>
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<td></td>
</tr>
</tbody>
</table>
Exercise program

Set (1)

exercise(1) (1min) exercise (2) (1min) exercise (3) (1min)
exercise (4) (1min) exercise (5) (1min) exercise (6)

Rest (3min)

Set (2)

exercise(1) (1min) exercise (2) (1min) exercise (3) (1min)
exercise (4) (1min) exercise (5) (1min) exercise (6)

Rest (3min)

Set (3)

exercise(1) (1min) exercise (2) (1min) exercise (3) (1min)
exercise (4) (1min) exercise (5) (1min) exercise (6)
One-Repetition Maximum (1-RM) Protocol

Subject: ___________  Age: _______  Weight: ___________

Date: ___________  Time: _______  Dominant Leg: R or L

<table>
<thead>
<tr>
<th>Weight</th>
<th>Repetitions</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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<td></td>
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</tbody>
</table>

1-RM: 
80% 1-RM: 

(Resistance exercise)
APPENDIX 8
Borg Rating of Perceived Exertion Scale

6

7 Very, Very Light

8

9 Very Light

10

11 Fairly Light

12

13 Somewhat Hard

14

15 Hard

16

17 Very Hard

18

19 Very, Very Hard
**Food Record – Day 1**

Name ___________________________ Day/Date ___________________________

<table>
<thead>
<tr>
<th>Serving Size / Food &amp; Beverage Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breakfast</td>
</tr>
<tr>
<td>Lunch</td>
</tr>
<tr>
<td>Dinner</td>
</tr>
<tr>
<td>Snacks</td>
</tr>
</tbody>
</table>

Estimated Daily Water intake = ______ ml/ounces/cups

Was this a typical day’s intake? (Y/N. If no, please explain.) ___________________________

Proceed to Food Record Day 2
Food Record – Day 2

<table>
<thead>
<tr>
<th>Serving Size / Food &amp; Beverage Description</th>
<th>Time of Day: AM/PM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breakfast</td>
<td>AM/PM</td>
</tr>
<tr>
<td>Lunch</td>
<td>AM/PM</td>
</tr>
<tr>
<td>Dinner</td>
<td>AM/PM</td>
</tr>
<tr>
<td>Snacks</td>
<td>AM/PM</td>
</tr>
</tbody>
</table>

Estimated Daily Water intake = ______ ml/ounces/cups

Was this a typical day’s intake? (Y/N. If no, please explain.)

Proceed to Food Record Day 3
### Food Record – Day 3

<table>
<thead>
<tr>
<th>Serving Size / Food &amp; Beverage Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Breakfast</strong></td>
</tr>
<tr>
<td><strong>Lunch</strong></td>
</tr>
<tr>
<td><strong>Dinner</strong></td>
</tr>
<tr>
<td><strong>Snacks</strong></td>
</tr>
</tbody>
</table>

Estimated Daily Water intake = _____ ml/ounces/cups

Was this a typical day's intake? (Y/N. If no, please explain.) __________________________