INFLUENCE OF AVERAGE PEDALLING RATE UPON THE MAGNITUDE OF THE MECHANICAL AND BIOCHEMICAL CHANGES ARISING FROM HIGH-INTENSITY EXERCISE.

by

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CONTENTS

SUMMARY

The process of fatigue during 30 s of high-intensity exercise results in rapid and substantial mechanical, electrical and biochemical changes in muscle fibres. This thesis describes a series of experiments, performed upon a friction-loaded cycle ergometer, which investigated whether the magnitude of the mechanical and biochemical changes is affected by changes in average pedalling rate. The ability to generate peak power in a subsequent sprint of 6 s duration was used to assess the magnitude of the mechanical changes. Changes in the concentrations of blood and muscle metabolites pre- and post-exercise permitted some of the biochemical changes to be measured.

The results demonstrated that the ability to generate peak power 1s and 3s after performing 30s of high-intensity exercise was dependent upon the pedalling rate at which prior high-intensity exercise was performed; the ability to generate peak power being greater following exercise at high, as opposed to low, pedal rates. However, there was an initial, rapid and substantial recovery of power output which was independent of pedal rate, so much so that peak power output recovered to 60-65% of the unfatigued value (from 40%) within 6s of completing high-intensity exercise. Thereafter, until 300s of recovery had elapsed, peak power output continued to recover independently of prior pedal rate, albeit at a much slower rate compared with the initial seconds of recovery. The magnitude of the changes in the concentration of selected blood and muscle metabolites was also independent of pedal rate.

The differential recovery of power output in the first 3s after performing high-intensity exercise suggests that the extent of the mechanical changes is affected by the pedal rate at which such exercise is performed. The muscle metabolite data were unable to suggest a mechanism for this differential recovery of power. Also the initial recovery of peak power output appears too rapid to be due primarily to the restitution of muscle metabolites. It is speculated that the rapid phase of recovery represents the reversal of changes which took place during exercise in the excitation-contraction coupling system. The initial differential recovery of power may be the result of a greater $[K^+]_{extra}$ and depolarization at fast, compared with slow, pedalling rates. The slower phase of recovery is probably closely allied to the resynthesis of muscle metabolites.

The experiments described herein have been reported elsewhere and the following is a list of relevant publications and conference presentations:-

Cherry, P.W., Fletcher, R.J., Lakomy, H.K.A. and Nevill, M.E. (1996). All-out and even paced strategies: a comparison of the performance and metabolic effects of maximal constant work exercise lasting 30 s. *Journal of Sports Sciences,* **14(1),** 70.

Cherry, P.W., Lakomy, H.K.A., Nevill, M.E. and Maddox, N. (1996). Effect of fatiguing high intensity exercise on subsequent peak power output. Paper presented at *"Le Premiere Congres Annuel du College Europeen de Sciences du Sport"* Nice, France, May 28-31 1996.

Cherry, P.W., Lakomy, H.K.A., Nevill, M.E. and Fletcher, R.J. (1997). Constant external work cycle exercise - the performance and metabolic effects of all-out and even-paced strategies. *European Journal of Applied Physiology,* **75,** 22-27.

Cherry, P.W., Lakomy, H.K.A., Maddox, N.L. and Nevill, M.E. (1997). Effect of the preceding number of muscle actions on subsequent peak power output. *Journal of Sports Sciences,* **15(2),** 201-206.

Cherry, P.W., Lakomy, H.K.A., Boobis, L.H. and Nevill, M.E. (1997). Rapid recovery of power output in females. *Acta Physiologica Scandinavica* (Submitted for publication).

Cherry, P.W., Lakomy, H.K.A., Boobis, L.H. and Nevill, M.E. (1997). Rapid recovery of power output following maximal exercise. Paper presented at *"The 10th International Biochemistry of Exercise Conference"* Sydney, July 15-19 1997.

Cherry, P.W., Lakomy, H.K.A., Nevill, M.E. and Boobis, L.H. (1997). Influence of pedalling rate upon the magnitude of the biochemical and mechanical changes resulting from maximal exercise. Paper to be presented at *"The Second Annual Congress Of The European College Of Sports Science"* Copenhagen, August 20-23 1997.

Cherry P.W. (1997). Factors influencing the short-term recovery of power output. Abstract accepted for *"The Achievements In Physiology and Medicine for the Future of Sport Science"* Kaunas, September 25-26 1997.

CHAPTER 1

GENERAL INTRODUCTION

A widely accepted definition of fatigue is, "the failure to maintain the required or expected power output" (Edwards, 1983). In sport and exercise, fatigue manifests itself at the point when the athlete can no longer maintain a particular exercise intensity and so has to reduce the pace. The point at which this occurs will depend upon the nature of the activity. Athletes competing in events relying heavily on endurance, such as a marathon or the Tour de France, will not have to slow the pace for some minutes or even hours. This is because to complete the race in the fastest time, a sustained sub-maximal intensity of exercise needs to be maintained. At the other extreme, some events such as weightlifting require a comparatively short burst of maximal, or near maximal, effort and fatigue can quickly manifest itself. In between these two extremes are many other sports, particularly field games such as hockey and soccer, where intermittent bursts of high-intensity exercise are separated by longer periods of lower intensity exercise. In these sports, requiring multiple bursts of high-intensity exercise (Williams, 1990), the recovery from fatigue is vital for the maintenance of performance. Although during exercise many changes take place concurrently, they recover at different rates after exercise. Therefore, the study of their pattern of recovery may permit a greater understanding of the factors which produce fatigue (McComas, 1996).

The process of fatigue during intense muscle stimulation results in rapid and substantial mechanical, electrical and biochemical changes. Thus, following a period of high-intensity exercise such as a 30 s "Wingate Cycle Test" (Bar-Or, 1978), some of the changes arising during exercise may be expected to be reversed quickly afterwards. If this is so, some important changes are likely to take place in the early stages of recovery, particularly in the first few seconds. Therefore, it is surprising that the ability to generate power output in the first few seconds after high-intensity dynamic exercise, as a means of assessing the extent of the mechanical changes, has only recently been investigated (Buttelli *et al.*, 1996). However, the short-term recovery (approximately 2-3 s post-exercise) of maximal power output following submaximal exercise has been investigated (Beelen and Sargeant, 1991; 1993). An interesting finding of these studies was that the recovery of power was influenced by the average pedalling rate at which prior exercise was performed. It was shown that greater fatigue,

reflected by a lower peak power output in a maximal sprint, arose following prior exercise at high as opposed to low pedal rates. Clearly, if the recovery from high-intensity exercise is also influenced by the pedal rate this may be reflected by a differential rate of recovery of power output and possibly blood and muscle metabolites in the first few seconds after exercise.

Thus, the purpose of this thesis is to assess whether the extent of the mechanical changes which take place in the recruited muscle fibres during high-intensity exercise is influenced by the average pedalling rate adopted. The term "average pedalling rate" as opposed to simply "pedalling rate" is appropriate because during a 30 s cycle sprint, it is likely that there will be a steady decline in pedalling rate as exercise takes place. Also, within each pedal cycle there will be variation in the external angular velocity. The reader should note that occasionally in this thesis, to avoid repetition, the word "average" is dropped. The changes in the concentration of selected blood and muscle metabolites resulting from high-intensity exercise at different average pedalling rates may suggest a mechanism for any differences in the extent of the mechanical changes that arise. The thesis is comprised of seven further Chapters.

- Chapter 2 reviews the literature. The mechanical, electrical and biochemical changes associated with high-intensity exercise are outlined. The possible effects of manipulating the pedal rate upon the magnitude of these changes are explored. The Chapter concludes with the experimental hypotheses.

- Chapter 3 entitled "General Methods" describes the equipment and procedures used in more than one of the experimental studies. It also includes details of a study where the aim was to assess whether females had a greater variation of performance during short-term high-intensity exercise compared with males. This aspect was considered important because of the hormonal changes associated with the female menstrual cycle.

Chapters 4-7 contain details of the studies which have been performed to test the hypotheses.

- The aim of Chapter 4 was to describe some of the metabolic responses to, and the effect on the short-term (2-3 s) recovery of power output, of constant external work exercise performed with an all-out or paced strategy. The

Chapter contains details of a minor pilot study as well as a more extensive main study.

- The second main study (Chapter 5) considered in more detail how the short term recovery of power output following high-intensity exercise is influenced by the pacing strategy and average pedalling rate adopted.

- The third main study (Chapter 6) describes the changes in the concentration of certain muscle and blood metabolites when 30 s of maximal exercise is performed at different average pedalling rates.

- The fourth and final main study (Chapter 7), investigated the pattern of recovery of power in more detail (up to 300 s post-exercise) following maximal exercise where average pedalling rate has been manipulated by altering the applied resistance.

- Chapter 8 draws together the findings from the experiments, discusses their implications for sport and exercise and suggests possible areas for further research.

CHAPTER 2

REVIEW OF LITERATURE

2.1. INTRODUCTION

This Chapter begins with a brief overview of the factors which may give rise to fatigue and highlights those which have been cited as being the most likely causes of the decline in power which occurs during high-intensity exercise. The majority of the remainder of the Chapter is divided into three main sections relating to the mechanical, electrical and biochemical events associated with fatigue during high-intensity exercise. It must be emphasised that, to some extent, the partitioning of changes into three discreet categories is merely for clarity of presentation. Each section should not be considered to be an independent entity separate from the other two and some individual changes such as the ionic shifts of potassium, sodium and other ions, could arguably be placed in more than one section. In each section, the nature and extent of the changes are assessed. Consideration is given to their possible role in the fatigue process and whether their magnitude may be influenced by manipulating the average pedalling rate at which exercise is performed. The mechanisms by which the changes are reversed during recovery are also reviewed and how they may be influenced by changes in average pedal rate. Wherever possible reference is made to human data although in some instances the only available data are from experiments on non-human muscle. The Chapter concludes with the experimental hypotheses.

2.2. OVERVIEW OF FATIGUE AND HIGH-INTENSITY EXERCISE.

In this thesis the term 'high-intensity exercise' refers to exercise at an intensity at, or very close to, the maximal which can be maintained for a short period of time, typically 30 s. This can result in external power outputs two to four times those elicited at maximal oxygen uptake (Spreit, 1995). This point is very relevant because the term is also used by many authors when referring to a sustained period of exercise at, or below, a power output required to elicit maximal oxygen uptake (Carnevale and Gaesser, 1991). Often the term 'supramaximal' is used to describe short lasting high-intensity exercise (Gastin *et al.*, 1995; Jacobs *et al.*, 1983). High-intensity exercise is performed in many sports and athletic events. Less obviously perhaps, many sedentary people also

regularly perform this type of exercise as they run for the bus or climb a steep flight of stairs (Williams, 1987). Therefore, it is vital that the underlying mechanisms causing fatigue are well understood.

Voluntary muscular contraction is a complex series of events and fatigue can occur at various stages in the chain from the brain to force generation. A distinction is usually made between central fatigue, where the impairment is located in the central nervous system and peripheral fatigue, where the impairment is located in the peripheral nerve or contracting muscle (Sahlin, 1992). Thus, the cause, or causes, of fatigue could be due to a partial, or total failure at any stage, or stages (Fig. 2.1). Clearly, the underlying mechanisms for fatigue will depend upon the task which the muscles have to perform. It is unlikely that the changes or failures which can be regarded as causing fatigue will be the same for athletes competing in events as diverse as the marathon and the 100 m.

Central Fatigue Is Not A Contributing Factor To The Fatigue Arising From Maximal Stimulation Of Short Duration.

A number of methods have been used to establish, firstly, whether human subjects are able to recruit all motor units during a voluntary movement and, secondly, whether they are able to fire at their optimum frequency for maximal force development. One method has been to compare the force of a maximal voluntary contraction (MVC) with that which could be attained by tetanic stimulation of the of the same muscle. Using such a technique on the adductor pollicis muscle, it has been shown in well motivated and practised subjects, that the force output resulting from both voluntary and brief periods of tetanic stimulation lasting 40-120 s run in parallel (Bigland-Ritchie *et al.*, 1982; 1983). This indicates that all motor units can remain fully activated throughout a sustained voluntary contraction.

Tetanic nerve stimulation can be extremely painful and an alternative technique used to investigate the extent of motor unit activation during effort is that of twitch interpolation. This method relies on the fact that if any motor units are not fully activated during the course of a strong contraction, then stimulation should give an additional twitch response. Using this technique, it was found that during extreme effort, full activation of motor units was achieved easily in tibialis anterior during dorsi-flexion of the ankle but only with difficulty in the plantar flexors (Belanger and McComas, 1981). The same

method, that of twitch interpolation, has been attempted during a maximal effort involving a more complex movement, namely, isokinetic cycling (Beelen *et al.*, 1995). However, the authors reported that they were unable to detect single twitches or trains of high frequency stimulation reliably. Nevertheless, they were able to compare the recovery of maximal force following a 25 s maximal effort under either voluntary or electrically stimulated (via percutaneous electrodes) conditions. In agreement with the findings using other methods, their results found no difference between the voluntary and electrically stimulated force.

According to James *et al.* (1990), whilst it cannot be argued that central fatigue is never a cause of the loss of performance, the desire to reduce activity can be largely overridden. Thus, the evidence seems clear that central fatigue is not important to the fatigue arising during the performance of high-intensity exercise performed over a short duration (<2 minutes).

Many Peripheral Stages Are Not Regarded As The Primary Causes Of Fatigue

Insofar as the peripheral factors are concerned, when muscle is maximally activated, either voluntarily or following physiological rates of stimulation, the reduction in force begins when there is a failure of a cellular mechanism subsequent to the muscle fibre action potential; that is, the failure must either involve excitation-contraction (E-C) coupling or the contractile machinery (McComas, 1996). The extent to which a failure of these two processes, E-C coupling or the mechanical performance of the myofibrils, is the primary cause of fatigue has been debated for many years.

The first evidence to indicate that it may be a failure of E-C coupling was provided by Eberstein and Sandow (1963) who showed that a fatigued muscle was able to achieve almost maximal force when subjected to caffeine. Lannergren and Westerblad (1989) also found that where the normal activation process was by-passed by application of caffeine, that fatigued single fibres were able to generate tensions similar to controls. A muscle fibre cannot shorten below its slack length unless it is activated and further evidence suggesting fatigue is due to a failure of processes connected with the E-C coupling system has come from experiments which have looked at the appearance of the myofibrils. Interestingly, the results from some such experiments have found that the peripheral layers of the fibre are straight, suggesting that they have shortened actively, whilst the central myofibrils have

a wavy appearance (Edman, 1992). These findings suggest that fatigue is due to a failure of the inward spread of activation into the muscle fibre.

Conversely, there is also ample evidence that the cause of fatigue lies within the contractile apparatus. The results of some studies have shown no increase in tetanic force with caffeine indicating that the failure lies somewhere in the contractile machinery (Edman and Lou, 1990). To establish whether the decrease in force during fatigue is due to a decrease in the number of cross-bridges formed or a reduction in the force produced by each cross-bridge, the measurement of muscle stiffness has been used (Edman, 1992). Muscle stiffness can be presumed to reflect the number of attached cross-bridges (Ford *et al.*, 1977) but was only reduced by 9 % in fatigued fibres compared with the resting value despite a much larger (25 %) depression in tetanic force (Edman and Lou, 1990). These results suggest that although there may be a slight decrease in the number of cross-bridges, the most important cause of the force decline during fatigue is reduced ability of the individual cross-bridge to produce force. Thus, the muscle stiffness experiments indicate that fatigue is due to altered kinetics of cross-bridge function with no significant change in the state of excitation of the contractile system (Edman, 1992).

According to Edman (1996), failure of the inward spread of activation is only important during intense stimulation of muscle and moderate levels of fatigue are the result of a failure in the cross-bridge machinery. The important, as yet, unanswered question is whether voluntary effort can result in a sufficient degree of stimulation to result in fatigue caused primarily by a failure of activation. It is easy enough, of course, to stimulate an isolated muscle preparation until some aspect of the E-C system, such as a failure of activation to the central myofibrils, fails, but it remains uncertain if this stage is ever reached *in vivo* (Edman, 1992). The model of high-intensity exercise performed by well trained subjects may provide the answer.

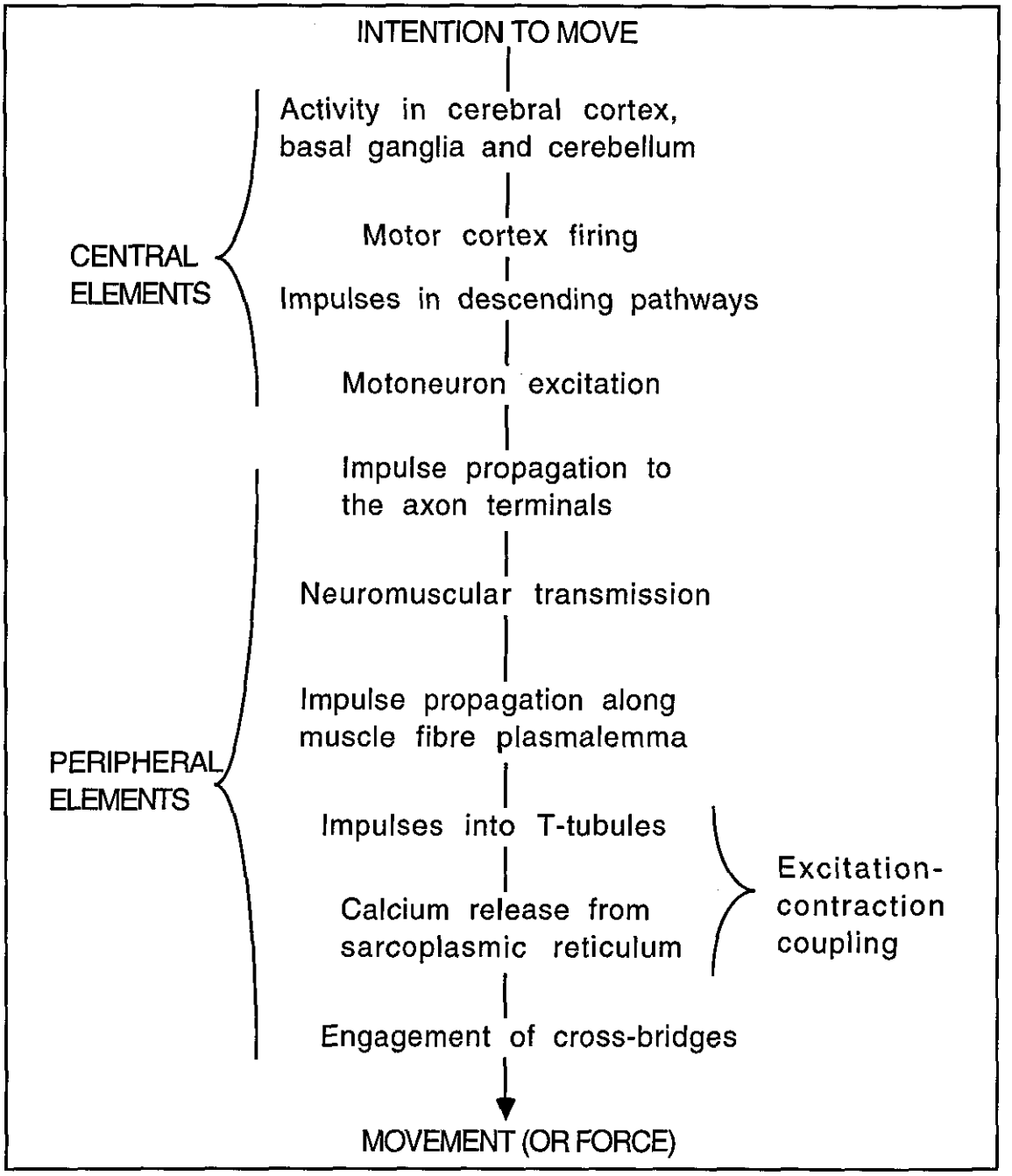

Fig. 2.1. Successive stages in the production of a voluntary movement (in McComas, (1996)).

2.3. CHANGES ARISING FROM HIGH-INTENSITY EXERCISE

2.3.1. MECHANICAL CHANGES

2.3.1(a). Mechanical Properties Of Unfatigued Muscle

For the performance of high-intensity exercise which requires the maintenance of high power outputs, the muscle has to have the functional ability to generate the necessary force and velocity. Peak force and power output of a muscle depend upon numerous factors but the most important are muscle size, force per cross sectional area, the peak rate of force development (dP/dt) and the maximal speed of shortening (V_{max}) (Fitts, 1992). These factors vary considerably for different fibre types and the force-velocity characteristics of mixed muscle and single fibres have been reported in the literature.

Historical Review-The Classic Force-Velocity Relationship Of Muscle

A.V. Hill (1922) came to the conclusion that a muscle could be represented mechanically by a spring working in a viscous medium. As the speed of shortening increased, so the tension exerted decreased according to the following equation:-

$$W=W_O - kv \qquad \text{(Hill, 1922)}$$

Where :-

W is the work at a specified velocity
W_O is the work at zero velocity i.e. an isometric action
v is the velocity of shortening
k is a constant

Two points are apparent from this equation. Firstly it describes muscle shortening and is thus restricted to concentric muscle actions. Secondly, it is a straight line equation where the gradient is given by -k, suggesting that force is inversely and linearly related to the velocity of a concentric action.

At around the same time, W. O. Fenn was conducting experiments relating the work performed and heat liberated during muscle shortening (Fenn, 1923). In these experiments, it was found that the rate of heat liberation was higher in a

muscle undergoing shortening compared with one developing isometric tension. This phenomenon is known as the "Fenn Effect". During an isometric action, the muscle does no work as the distance moved is zero; all the energy is liberated as heat. Figure 2.3(b) which is taken from Bagshaw (1993) shows the rate of energy liberation (heat + work) and compares this with power output (work rate) as a function of isotonic shortening velocity. Fenn and Marsh (1935) suggested an equation which related force to velocity in an exponential fashion and concluded that this relationship was connected in some way with the processes associated with developing extra energy for the work of shortening. However, at that time they could not explain these observations. The Fenn effect is allied with the concept of mechanical efficiency which was represented by Hill (1922) in the following equation:-

$$E = W / H$$

(Hill, 1922)

Where:-
E is the mechanical efficiency
W is the external work done
H is heat production

It is more common today to express mechanical efficiency in terms of energy rather than heat production:-

$$E=We/(Et-Er)$$

(Bangsbo, 1996a)

Where:-
E is the mechanical efficiency
e is the external mechanical work
Et is the total energy consumption
Er is the resting energy consumption:-

The influence of speed on mechanical efficiency was reviewed by Cathcart *et al.* (1924) who concluded that when an amount of work was performed, mechanical efficiency was low with very fast and slow rates of movement and high with medium rates. A few years later the mechanical efficiency of bicycle pedalling at different rates of shortening was investigated (Dickinson, 1929). At

first glance, a rather more complex equation was used to define the mechanical efficiency of a muscle movement:-

$$E = 1-(k/t)/a(1+bt)$$

(Dickinson, 1929)

Where :-

E is the mechanical efficiency
k represents the theoretical maximum time of the action which would be attained only if no external work were done.
t is the time occupied by the action
a and b are constants where a represents the energy required to set up a action capable of doing one unit of work under maximal conditions: the product ab is the energy required per stimulus to maintain that action.

According to Dickinson, if the above equation held for any muscular movement, then efficiency would be independent of the force overcome and would vary with change of the time occupied by the movement i.e. efficiency would vary with the speed of shortening which, in this case, was the rate pedalled by the subjects. Also, the efficiency would tend to zero as t tended towards zero or infinity, and there would be a maximum at some intermediate speed.

The results of her study largely confirmed these predictions. Namely, that efficiency was a function of the speed of shortening and not the load. Also, high and low pedalling rates were found to be very inefficient with the optimum being at approximately 0.9 s per half pedal revolution which is equivalent to 3.5 $rad \cdot s^{-1}$ or 33 $rev \cdot min^{-1}$. Interestingly, efficiency was hardly affected by a range of pedalling rates about the optimum, suggesting that a plateau, rather than a specific maximum, best described the power-velocity relationship of the muscles involved.

A.V. Hill (1938) considered mechanical measurements in conjunction with thermal measures of heat release. He demonstrated that the force-velocity relationship was non-linear and was governed by the way in which energy was released during shortening and accorded to the following equation:-

$$(P+a)(V+b)=(P_o+a)b$$

(Hill, 1938)

Where:-

P=the force of action
V=the velocity of shortening
P_o=the maximal isometric force
a and b are constants with units of force and velocity respectively.

Hill's equation describes the relationship between force and velocity for concentric actions only and is shown in Figure 2.2. It was clear at the time that the equation could not be applied to eccentric muscle actions (Katz, 1939). Later, a series of experiments were conducted on intact muscle in human subjects who performed flexion of the elbow (Wilkie, 1950). These experiments showed, subject to a number of provisos, that the force-velocity curve could be satisfactorily represented by Hill's equation for intact human muscle. In sporting situations requiring high-intensity exercise, athletes are concerned with maximising the rate at which work is performed. Therefore, power, which is a function of velocity as well as force is the prime consideration. A power-velocity curve can easily be constructed from a force-velocity relationship (Fig. 2.3(a)).

The Force-Velocity Characteristics Of Individual Muscle Fibre Types

Human muscle is heterogeneous and mechanical properties vary between different fibre types. Much research, using skinned fibres and similar preparations, has concentrated on the measurement of isometric tension (P_o), or the maximum rate at which cross-bridges can be cycled; the latter being measured either as the maximum unloaded shortening velocity (V_o) by the slack-test technique, or as the maximum shortening velocity (V_{max}) determined by extrapolation of force-velocity curves (Josephson and Edman, 1988).

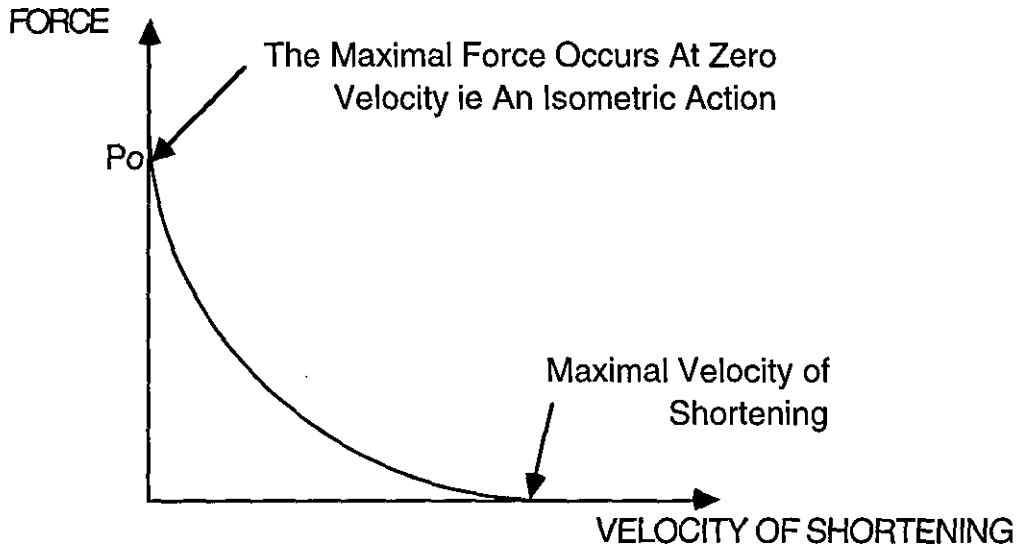

Fig. 2.2. The classical force-velocity curve for concentric muscle actions as described by Hill's equation.

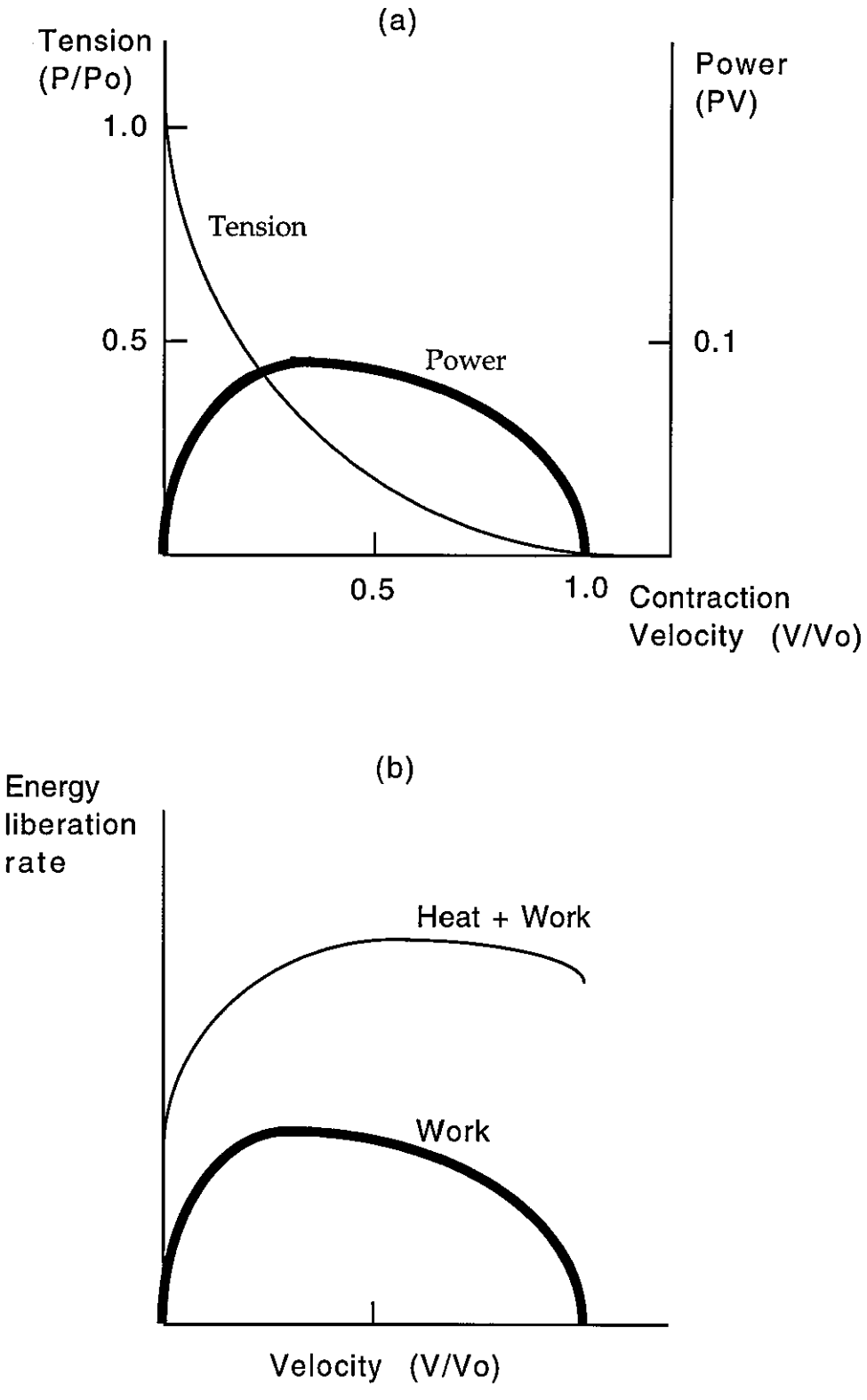

Fig. 2.3. (a) power output (work rate) as a function of shortening velocity and (b) rate of total energy liberation (heat + work) (in Bagshaw, 1993).

This concentration upon these two extremes, V_{max} and P_o, is unfortunate as the performance of high-intensity exercise, where the performer is trying to maximise power output, usually requires the muscle to generate isotonic force at a velocity far less than maximal rate of shortening.

The force-velocity properties of muscle fibre types have been represented by a mathematical model (MacIntosh *et al.*, 1993) based on the previous *in vitro* findings of Faulkner *et al.*, 1986. However, recently the force-velocity properties of different human muscle fibre types has been described in a manner which has attempted to relate these properties to factors which might be of critical importance *in vivo* (Bottinelli *et al.*, 1996). The study involved the examination of 156 human skeletal muscle fibres taken from the vastus lateralis muscles of six male subjects. The fibres were typed in five categories based on their myosin heavy chain isoform composition determined by sodium dodecyl sulphate polyacramide gel electrophoresis. This resulted in 37 % of the fibres being categorised as type I, 36 % as IIA, 10 % as IIB, 9 % as intermediate I/IIA fibres and the remaining 8 % as intermediate IIA/IIB, although results were only presented for types I, IIA and IIB.

The results demonstrated that V_{max}, V_o, maximum power output and the velocity at which maximal power was produced were dependent on the myosin heavy chain isoform. In all cases, values were significantly lower in type I than both type IIA and IIB fibres. For the type II fibres, the values were lower in the type IIA fibres compared with the type IIB. Examples of the force-velocity and power-velocity relationships between fibre types are shown in Figure 2.4. The ratio of V_{max} for type I and IIB fibres was approximately 1:4. The velocity at which maximal power occurs is related to V_{max} and for all fibre types this is approximately at 0.3 V_{max}. It was also apparent that the maximal power output of type II fibres, particularly the IIB, is many times that of the type I. Notwithstanding the significant differences in V_{max} and other measures for each fibre type, the results from the study demonstrated that within a group of fibres containing the same myosin heavy chain, there was a large variation between fibres. Also, between different types of fibres there was a degree of overlap. These two findings suggest that although fibres are classically typed into discrete subsets by histo-chemical methods, it is more likely that there is a continuum and that classification is a little misleading.

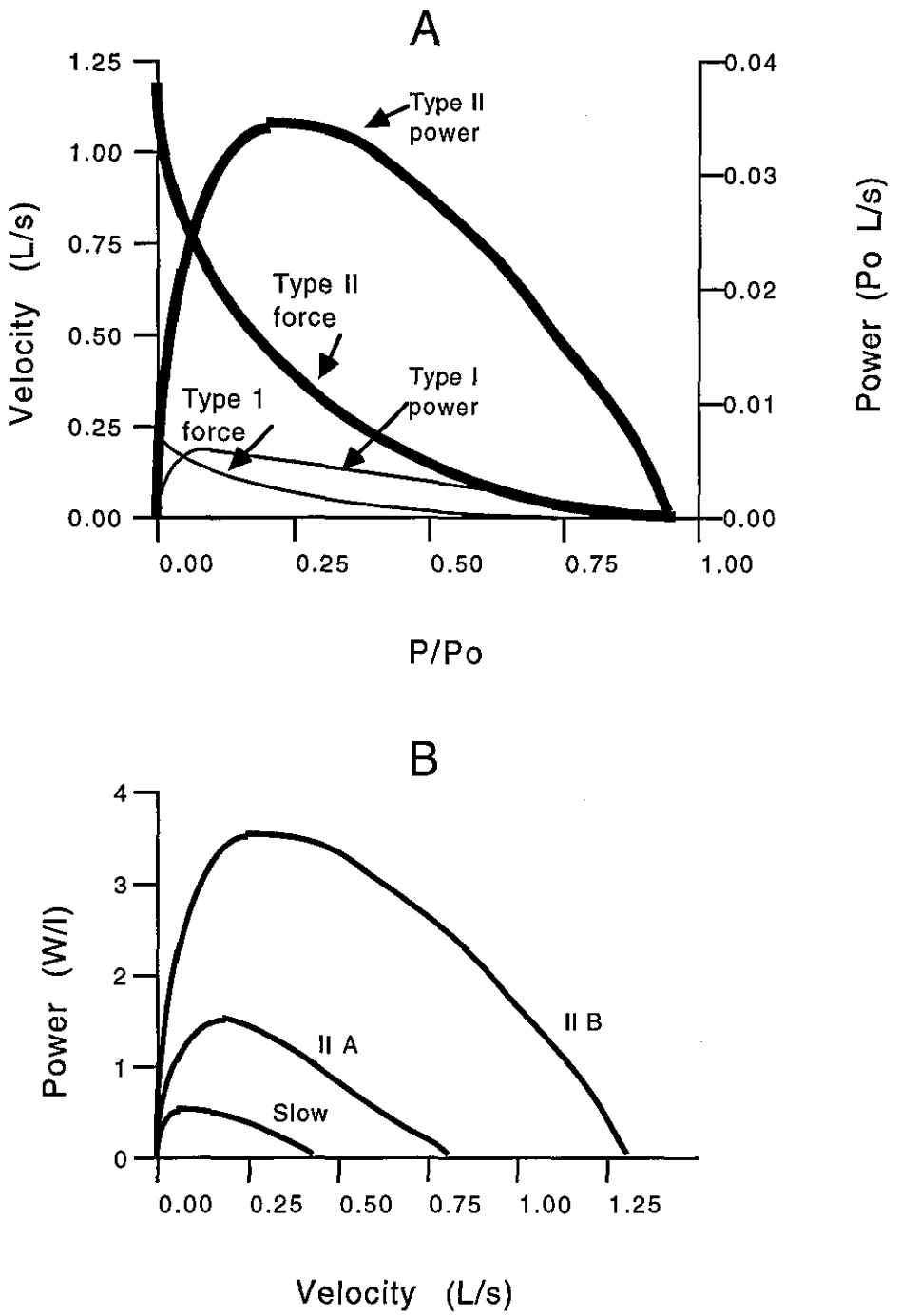

Fig. 2.4. A; force-velocity and force-power curves of a representative type I and IIB fibre. Velocity is expressed in fibre lengths per second ($L \cdot s^{-1}$). Load and power are expressed relative to P_o, i.e. load as P/P_o and power as P_o $L \cdot s^{-1}$. V_{max} of type IIB fibre = 1.239 $L \cdot s^{-1}$ and of type I fibre is 0.309 $L \cdot s^{-1}$. B; velocity-power curves obtained on the basis of the parameters of Hill's equation and of average specific tension for type I, IIA and IIB fibres. Power is expressed in absolute values $W \cdot l^{-1}$ (in Bottinelli *et al.*, 1996).

The potential contribution to power output of different muscle fibre types at muscle action speeds which may be expected to be used by humans performing intense exercise has been widely investigated by Sargeant and various co-workers. In many recent reviews of this work, a model has been presented which attempts to describe the impact of fibre type variability on power output (Sargeant and Beelen, 1993; Sargeant, 1994; Sargeant and Jones, 1995). The model is described in detail below although a number of general points need emphasising.

Firstly, as previously mentioned, although muscle fibres are often placed into discreet groups from histochemical staining techniques, it is more probable that fibres show a continuum of properties across types. Secondly, the model only relates to concentric muscle actions. Although eccentric muscle actions form a major part of muscle function in many activities such as walking and running, the role of different fibre types in the generation of eccentric force is not well understood. Lastly, the model is just that, a model. It is of great assistance in relaying general principles in a visual manner but reference should always be first and foremost to the studies themselves reviewed in this section and elsewhere.

To simplify the model and aid illustration four assumptions and simplifications are made. The first assumption is that only two types of fibre exist, type I and II, with a ratio of 1:4 for their maximal velocity of shortening. Secondly, the model assumes that each type generates one half of the whole muscle maximum isometric force once account has been taken of their relative cross-sectional areas. Sargeant acknowledges that this assumption is arbitrary but suggests that it is not unreasonable for human muscle of mixed composition. The third assumption is that both fibre types have the same length tension relationship relative as the whole muscle. The final assumption is that the constant a/P_O, which describes the shape of the force-velocity relationship, is similar for both fibre types. Once these assumptions are made then relative force-velocity and power-velocity curves as shown in Figure 2.5 can be constructed.

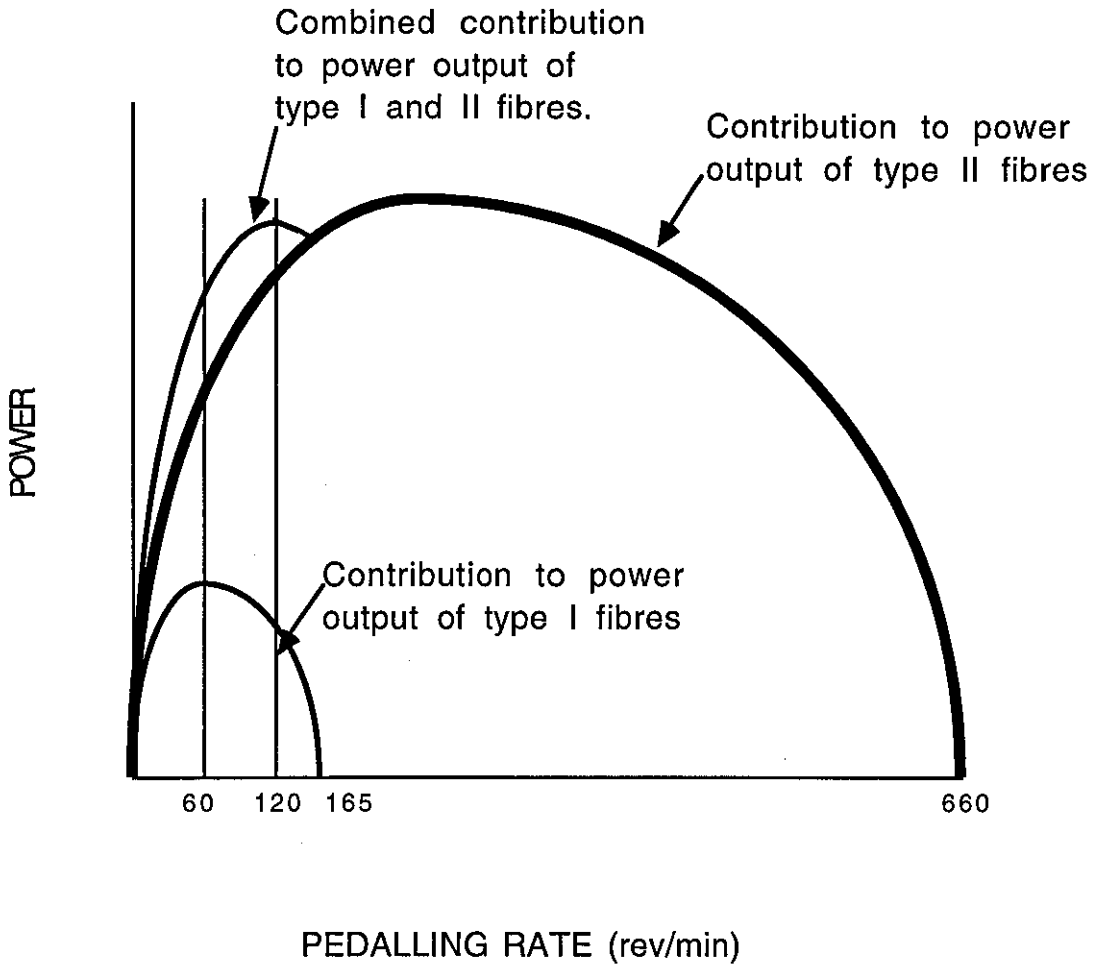

Fig. 2.5. The component and combined power velocity relationship for whole muscle (Adapted from Sargeant, 1994).

The whole muscle maximal power output that can be generated is the combined sum of the individual power curves for type I and II fibres. At a pedalling rate of approximately 6.3 rad·s^{-1} (60 rev·min^{-1}), the model predicts that the type I fibres are able to make their highest contribution to power output but as the rate increases their contribution declines. At pedalling rates in excess of approximately 17.3 rad·s^{-1} (165 rev·min^{-1}), where the maximal speed of shortening of the type I fibres is exceeded, they can make no contribution to power output. For mixed muscle, the model indicates that the optimal rate for peak power output is in the region of 12.6 rad·s^{-1} (120 rev·min^{-1}). The model also suggests it may be possible to achieve the same or slightly higher maximal power at approximately twice this rate where only the type II fibres contribute to power output, but such rates (approximately 26.2 rad·s^{-1}(250 rev·min^{-1})), are not within the realms of normal human locomotion and thus the model may not be a true reflection of the ability to generate power at different rates.

2.3.1(b). Extent Of The Mechanical Changes During High-Intensity Exercise

The mechanical changes that are associated with muscular fatigue are depicted in Figure 2.6 and each (isometric and isotonic) is dependent on specific cellular and molecular events associated with excitation-contraction coupling and/or the cross-bridge cycle (Fitts and Balog, 1996). Specifically, the mechanical changes arising during high-intensity exercise are a decline in power output, a reduction in the maximal rate of cross-bridge cycling and a slowing of relaxation. These factors change the force-velocity and power-velocity characteristics of the muscle.

The relationship between power output and the speed of shortening has been widely studied using a number of experimental techniques. All seem to demonstrate a parabolic relationship and a region at muscle action speeds of approximately 0.3 of the maximal velocity of shortening (V_{max}) where the ability to generate power is maximised. This is a critical factor that cyclists should be aware of when selecting the gearing ratio to be used (Burke, 1986). Fatigue appears to result in a reduction in the maximal velocity of shortening so that the force-velocity curve for the whole muscle will be shifted to the left. This results in a reduction in the velocity at which maximal power can be attained as this remains constant at about 0.3 V_{max}. In addition there is a vertical shift as the magnitude of power output at the optimal velocity in the fatigued state is also reduced, compared with that which can be achieved at the higher optimal velocity in the unfatigued state.

CONTRACTILE PROPERTIES OF FATIGUED MUSCLE

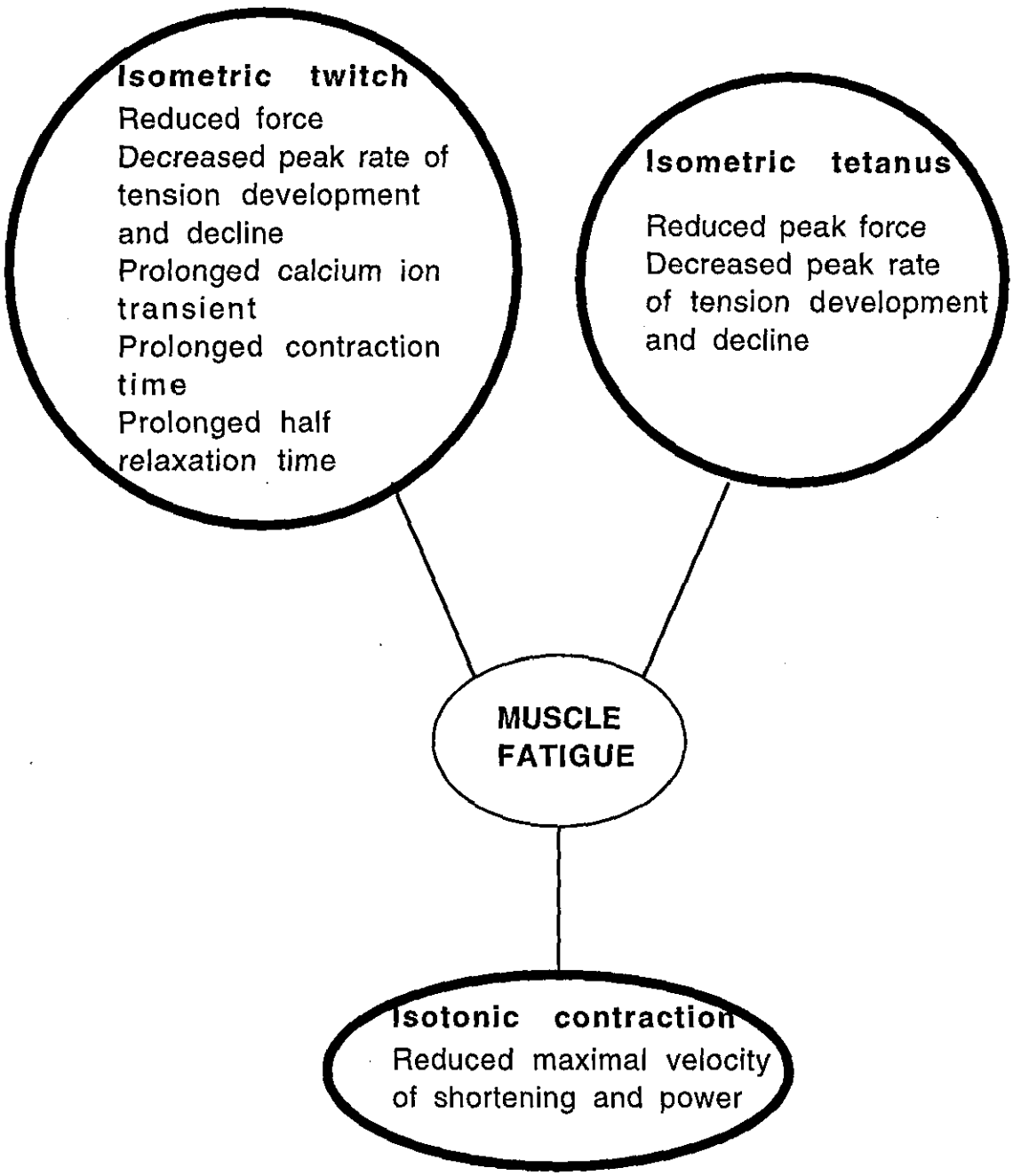

Fig. 2.6. A summary of the known changes in the isometric twitch and tetanus, and isotonic contractions with fatigue (in Fitts and Balog, 1996).

This two-way (vertical and horizontal) shift of the power-velocity curve was clearly demonstrated in the results of a study conducted by de Haan *et al.* (1989). In that study, the force-velocity properties of rat medial gastrocnemius muscle were investigated in the unfatigued, fatigued and recovered state. Initially measurements were taken of fresh unfatigued muscle and then fatigue was induced by a 15 s contraction at 60 Hz. The muscle was then allowed to recover for 15 minutes before further measurements were taken during a 1 s action. For the fatigued muscle there was a reduction in maximal isometric force (referred to in the paper as F_0 instead of the more common P_0), maximal velocity of shortening (V_{max}) (Fig. 2.7) and a slowing of relaxation. These results indicate that assessments of fatigue based on the loss of isometric force alone can seriously underestimate the functional impairment because of the accompanying reduction of maximal shortening velocity. In explanation of their results, the authors cited a slowing in the rate of cross-bridge cycling. Furthermore, whilst it may have been that fatigue resulted in a slowing of all the acting fibres, the authors felt that this was unlikely and suggested that there was a selective fatigue of the fastest fatiguing fibres. This would change the characteristics of the muscle as a whole towards those exhibited by the slower more fatigue resistant fibres.

Lannergren and Westerblad (1989) investigated the maximum tension and force-velocity properties of fatigued, single *Xenopus* muscle fibres. The fibres were fatigued at 70 Hz to approximately 40 % of the unfatigued P_0 which resulted in three distinct types of fibre which the authors referred to as easily fatigued, fatigue resistant and very-fatigue resistant. When the normal activation processes were by-passed by application of caffeine to the bathing medium, all fibre types were able to develop tensions similar to those of controls. Force-velocity measurements revealed that the maximum shortening velocity of the easily fatigued fibres was reduced to 25 % (range 17-40 %) of the unfatigued value but in the other two types of fibres there was no significant depression. The authors concluded that the cross-bridges of fatigued fibres can produce full tension, provided that they can be fully activated although the rate of cross-bridge cycling is reduced. Similarly, Crow and Kushmerick (1983) also found that there is a decrease in the cross-bridge turnover rate in mouse fast-twitch EDL muscle during a maintained tetanus but not in the slow-twitch soleus muscle. However, they concluded that the force generating capacity of fully activated cross-bridges is markedly depressed in fatigued fibres.

Lodder *et al.* (1991) investigated the effect of the velocity of shortening upon the energy cost of repeated muscle actions in rat EDL. The protocol involved 40 muscle actions at three different shortening velocities. The shortening velocities chosen were the optimal velocity at which peak power could be achieved (approximately 50 $mm \cdot s^{-1}$) and also at 50 % (approximately 25 $mm \cdot s^{-1}$) and 150% (approximately 75$mm \cdot s^{-1}$) of this velocity. The experiment was designed so that total work was similar at the three velocities. Efficiency was highest at the optimum velocity yet greater fatigue occurred at the highest velocity of shortening. The authors attributed this to a higher metabolic flux and a greater contribution to power output at high velocities from the fastest fibres.

In summary, the experiments on animal muscle suggest that in order to remain at the optimal velocity for external power production (0.3 V_{max}) athletes should adopt a pacing policy where muscle action speed is gradually reducing. The changes in the power-velocity characteristics of the whole muscle may be primarily due to a selective fatigue of a minority of fibres which may undergo greater fatigue at high, compared with low, muscle action speeds. According to Lodder *et al.* (1991), to explain the underlying mechanisms of fatigue, electrically stimulated isolated muscle or similar preparations are required as opposed to dynamic exercise. Nevertheless, researchers have recently tried to investigate the effect of fatigue on the force-velocity relationship of muscle using techniques involving dynamic exercise.

A number of studies have attempted to describe the force-velocity relationship and the impact of fatigue upon it by conducting experiments *in vivo* on human muscle often with just a single, or small number, of actions (Hill, 1922; Coyle *et al.*, 1979). However, it became clear that such experiments possibly did not reflect the situation where high-intensity dynamic exercise was performed involving a large muscle mass acting over a number of joints. A suitable model needed to be developed and a number of research groups began to develop a cycle based technique in which both power output and the force-velocity relationship of muscle could be investigated.

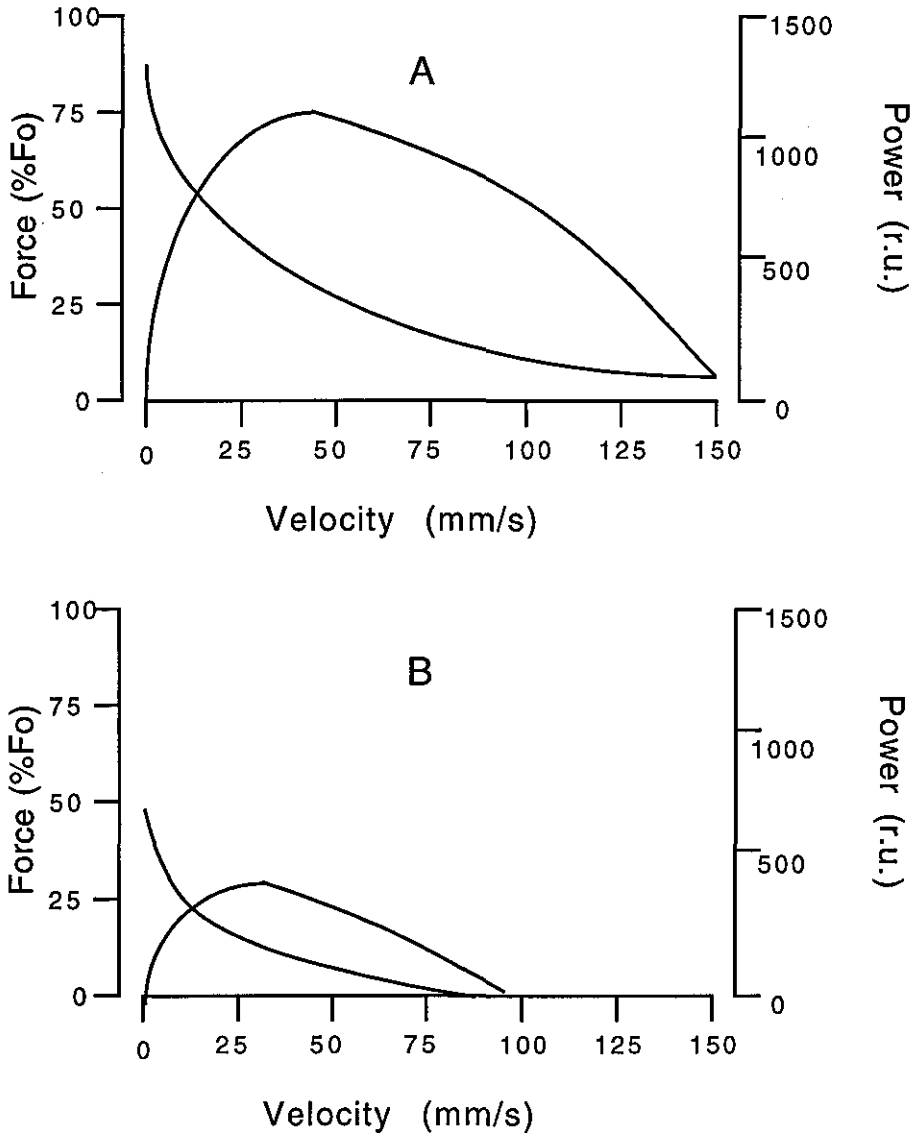

Fig. 2.7. Force and power in relation to velocity for (A) un-fatigued and (B) fatigued rat medial gastrocnemius muscle where fatigue was induced by a 15s electrically stimulated contraction. Fatigue resulted in a reduction in V_{max} (156 to 96 mm·s^{-1}), maximal power (1,141 to 421 relative units (r.u)) and the velocity at which maximal power was attained (45.9 to 30.6 mm·s^{-1}). Data in brackets compares figure (A) with (B) (adapted from de Haan *et al.*, 1989).

The chosen techniques used either an isokinetic or constant velocity ergometer in preference to friction-loaded ergometers, such as that used by the Wingate protocol (Bar-Or, 1978), which required an accelerating effort where the muscle only spent a small time at its optimal velocity. Such an isokinetic model is described by Sargeant (1980) and in more detail by McCartney *et al.* (1983b). Briefly, the ergometer contains a small motor which drives the cranks and a gearing system which allows the experimenter to set the rate, within a certain range, at which exercise is performed. Despite the maximal effort of the subject the set pedal rate cannot be exceeded and strain gauges, attached to the pedals, record the effective force from which power output can be calculated.

Using an isokinetic ergometer, it has been found in five subjects (4 male and 1 female) that force and velocity at a range of pedalling rates from 2.4-17.9 $rad \cdot s^{-1}$ (23-171 $rev \cdot min^{-1}$) are inversely and linearly related (Sargeant *et al.*, 1981). From the force-velocity data, the resultant power-velocity curve was a parabola, with the 'apex' at approximately 11.5 $rad \cdot s^{-1}$ (110 $rev \cdot min^{-1}$). A more recent study also found a linear force-velocity relationship between 6.3 and 16.8 $rad \cdot s^{-1}$ (60 and 160 $rev \cdot min^{-1}$) (McCartney *et al.*, 1983a). The authors suggested that the linear relationship during cycling experiments conflicted with the curvilinear force-velocity relationship found from isolated muscle experiments was because of the relatively more complex nature of cycling which involves the interaction of different muscles across more than one joint. There was a large degree of inter-subject variability probably because of fibre type differences between the subjects, which resulted in a plateau rather than an apex between 12.6 and 16.8 $rad \cdot s^{-1}$ (120 and 160 $rev \cdot min^{-1}$).

A key feature of the isokinetic system is that it enables the experimenter to select a pedalling rate which allows the muscles to operate at a velocity close to optimal as described from the power velocity relationship of muscle (Wilkie, 1960). From the results of the studies described above it is clear that the optimal pedalling rate for the production of short term maximal power where muscle is in the unfatigued state is in the range of 11.5-17.8 $rad \cdot s^{-1}$ (110-170 $rev \cdot min^{-1}$). However, the experiments on animal muscle suggest that these pedalling rates will only be optimal for a relatively short time because of the reduction in the optimal velocity for power production in the fatigued state.

The extent of the mechanical changes in a single sprint are often given in terms of a "fatigue index" which can be represented by the following equation:-

$$\text{Fatigue index (FI)} = \frac{\text{initial power - final power}}{\text{initial power}} \times 100$$

(in McCartney *et al.*, 1983a)

For a 30 s sprint on an isokinetic cycle ergometer at 14.7 rad·s^{-1} (140 rev·min^{-1}) the FI has been quoted as 59.3 % (McCartney *et al.*, 1983a). However, although a pedalling rate of this magnitude is near optimal in the unfatigued state, it is very likely that it will be far from optimal in the latter stages of the sprint because of the shifts in the power-velocity relationship of muscle. Therefore, the use of the FI as a measure of the extent of the mechanical changes during a single 30 s sprint may be unsatisfactory. A more satisfactory method may be to compare peak power output in an almost immediate second sprint with the peak power attained in the first.

2.3.1(c). Effect Of Changes In Pedalling Rate Upon The Extent Of The Mechanical Changes

Average pedalling rate during cycle exercise can be regarded as a good indicator of the total number of muscle actions which take place. Sargeant and Dolan (1987) published the results of a study comprised of three parts, which examined the effect of prior exercise of differing durations and/or power outputs on the ability to generate maximal short-term power at a velocity of 11.7 rad·s^{-1} (112 rev·min^{-1}). In one part of the experiment six male subjects were required to perform a 20 s maximal effort following prior exercise at 98 % of maximal oxygen uptake which lasted either 0.5, 1, 3 or 6 minutes. As the duration of prior exercise was increased, subsequent maximal power became reduced to a greater extent; 6 minutes of prior exercise resulted in a 30 % lower maximal power compared with control. In another part, the duration of prior exercise was fixed at 6 minutes but the intensity was varied at a number of power outputs which elicited 32 to 100 % of maximal oxygen uptake. Six minutes of low intensity exercise, at power outputs equivalent to 39 and 56 % of maximal oxygen uptake enhanced subsequent short term power output by 15.0 % and 10.5 % respectively compared with control, possibly due to an increase in muscle temperature. This contrasted with the effect of exercise at power outputs which elicited more than 60 % of maximal oxygen uptake which

reduced subsequent maximal power compared with control; the extent of this reduction increasing as the intensity increased. The final part followed the pattern of recovery of power output following 6 minutes of prior exercise at a power output eliciting 87 % of maximal oxygen uptake and found that there was a rapid recovery with a half-time of approximately 32 s which the authors attributed to the kinetics of phosphocreatine resynthesis.

In an extension to Sargeant and Dolan's work, the results of a study considering the effect of fatigue on maximal power output at different muscle action velocities was published by Beelen and Sargeant (1991). This study required six male subjects to perform a maximal 25 s effort on an isokinetic ergometer at five different pedalling rates 6.3, 7.9, 9.4, 11.0, and 12.6 rad·s^{-1} (60, 75, 90, 105 and 120 rev·min^{-1}), in contrast to Sargeant and Dolan (1987) which used only one pedalling rate, following 6 minutes of prior fatiguing exercise at 90 % of maximal oxygen uptake. The authors speculated that fatigue may have a velocity dependent effect as animal studies had shown a greater effect on maximal power at high compared with low muscle action speeds (Crow and Kushmerick, 1983; De Haan *et al.*, 1989). Prior exercise did not reduce maximal power at 6.3 and 7.9 rad·s^{-1} (60 and 75 rev·min^{-1}) compared with control, but did so at the three higher velocities supporting their hypothesis. To explain these findings the authors speculated that there may be a greater, or selective, fatigue of fast fatigue sensitive fibres. The effect of this selective fatigue would be relatively minor at slow velocities where all muscle fibre groups would be able to contribute to power output but more pronounced at higher velocities where the relative contribution to power output the fastest fibres would be greater.

Clearly the results of the study performed by Beelen and Sargeant (1991) suggest that the pedalling rate at which exercise is performed may impact upon subsequent maximal power output. This question was addressed using a protocol of 6 minutes of sub-maximal exercise followed, after a short delay of 2-3 s, by a maximal 25 s effort (Beelen and Sargeant, 1993). The results of the study found that 6 minutes of sub-maximal exercise performed at 12.6 rad·s^{-1} (120 rev·min^{-1}) reduced subsequent short term power output at 12.6 rad·s^{-1} (120 rev·min^{-1}) to a greater extent than when the first bout of exercise was matched for external work production at a lower constant average pedalling rate of 6.3 rad·s^{-1} (60 rev·min^{-1}). The authors again speculated that these results suggested a greater involvement of fast-fatiguing type II fibres at high pedalling rates. No

studies have considered the influence of pedalling rate upon the ability to generate maximal power where prior exercise is of a high-intensity.

2.3.2. ELECTRICAL CHANGES

A motorneuron and all of the muscle fibres that it innervates form a motor unit. The muscle fibres within each unit will have very similar properties although some variation will undoubtedly arise due to the different spatial arrangement within the muscle. However, the very fact that motor units exhibit such striking differences amongst themselves in terms of size, speed of contraction and biochemistry suggests that their pattern of involvement in the generation of tension is unlikely to be random but suited to the task demanded of them (McComas, 1996). Since the introduction of the needle electromyography (EMG) technique (Adrian and Bronk, 1929) it has been possible to measure the electrical activity of motor units. Not only can the technique indicate the extent of motor unit activation, it can also provide valuable information as to the frequency of impulse firing in motor nerves. This section will firstly consider the roles of recruitment and firing rate modulation in the generation of force in unfatigued muscle. It will then examine the extent of the changes that take place during intense stimulation. Finally, the section will consider whether the magnitude of the changes may be influenced by changes in average pedalling rate during high-intensity exercise.

2.3.2(a). Properties Of Unfatigued Muscle

Since the advent of needle electromyography (EMG), it has been evident that muscles are able to increase the force of their contractions by recruiting more units ("recruitment") or increasing the firing frequency of the motorneurones ("firing frequency") (Adrian and Bronk, 1929). The authors of that study suggested that of the two processes, recruitment was the major means of regulating force at low force levels. However at higher forces, an increase in the firing frequency became more important. Research in the intervening years has suggested that this opinion was correct although the underlying mechanisms are now better understood.

A positive linear relationship between work rate and the integrated EMG (iEMG) has been well documented from Lippold (1952) onwards. For example, the recorded iEMG at twelve sub-maximal work rates from 0-1980 $kpm \cdot min^{-1}$ (0-330 W) at 6.3 $rad \cdot s^{-1}$ (60 $rev \cdot min^{-1}$) on a friction loaded cycle ergometer

increased linearly as the work rate was increased (Kobayashi *et al.*, 1976). Henneman *et al.* (1965) found that in the cat, smaller motor units had lower thresholds than larger units. They suggested that at low forces the small units were recruited, but as the force requirement increased so more units were recruited in an hierarchical manner based on size. Subsequently, it has been shown in humans that even during the least automatic goal-directed finger movements, the action of the motor units seems rigidly determined. Experiments on single motor units recorded from human hand and forearm muscles during voluntary contractions support the 'size principle' hypothesis derived from animal experiments by Henneman (Freund, 1983).

The relationship between muscle force and the extent of fibre recruitment was examined by Greig *et al.* (1986). Their findings are shown graphically in Figure 2.8 and highlight that at a work rate required to elicit maximal oxygen uptake, virtually all of the fibres are recruited even though only approximately 50 % of the maximum force is generated. Thus, it is reasonable to assume during high-intensity exercise, which results in power outputs 2-4 times that elicited at maximal oxygen uptake (Spreit, 1995), that all units are recruited. This contrasts with exercise performed at a sub-maximal work rate (i.e. at a power output eliciting only a part of the maximal oxygen uptake) where not all of the motor unit pool may be recruited. In these circumstances, any increase in work rate will require the muscle to recruit more units. Thus, the literature suggests that all motor units in the appropriate muscles will be recruited during high-intensity exercise and any minor changes in the extent to which the muscle generates tension will be solely modulated by adjusting the firing frequency (Fig. 2.9).

It is, therefore, of importance to be able to ascertain the minimum and maximum and firing frequencies of individual motor units. According to Freund (1983) the lowest possible maintained firing rates are 6-8 Hz without any significant differences between units of different size. Conversely, there is more variation concerning the maximal firing frequency which is dependent upon the type of unit. During strong ballistic contractions of human tibialis anterior muscle, rates as high as 120 Hz have been reported although variation between subjects was large (60-120 Hz) (Desmedt and Godaux, 1977). The authors pointed out that the high rates were only exhibited early in the burst and that the firing frequency decreased thereafter. The literature, therefore, indicates that during a maximal effort, such as a high-speed 30 s sprint on a

cycle ergometer very high firing rates may be achieved in the early stages of the bout.

2.3.2(b). Magnitude Of The Electrical Changes During High-Intensity Exercise

The fatigue associated with high-intensity exercise reduces EMG activity. It is important to establish whether the reduction in EMG activity relates to a modifications in either, or both, recruitment and firing frequency.

The changes in motorneurone firing rates during sustained maximal voluntary contractions (MVC) of human adductor policis muscle was investigated by Bigland-Ritchie *et al.* (1983). In their study, five subjects (male and female; numbers of each not specified) performed a 10 s MVC and a number of sustained contractions lasting 40-120 s. Their results found a range of firing rates from 10-50 Hz during the MVC with a mean of 26.9 Hz. During the sustained efforts, the mean firing rate declined with time. Moreover, there was little change in the minimum rates which remained at approximately 8 Hz but a marked decline in the number of units firing at high frequencies suggesting that the units with the highest initial maximum firing rates tend to decline rapidly. Interestingly, all the motor units remained activated despite the reduction in the mean firing frequency. This apparent paradox was attributed to the slowing of relaxation with fatigue. The slower that a muscle is, or rather the longer it takes to relax, the lower is the excitation rate required to maintain tetanic force. This "rate coding" phenomenon whereby maximal firing rates are reduced is depicted in Figure 2.10 and is the mechanism behind the reduction in electrical activity exhibited during high-intensity exercise.

2.3.2(c). Effect Of Changes In Pedalling Rate Upon The Extent Of The Electrical Changes During High-Intensity Exercise

For many years a direct relationship between the integrated EMG of a muscle, the work load and speed of muscle action has been evident (Lippold, 1952; Bigland and Lippold, 1954). The iEMG at sub-maximal intensities of exercise performed at various loads, pedalling rates and equivalent external powers has been investigated.

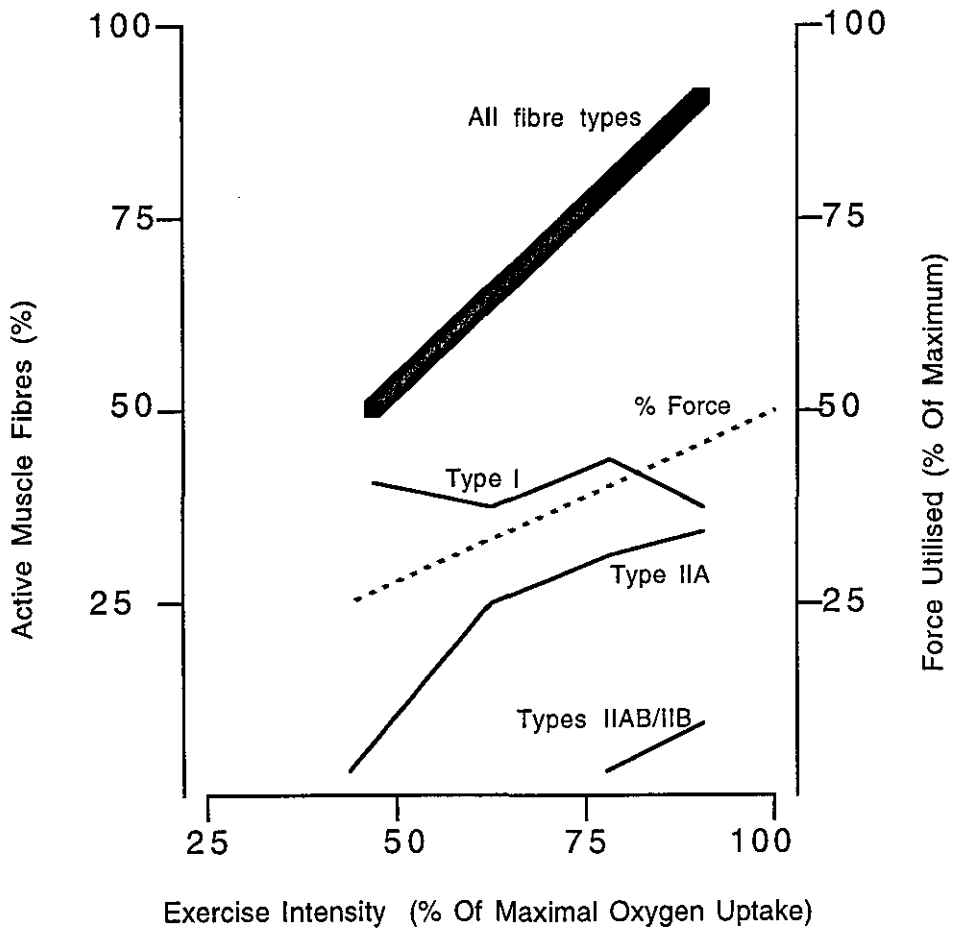

Fig. 2.8. Proportion of maximal force utilised (dotted line) and of active muscle fibres (thick line) in relation to exercise intensity (in Greig *et al.*, 1986).

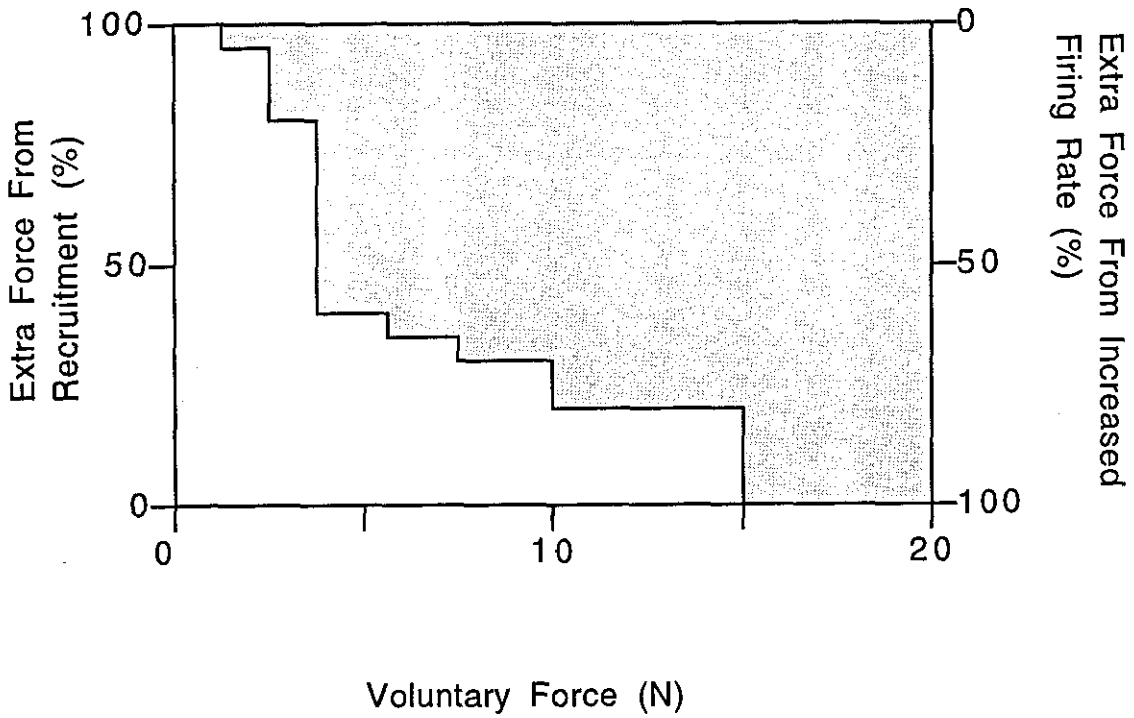

Fig. 2.9. The relative importance of recruitment of additional motor units and of increase in firing frequency during increasingly strong contractions of the first dorsal interosseous muscle (in McComas, 1996).

In a study by Goto *et al.* (1976), exercise was performed at a fixed load on a friction-loaded cycle ergometer (either 0, 1, 2 or 3 kg) where pedal rate was manipulated (4.2, 6.3, 8.4 and 10 .5 rad·s^{-1} (40, 60, 80 and 100 rev·min^{-1})); or at a fixed pedal rate where the load was manipulated; or at a constant external power where both were manipulated. The iEMG was recorded from the gluteus maximus, vastus lateralis, gastrocnemius and tibialis anterior muscles for 10 s. Their results found that the iEMG in all muscles increased with work load and pedalling rate in either a linear or curvilinear fashion. Although the increase in iEMG represented an increase in excitation of motor units, the results did not show whether this was due to an increase in the number of units recruited or firing rate. Interestingly, at an equivalent power output, the iEMG was more affected by pedalling rate than work load.

As the speed of muscle action is increased, for example by an increase in average pedal rate during cycle exercise, there will be a reduction in the threshold force at which motor units are recruited. This means that for a given force more motor units will be recruited with increasing muscle action speed (Freund, 1983). Note that in such instances, although the force remains constant, external power output or work production will increase because of the increase in speed of the movement. Similarly, there will be an increase in the number of units recruited from slow walking to rapid running (Grimby, 1984). Indeed, when a subject attempts to perform a ballistic contraction as fast as possible, the motor units are recruited before any force production is recorded and the threshold force could thus be said to approximate zero (Desmedt and Godaux, 1977).

This poses a problem when conducting experiments involving sub-maximal exercise. During such exercise, where the entire motor unit pool may not be recruited, it is difficult to discriminate between the extent which recruitment and firing rate modulation contribute to the increase in iEMG with increasing muscle action speed (Goto *et al.*, 1976). Exercise at a high-intensity does not suffer from the same problem as force is modulated solely by adjusting firing frequency. However, irrespective of whether exercise is performed at sub-maximal intensity or at a high-intensity, an increase in the speed of the movement may result in more muscles from various parts of the body being brought into action (Goto *et al.*, 1976). The potential involvement of additional motor units to provide, for example, greater stability to the trunk as muscle action speed is increased is a possible drawback of using exercise involving a large muscle mass.

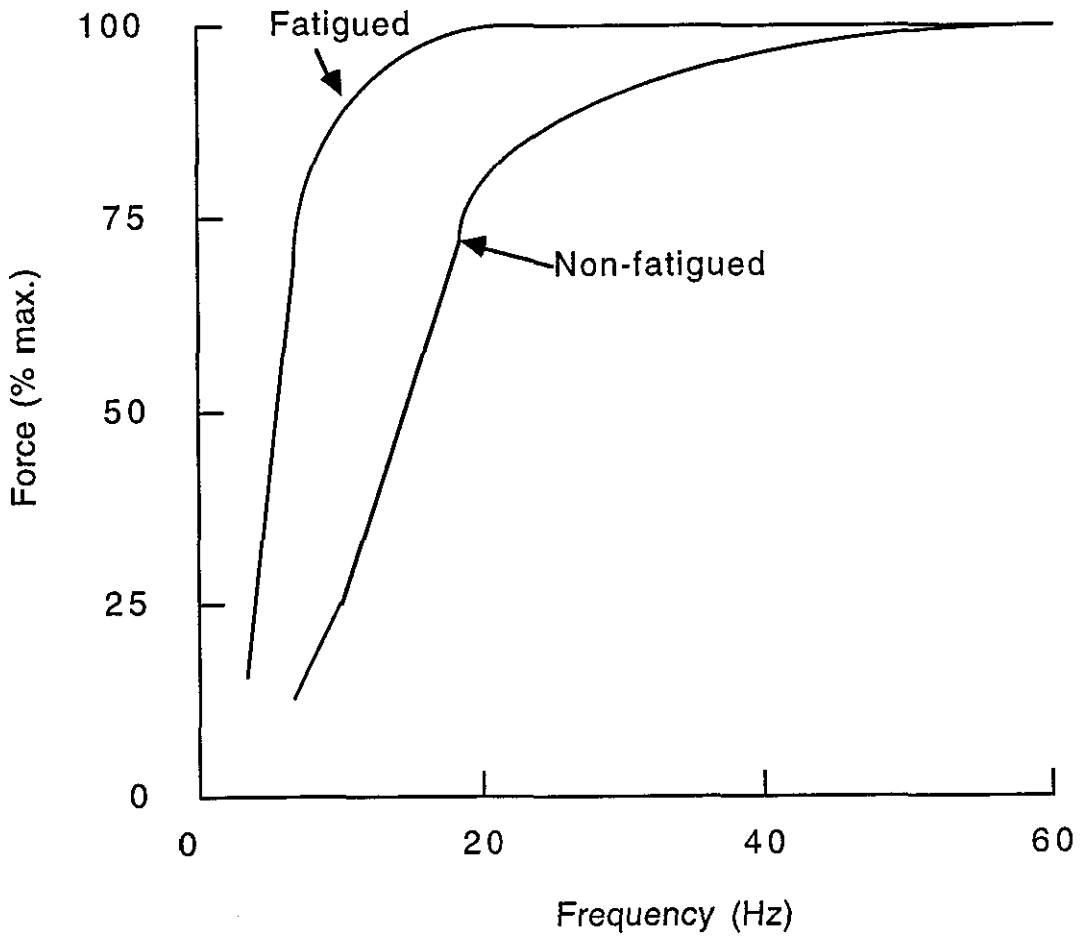

Fig. 2.10. Theoretical force frequency curves for muscle. For effective rate coding of contraction strength, the range of motorneurone firing rates must be upon the steep part of the curve. In the fatigued state, the curve is displaced to the left so that the steep part moves to lower frequencies (in Bigland-Ritchie *et al.*, 1983).

Another potential problem of changing pedalling rate is that there is some evidence that during movements involving high muscle action speeds that the normal recruitment order based on size may be violated. High threshold motor units are not recruited during normal walking but are in short bursts during corrective movements (Grimby, 1984). This may indicate that during briskly executed movements there is a modification or even reversal of the normal recruitment order (Grimby and Hannerz, 1968; 1973: Hannerz, 1974). However, the evidence suggesting that there is a change in recruitment order has generally only been found where the nature of the task has been altered in some way (Gielen and Denier van der Gon, 1990). Where the nature of the task remains essentially the same the recruitment order is preserved (Desmedt and Godaux, 1977). Thus, it seems unlikely that changes in pedalling rate will alter the recruitment order of the motor units.

Leg power, muscle strength and peak EMG activity during maximal isokinetic knee extension at 60, 180 and 360° s^{-1} were investigated by Andersen *et al.* (1979). Although isometric torque was different at each of the three external angular velocities, being 2-3 times greater at the slowest velocity compared with the fastest, peak EMG activity was similar at all speeds. These results accorded with those of a previous study which found that within the velocity range of the dynamometer which was used, the relationship between iEMG and muscle tension was similar irrespective of the velocity of contraction selected (Komi, 1973). Thus, it seems unlikely that the available technology is sophisticated enough to detect any changes in the extent of the electrical changes which arise through changes in pedal rate.

In summary, this section has demonstrated that manipulation of the pedalling rate potentially creates a number of problems. A harness can be used to restrict exercise to the lower limbs but, despite this, an increase in pedalling rate may involve the recruitment of additional motor units for extra stability. Also, there is some evidence that the normal recruitment order based on size may be modified but this is considered unlikely as the movement of cycle pedalling is essentially the same irrespective of pedalling rate. Unfortunately there is one problem that is considered insurmountable and this is the subtle changes in the extent of electrical activity may not be detectable. In particular, it is very unlikely that any differences would be detectable using surface EMG electrodes during dynamic high-intensity exercise where force is solely modulated by adjusting the firing frequency.

2.3.3. BIOCHEMICAL CHANGES

This section concerning the biochemical changes which result from high-intensity exercise is divided into two parts. The first deals with the changes in the concentration of muscle metabolites, whilst the second considers the movement of ions, particularly sodium and potassium, across cell membranes that occur during high-intensity exercise.

2.3.3.1. Metabolic Changes

High-intensity exercise results in power productions many times that required to elicit maximal oxygen uptake (Fig. 2.11; Spreit, 1995). This requires very high levels of activity of the actomyosin, Ca^{2+} and Na^{+}-K^{+}, ATPases which hydrolyse ATP.

$$\text{ATP} \xleftarrow{\text{ATP ases}} \text{ADP} + P_i + H^+$$

The rate of ATP resynthesis is high during high-intensity exercise. For example, during a 6 s sprint, the total ATP turnover rate from anaerobic sources was 10.4 mmol·kg dry muscle^{-1}· s^{-1} (Boobis, 1987) although higher rates of 15 mmol·kg dry muscle^{-1}· s^{-1} have been reported (Gaitanos *et al.*, 1993) for similar exercise. The muscles only possess a small reserve of stored ATP (approximately 25 mmol·kg dry muscle^{-1}) and so to achieve this high demand for energy, ATP must be resynthesised at a very high rate. This requires a large contribution to energy provision of anaerobic metabolism. The pathways by which ATP can be resynthesised anaerobically can be summarised in the following equations:-

$$\text{PCr} + \text{ADP} + H^+ \xleftarrow{\text{CPK}} \text{ATP} + \text{Cr}$$

$$\text{Glycogen} + 3\text{ADP} + 3P_i \longrightarrow 3\text{ATP} + 2\text{Lactate}^- + 2H^+$$

$$2\text{ADP} \xleftarrow{\text{AK}} \text{ATP} + \text{AMP}$$

$$\text{AMP} + H^+ \xleftarrow{\text{AMP deaminase}} \text{IMP} + NH4^+$$

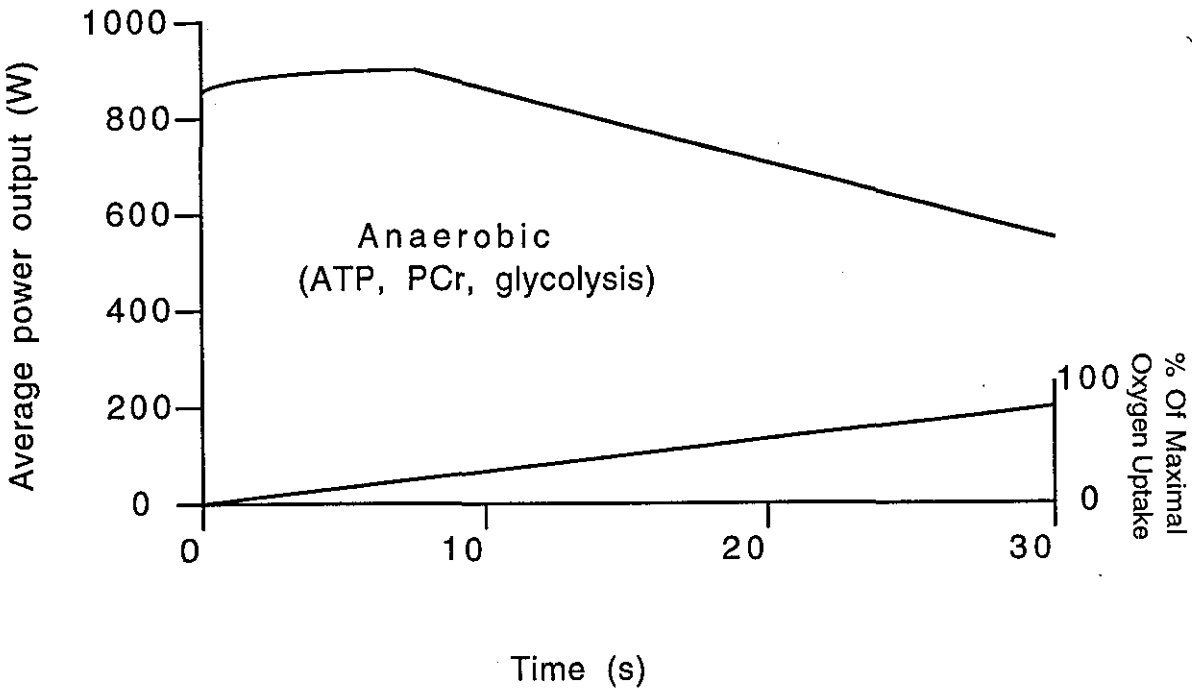

Fig. 2.11. Average power output during 30 s of maximal isokinetic cycling at 10.5 rad·s^{-1} (100 rev·min^{-1}). The generated power in the initial seconds is 2.5-3 times that required to elicit maximal oxygen uptake. The average power over the whole of the 30 s was 2.5 times the power required to elicit maximal oxygen uptake (in Spreit, 1995).

Authors	Bogdanis *et al.* (1995)	Bogdanis *et al.* (1996)	Casey *et al.* (1996a)	Greenhaff *et al.* (1994)	Jacobs *et al.* (1982)
Subjects	8 M	8 M	9 M	5 M; 1 F	9 F
Glycogen	321.5±18.2	327.5±14.3	----------	417±33	360±25
PCr	77.1±2.4	75.2±4.4	83.9±3.6	81.2±4.2	62.7±3.7
Cr	30.7±1.8	42.3±2.1	43.6±0.9	----------	----------
Pi	2.9	2.9	----------	----------	----------
ATP	25.6±0.4	27.0±0.8	23.9±0.4	25.7±0.5	20.9±0.6
ADP	2.2±0.2	----------	----------	----------	----------
Glucose	1.7±0.2	1.7±0.2	----------	1.97±0.34	----------
G-1-P	0.12±0.01	0.2±0.1	----------	0.22±0.09	----------
G-6-P	1.21±0.2	1.4±0.1	2.5±0.9	2.58±1.37	----------
F-6-P	0.13±0.02	0.3±0.1	----------	0.67±0.41	----------
Pyruvate	0.95±0.1	0.5±0.1	----------	----------	----------
Lactate	3.8±0.3	5.8±0.9	4.4±0.5	3.1±1.1	9.0±1.0
Gly-3-P	----------	----------	----------	0.86±0.61	----------

Table 2.1. Unfatigued mixed muscle metabolite concentrations (mmol·kg drymuscle^{-1}; mean ± SE) from the vastus lateralis. Values have been obtained from biopsy samples taken from human subjects prior to the performance of high-intensity exercise.

2.3.3.1(a). Concentrations In Unfatigued Muscle Of Metabolites Involved In The Anaerobic Pathways

A number of studies have analysed biopsy samples from the human quadriceps, usually from the vastus lateralis. A selection of results from recent such studies highlighting the resting mixed muscle concentrations of metabolites involved in the anaerobic pathways is shown in Table 2.1. The concentration of a few metabolites has also been reported in single fibres. The pre-exercise concentrations of phosphocreatine (PCr) and glycogen were higher in type II fibres (79.3±2.7 and 472±35 mmol·kg dry muscle^{-1} respectively) compared with type I fibres (71.3±3.0, $P<0.01$ and 375±25 mmol·kg dry muscle^{-1}, $P<0.01$, respectively) (n=6, 5 M; 1 F). There was no difference in the resting concentration of ATP in the two fibre types (Greenhaff *et al.*, 1994). A lower concentration of PCr in type 1 fibres (66.6±4.2) compared with type II (79.3±1.5) (both expressed in mmol·kg dry muscle^{-1}) has also been reported by Casey *et al.* (1996b) (n=9 M).

2.3.3.1(b). Extent Of The Changes In The Concentration Of Muscle Metabolites In The Anaerobic Pathways With High-Intensity Exercise

The regulation and rate limiting steps of the anaerobic metabolism of muscle glycogen and glucose to lactate have been reviewed on many previous occasions (for example see Bangsbo, 1996b). Thus, these aspects will not be covered in this section which will, instead, concentrate on recent research which has focused on whether the extent of the changes in the concentration of the substrates for the anaerobic pathways may limit performance during high-intensity exercise.

There are a number of direct and indirect methods for determining the contribution of the anaerobic pathways to energy supply during intense exercise. These are (i) oxygen deficit, (ii) debt, or (iii) the lactate produced combined with the depletion of ATP and PCr concentrations of muscle (Bangsbo *et al.*, 1990). At the present time, analysis of muscle metabolites from muscle samples taken pre- and post-exercise, often in conjunction with blood metabolite data, seems to be the most reliable method. This section will draw predominantly upon data obtained in this way from human subjects performing dynamic exercise.

However, the muscle biopsy technique is invasive and requires medical assistance. As a result, non-invasive techniques have been proposed at various times such as the oxygen debt and oxygen deficit (Krogh and Lindhard, 1919/1920). Of the two, recent studies have indicated that the deficit technique is the more useful (Spreit, 1995). For example, the contribution of the anaerobic pathways to energy provision during high-intensity cycle exercise determined by the oxygen deficit technique was similar to that calculated from the changes in the concentration of muscle metabolites before and after exercise (Medbo and Tabata, 1993). However, it has been suggested that the oxygen uptake-power output relationship is non-linear over the whole range of power outputs from rest to maximum (Whipp, 1986). In particular, a non-linear relationship at power outputs at, or above 60 % of maximal oxygen uptake has been reported (Zoladz *et al.*, 1996). In view of these observations it has been suggested that a simple linear extrapolation of a the submaximal relationship between power output and oxygen uptake (Hermansen and Medbo, 1984; Medbo *et al.*, 1988; Medbo and Tabata, 1989; Medbo and Burgers, 1990) is invalid to work intensities above maximal oxygen uptake (Green and Dawson, 1995; Barnett *et al.*, 1996).

Another promising non-invasive approach is the determination of muscle metabolites by nuclear magnetic resonance (NMR) techniques. However, the main disadvantage of NMR is its lack of sensitivity (Bagshaw, 1993). It takes several minutes to build up sufficient signal to detect components in the millimolar range. Also, at the present time, only exercise of a very limited nature can be performed so the technique is not suitable for short lasting dynamic high-intensity exercise.

The examination of muscle metabolites permits the metabolic changes that take place as a consequence of a single bout of exercise to be quantified. A 30 s maximal sprint results in a reduction of approximately 32 % in glycogen, 67 % in PCr and 28 % in ATP, although the majority of these changes take place in the first 10 s (Figure 2.12; Nevill *et al.*, 1996). Although both PCr and anaerobic glycolysis contribute ATP from the start of exercise (Balsom *et al.*, 1992), the maximal rate of the former occurs in the first second whilst it takes approximately 3 s for glycolysis to reach its maximal rate (Table 2.2; Greenhaff *et al.*, 1996). This emphasises the ability of PCr to act as a buffer for the rapid increase in the rate of ATP hydrolysis at the start of exercise.

The vast majority of studies which have investigated the changes in muscle metabolites have used predominantly male subjects. Consequently, data for females performing high-intensity exercise is scarce. However, one study has quantified the changes in selected metabolites associated with anaerobic metabolism during a 30 s Wingate Test using solely female subjects (Jacobs *et al.*, 1982). The results found that the concentration of ATP decreased from 20.9 to 13.8 (-34%); PCr decreased from 62.7 to 25.1 (-60 %); lactate increased from 9.0 to 60.5 (+570 %); and glycogen decreased from 360 to 278 (-23%); (all concentrations are mmol·kg dry muscle^{-1}). The extent of these changes are of the same order of those quoted previously (Nevill *et al.*, 1996) which were based predominantly on samples obtained from male subjects.

Although examination of muscle and possibly blood metabolites during a single brief period of high-intensity exercise can reveal much information concerning the extent to which various metabolic pathways are involved in energy provision, it is not a very helpful model for establishing whether the changes in the concentration of a particular metabolite are responsible for fatigue. A further method is to use the model of repeated exercise, whereby the relationship between muscle metabolites and performance can be examined by investigating the recovery of muscle metabolites and power output (Nevill *et al.*, 1996). According to Nevill *et al.* (1996), this experimental design has the advantage that some of those variables that may be important in fatigue may recover at different rates. However, reductions in external power output and changes in muscle metabolites may have a common cause rather than a cause-and-effect basis (McCartney *et al.*, 1986). Despite these reservations expressed by McCartney and co-workers, recent research has used such an approach to establish whether muscle PCr or glycogen availability may limit performance.

Using the model of repeated exercise in conjunction with the muscle biopsy technique, Bogdanis *et al.* (1995) investigated the recovery of power output and muscle metabolites following 30 s of maximal sprint cycling. The study required fourteen male subjects to perform two 30 s sprints on a friction loaded cycle ergometer, both against an applied resistance of 75 N·kN^{-1} body weight, on three separate occasions with recovery intervals of 1.5, 3 and 6 minutes. On a further occasion, the subjects performed only a single 30 s sprint and muscle biopsies were taken at rest, immediately after exercise and during recovery at the same time points as the second sprints commenced in the other three trials.

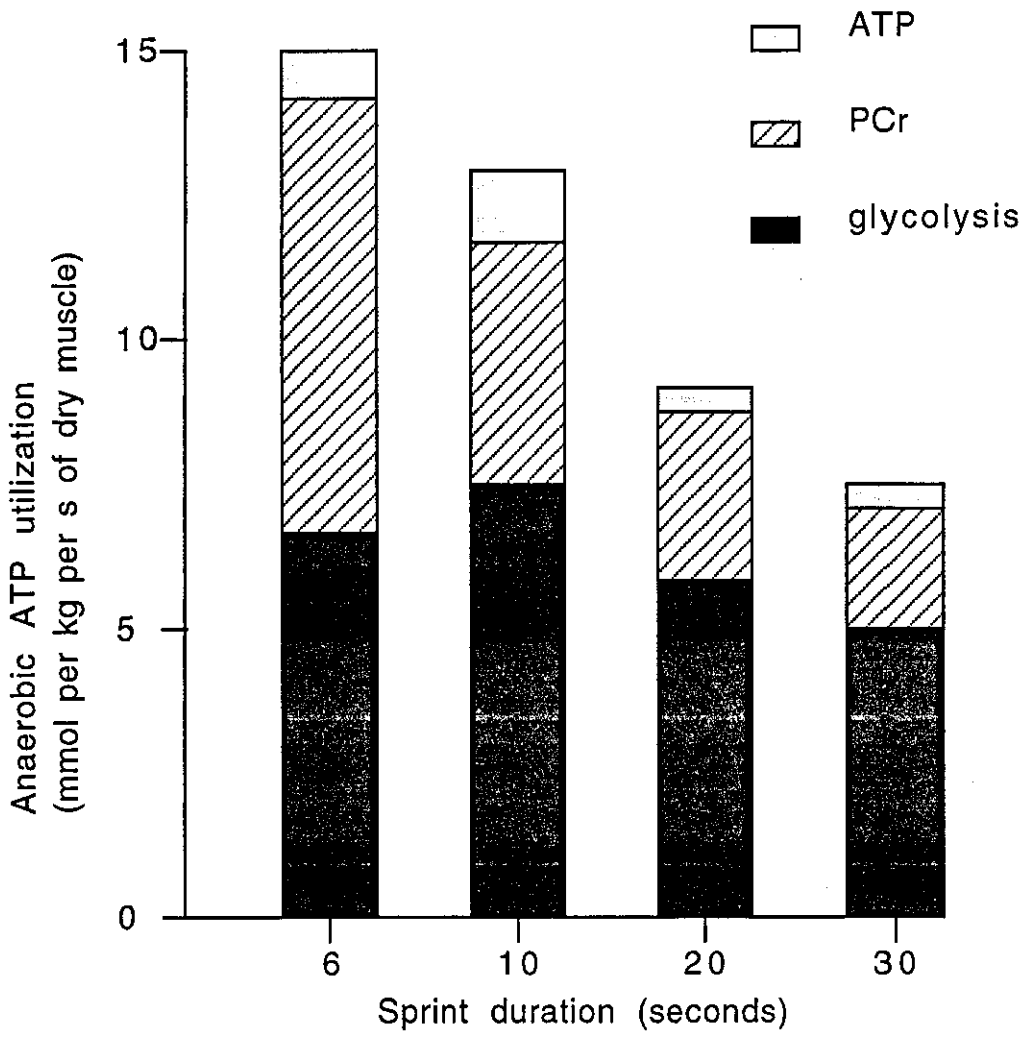

Fig. 2.12. Utilisation of ATP derived from anaerobic metabolism during maximal sprint cycling of 6, 10, 20 and 30 s duration (in Nevill *et al.*, 1996).

Duration of stimulation (s)	ATP production ($mmol \cdot s^{-1} \cdot kg^{-1}DM$) PCr	Glycolysis
0-1.3	9.0	2.0
0-2.6	7.5	4.3
0-5	5.3	4.4
0-10	4.2	4.5
10-20	2.2	4.5
20-30	0.2	2.1

Table 2.2. Rates of anaerobic ATP production from PCr and glycolysis during an intense intermittent electrically evoked isometric action (in Greenhaff *et al.*, 1996).

The results showed that at the end of the 30 s sprint, PCr and ATP had been reduced to 19.7 and 70.5 % of the resting concentrations. During recovery, PCr resynthesis increased rapidly to 65 % of the resting value by 1.5 minutes but the rate slowed somewhat thereafter only reaching 85.5 % of the resting value after 6 min of recovery. The recovery data enabled a model of PCr resynthesis to be constructed which gave an average half-time of approximately 57 s (Bogdanis *et al.*, 1995; Nevill *et al.*, 1997). Moreover, when combined with the performance data from the second sprints, a high correlation (r^2=0.74) was found between the extent of PCr resynthesis and the restoration of peak power, peak pedal speed and mean power in the first 6s of the second 30s sprint.

The importance of muscle PCr during intermittent maximal cycling has also been investigated by Trump *et al.* (1996). This study required seven male subjects to perform 3 bouts of isokinetic cycling at 10.5 rad·s^{-1} (100 rev·min^{-1}) with 4 minutes recovery between bouts. After bout 2, blood flow to one leg was occluded to prevent resynthesis of PCr; the circulation to the other leg remaining intact. The cuff preventing blood flow was removed just before bout 3 and muscle biopsies were taken from both legs prior to and immediately after bout 3. The results found that total work during bout 3 was reduced by 15 % in the cuffed leg compared with control and, in the absence of any other differences in muscle metabolites apart from PCr between the two legs, the authors concluded that PCr resynthesis and the recovery of power output were closely allied.

In view of the results from muscle preparation studies which suggested that power production was closely linked to the contribution to energy production of type II fibres (Faulkner *et al.*, 1986), the single fibre responses to maximal exercise were investigated by Greenhaff *et al.* (1994). The study required six subjects to perform a single maximal 30 s sprint on a non-motorised treadmill (Lakomy, 1987) with muscle biopsies taken pre- and post-exercise. The rates of PCr and glycogen utilisation were higher in type II fibres compared with type I; PCr being almost totally depleted in the type II fibres. The authors speculated that fatigue during such exercise may be due to an inability to maintain a high rate of ATP resynthesis in type II fibres.

Further support for such a hypothesis has recently come from the results of a study which investigated the single fibre responses during repeated bouts of maximal exercise (Casey *et al.*, 1996b). Nine male subjects performed two bouts of 30 s maximal isokinetic cycling at 8.4 rad·s^{-1} (80 rev·min^{-1}) separated by 4

minutes. During the recovery period, the extent of resynthesis of PCr in mixed muscle was 88 % and, in agreement with other studies (Bogdanis *et al.*, 1995; Trump *et al.*, 1996) was highly correlated with the restoration of work in bout 2. However, the recovery period of 4 minutes enabled almost complete resynthesis in type I fibres but was insufficient to allow the same extent of resynthesis in type II fibres. Further evidence implicating the importance of PCr has come from studies which have manipulated the PCr/Cr pool by dietary supplementation.

Creatine Supplementation And Fatigue

In human muscle, creatine (Cr) is present at a concentration of approximately 125 mmol·kg dry muscle^{-1} of which about 60 % is in the form of PCr (Table 2.1). In view of the close correlation between the recovery of power output and the extent of resynthesis of PCr, it has been suggested that any mechanism capable of increasing the intramuscular total Cr store might increase muscle PCr availability. This may delay PCr depletion, the rate of ATP degradation during exercise and may influence PCr resynthesis during recovery (Greenhaff *et al.*, 1996). One such mechanism is to supplement a person's normal diet with Cr (frequently creatine monohydrate). Harris *et al.* (1992) showed that 5 g of Cr taken four times daily for five days increased the total Cr content by 20-50 %; importantly, 20 % of this increase was in the form of PCr. More extensive research on 31 male subjects has found that a 6 day regimen of Cr supplementation increased total Cr content by approximately 20 % and that this higher content could thereafter be maintained by a reduced daily intake of 2 g per day (Hultman *et al.*, 1996). A common finding of the studies which have been concerned with the skeletal muscle Cr content is that there is a large inter-individual variation between subjects. Also, there exists an upper limit of about 145-160 mmol·kg dry muscle^{-1} above which supplementation has no effect (Greenhaff *et al.*, 1996).

Although discovered in 1832, it was not until the 1960's that it was widely recognised that PCr was a key energy substrate during high-intensity exercise (Balsom, 1985). The first major study to assess whether Cr supplementation enhanced performance during repeated bouts of maximal exercise was published by Greenhaff *et al.* (1993). In their study twelve active, but untrained, subjects (9 male; 3 female), performed five bouts of 30 maximal voluntary isokinetic contractions on a Cybex dynamometer at 3.14 rad·s^{-1} (180°s^{-1}) with 1 minute of recovery between bouts. The exercise was repeated after five days of

Cr supplementation or placebo. The results found that muscle torque was enhanced after Cr supplementation in the latter stages of bouts 1 and 5 and throughout bouts 2, 3 and 4. In addition, plasma ammonia concentration was lower post-exercise after Cr ingestion. The authors suggested that these results demonstrated the important link between the extent of fatigue and PCr availability. A number of other studies have been published which have also showed enhanced performance after a period of Cr supplementation (for example Balsom *et al.*, 1993; Birch *et al.*, 1994; Balsom *et al.*, 1995).

Although the exact mechanism by which short term Cr ingestion may improve performance during repeated bouts of maximal exercise is not clear, the available data indicate that it may be related to the stimulatory effect that Cr has on pre-exercise PCr availability and resynthesis during recovery. Interestingly, it has been speculated that during the initial stages of recovery, it is unlikely that the rate of PCr formation will be dependent upon the availability of free Cr. However, as recovery proceeds the extent of PCr resynthesis does seem to be dependent upon Cr availability. In support of such a hypothesis it has been shown that the rates of PCr resynthesis were almost identical during the initial 20 s of recovery when comparing values before and after Cr supplementation but was greater thereafter when the total pool had been increased by Cr supplementation compared with control (Fig. 2.13; Greenhaff *et al.*, 1996).

Thus, if PCr availability is the major determinant of the ability to generate peak power, then the short-term (<20 s) recovery of power should be unaffected by Cr supplementation. Certainly during intermittent exercise, the magnitude of any ergogenic effect does seem to be dependent upon the length of the period of recovery between exercise bouts. For example, 6 days of Cr supplementation resulted in a 23 % enhancement of total torque during repeated bouts of maximal isokinetic knee extensions separated by 120 s but with only 20 s of recovery performance was similar to the control (Vandenberghe *et al.*, 1996).

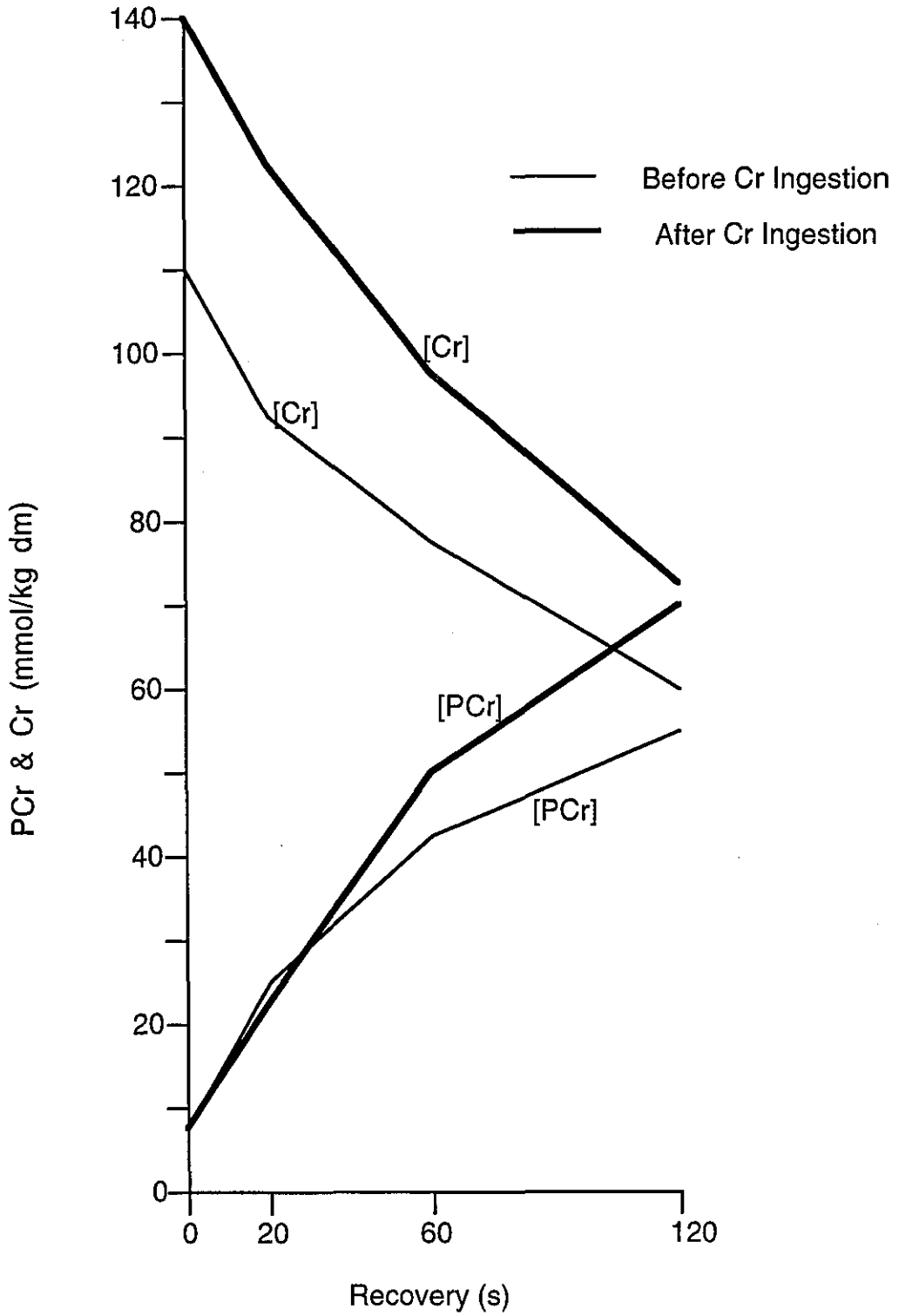

Fig. 2.13. PCr and Cr concentrations measured in muscle biopsy samples obtained after 0, 20, 60 and 120 s of recovery from intense contractions, before and after a period of Cr supplementation. During the first 20 s of recovery the resynthesis of PCr in independent of the total Cr pool. However, from 20-120 s of recovery PCr resynthesis is enhanced following a period of Cr supplementation (in Greenhaff *et al.*, 1996).

Glycogen Availability And Fatigue During High-Intensity Exercise

In addition to the degradation of PCr, the anaerobic degradation of muscle glycogen also makes a major contribution to energy provision during high-intensity exercise. With this in mind, a number of recent studies have also investigated whether the availability of muscle glycogen may limit performance. According to Fitts (1992), if only a single bout of high-intensity exercise is involved the answer is undoubtedly no. This view seems to be supported by muscle metabolite data which shows that a single sprint does not reduce the muscle glycogen concentration to extremely low levels. For example, a maximal 30 s sprint reduces glycogen concentration from 320 to 210 mmol·kg dry muscle^{-1} (Bogdanis *et al.*, 1995).

However, the situation is less clear where repeated bouts of high-intensity exercise are performed (such as the multiple sprint sports) or where a single sprint is performed from a low initial concentration of muscle glycogen (such as a sprint at the end of a marathon). The relationship of glycogen availability to performance and blood metabolite accumulation during repeated bouts of maximal knee extensions (Bangsbo *et al.*, 1992), sprint running (Nevill *et al.*, 1993) and cycle exercise has been investigated (Jenkins *et al.*, 1993; Casey, 1995; Casey *et al.*, 1996a).

Nevill *et al.* (1993) found that the performance of eighteen games players (9 male; 9 female) was enhanced, and the disturbance to blood lactate and glucose was attenuated, when 30 maximal 6 s sprints were performed on a non-motorised treadmill, following a high compared with a low carbohydrate diet. Similarly, Jenkins *et al.* (1993) found that three days on a low carbohydrate reduced total work during five 60 s maximal sprints separated by 5 minutes of recovery but work was marginally increased where the diet was comprised of a moderate or high proportion of carbohydrate. However, these findings seem to be the result of a deterioration in performance of just a single subject in the low carbohydrate group. Further evidence for a possible role of glycogen availability limiting performance are the findings of a study which required 11 male subjects to perform four bouts of maximal isokinetic cycling on an isokinetic cycle ergometer at a pedalling rate of 10.5 rad·s^{-1} (100 rev·min^{-1}). Glycogen stores were reduced by intense exercise and then the subjects were split into two groups; one consuming a high CHO (82 % of energy) and the other a low CHO (8 % of energy) diet for three days. On day 4, the subjects repeated the four bouts of maximal exercise. Performance in the first three

bouts was impaired to a greater extent in the low CHO group compared with the high CHO group, but not in the fourth bout (Casey *et al.*, 1996a). In explanation of the similar performance of the two groups in the fourth bout of exercise, the authors stated that fatigue at this point was due to mechanisms other than energy provision. However, exactly how the authors were able to make this assumption for the fourth bout of exercise and not the previous ones is difficult to ascertain. According to the authors, their exercise and diet regimen would have reduced glycogen concentration to approximately 58 mmol·kg drymuscle^{-1} in the low CHO group. This is considerably lower than previous studies which have attempted to reduce muscle glycogen concentrations to low levels. In these previous studies a reduced glycogen concentration of approximately 150 mmol·kg dry muscle^{-1} did not impair performance (Symons and Jacobs, 1989; Ren *et al.*, 1990). Thus, the literature suggests that glycogen availability may only limit performance where many bouts of sprint exercise are performed, or where at the start of exercise, glycogen concentrations are at a very low level. Although these findings have important implications for those competing in multiple sprint sports, especially when required to perform in tournaments with only a short gap between matches, it seems doubtful that glycogen availability is an important cause of fatigue during a single sprint

Thus, there is some evidence that energy supply, particularly the pre-exercise availability of PCr, may be an important limiting factor in the maintenance of power output. Furthermore it seems that energy supply is more important than the direct effect of H^+ on the contractile mechanism. However, whether or not energy supply is the cause of fatigue during sprinting remains in question (Nevill *et al.*, 1996). According to Balsom (1995), although substrate availability may be a limiting factor for performance, the studies completed so far have not elucidated a mechanism for the actual cause of fatigue. Indeed, Hermansen (1981) states that there is no biochemical basis for postulating that depletion of PCr stores is directly responsible for the reduction in tension development or fatigue.

These cautionary notes regarding the role of substrate availability may be well founded. The results from some studies have shown no statistically significant enhancement of performance with dietary supplementation of Cr during single (Rossiter *et al.*, 1996) and repeated bouts of high-intensity exercise (Odland *et al.*, 1994; Cooke *et al.*, 1995; Mujika *et al.*, 1996; Barnett *et al.*, 1996a; Odland *et al.*, 1997). Interestingly, where the Cr content of skeletal muscle is enhanced by

supplementation, any ergogenic effect is counteracted by caffeine (Vandenberghe *et al.*, 1996). A number of studies have also failed to verify that glycogen availability influences performance in intermittent high-intensity exercise (for example, Bangsbo, 1996b).

2.3.3.1(c). Effect Of Changes In Pedalling Rate Upon The Metabolic Changes Arising From High-Intensity Exercise

Upon a friction-loaded cycle ergometer, changes in average pedalling rate during high-intensity exercise can be achieved by two methods. The first method is to impose a pre-set constant average rate. This "paced" strategy will result in the even production of external work, provided the set rate can be maintained. The second method is to adopt an "all-out" strategy where average pedalling rate will be a function of the applied resistance. Whichever method is used, the speed and total number of muscle actions can be manipulated.

During an all-out sprint it has been shown that peak power output is achieved in the first few seconds of exercise (Lakomy, 1988). Conversely, if pedalling rate is affected by imposing a pedalling rate with the resultant even production of external work, it follows that in the first few seconds of exercise, a sub-maximal power output is produced. It is also possible, depending on the required power output and imposed rate, that the recruited muscles will be generating less than their maximal possible tension. A muscle regulates tension by the recruitment of additional motor units or by rate coding but the latter is the most important method of regulating force during intense muscle stimulation. Thus, during a paced strategy there is likely to be a reduction in the firing frequencies of some units compared with all-out because of the reduced power output in the initial stages.

Electrical stimulation data suggest the extent of the changes in the concentration of muscle metabolites is unaffected by the frequency of stimulation. The changes in muscle metabolites during intermittent electrical stimulation (1.6 s contraction and 1.6 s rest) at two frequencies (20 and 50 Hz) have been reported (Hultman and Sjoholm, 1983). The initial force and ATP turnover rate was higher at 50 compared with 20 Hz. Over the course of 25 contractions (total contraction time 40 s) there was a greater loss of force at 50 Hz, so that at the end, force was lower compared with 20 Hz. However, despite these differences in force, the magnitude of the changes in the concentrations of metabolites (ATP, ADP, PCr, Pi and lactate) were similar.

Using the same two frequencies and intermittent stimulation pattern of 1.6s contraction and rest, the changes in muscle metabolites in type I and II human muscle fibres have been reported (Soderlund *et al.*, 1992). Muscle biopsies were taken from seven subjects (four women and three men) at rest and after 10 and 20 seconds of stimulation; 6 and 12 contractions respectively. With these shorter stimulation periods, the decrease in force, expressed as a percentage of the initial force, was similar at the two frequencies. The PCr degradation rate was higher in type II fibres compared with type I at both frequencies. However, in accord with the mixed muscle findings, there was no difference in the PCr degradation rate, or in any other measured metabolite between the two frequencies.

The muscle metabolite responses of three male subjects to exhausting exercise at different initial power outputs were examined by Karlsson and Saltin (1970). Exercise took place on a Krogh cycle ergometer at a pedalling rate of 6.3 $rad \cdot s^{-1}$ (60 $rev \cdot min^{-1}$) and each subject completed a total of 6 trials. One trial was against a "heavy" resistance which exhausted the subjects in 2-3 minutes. Two trials were performed against a "medium" resistance; one to exhaustion which took 5-7 minutes and the other was terminated at the point of fatigue in the "heavy" trial. Three trials were performed against a "light" resistance; one trial to exhaustion (15-20 minutes) and the other two trials were stopped at the points of exhaustion against the heavy and medium resistances. In each trial, a muscle biopsy was taken pre- and post-exercise.

The results found that the changes in ATP and PCr were identical after 2-3 minutes irrespective of the load despite the fact that the total exercise times were very different between the three resistances (Fig. 2.14). Unsurprisingly, the lactate production and measured oxygen deficit were both dependent upon work rate, being greatest against the heaviest resistance. Nevertheless, the muscle metabolite responses of this study were surprising bearing in mind the different work rates and duration of exercise.

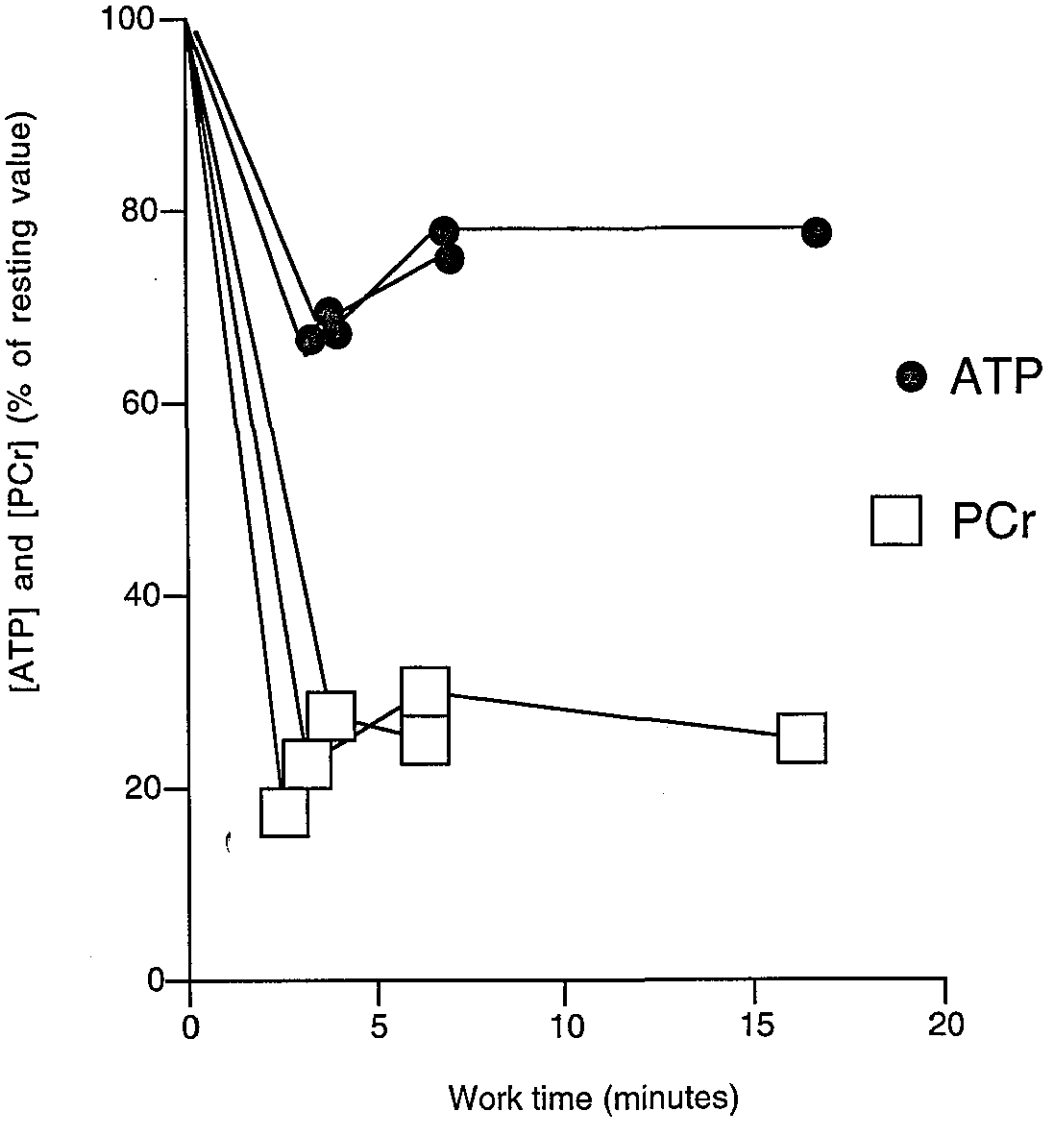

Fig. 2.14. ATP and PCr concentrations after exhaustive exercise lasting at three different work rates which could be sustained for 2-3, 5-7 and 15-20 minutes respectively (in Karlsson and Saltin, 1970).

Similar results have been reported where the concentration of muscle metabolites has been determined by NMR as opposed to the analysis of muscle biopsies. Using this technique in conjunction with force recordings from a Cybex dynamometer, the effects of length and stimulation frequency on fatigue and changes in the concentration of metabolites in the tibialis anterior muscle have been investigated (Sacco *et al.*, 1994). The muscle metabolite responses to four 15s tetanii separated by 5s rest were similar at a muscle length which was optimal for force production and at a shorter length where the measured force was less 60% of optimal. The authors concluded that any difference in the rate of fatigue at the two muscle lengths could not be explained by different energy costs of the contractions despite previous suggestions (Fitch and McComas, 1985) that this may be the case. The same protocol repeated at two stimulation frequencies, 15 and 60 Hz, also resulted in muscle metabolite changes of a similar magnitude despite a different force profile.

Even before phosphate metabolites were identified, measurements of heat changes demonstrated an important property of the energy transduction mechanism in muscle (Bagshaw, 1993). In particular, the rate of heat liberation was found to be higher in a muscle undergoing shortening compared with one developing isometric tension and the total energy liberation rate was a function of the speed of shortening (Section 2.3). Subsequently, it has been shown that at high velocities of shortening, ATP consumption is relatively low although heat output is high (Woledge *et al.*, 1985). According to Bagshaw (1993), a muscle behaves like a car where fuel consumption is related to speed and load. He also states that the analogy may be taken further. An isometrically contracting muscle hydrolyses considerably more ATP than a resting muscle, but all the energy is liberated as heat. Likewise a motor car may be held on an incline at the expense of fuel and a hot clutch, but no external work is performed. In species such as the mollusc, a 'handbrake' mechanism is found where their muscles can show a high economy but vertebrate skeletal muscle can only develop a high tension at zero cost in the state of rigour. At high velocities of shortening the ATP consumption declines suggesting that the number of effective actin-myosin interactions falls. Car manufacturers have also recognised that cruising at high speeds does not demand as much fuel as during acceleration and have devised fuel injection systems which automatically reduce the number of cylinders which are fed. Thus, it appears that altering the speed of muscle actions may affect the muscle metabolite responses. Specifically, as muscle action speed is increased, it might be anticipated that the extent of the metabolic changes would be reduced.

However, in rat EDL, it was shown that 40 contractions performed at three speeds of shortening (25, 50 and 75 mm·s^{-1}) result in a similar reduction in PCr and lactate (Lodder *et al.*, 1991). In human subjects performing dynamic exercise, glycogen depletion studies have shown a sequential depletion of glycogen in histochemically stained individual muscle fibre types as the intensity and/or duration of exercise proceeded (Vollestad, 1984). Using this technique Beelen *et al.* (1993) examined the pattern of glycogen depletion in four subjects performing exercise at a power output equal to 90% of maximal oxygen uptake at 6.3 and 12.6 rad·s^{-1} (60 and 120 rev·min^{-1}). At each rate, the subjects performed two periods of exercise lasting 6minutes with 10minutes recovery. Their results showed that despite less external work production at the higher rate there was a greater depletion of glycogen in all fibre types thereby suggesting that total ATP utilisation increased with an increase in muscle action speed. Interestingly, their results also refuted previous suggestions of a selective fatigue of type IIB fibres at high average pedalling rates (Section 2.3.).

The results of a number of other studies have also found that changes in muscle action speed have little effect on the magnitude of the metabolic responses. At a sub-maximal intensity of exercise, many studies have obtained blood and/or muscle metabolite data from subjects performing dynamic cycle exercise at different average pedalling rates (for example Lollgen *et al.*, 1980) but data are rare for high-intensity exercise. A maximal 30 s sprint performed on an isokinetic cycle ergometer at 6.3, 10.5 and 14.7 rad·s^{-1} (60, 100 and 140 rev·min^{-1}) resulted in a greater fatigue index with increasing pedalling rate but there was no difference in the pattern of recovery of blood lactate post-exercise at the three pedalling rates (McCartney *et al.*, 1983a). In a later study, the same research group examined the changes in muscle metabolite concentrations caused by 30 s of maximal isokinetic cycling at fast 14.7 rad·s^{-1} (140 rev·min^{-1}) and slow 6.3 rad·s^{-1} speeds (60 rev·min^{-1}) in five male subjects. The results found that external work was similar at the two speeds but that the fatigue index was greater at the fast pedalling rate. The changes in concentration of most of the selected muscle metabolites were similar at both speeds. However, post-exercise concentrations of lactate were higher at the fast speed but the authors concluded that despite this, their results were not suggestive of any greater energy cost of the exercise at the faster rate (Jones *et al.*, 1985).

Thus, although the literature suggests that the ATP cost per muscle action may decline with increasing muscle action speed, studies which have investigated

the metabolic responses during dynamic exercise have found this not to be so. A possible reason for this conflict could be that in attempting to control the speed of muscle actions by, for example manipulating average pedalling rate on a cycle ergometer, research groups often overlook the fact that for exercise of a given duration, an increase in the speed of muscle actions will also result in a greater number of actions being performed. The relationship between the energy cost and fatigue where the number of muscle actions has been manipulated has been described by Bergstrom and Hultman (1988). Two regimens of electrical stimulation with a work to rest ratio of 1:1 involving periods of either 0.8 s or 3.2 s were used to give a total contraction time of 51.2s. The shorter period resulted in a total of 64 contractions and the longer in 16, with biopsies taken after 22.4 s and at the end of stimulation. The results showed force declined more rapidly with the regimen involving more muscle actions of short duration. Despite very different force levels at the end of stimulation, the concentration of PCr was similar and there was no difference in the anaerobic ATP utilisation between the two protocols. However, the ATP cost per unit of work was different being greater when similar work was produced with a greater number of actions. This was attributed to approximately 37 % of the energy cost of a 1 s tetanus being attributable to activation and relaxation.

In summary, the literature suggests that manipulation of average pedalling rate and the associated changes in the speed and number of muscle actions, may affect the magnitude of the metabolic changes. However, the limited number of studies which have attempted to measure muscle metabolite responses during intense exercise have found no difference in the magnitude of the changes at different muscle action speeds. Furthermore, data from studies involving high-intensity exercise, particularly using exercise models where the limbs are free to accelerate, such as friction-loaded cycle ergometry cannot be found in the literature. At the beginning of this section it was stated that during high-intensity exercise there was a high activity of the three ATPases which hydrolyse ATP. The existence of more than one ATPase emphasises that several steps in the contractile processes are known to be energy dependent (Fig. 2.15). These include maintenance of the Na-K gradient over the sarcolemma, re-uptake of Ca^{2+} by the sarcoplasmic reticulum (SR), cross-bridge cycling and coupling between t-tubular depolarization and calcium release from the SR (Sahlin, 1992).

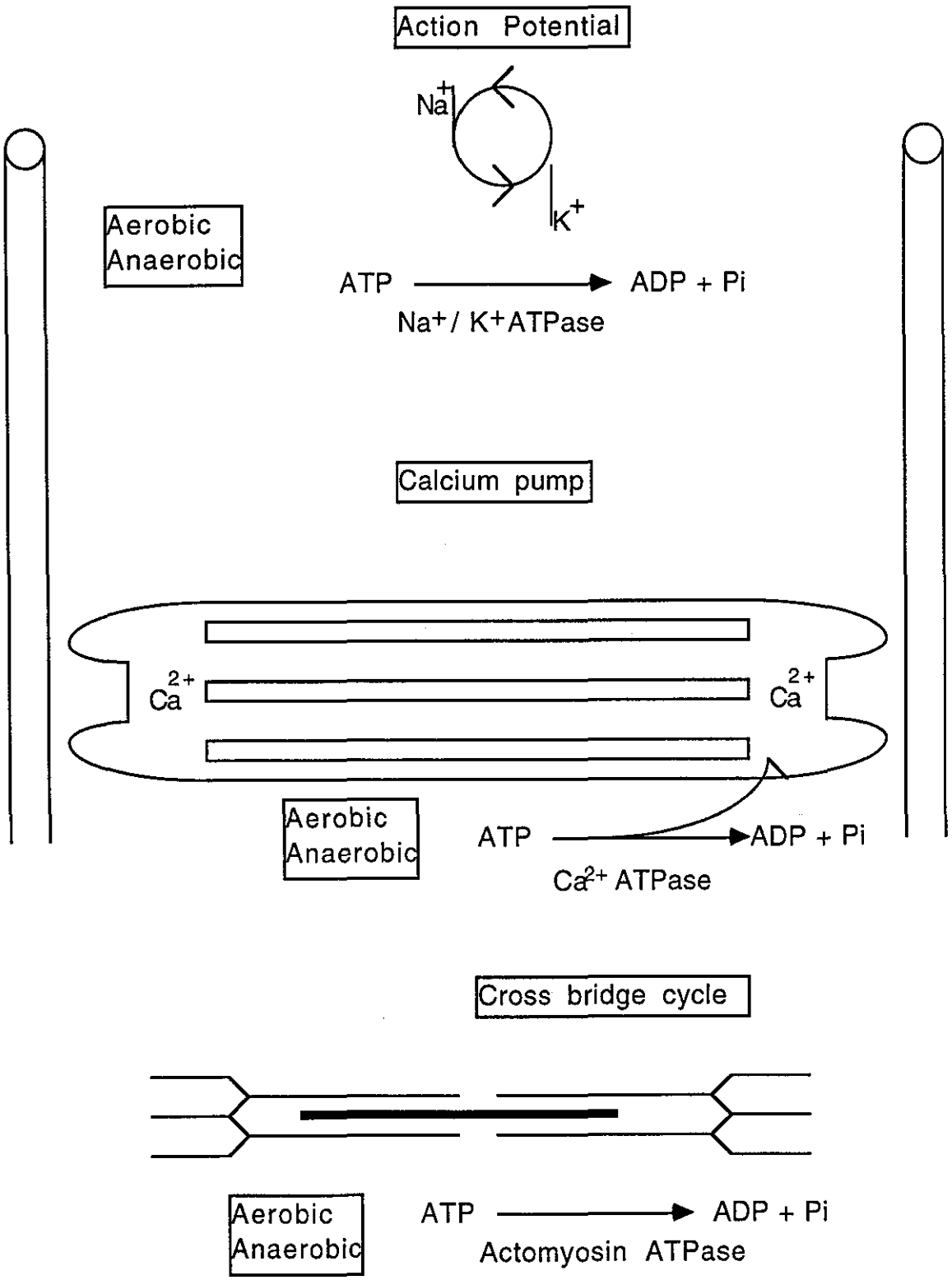

Fig. 2.15. Major sites of ATP utilisation used to sustain excitation and contraction in skeletal muscle. There are three ATPases; sarcolemma Na^+-K^+ ATPase, the sarcoplasmic reticulum ATPase and the actin myosin ATPase (in Green, 1995).

Muscle (Mouse)	Fibre type	ATP consumption $\mu mol \cdot g\ muscle^{-1} \cdot s^{-1}$	
		Calcium handling[a]	Total[b]
EDL	Fast	1.2	4
Soleus	Slow	0.4	1.3

Table 2.3. ATP consumption in fast and slow mouse muscle. [a] Proportion of total ATP consumption required for calcium uptake and release from the sarcoplasmic reticulum [b] Total ATP consumption at 2.2 μm sarcomere length (in Ruegg, 1987).

The energy requirements of these processes also vary between fibre types. During the re-uptake of calcium by the sarcoplasmic reticulum, the transport of two calcium ions requires one molecule of ATP to be split. Type I fibres have a slower rate of calcium release and re-uptake into the sarcoplasmic reticulum compared with type II. As a result, the energy requirement for calcium handling is about three times lower in slow compared with fast muscle (Table 2.3; Ruegg, 1987). Maintenance of the membrane potential requires the expenditure of energy and the increase in intracellular Na^+ and extracellular K^+ at fatigue may be a consequence of energetic deficiency (Sahlin, 1992). In addition, the energy required for cross-bridge cycling is also higher in fast compared with slow muscle since this depends on the rate constants of cross-bridge attachment and detachment.

2.3.3.2. IONIC CHANGES

The ability of ions to pass across membranes is fundamental to the activity of muscle fibres. A large number of anions and cations are found inside and outside muscle cells and many have been implicated as primary causes of fatigue. The effect of an elevated concentration of H^+ and P_i has been widely studied (for example Cooke and Pate, 1990; Jones and Heigenhauser, 1992). However, recently fatigue has been shown to be independent of muscle pH but always occurred at the same plasma $[K^+]$ suggesting that accumulation of potassium may be an important factor in the development of fatigue (Bangsbo *et al.*, 1996). Thus, this review will focus upon the movements of Na^+ and K^+ between the intra- and extra- cellular environment.

Upon stimulation of the fibres, the extent of these movements may be such that ionic balance cannot be restored before the arrival of the next action potential. Moreover, during intense stimulation, the extent of ionic imbalance and associated depolarization may be sufficiently great to interfere with excitability (Everts and Clausen, 1994). It has been known for many years that a muscle exposed to a high extracellular K^+ (>8-10 mM) loses excitability (Hodgkin and Horowicz, 1959) due to depolarization and inactivation of voltage dependent Na^+ channels (Ruff *et al.*, 1988). This in turn leads to a reduction in the amplitude of action potentials and a loss of contractile force (Juel, 1988). The critical question is whether the ionic changes can give rise to fatigue during high-intensity exercise.

2.3.3.2(a). Ionic Balance In Unfatigued Muscle

The resting membrane potential can be accounted for by the fact that there is a net excess of negative ions inside the cell and a net excess of positive ions outside. The magnitude of the potential at any time is determined mainly by two factors (1) the difference in ion concentrations of the intracellular and extracellular fluids and (2) the permeability's of the plasma membrane to the different ionic species (Vander *et al.*, 1990). Sodium, potassium and chloride ions are present in the highest concentrations and it is their distribution which is primarily responsible for the resting membrane potential. K^+ ions are more concentrated inside the cell than outside; the reverse is true for Na^+ and Cl^- (McComas, 1996). At rest, the membrane is more permeable to K^+ and Cl^- than to Na^+. In the extracellular fluid, the Na^+ ions are present in a similar concentration to the anions but in the intracellular fluid, K^+, although attracted into the cell by the electrical attraction of the anions there, has to move against its concentration gradient. As a result, the internal K^+ concentration is a little less than that of the anions so the inside of the cell which is negative with respect to the outside. The size of this potential difference can be calculated from the Nernst Equation and assuming values of 4.5 and 160 mM respectively for the external and internal concentrations of K^+ gives a theoretical resting potential of -95 mV (McComas, 1996). The actual resting membrane potential of mammalian biceps brachii and quadriceps muscle has been shown to be a little less than the theoretical value with a mean of -84 mV and range of -64 to -98 mV (McComas *et al.*, 1968).

There are three mechanisms by which ions can pass across muscle membranes namely (i) pumps requiring energy (ii) channels and (iii) carrier proteins. An action potential is initiated by the opening of Na^+ channels and the influx of sodium ions. This is closely followed by the opening of potassium channels and the efflux of potassium ions. For further action potentials to take place these processes must be reversed and this is primarily achieved by the action of the Na^+-K^+ pump which carries 3 Na^+ out of, and 2 K^+ into, the cell during the splitting of ATP. The capacity of the pump is determined by the number of available pumps and the extent of Na^+ movement. At rest and during moderate intensity exercise, the capacity of the transport mechanisms, particularly the Na^+-K^+ pump is adequate to maintain ionic balance sufficiently for it not to be a cause of fatigue (Sejersted, 1992).

2.3.3.2(b). Ionic Changes With High-Intensity Exercise

There is a multitude of ionic changes in the cytosol caused by and causing increased translocation of ions across the sarcolemma and the most pronounced changes are seen with high-intensity exercise (Sejersted, 1992). Therefore, both the extracellular and intracellular fluid compartments must be regulated in an integrated manner if excessive changes in the concentrations of ions are to be prevented or reduced so that muscle function can be maintained. During high-intensity exercise, it has previously been stated that the all motor units are recruited and high mean firing frequencies are achieved. This may place too great a demand on the mechanisms available which permit ions to pass across muscle membranes. During such exercise there is increasing evidence that K^+ mediated events in the sarcolemma may be a key factor integrating the cellular sites of fatigue (Lindinger *et al.*, 1995).

There are very little data where the direct measurement of the concentration of ions in the muscle has been made largely because of methodological difficulties. However, in one such study which employed a potassium sensitive electrode, extracellular potassium concentration rose from 4.5 to 9.5 mM during maximal voluntary contractions (Vyskocil *et al.*, 1983). During a 30 s maximal cycle sprint on an isokinetic cycle ergometer at 10.3 $rad \cdot s^{-1}$ (100 $rev \cdot min^{-1}$), the intracellular $[K^+]$ decreased by 19 mM (16 %) from 138±8 mM. The rate of K^+ release from skeletal muscle depends on the intensity of contraction and the number of action potentials per unit time (Lindinger *et al.*, 1995).

It is more usual for plasma concentrations to be measured and an increase to 8 mM follows high-intensity exercise (Sejersted *et al.*, 1984). The plasma potassium concentration changes in response to a single bout of exercise performed on an inclined treadmill at a speed causing exhaustion in 60 s were reported by Medbo and Sejersted (1990). In this study, catheters were inserted into the femoral veins and arteries in twelve male subjects (6 endurance and 6 sprint trained). Resting femoral-venous concentrations were 3.8 mM, which more than doubled during exercise to reach a peak of 8.3 mM. In the femoral artery, a similar peak concentration of 8.2 mM was obtained and both the venous and arterial concentrations decreased exponentially during recovery with a half-time of 25 s with little inter-subject variability. Interestingly, at 3-9 minutes into recovery there was an undershoot of approximately 0.5 mM compared with the resting values. Those subjects with the highest resting concentrations reached the highest peak and had the greatest undershoot

during recovery. These findings were later endorsed and expanded upon in a subsequent publication (Medbo and Sejersted, 1994). The authors reported that the rapid changes which took place in the immediate 10-20 s post-exercise period made precise temporal matching of the venous and arterial values impossible.

Muscle cells lose K^+ and gain Na^+ during the development of fatigue (Fitts and Balog, 1996). However, the concentration of both ions has been shown to increase in plasma during exercise and be restored much more rapidly than the restoration of plasma volume (Sejersted *et al.*, 1986). This somewhat surprising rise in the plasma concentrations of both Na^+ and K^+ ions, bearing in mind that the two ions move in opposite directions during an action potential, points to the importance of plasma volume shifts. During short lasting high-intensity exercise the active muscle takes up fluid and the haemoconcentration is related to the intensity of exercise. Reductions in plasma volume up to 20 % are often encountered during high-intensity exercise (Sejersted, 1992). The increase in plasma sodium concentration can be readily attributed solely to changes in plasma volume (Sejersted *et al.*, 1986; Sejersted, 1992). However, the changes in plasma potassium greatly exceed that which can be attributed to changes in plasma volume and has led to speculation that fatigue and potassium may be connected. Specifically, it has been suggested that because there is a low density of Na-K pumps in the t-tubular system, potassium ions may accumulate there so causing a depolarization block due to inactivation of the sarcolemmal channels (Sjogaard, 1990; Sejersted, 1992).

A doubling of the extracellular potassium concentration to approximately 8 mM would theoretically depolarize the cell membrane by 18 mV (Medbo and Sejersted, 1990). It has been hypothesised that a depolarization of this magnitude results in a reduced action potential amplitude and in some muscle cells complete inactivation (Sjogaard, 1990; Lindinger and Sjogaard, 1991). According to Edman (1992) an increased concentration of potassium in the t-tubules, possibly in conjunction with osmotic changes in the t-tubular membrane, will impair the inward spread of the action potential and so make the inner parts of the fibre inaccessible for any stimulus. Such a hypothesis was first described by Bezanilla *et al.*, (1972). However, whether such a situation ever occurs *in vivo* is uncertain.

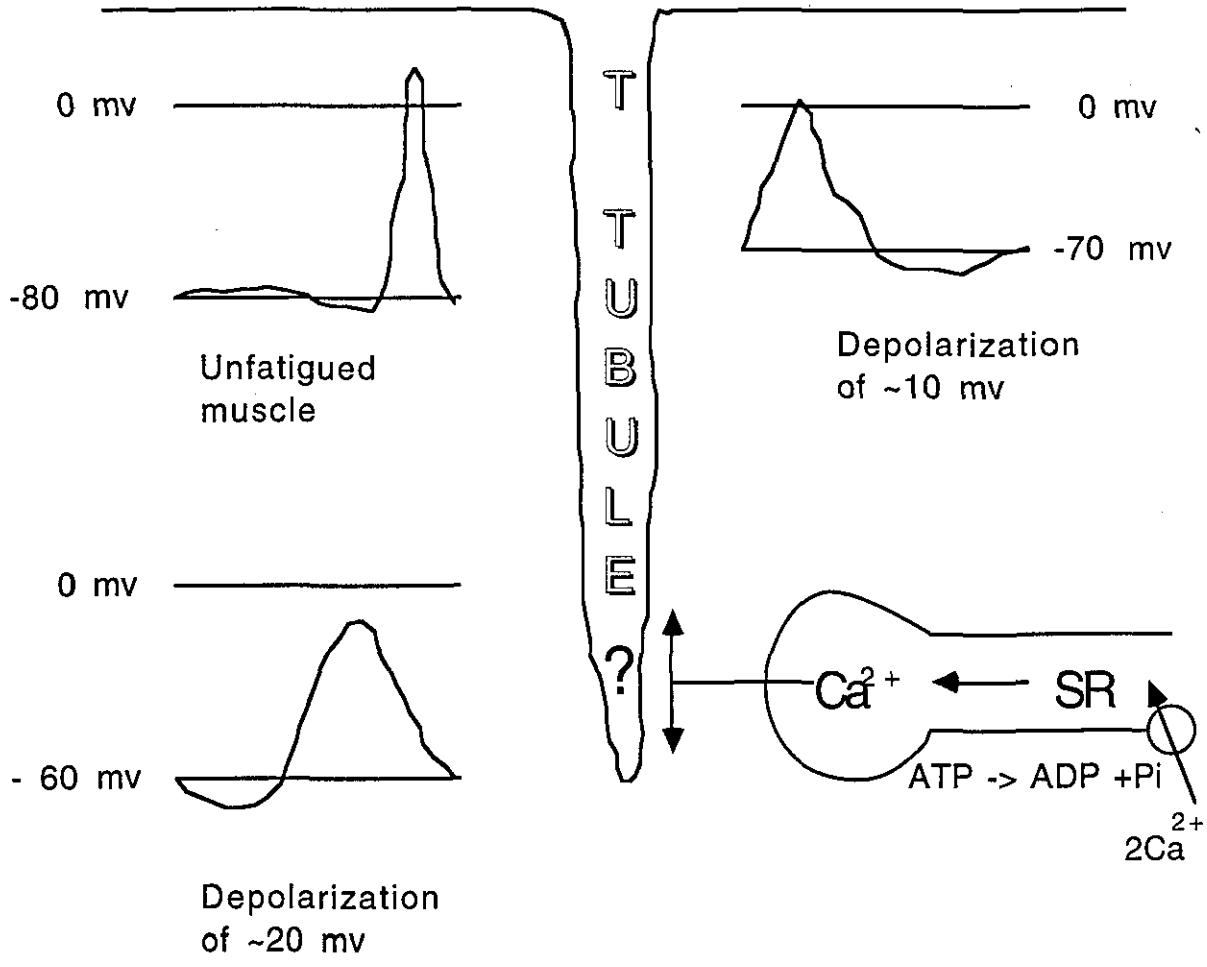

Fig. 2.16. A schematic representation of E-C coupling showing a sarcolemma action potential in the unfatigued state (top left potential) and following fatigue (top right). In the unfatigued state the resting membrane potential is approximately -80mV and with activation the spike height approaches +20mV. The action potential duration is short (1-1.5ms). With the development of fatigue, the resting membrane potential depolarizes by about 10mV, the action potential spike height declines by a similar magnitude and the duration becomes prolonged. It is unknown how fatigue affects the action potential in the depths of the t-tubule (bottom left) and the depolarization of approximately 20 mv is purely theoretical. The question mark indicates that the composition of the extracellular fluid in the depths of the t-tubule is currently unknown (Adapted from Fitts and Balog, 1996).

This hypothesis, that of accumulation of potassium in the t-tubules and associated depolarization, is shown diagrammatically in Figure 2.16. Using isolated bundles of fibres and whole soleus muscles from the rat, the relationship between extracellular potassium concentration, membrane potential and tetanic force has been described (Cairns *et al.*, 1995). An increase of the extracellular potassium concentration from 4 to 8 mM resulted in a depolarization from -74.1 to -62.2 mV but had little effect on tetanic force. However, further 1 mM increases in the extracellular potassium concentration resulted in small additional depolarizations of 1-2 mV but a large loss of force. At an extracellular potassium concentration of 12 mM, peak tetanic force was reduced by approximately 55 % and 90 %, in the bundles and whole muscles respectively, compared with that elicited at 4 mM.

The actual mechanism by which an increase in extracellular [K^+] and associated depolarization lead to such a dramatic loss of force is not fully understood. However, Ruff *et al.* (1988) have suggested that following mild depolarization there is a slow inactivation of of the inward Na^+ current which reduces excitability. Subsequently, Ruff and Whittlesey (1992, 1993) made similar observations on human muscle and also demonstrated that fast twitch fibres have a greater susceptibility to a loss of excitation through the slow inactivation of Na^+ channels, compared with slow twitch fibres. They demonstrated that in human intercostal muscle fibres, the membrane potential at which 50% of Na^+ channels were closed ($V_{s,1/2}$) due to slow inactivation was more negative in fast compared with slow fibres (-93.3mV for the fast vs. -71.6mV for the slow).

The Na^+-K^+ pump plays a vital role in restoring the concentration of intra- and extra- cellular potassium. During intense stimulation a failure of the pump to cope fully with the shifts could be a major contributing factor of the depolarization which arises. Its function has been extensively studied and reviewed (Clausen, 1986; Clausen *et al.*, 1997). According to Clausen *et al.* (1997), as long as the Na^+-K^+ can keep pace with the passive leaks arising from action potentials ionic balance will be maintained. Indeed in some instance a hyperpolarization may develop. The effectiveness of electrical propagation across the neuromuscular junction can be assessed by recording the muscle mass action potential (M-wave) evoked when single maximal shocks are to the motor nerve are superimposed during voluntary contractions. Using such a technique, stimulation of the human biceps brachii at 10-20 Hz enlarged the M-wave recorded from the muscle suggesting that neuromuscular transmission or muscle membrane excitability was enhanced (Cupido *et al.*, 1996). However at

higher frequencies (300-500 Hz), depolarization of rat gastrocnemius muscle developed (Locke and Solomon, 1967).

The short and long term regulation of the pump is under the control of a number of factors and these are shown in Figure 2.17. Therefore, using muscle preparations, the capacity of the pumps can be affected by changing one or more of the long and short term regulators of pump activity, or by using the inhibitor ouabain. The effect on pump function of pump stimulators, salbutamol, insulin and calcitonin gene-related peptide (CGRP), and the inhibitor, ouabain, at extracellular potassium concentrations of 4 and 11 mM in rat soleus muscle has been reported (Cairns *et al.*, 1995). Incubation of the muscle in the stimulators restored tetanic force by 26-55 % depending on the stimulant and $[K^+]$extr. Conversely, the inhibitor, ouabain, as expected reduced tetanic force at an extracellular potassium concentration of 11 mM but had no effect at 4 mM. Similarly, over the first 30 s of 30 Hz stimulation, pre-incubation in ouabain at concentrations of 10^{-5} and 10^{-3} M increased the rate of force decline by 200 and 360 % compared with muscle which had not been pre-incubated in ouabain (Nielsen and Clausen, 1996).

The authors concluded that Na^+-K^+ pump capacity and the run down of transmembrane gradients of sodium and potassium play important roles in determining the contractile performance of skeletal muscle.

To test this hypothesis, rat soleus muscles were stimulated at 90 Hz until such time as isometric force was reduced to 20 % of the unfatigued value. The recovery of force at 0.5, 1 and 5 minutes of recovery was tested. Muscles that had been incubated in ouabain, which inhibited approximately 80 % of the available pumps, had a 102 % faster rate of decline in force and had a 81 % slower initial rate of recovery compared with the controls (Fig. 2.18; Harrison *et al.*, 1996).

According to Clausen *et al.* (1997), these experiments have refuted previous assumptions that the Na^+-K^+ pump is a slow, long-term mechanism for the compensation of the excitation-induced rundown of Na^+, K^+ gradients and show that, in fact, it restores excitability by undergoing prompt and large scale activation. Skeletal muscle Na^+-K^+ pump activity during exercise *in vivo* may be estimated in two ways: from measures of arteriovenous $[K^+]$ difference immediately on cessation of exercise or from increases in intracellular muscle $[K^+]$ during the first minutes of recovery (Lindinger *et al.*, 1995).

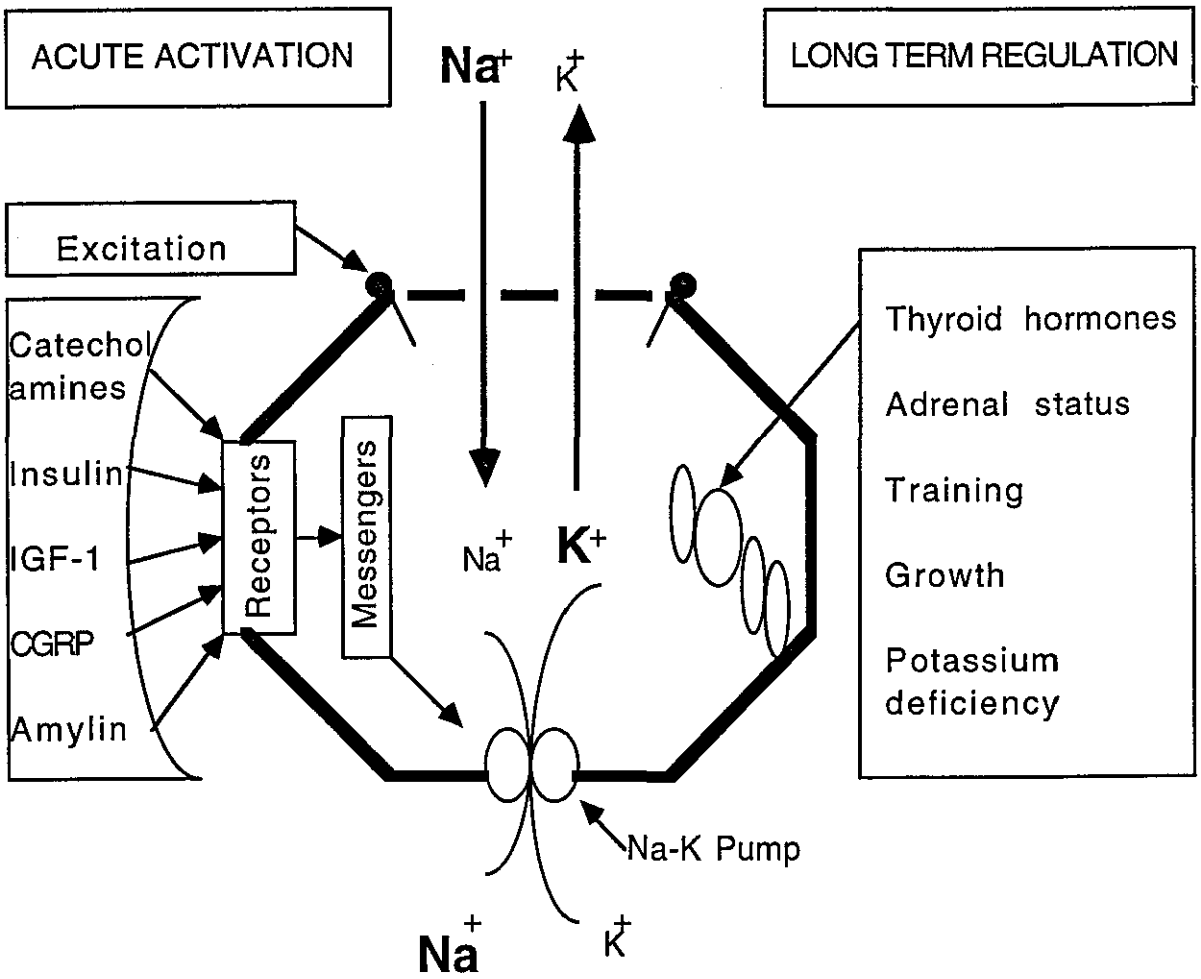

Fig. 2.17. Acute activation and regulation of the synthesis of Na-K pumps in skeletal muscle (Adapted from Clausen, 1996).

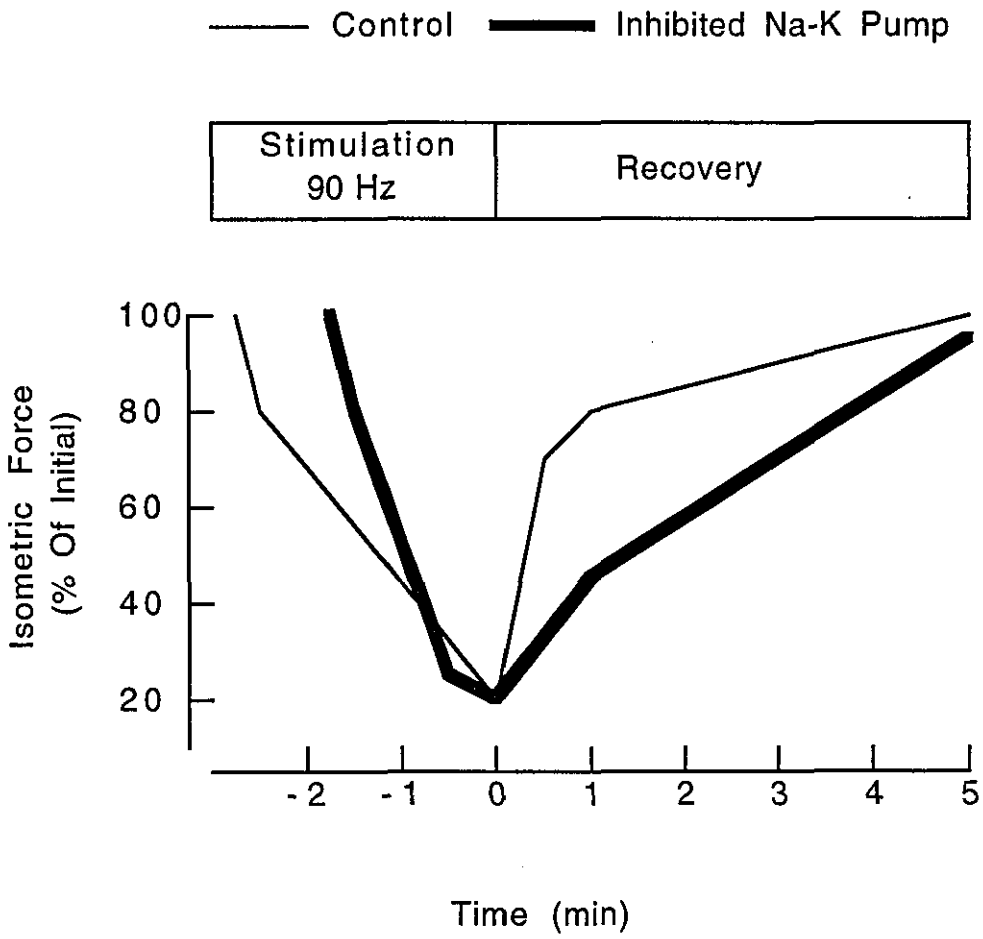

Fig. 2.18. Contractile performance of soleus muscles from 4-wk-old rats with (thick lines) or without (thin lines) ouabain pre incubation. (in Harrison *et al.*, 1996).

Despite the results of the animal experiments at similar extracellular potassium concentrations to those encountered during high-intensity exercise, it has been questioned whether cell depolarization is a cause of fatigue during voluntary human activity because of the very high stimulation frequencies required (Hicks and McComas, 1989). Such fatigue, termed high frequency fatigue, can be recognised by the following features (Jones, 1996):-

(i) Loss of force is accompanied by a loss of amplitude and slowing of the waveform of the muscle action potential.
(ii) The rapid loss of force encountered during stimulation at high frequencies is reversed rapidly by reducing the frequency.
(iii) Loss of force is exacerbated if extracellular [Na^+] is decreased or [K^+] increased.

The first feature of HFF, namely a change in the wave form of the muscle action potential, suggests that force loss may be due to failure of electrical propagation (Jones and Bigland-Ritchie, 1986). The second feature relates to the recovery of force. When frog and mouse muscle is electrically stimulated, force recovery is extremely rapid after high-frequency stimulation and may even occur during stimulation if the impulse frequency is lowered (Lannergren and Westerblad, 1986; Lannergren, 1992). Moreover, force recovers more rapidly than metabolite resynthesis could occur (Jones and Bigland-Ritchie 1986; Westerblad *et al.*, 1991). Indeed, according to Lannergren (1992), this rapid recovery of force suggests that metabolic changes are unlikely to be responsible and is probably due to the restoration of normal ionic composition in the t-tubules. The time-course of recovery of force in *Xenopus* fibres was investigated by Lannergren and Westerblad (1986). They reported that after continuous stimulation at 70 Hz force recovered in two phases, an initial rapid phase to 45-80% of the unfatigued value, followed by a much slower recovery, sometimes requiring up to an hour for completion. Interestingly, the half-time of the initial recovery was 1 s. The results from other studies have also shown that muscle recovers from fatigue in two distinct phases; a rapid phase complete within 2 minutes and a slower phase lasting 30-60 minutes (Fitts and Holloszy, 1978; Thompson *et al.*, 1992). These kinetically distinct phases of recovery may represent at least two distinct mechanisms of fatigue. According to Fitts and Balog (1996) the second phase is related to the increases of H^+ and P_i whilst the initial rapid phase probably involves one or more steps in the E-C coupling process.

Although the features of high-frequency fatigue have been widely studied in animal muscle, there is less in the literature where the data have been obtained from humans. However, one of the features of HFF is a rapid recovery of force and the pattern of recovery of force following four 15 s tetanic contractions, separated by 5 s rest, of human tibialis anterior muscle at 15, 30 and 60 Hz has been reported (Sacco *et al.*, 1994). During the first two minutes of recovery, the extent of recovery was dependent on the prior stimulation frequency with a rapid recovery at 60 Hz but no recovery during this period following stimulation at 15 Hz (Fig. 2.19). The authors speculated that the high frequency stimulation caused the greatest disturbance to the extracellular cation concentrations and the extent of action potential failure would be dependent upon on a number of factors including the frequency of stimulation. These changes would be expected to be reversed rapidly and the differential rates of force recovery could reflect the extent of the ionic disturbance during prior stimulation.

Interestingly there are also some reports in the literature which suggest that HFF, although not a feature of normal activity, may play a role in the fatigue during high-intensity exercise. For example, Lindinger *et al.* (1995) state that a decreased intracellular [K^+] and increased extracellular [K^+] resulting from high-intensity muscle contraction are associated with decreased muscle E_m and function. While maximal *in vivo* activation of the Na^+-K^+ pump may occur within 10-30 s, this is insufficient time to restore intracellular [K^+] during 30 s of maximal isokinetic cycling.

2.3.3.2(c). Effect Of Changes In Pedalling Rate Upon The Extent Of The Ionic Changes

If HFF does play a role during high-intensity cycle exercise, then the previous section has emphasised that there should be an initial rapid recovery of force and possibly power output in the first few seconds after exercise. If this is so, it might be expected that changes in pedalling rate thereby impacting upon the total number of muscle actions might affect the pattern of this initial recovery phase.

Medbo and Sejersted (1990) considered whether manipulation of the experimental conditions might affect the extent of the movements of K^+. Eight male subjects were divided into two groups (four endurance trained, the others being sprint trained) for both strategies. In one strategy, they completed 5 x 1

minute exhausting bouts of inclined treadmill running with 4 minutes recovery in-between bouts. If a subject could not maintain the pace during a particular bout then he could stop but had to attempt all 5 bouts. Although the total distance run was similar between the two groups over the course of the five bouts, the pattern of work production was not. Both groups could maintain the pace (5 $m \cdot s^{-1}$ and 5.6 $m \cdot s^{-1}$ for the endurance and sprint groups respectively) for virtually the whole minute for the first two bouts but by bout 3, the endurance athletes maintained a longer duration of exercise (56 s) compared with the sprint group (35 s); this pattern continued through to bout 5. Namely, there was an increase of 3.6-3.8 mM for both groups during bouts 1 and 2 but during bouts 3, 4 and 5, the concentration in the sprint trained group was lower showing that the changes in plasma potassium reflected the exercise duration of each bout. The authors also found that manipulating the treadmill speed, whilst keeping the duration fixed, also influenced the extent of the changes in plasma $[K^+]$. There are no data which have considered whether changes in pedalling rate affect the magnitude of the ionic movements during high-intensity exercise.

It has already been stated that an increased extracellular potassium (and increased intracellular sodium) concentration could reduce force by making some fibres inexcitable (Fig. 2.16). It has, therefore, been suggested that when muscle fibres are electrically stimulated, strong electrical pulses may result in a greater 'safety margin' for action potentials compared with weaker pulses as fewer fibres would become inexcitable (Cairns *et al.*, 1995). To test this hypothesis, the effect of manipulating the electrical pulse frequency and amplitude upon the performance of rat soleus muscle at an extracellular potassium concentrations of 4 and 11 mM was investigated. Manipulation of the pulse duration from 0.1 ms to 1.0 ms had no effect at an extracellular potassium concentration of 4 mM but at 11 mM the force reduction was almost doubled with the shorter duration pulses compared with the longer. At the higher concentration, doubling the voltage strength increased force by as much as 35 %.

Changes in pedalling rate would affect the average duration of each muscle action and the total number of actions. Particularly during high-intensity exercise, where the stimulation frequencies are high, changes in average pedalling rate may greatly affect the extent of the ionic movements resulting in greater fatigue at high as opposed to low rates.

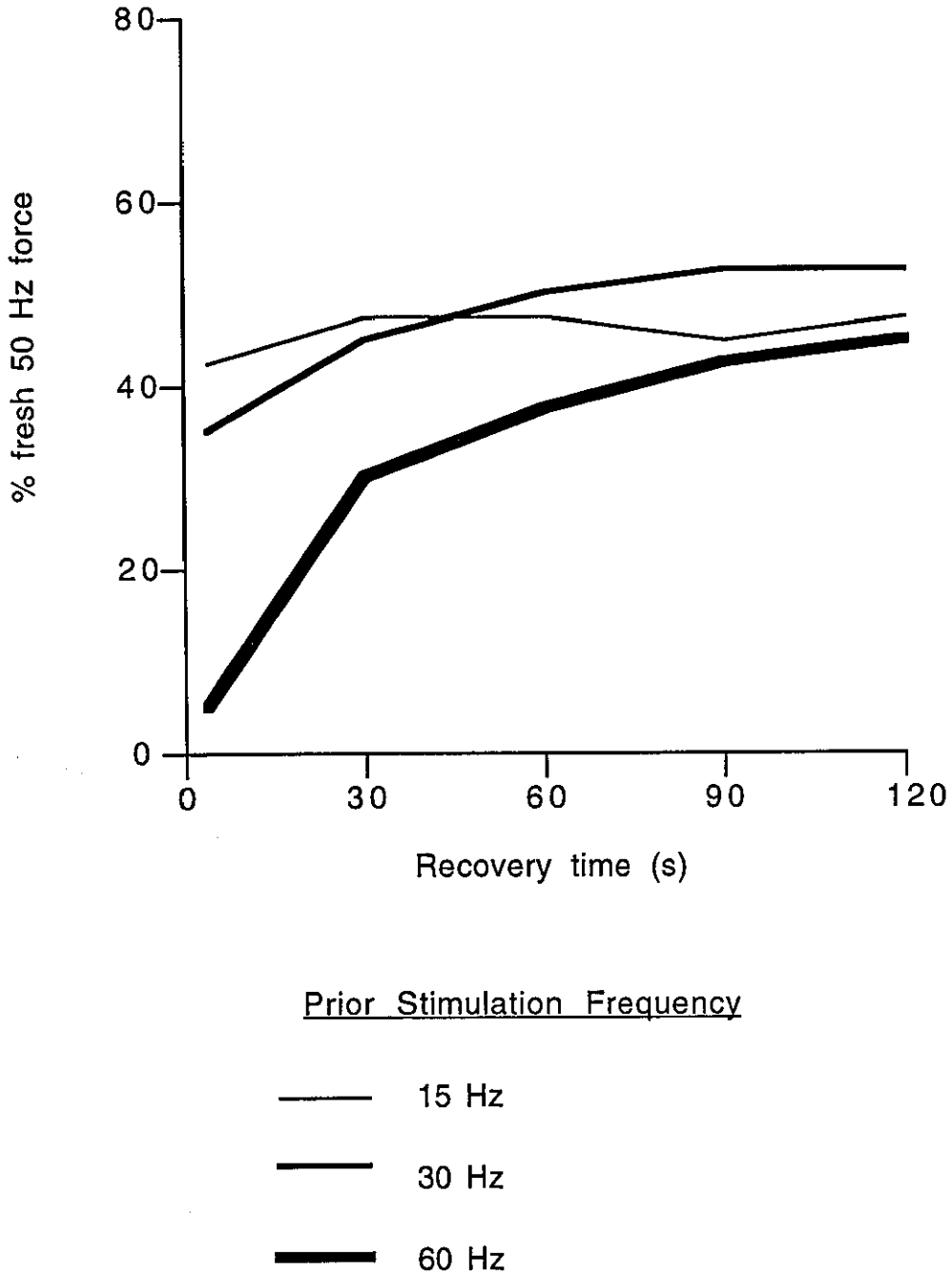

Fig. 2.19. Force recovery after previous electrical stimulation of 4 x 15 s tetanii with 5 s rest in human tibialis anterior muscle at different stimulation frequencies. During the first few seconds of recovery there is a rapid recovery of force following prior stimulation at 60 Hz but not when the stimulation frequency had been lower (in Sacco *et al.*, 1994).

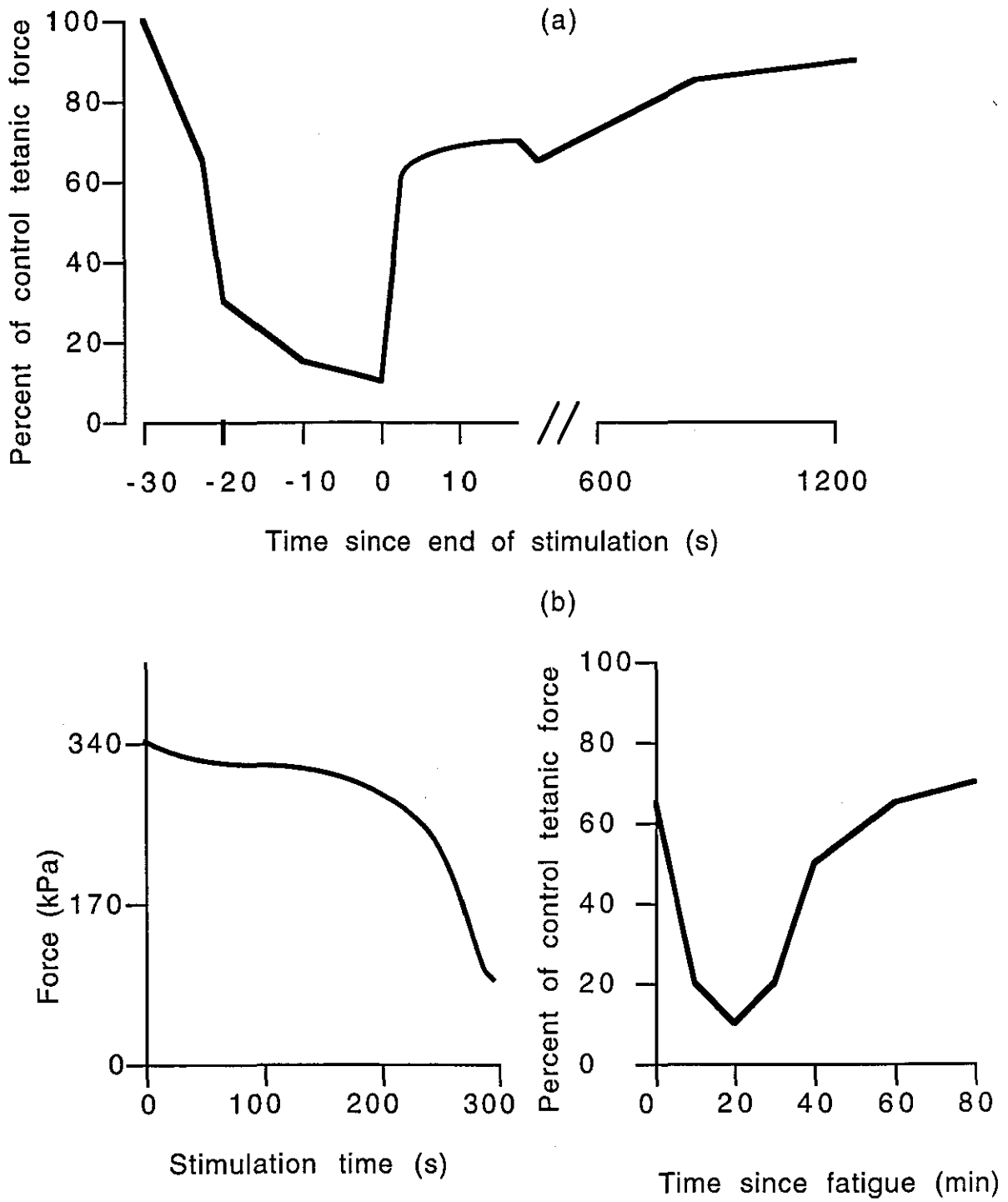

Fig. 2.20. Decline and recovery of tetanic force depends upon the stimulation pattern used to induce fatigue. (a) continuous 70 Hz stimulation for 30 s causes a rapid, large fall in tetanic force which recovers rapidly (b) Intermittent stimulation at 70 Hz where pulses are separated by 4 s results in a gradual loss of force which continues into recovery (in Lannergren and Westerblad, 1996).

2.4. HYPOTHESES

This review has shown that intense muscle stimulation such as that encountered during high-intensity exercise results in extensive mechanical, electrical and biochemical changes. Changes in average pedalling may influence the magnitude of these changes although it is unlikely that the available technology is sophisticated enough to detect any differences in the magnitude of the electrical changes where the movement involves a large muscle mass. Thus, the experiments described in this thesis will try to establish whether the magnitude of the mechanical and biochemical changes is affected by changes in average pedalling rate.

2.4.1. Mechanical Changes

The model of repeated exercise has many advantages compared with a single bout of exercise as a tool for investigating the aetiology of fatigue during high-intensity exercise. In view of the rapid and substantial mechanical changes that take place during exercise it is possible that some are reversed in a similar fashion. The extent of the mechanical changes will be assessed by considering the very short term (2-3s) recovery of power and the pattern of recovery of power output from 0-300s following high-intensity exercise at different average pedalling rates. It is hypothesised that the average pedalling rate at which high-intensity exercise is performed will influence the extent of the mechanical changes. It is also hypothesised that the mechanical changes will be reversed very rapidly and that some of the features of high-frequency fatigue will be exhibited.

2.4.2. Biochemical Changes

(i) Metabolic Changes

Previous research has suggested that the ability of muscle to generate power during high-intensity exercise is highly correlated with the concentration of PCr but only poorly correlated with other metabolic indicators. However, there is no evidence to suggest that the extent of PCr utilisation is affected by changes in average pedalling rate. It is hypothesised that the magnitude of the metabolic changes will be unaffected by changes in average pedalling rate but that the magnitude of the mechanical changes will be influenced by the speed and number of preceding muscle actions.

(ii) Ionic Changes

Research on muscle preparations has suggested that after intense stimulation, there is an initial rapid recovery of force connected with the restoration of ionic balance. Changes in average pedalling rate may influence the magnitude of the ionic disturbance. The cause of high frequency fatigue is thought to be largely ionic and it is hypothesised that there will be a close link between the magnitude of the mechanical and ionic changes.

CHAPTER 3

GENERAL METHODS

This Chapter describes the methods which are common to more than one of the experimental Chapters (4-7) of this thesis. It begins with a description of the equipment, instruments and calibration routines used. The procedures employed to collect, handle and analyse the blood samples obtained in Chapters 4 and 6 are then described. Further details regarding the blood assays can be found in Appendix B. Muscle samples were only obtained in one study and the procedures relating to their collection and handling can be found in the relevant Chapter (Chapter 6). Full details of the assays used to analyse for mixed muscle metabolites and validate standards can be found in Appendix C.

3.1. SELECTION OF A SUITABLE MODE OF EXERCISE

A number of techniques have been developed to measure power output during high-intensity exercise. Some have permitted measurements for movements that actually take place in sports events whilst others have concentrated on a specific movement about a single joint. Examples of equipment developed to measure movements in sports include a specialised multi-purpose ergometer which permits measurements during rowing (Harrison, 1970); a non-motorised treadmill for sprint running (Lakomy, 1987) and many cycle ergometers where the external angular velocity is controlled (McCartney *et al.*, 1983b), or not (Martin, 1914; Williams *et al.*, 1988). For movements about a specific joint, isokinetic dynamometers have become a favoured method for the assessment of dynamic muscle function in both sports environments and clinical research (Gleeson and Mercer, 1996).

The muscles surrounding a joint can generate mechanical energy and absorb energy by concentric and eccentric actions respectively. It was emphasised in Chapter 2 that whilst the force- and power-velocity relationships of concentric muscle actions have been extensively investigated, little is known concerning the same properties of muscle lengthening. Therefore, in a thesis concerned with the effect of changes in average muscle action speed upon a number of parameters, it is vital that the contribution to power output of the mode of exercise should be wholly or predominantly provided by concentric muscle actions. Such movements can be performed on an isokinetic dynamometer but

according to Beelen *et al.* (1994) they are only a reliable tool for measuring maximal power at low external angular velocities up to about a quarter of the maximal voluntary velocity. This is because the muscles must accelerate from a resting position to achieve the controlled velocity. During walking and running, the hip and knee muscles work both concentrically and eccentrically and the knee extensors absorb (eccentric) approximately 3.5 times as much energy as they generate (concentric) during slow jogging (Winter, 1983). Conversely, the power output and work in different muscle groups during ergometer cycling has been shown to be predominantly concentric (Table 3.1; Ericson *et al.*, 1986). However, the point should be made that cycling is not exclusively a concentric work exercise as is commonly stated, although the eccentric work is comparatively small.

3.2. HIGH-INTENSITY EXERCISE AND CYCLE ERGOMETRY

The use of cycle ergometry as a laboratory tool for the testing of dynamic muscle function has a long pedigree. Dickinson (1928) described a friction loaded cycle ergometer which had been designed by Martin in 1914 to investigate the dynamics of cycle pedalling. In her study, the subjects pedalled as fast as possible for about 10 seconds from a stationary start against a range of applied loads. The peak pedal rate at each load declined as the applied load increased. From these data, a linear force-velocity curve was constructed which was extrapolated to the axes to give the maximal force during an infinitely slow action (analogous to P_O) and the maximum rate at which the subjects could pedal (analogous to V_{max}). She concluded that it would be possible to calculate an optimal velocity at which power would be optimised. Interestingly, in the same paper mention was made that during friction loaded cycle ergometry, the force required to move the ergometer had two components; a frictional element to overcome the force exerted at the rim of the wheel and a correctional force equal to the change of momentum of the moving parts multiplied by a suitable constant.

Many decades later, a simple cycle ergometer test was introduced (Cumming, 1974) to assess high-intensity exercise, developed and culminated in the 'Wingate' protocol (Bar-Or, 1978). This protocol required the subject to pedal as fast as possible on a friction-loaded ergometer, a resistance, usually equal to 75 $N \cdot kN^{-1}$ body weight was then applied. The subject continued to pedal as fast as possible for a period of 30 s and external work was measured, simply as the frictional force applied to the belt, multiplied by the distance travelled by the

flywheel, over 5 s averaging periods. The anaerobic capacity has been defined as the maximal amount of ATP which can be formed by the anaerobic processes during exercise (Medbo *et al.*, 1988) and the Wingate protocol has often been used with the intention of measuring a subject's maximal anaerobic capacity. However, this practice has been questioned (Bulbulian *et al.*, 1996) and other tests developed involving different durations of exercise and applied resistances (Katch *et al.*, 1977; Gastin *et al.*, 1991; Gastin and Lawson, 1994). However according to Saltin (1990), an accepted method of measuring a person's anaerobic capacity is not yet available.

In addition to measuring total external work production, high-intensity cycle tests also provide an opportunity to measure a persons ability to generate their maximal or peak power output. However, the original Wingate protocol did not give an accurate reflection of their ability to generate peak power. The method assumed that the flywheel was revolving at a constant external angular velocity during the averaging period whereas, in fact, this was not the case. For the correct measurement of muscle function, the measurement must be made at a velocity at which the force-velocity relationship is optimised (Wilkie, 1960). Due to the rapid changes in pedalling speed, this is only the case for a minor part of the time during a 5 s averaging period. This shortcoming led to the development of the isokinetic or constant velocity cycle ergometer described in Chapter 2. These ergometers permitted the experimenter to choose a velocity which was optimal as defined by the power-velocity relationship of muscle. Force data is collected via the strain gauges on the pedals and averaged over a short time span such as a half-pedal revolution.

To overcome the problems inherent with the measurement of peak power on friction-loaded cycle ergometers, two different types of procedures were developed which can be categorised as optimisation and correction procedures (Winter *et al.*, 1996).

	Concentric work			Eccentric work		
	Peak power (W)		% of total concentric work	Peak power (W)		% of total eccentric work
	mean	SD		mean	SD	
Hip extensors	74.4	29.4	27	8.9	6.7	32
flexors	18.0	6.6	4	0.0	0.0	0
Knee extensors	110.1	25.9	39	4.5	2.1	7
flexors	30.0	10.5	10	8.3	8.6	4
Ankle plantar flexors	59.4	12.1	20	11.4	4.1	57
TOTAL	291.9		100	33.1		100

Table 3.1. Peak power output from concentric and eccentric muscle work during ergometer cycling at 120 W, 6.3 rad·s^{-1} (60 rev·min^{-1}), (n=6 male subjects) (in Ericson *et al.*, 1986).

Optimisation Procedures

These procedures have been presented in a number of papers (Nakamura *et al.*, 1985; Vanderwalle *et al.*, 1985; Winter *et al.*, 1991) and rely on the inverse linear relationship between applied load and peak pedalling rate first demonstrated by Dickinson (1928; 1929). This relationship can be described by the following equation (Winter *et al.*, 1996):-

$$R = a + bL$$

Where:-
R is the peak pedal rate
L is the applied load
a is the intercept
b is the gradient

From this equation an expression for power output can be produced which, in turn, can be differentiated to obtain the gradient of the power-load curve. This procedure, described in more detail by Winter *et al.* (1996), enables the optimised peak power as well as the optimal load and pedalling rate to be determined.

Correction Procedures

The calculation which assumes that power output is the product of applied resistance and the number of flywheel revolutions at a constant average angular velocity and has no moment of inertia is invalid (Lakomy, 1986; Bassett, 1989). Thus, corrective procedures have been introduced which take into account the additional work that is required to accelerate the flywheel. This was clearly demonstrated by Lakomy (1986) who compared peak power output attained during a standard 'Wingate' protocol with one corrected for the acceleration of the flywheel. The protocol made use of a micro computer so that power values could be recorded over intervals as short as 0.1 s (cf. 5 s in the 'Wingate' protocol). The results found that when no correction was made peak power was greatly (approximately 32 %) underestimated. Also, the time to reach peak power was reduced when the correction was applied. Total external work over 30 s was similar although the power values averaged over 5 s intervals were different. The important aspect of the procedure was an 'accelerating balancing load' which was the force required at any instant to stop the subject accelerating the flywheel. The basis of the corrected methods can be summarised in the following equation:-

$$T_{tot} = T_B + T_K \qquad \text{(in Seck } et\ al.\text{, 1995)}$$

Where:-

T_{tot} = total torque

T_B = the braking torque

T_K = the torque necessary to accelerate the flywheel.

Once the corrected method is employed, the recorded peak power output values accord very closely with those attained with other methods such as the isokinetic system, provided the muscle is able to pass through its optimal velocity (Seck *et al.*, 1995). Indeed, isokinetic, or non-isokinetic cycle ergometers where either optimisation or correction procedures are employed, can both be used in different studies to measure maximal performance and individual force- and power-velocity relationships. For example, similar linear relationships between force and velocity to those found on isokinetic systems have been found on friction loaded ergometers (Vanderwalle *et al.*, 1985; Arsac *et al.*, 1996; Buttelli *et al.*, 1996). Despite these advances in methodology, it must be borne in mind that both the isokinetic and non-isokinetic systems are not perfect models. On an isokinetic ergometer, the left shift of the force-velocity relationship of muscle with fatigue will mean that the imposed pedal rate will not be optimal from the start to the end of a fatigue test. Similarly, on a friction loaded ergometer, T_B will not be optimal throughout because of the decrease in P_o with fatigue (Buttelli *et al.*, 1996). The choice between them must be decided by the necessity for controlling experimental conditions or for being as close as possible to normal sprint conditions (Hautier *et al.*, 1996). In this thesis, the friction loaded ergometer was chosen as the limbs are free to accelerate in a manner similar to that found in sport and exercise. However, the cycle ergometer should not be regarded as exact replication of conditions encountered in cycle racing where 'drag' and rolling resistance are major factors influencing the ability to generate power (De Groot *et al.*, 1994; 1995).

3.3. EQUIPMENT

In all the studies in this thesis a modified friction-loaded cycle ergometer (Monark, model 864), interfaced to a micro-computer (BBC, model B), was used in order to calculate the "corrected" power output (Lakomy, 1986). An electric generator was connected to the ergometer flywheel, which gave an analogue signal proportional to the external angular velocity of the flywheel. The signal was logged by the micro-computer via an analogue-to-digital (A-D) converter,

combined with a timing signal derived from the computer's internal clock. The sampling rate was 20 Hz.

Before and at the end of each experiment and at the beginning of each day's trials, the relationship between the flywheel external angular velocity and the output from the generator and A-D converter was calibrated. The ergometer was pedalled for approximately 100 s at 6.3 $rad \cdot s^{-1}$ (60 $rev \cdot min^{-1}$) and pedal revolutions counted. The deceleration time of the flywheel for three different loads was also determined by pedalling in excess of 12.6 $rad \cdot s^{-1}$ (120 $rev \cdot min^{-1}$), ceasing pedalling, and recording the time it took the flywheel to stop. These measures enabled a linear regression equation of load against flywheel deceleration to be obtained from which the "accelerating balancing load" could be calculated (for a full explanation, see Lakomy, 1988).

The magnitude of the measured power output value may be influenced by many factors in addition to those which the experiments attempted to manipulate. These include crank length, pelvic inclination, saddle height (Nordeen-Snyder, 1977; Yoshihuku and Herzog, 1996); muscle (Sargeant, 1994) and ambient (Ball *et al.*, 1997) temperature. In order to control to some extent for these variables, a number of procedures were standardised for all trials. For each subject, the same saddle height was used so that at the bottom of the pedal stroke slight flexion of the knee occurred. Also, a restraining harness was passed around the subject's hips to restrict exercise to the lower limbs and prevent subjects rising from the saddle. This was especially important as the degree of pennation influences the size of the torque which muscle can produce at a joint (Herbert and Gandevia, 1995). The harness was positioned so that it did not interfere with the subject's breathing and did not generate too much pressure on the subject's stomach. The two side straps of the belt were fixed to a hook which, in turn, was fixed to a metal rail bolted to the floor behind the ergometer. A number of holes were drilled in the metal rail so that the tension could be adjusted; the same setting was used for all trials. Unfortunately, the testing laboratory was not air-conditioned so it was difficult to control accurately for ambient temperature but all trials were preceded by a standardised warm-up and at a similar time of day so that the inter-trial variation in muscle temperature was probably not too great.

3.4. SUBJECTS

Because of the demanding nature of the exercise performed in the studies described in this thesis, only subjects regularly participating in sport and exercise were recruited. Previously, research involving high-intensity exercise has been reported largely upon data from male subjects. The changes in the isometric force which can be generated by the quadriceps during the menstrual cycle have been reported (Sarwar *et al.*, 1995). The results showed that there was an 11 % increase in isometric force between the 1 st and 14 th day of the cycle. Phillips *et al.*, 1996 recorded the maximum voluntary force (MVF) that was achieved by the adductor policis muscle over a period of six months. Their results showed that there was a significant increase in MVF in both trained and untrained female subjects of about 10 % during the follicular phase, when oestrogen levels are high, followed by a significant decrease at around the time the subjects were ovulating, when oestrogen levels are low. These cyclical changes in MVF were not found in either male subjects or a separate group of female subjects who were taking oral contraceptives. *Prima facie,* these results suggest that the use of female subjects for tests requiring them to exert a maximal, or near maximal, effort should be avoided unless all tests are carried out at a similar stage of their menstrual cycle. One method of determining a females menstrual cycle is to ask them to record their temperature each morning. However, this requires a significant degree of co-operation of the subjects and may be an unreliable procedure for trained athletes who may have irregular menstrual cycles.

With these problems in mind, a pilot study was performed which measured the coefficient of variation of peak power output and average pedalling rate during 10 separate sprints performed by 6 male and female subjects. This study is presented in full at the end of this section and the results refute suggestions in the literature that the performance of highly-trained females is influenced by hormonal changes in their menstrual cycle. In view of the findings of this pilot study, in all the main studies described in this thesis, at least half of the subject group is female. Participation was voluntary and no remuneration, this includes travel and other expenses, was given to any of the subjects, although they were provided with free post-trial food and drink.

A PILOT STUDY
THE COEFFICIENTS OF VARIATION FOR REPEATED SPRINTS FOR MALE AND FEMALE SUBJECTS

Introduction

Animals are often used as subjects for experiments because the potential risks and hazards are too great to recruit humans. However, in other experiments where there is a choice, animals are still the preferred model because of the many practical advantages they have over humans. Where humans are used, the vast majority of studies which have investigated short-term high-intensity exercise have recruited mostly or totally male subjects. Indeed a considerable number of articles have tried to establish whether there is a sex difference (see Kanehisa *et al.*, 1996) but it is still a subject of debate. Also, it has been widely suggested that the female menstrual cycle and the associated hormonal and mood changes make repeated measures of performance in females very unsatisfactory (Phillips *et al.*, 1996).

The purpose of the present study was to measure the coefficients of variation for a number of measures frequently quoted in the main studies described in this thesis and to ascertain the extent of any sex difference.

Methods

Twelve subjects volunteered to take part in the study and their physical characteristics are shown in Table 3.2. All the subjects were physically very active, performing or training on at least 6 occasions per week. After familiarisation, the subjects came to the laboratory on 2-3 occasions per week over a four week period so that a total of 10 main trials were completed. On each visit, the subjects completed a maximal 30 s sprint from a standing start against an applied resistance of 75 $N \cdot kN^{-1}$ body weight. The start and finish of each sprint were relayed to the subject by loud computer generated sounds (for full details refer to Chapter 7 "Methods"). The coefficients of variation for peak power output and the number of muscle actions were calculated for each subject. The formula for the coefficient of variation is given by the formula below where S.D. is the standard deviation and M_S is the mean of the sample :-

$$V = \frac{100\ S.D.}{M_S}\ \%$$

(in Cohen and Holliday, 1982)

SUBJECT	AGE	HEIGHT (m)	MASS (kg)
1-Male	21	1.86	88.7
2-Male	30	1.79	69.3
3-Male	25	1.88	89.6
4-Male	34	1.75	80.0
5-Male	31	1.74	81.5
6-Male	29	1.72	69.0
7-Female	22	1.66	62.1
8-Female	23	1.55	52.3
9-Female	22	1.59	51.0
10-Female	20	1.71	57.9
11-Female	21	1.72	74.6
12-Female	20	1.57	55.0
Average ALL	25	1.71	69.3
SD ALL	5	0.10	13.2
Average MALE	28*	1.79*	79.6*
SD MALE	5	0.06	8.2
Average FEMALE	21	1.63	58.8
SD FEMALE	1	0.07	7.9

Table 3.2. Physical characteristics of the subjects (*=significantly greater than females)

Results

The individual data are depicted in Tables 3.3-3.6. The group data are summarised in Table 3.7. The coefficients of variation for peak power output were higher ($P<0.05$) compared with the coefficients of variation for the total number of muscle actions. For both measurements, there was no sex difference although values tended to be lower in the females.

Discussion

The coefficient of variation for power output of 5.5% is a little higher than the test-restest value of 2.6% reported by Winter *et al.* (1996). However, the value in the present study of 2.8% for the number of muscle actions compares favourably with the 5.5% figure for pedalling rate also reported by Winter and colleagues. The range of inter-subject variation was quite small despite differences in the subjects physical characteristics, although the coefficients of variation for peak power output of some individual subjects are a little high (for example male subject 3 at 9.2%) Nevertheless, for the group as a whole the quite low coefficients of variation for the two measured variables suggest that values obtained using highly trained subjects can be confidently treated as being close to the true value.

The findings do not provide any support for the argument that the performance of females is subject to greater variability compared with males because of the changes associated with their menstrual cycle. Indeed, the females achieved marginally lower coefficients for both measured variables. It should, however, be stressed that the female subjects used in this study were highly active and were accustomed to intense physical activity on most days. Therefore, the use of similarly highly active female subjects in repeated tests of high-intensity exercise would appear to be a satisfactory experimental model.

PEAK POWER OUTPUT-MALE

	SUBJECT NUMBER					
SPRINT NUMBER	1	2	3	4	5	6
1	1410	1054	1428	1138	1257	1196
2	1678	1056	1745	1127	1063	1235
3	1644	1054	1545	1228	1045	1252
4	1731	1031	1784	1114	1105	1275
5	1513	1062	1661	1125	1138	1378
6	1641	994	1512	1194	1106	1292
7	1679	986	1844	1199	1089	1389
8	1768	1032	1388	1174	1109	1213
9	1467	955	1557	1034	1048	1322
10	1640	1022	1501	1073	1176	1265
MEAN	1617	1025	1597	1141	1114	1282
SD	110	34	147	60	61	62
CV(%)	6.8	3.3	9.2	5.3	5.5	4.8

Table 3.3. Peak power output (W) achieved by 6 male subjects on 10 separate occasions.

PEAK POWER OUTPUT-FEMALES

	SUBJECT NUMBER					
SPRINT NUMBER	7	8	9	10	11	12
1	741	732	721	818	1179	766
2	724	672	737	723	1287	892
3	752	624	768	779	1200	853
4	727	602	745	717	1178	923
5	776	700	759	723	1353	831
6	740	697	750	715	1173	782
7	708	616	649	760	1293	848
8	703	631	779	708	1151	762
9	686	627	716	705	1299	821
10	692	639	726	672	1290	818
MEAN	725	654	735	732	1239	830
SD	27	41	35	40	66	50
CV(%)	3.7	6.3	4.8	5.5	5.3	6.0

Table 3.4. Peak power output (W) achieved by 6 female subjects on 10 separate occasions.

NUMBER OF MUSCLE ACTIONS-MALES

	SUBJECT NUMBER					
SPRINT NUMBER	1	2	3	4	5	6
1	119.6	111.1	111.1	107.2	112.2	119.8
2	134.1	113.2	120.9	102.2	108.7	115.7
3	123.0	106.8	111.5	103.0	107.9	110.9
4	124.8	110	119.4	103.3	111.4	113.1
5	125.8	112.3	108.7	101.5	107.8	118.5
6	124.7	109.3	109.1	100.7	109.7	114.6
7	122.9	112.4	120.2	104.9	105.1	120.7
8	128.4	115.5	110	107.3	106.7	118.2
9	129.6	110.5	120.7	108.9	103.9	119
10	126.0	112.4	118.0	105.1	107.0	116.4
MEAN	125.9	111.3	115.0	104.4	108.0	116.7
SD	3.8	2.3	5.0	2.6	2.5	3.0
CV(%)	3.0	2.1	4.3	2.5	2.3	2.6

Table 3.5. The total number of muscle actions regarded as the mean average pedalling rate in rev·min^{-1} achieved by 6 male subjects during a 30 s maximal sprint against an applied resistance of 75 N·kN^{-1} body weight on 10 separate occasions.

NUMBER OF MUSCLE ACTIONS- FEMALES

	SUBJECT NUMBER					
SPRINT NUMBER	7	8	9	10	11	12
1	86.7	98.0	105.0	102.8	116.8	112.2
2	85.2	98.7	107.7	103.1	111.9	111.5
3	90.5	98.1	112.8	100.9	115.4	111.6
4	92.0	89.7	105.5	96.9	109.4	114.2
5	89.4	90.4	111.9	100.2	113.5	107.8
6	89.6	94.8	110.4	100.8	112.7	114.2
7	86.7	93.4	104.2	101.1	115.8	108.8
8	89.0	91	110.0	101.0	113.3	108.2
9	90.0	94.2	108.7	95.8	114.7	111.0
10	90.1	93.3	107.1	98.1	115.7	109.2
MEAN	88.9	94.2	108.3	100.1	113.9	110.9
SD	2.0	3.1	2.8	2.3	2.1	2.2
CV(%)	2.2	3.3	2.6	2.3	1.8	2.0

Table 3.6. The total number of muscle actions regarded as the mean average pedalling rate in rev·min^{-1} achieved by 6 female subjects during a 30 s maximal sprint against an applied resistance of 75 N·kN^{-1} body weight on 10 separate occasions.

SUMMARY OF GROUP DATA

	PEAK POWER OUTPUT	MUSCLE ACTIONS
ALL SUBJECTS	5.5±1.5%	2.8±1.2%
MALE SUBJECTS	5.8±1.8%	3.3±1.4%
FEMALE SUBJECTS	5.3±0.8%	2.4±0.5%

Table 3.7. Summary of the coefficients of variation for peak power output and total number of muscle actions (n=12; 6M,6F)

At the commencement of each study, subjects came to the laboratory for a number of preliminary visits. On the first visit, they were given verbal details of the purposes and protocol of the study, including details of any potential risks, and asked to complete a health history questionnaire (Appendix A). Any subject with any medical condition that might have been a potential problem was excluded. Written details of the study were handed to the subjects together with a consent form (Appendix A). The subjects were asked to return at a future date if they wished to participate and only to sign the consent form when they were positive they wished to take part. Once recruited, the subjects attended 2-7 (typically 3) more familiarisation sessions where they became acquainted with the equipment and experimental protocol. Usually this was achieved by performing an abridged version of the main trials until a consistent level of performance was achieved. On the day of, and in the days before each main trial, subjects were asked to perform, as far as possible, the same pattern of physical activity and repeat their diet, although the only strict directive in these respects was that they should not eat anything in the two hours before each trial.

3.5. PROCEDURES FOR THE SAMPLING, HANDLING AND ANALYSIS OF BLOOD SAMPLES

It is possible to extract arterial, capillary, arterialised venous and venous blood samples from exercising subjects. Arterial blood enables investigators to study many physiological mechanisms but cannulation of systemic arteries in healthy subjects can be both complicated and potentially dangerous (Gallen and MacDonald, 1990). Capillary blood samples can be relatively easily obtained but the quantity of blood which can be taken is limited which may restrict the number of assays which can subsequently be performed. According to Williams *et al.* (1992) differences in arterial and venous lactate concentrations have led some to question the use of the venous blood samples. With these factors in mind, heating techniques have been developed which increase blood flow and result in arterialisation of the blood which, for many purposes, can be assumed to equal arterial samples without serious error (McLoughlin *et al.*, 1992). The warm air-box described by Gallen and MacDonald (1990) is particularly useful for subjects performing dynamic exercise such as cycling as the box can surround the handle-bars. However, during intense treadmill exercise, no differences between venous and arterial blood lactate concentrations have been reported (Williams *et al.*, 1992). Therefore, it was not considered necessary to obtain arterialised blood samples during the

experiments described in this thesis which involved high-intensity exercise and venous blood samples, which permitted a large volume of blood to be obtained in relative safety, compared with arterial samples, were obtained.

Prior to those trials requiring blood samples, subjects rested for approximately 15 minutes on a couch before a cannula (Venflon 2, 18 gauge) was inserted under local anaesthesia (2 ml lignocaine (1% w/v)) into an antecubital vein in their forearm. To facilitate repeated sampling, a 3-way stop cock and tube (Connecta®) was connected to the cannula and strapped securely with plaster and surgical tape. The subject was then seated upon the ergometer and after completing a standardised warm-up, a venous blood sample of approximately 7-8 ml was obtained. All further samples, immediately after exercise and during recovery, were taken with the subject in a seated position either on the ergometer or an adjacent chair. After each sample had been taken, the cannula, attached tube and stop-cock were flushed with sterile heparinised saline (sodium chloride(0.9% w/v)). Where a muscle and blood sample were due at the same time, the muscle sample took priority and in Chapter 6 further reference is made to the importance of this aspect.

The samples were dispensed into three tubes, namely a calcium heparinised Eppendorf tube, a 5 ml serum tube and a 5 ml lithium heparinized tube. Full details of the procedures relating to dispensing and handling of the samples are shown diagramatically in Figure 3.1. Blood pH was measured as soon as possible after the sample was taken (within 1-2 minutes) whilst the plasma, which was subsequently analysed for ammonia, was centrifuged and placed in the freezer within 6 minutes of the sample being taken. In addition to the measurement of blood pH, haemoglobin concentrations and haematocrit values were determined on the same day as the sample. Changes in plasma volume were estimated from the changes in haemoglobin and haematocrit (Dill and Costill, 1974). Plasma ammonia concentration was determined within 48 hours of sampling. Lactate, Na^+ and K^+ (both measured only in Chapter 5), were determined at the end of each experiment. Full details of the blood assays can be found in Appendix B. The coefficients of variation for the blood and plasma assays and mixed muscle metabolite assays can be found in tables 3.8(a) and (b).

METABOLITE	CV (%)
Blood Lactate-	
Automated method	2.4
Non-automated Method	3.1
Serum Sodium	1.1
Serum Potassium	3.2
Plasma Ammonia	8.2
Blood pH	0.3

Table 3.8(a). Coefficients of variation for the blood metabolite assays obtained by way of 10 repeated measures on the same sample.

METABOLITE	CV (%)
PCr	2.4
ATP	4.1
Lactate	1.2
Pyruvate	6.0
Cr	3.0

Table 3.8(b). Coefficients of variation (%) for the mixed muscle metabolite assays determined from 10 duplicate homogenates obtained from rat muscle samples.

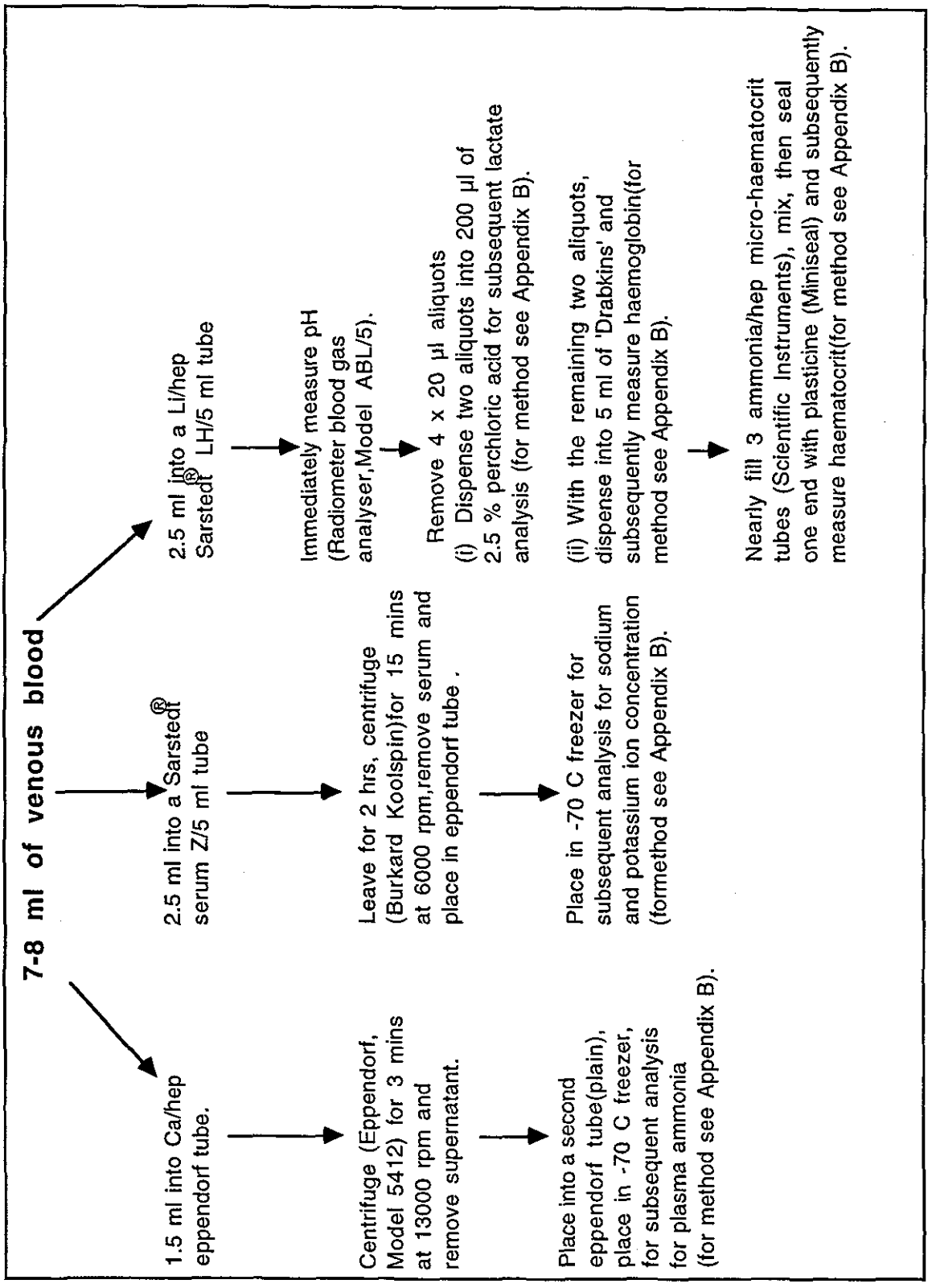

Fig. 3.9. Diagrammatic representation of the procedures for handling and storage of the venous blood samples.

CHAPTER 4

CONSTANT EXTERNAL WORK CYCLE EXERCISE - THE MAGNITUDE OF THE MECHANICAL AND METABOLIC CHANGES OF ALL-OUT AND EVEN-PACED STRATEGIES.

This Chapter contains the results from two studies. On a friction loaded cycle ergometer, one method of affecting pedalling rate is to impose a pre-set constant average rate. As a result, over the duration of exercise power output remains constant which contrasts with an all-out sprint where the initial maximal effort may lead to the generation of very high external power outputs. Both studies compared some of the metabolic changes of a 30 s all-out sprint with those arising from a strategy where a pre-set constant rate was imposed which, if maintained for the full 30 s, would result in the same total external work as an all-out effort. The second study also investigated the magnitude of the mechanical changes arising from 30s exercise. This aspect was assessed by the subjects ability to generate peak power in a sprint performed 2-3s after each 30s bout of exercise. The first study was a pilot study and subject numbers are small. As a result, the major part of this Chapter is devoted to the second of the two studies.

4.1. PILOT STUDY

Introduction

The purpose of this pilot study was to establish whether the external work achieved during a high-speed all-out sprint could be replicated during 30 s by adopting a constant work pacing policy and, if so, whether either strategy resulted in the attenuation of any of the metabolic disturbances arising from intense exercise.

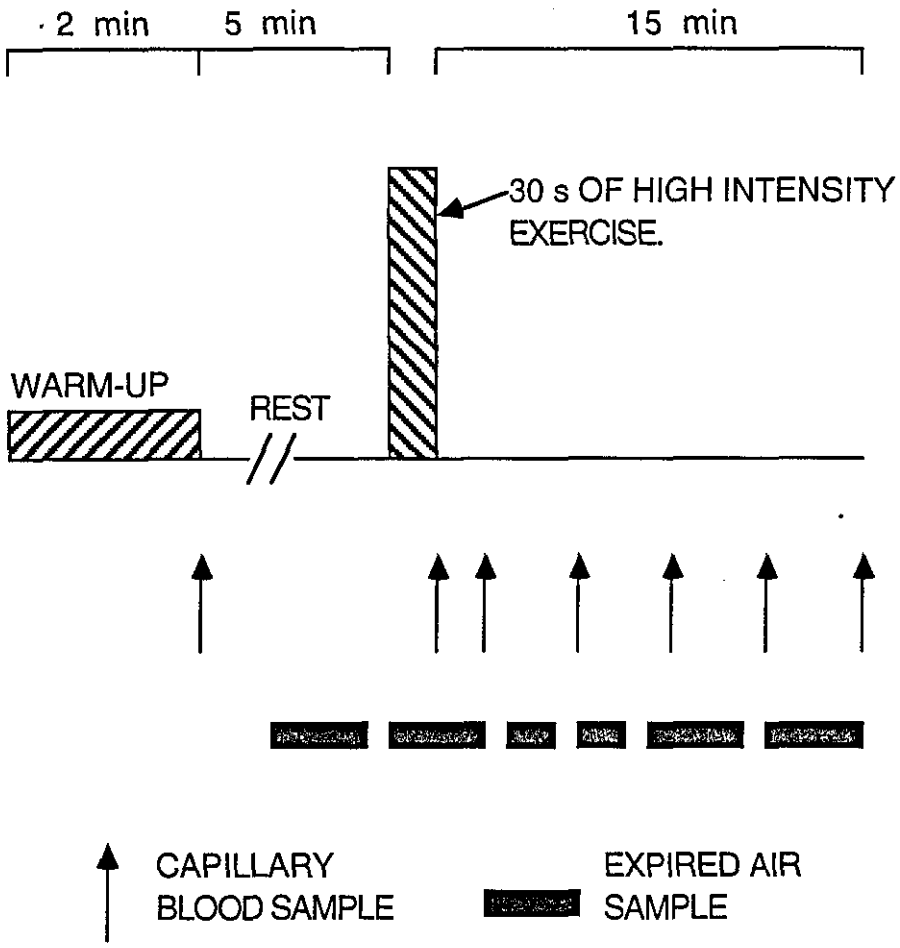

Fig. 4.1. Schematic representation of the protocol employed during the pilot study. After completing a standardised warm-up lasting 2 minutes, the subjects provided a 20 µl capillary blood sample and 3 minutes later an expired air collection of 60 s was taken. At 5 minutes post warm-up, the subjects performed 30 s of high intensity exercise. During the first trial, this was a maximal all-out effort and on two further occasions the subjects attempted to replicate the external work of trial 1 at a pre-set constant average pedalling rate. Further capillary blood samples were taken immediately after exercise and at 3, 6, 9, 12 and 15 minutes into recovery. Expired air was collected during each bout of exercise and at further intervals during recovery.

Methods

Five subjects (4 male; 1 female; age 21±1 year; body mass 72±3 kg) volunteered to take part in the pilot study. After a period of familiarisation, subjects came to the laboratory on three occasions and completed a main trial during each visit. A schematic representation of the experimental protocol is shown in Figure 4.1. The only variation for each trial related to the 30 s bout of high-intensity exercise. On their first visit, the subjects completed a maximal 30 s all-out effort against an applied resistance equal to 75 $N \cdot kN^{-1}$ body weight. During the second and third main trials, the subjects attempted to replicate the external work of trial 1 by adopting a paced strategy against applied resistances of 55 (FAST trial) or 95 (SLOW trial) $N \cdot kN^{-1}$ body weight in a random order. Against both applied resistances the subjects were assigned a pedalling rate where the initial external power output was below the maximal that the subjects could achieve. 20 µl capillary blood samples were analysed for lactate (Maughan, 1982) and glucose (Werner, 1970). Expired air samples taken during each bout of exercise and into recovery enabled the aerobic and anaerobic contributions to energy provision to be calculated by the oxygen-debt method (for full details of the procedures for collecting and analysing the expired air samples see the main study described in this Chapter). It was assumed that the active muscle mass during the sprint was 25 % and that 290 mM of ATP were supplied per litre of oxygen consumed.

Results

The subjects were able to replicate the external work of the all-out trial during the SLOW trial but were unable to do so in the FAST trial because they were unable to sustain the required rate for the full 30 s (Table 4.1). The relative aerobic and anaerobic contribution to energy provision was similar between the three trials (Table 4.1) as were the changes in blood glucose concentrations. However, the blood lactate concentrations during recovery were different between the three trials (Figure 4.2).

Discussion

A number of important findings arose from this small pilot study. The main finding was that a limited number of subjects were able to replicate the external work of a high speed all-out effort by adopting a paced strategy requiring

average pedalling rates of approximately 8.4 rad·s^{-1} (80 rev·min^{-1}) but could not do so at faster rates of 12.6 rad·s^{-1} (120 rev·min^{-1}).

For all three strategies the oxygen debt data suggest that the relative proportion of the aerobic and anaerobic pathways to energy provision was similar. The oxygen debt data suggest that approximately 75 % of the contribution to energy provision came from the anaerobic pathways and 25 % from aerobic sources. These proportions are similar to others reported in the literature in which there is a wide variation of reported values for 30 s of high-intensity exercise. Spreit (1995) suggests the relative anaerobic/aerobic split is 80/20, whilst using the oxygen deficit technique Medbo and Tabata (1989) suggest the aerobic contribution is as high as 40 %. The similarity of the results between all three trials is perhaps a little surprising as it has been frequently suggested that as pedalling rate is increased, so the contribution to external power output of type II muscle fibres is increased. Type I fibres are fatigue resistant and have a high oxidative capacity and if their contribution to power output had been greater during the slow paced trial compared with the faster paced trial and the high-speed all-out effort, then a higher aerobic contribution may have been predicted. Generally, many of the subjects were uncomfortable with the need for lengthy periods of gas collection before, during and particularly after exercise. In view of the similarity of the results, methodological problems and the broad assumptions made with the calculation, the oxygen debt method should not be pursued as a method for establishing whether changes in pedal rate affect the extent of the metabolic changes.

Nevertheless, there was some indication from the limited blood data that there may be differences in the magnitude of the metabolic changes between paced and all-out strategies. In particular, the blood lactate data suggested that similar external work produced by a slow paced strategy may attenuate the metabolic responses compared with a high speed all-out effort. This potential metabolic benefit may enable enhanced performance in a subsequent sprint. Therefore, the pilot study indicated that a more extensive study to compare the metabolic and performance effects of all-out and paced strategies was warranted.

	TRIAL		
	ALL-OUT SPRINT	PACED-SLOW	PACED-FAST
WORK (kJ)	15.8±1.4	15.8±1.2	14.1±1.5*
MEAN PEDAL RATE			
($rev \cdot min^{-1}$)	103±8	80±6*	122±8*
($rad \cdot s^{-1}$)	10.8±0.8	8.4±0.6*	12.8±0.8*
AEROBIC CONTRIBUTION (%)	22.0±1.4	24.8±2.2	21.8±2.0
ANAEROBIC CONTRIBUTION (%)	78.0±1.4	75.2±2.2	78.2±2.0

Table 4.1. Work, mean pedal rate, estimated aerobic and anaerobic contribution to energy provision arising from a 30 s maximal all-out effort against an applied resistance equal to 75 $N \cdot kN^{-1}$ body weight (ALL-OUT SPRINT) and during two further trials where the pedalling rate was set against applied resistances equal to 95 (SLOW) and 55 (FAST) $N \cdot kN^{-1}$ body weight. (n=5; Values are mean ± SEM; * $P<0.05$ compared with ALL-OUT SPRINT).

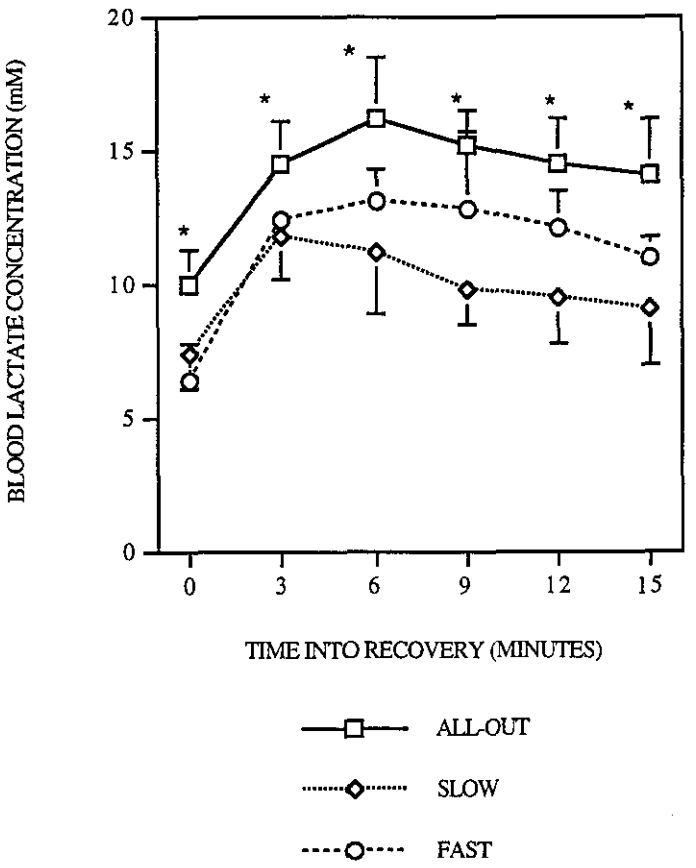

Fig. 4.2. Blood lactate concentrations (mM) during recovery from 30 s of high intensity exercise performed as an all-out sprint or at a fast and slow constant pace. (*P<0.05= interaction (time-trial) differences between all-out and at least one of the paced trials).

4.2. MAIN STUDY

Introduction

The results of the pilot study indicated that during 30 s of high-intensity exercise on a friction loaded cycle ergometer, the extent of the metabolic changes may be influenced by average pedal rate. In that study pedalling rate was affected by adopting two contrasting pacing strategies, all-out and paced. The post-exercise concentrations of lactate were lower in the paced trials compared with all-out, particularly in the slower of the two paced trials. It was suggested that these differences in the magnitude of the metabolic changes might influence the magnitude of any mechanical changes but this was not tested. However, the extent of the mechanical changes to a given bout of exercise can be assessed by measuring a subject's ability to generate peak power in a sprint performed shortly after prior fatiguing exercise.

In a sporting setting, athletes competing in sprint events almost universally adopt an all-out policy where maximal effort is applied from the outset. Similarly, in laboratory tests such as the Wingate protocol (Bar-Or, 1978), subjects are required to sprint maximally throughout. Such a policy involves constantly changing muscle action speeds and the rapid initial acceleration phase leads to the attainment of high limb velocities. This policy would appear to conflict with the optimum strategy based on a knowledge of the power-velocity relationship of muscle (Chapter 2), which suggests that there is an optimal velocity for the production of power, at approximately one third of the maximal velocity of shortening (V_{max}). Any substantial deviation from the optimal velocity, as occurs in a high speed all-out effort, will cause a marked reduction in the muscles ability to generate external power.

Previous research by McCartney *et al.* (1983a) has suggested that instead of a single optimal pedal rate, a range exists where the ability to generate power is maximised and this range, which can be represented as a plateau on the power-velocity relationship, encompasses pedalling rates of 12.6-16.8 rad.s^{-1} (120-160 rev.min^{-1}), for muscle in the unfatigued state. The suggested optimal rate for a 25 s bout is a little slower at 11.6 rad.s^{-1} (111 rev.min^{-1}) (Sargeant *et al.*, 1984), and studies requiring subjects to exercise at a high intensity for 20-30 s on a constant velocity ergometer, typically employ pedalling rates in the region of 11.5-13.1 rad.s^{-1} (110-125 rev.min^{-1}). Fatigue causes a reduction in the maximal velocity of shortening (Edman and Mattiazzi, 1981) which shifts the power-

velocity curve to the left. The optimal region remains at muscle action speeds of approximately $0.3V_{max}$, thus, it also will be shifted to the left and hence slower velocities will be optimal in the fatigued state. Thus, if high-intensity exercise is performed at a constant velocity which is considered optimal for unfatigued muscle, towards the end of exercise the ability to generate power at such a velocity will be greatly reduced (de Haan *et al.*, 1989). Similarly, if muscle action speed is not held constant and the athlete is able to attain very high pedalling rates, these will far exceed the optimal region, resulting in less than optimum power production. Thus, according to the power-velocity relationship a constantly declining pacing policy, from an initial velocity in the optimal region for unfatigued muscle, may allow the highest mean power to be generated. However, it is also possible that a constant velocity, which is slower than optimal for unfatigued muscle but becomes optimal as fatigue ensues, may allow the cyclist to spend more time at, or quite close to, the plateau, than either an all-out or a faster constant pace policy.

Cycling is a particularly good model for conducting experiments relating to the power-velocity relationship as pedalling rate can be considered to be representative of average muscle action speed. Also, cycling enables the athlete to attain the same external velocity despite markedly different rates of limb movements by varying the gear ratio (Hagberg *et al.*, 1981). In a not too dissimilar manner, similar external work can be produced on a friction loaded cycle ergometer by manipulating either, or both, the average pedalling rate and applied resistance.

The purpose of this study was to establish whether the performance of a 30 s all-out sprint could be replicated in two further trials, involving pre-set constant average pedalling rates so that external work was produced evenly during the exercise period. The effect of each strategy on the metabolic responses to exercise was determined, along with the impact upon subsequent sprint performance.

Methods

Subjects: Twenty-four university students (12 male; age 22 ± 2 years; bodymass 78.0 ± 8.7 kg; height 179 ± 14 cm : 12 female; age 20 ± 1 years; bodymass 66.0 ± 8.3 kg; height 165 ± 10 cm. Mean ± SD) volunteered for the study. Approval for the study, which was performed in accordance with the appropriate ethical standards laid down in the 1964 Declaration of Helsinki, was granted by the University Ethical Committee. Each subject completed a health questionnaire and voluntary consent form (specimen documents can be found in Appendix A), received a written and verbal explanation of the purpose of the study and was fully familiarised prior to the main trials. The subject group was sufficiently large enough to permit it to be divided into two groups and individual subjects were assigned to either a metabolic (MET) or performance (PERF) group each group containing 6 males and 6 females. This resulted in a reduction in the number of visits that each subject had to make to the laboratory and also reduced the amount of equipment required to collect blood samples, compared with a single group of 24 subjects.

All subjects completed three main trials which were separated by seven days. On each occasion, the subjects performed a warm-up (30s @103 W, 30s @ 118 W, 30s rest, 30s @ 162 W), followed by 5 min rest and then a 30 s bout of exercise. Each exercise bout attempted to achieve the same external work by adopting a different pacing strategy.

Trial 1 (all-out): Both groups completed a maximal 30 s sprint, from a rolling start of 7.3 rad·s^{-1} (70 rev·min^{-1}), against 75N·kN^{-1}body weight, upon a friction loaded cycle ergometer (Monark 864).

Trials 2/3 (constant pace): Seven and fourteen days later, the subjects attempted to replicate the external work achieved during trial 1 at a constant pre-set pace in 30 s. The resistance was calculated so that if the required pedalling rate was maintained for 30 s, the external work of trial 1 would be replicated. One constant pace trial required the subjects to pedal constantly at 55% of their peak pedalling rate achieved during trial 1 (SLOW trial). The other trial, necessitated the subjects to maintain 90% of the trial 1 peak (FAST trial). The order of trials 2 and 3 was randomly assigned.

All trials: A schematic diagram of the protocol is shown in Fig. 4.3. Venous blood samples were obtained via a cannula placed in an antecubital vein in the

subject's arm from the MET group only, post warm-up and at 0,3,6,9,12 and 15 min post-exercise and subsequently analysed for pH, lactate, ammonia and change in plasma volume. Full details of the sampling, handling and storage procedures of the blood samples can be found in Chapter 3 and the relevant analytical procedures are described in full in Appendix B. All blood samples were taken with the subject seated either in the saddle or on an adjacent chair. At the end of each 30s bout, the PERF group only, pedalled with no resistance for 3s and then performed a sprint of 6 s duration against 75N·kN^{-1}body weight.

Measurements: The ergometer was interfaced to a microcomputer (BBC) so that high frequency logging of the flywheel angular velocity could be obtained. The instantaneous power values were corrected so as to allow for changes in kinetic energy of the flywheel (Lakomy, 1986); power and pedalling rate values were calculated over 1 s intervals. Peak power output and peak pedal rate were specified as the highest values attained over a single second whilst mean pedalling rate and mean power output were the average achieved during 30 s. The total external work achieved during 30 s was also recorded. During the subsequent 6 s sprint peak power output and peak pedalling rate were recorded. Previous research has indicated that both are attained within a 6 s sprint (for example Lakomy, 1988).

Expired air samples were collected from both groups during each 30 s bout using the Douglas Bag method. From each bag, a small measured sample was analysed for the percentage oxygen content (Sybron Taylor Servomex Analyser OA 570) and a second for carbon dioxide (Lira Infrared Analyser Model 303). The volume remaining in each bag was then measured (Harvard Gas Meter). The total volume which had passed through the three analysers was corrected to 273 K and 760 mmHg. Using the Haldane transformation, the volume of inspired air was calculated. Oxygen uptake ($\dot{V}O_2$), carbon dioxide production ($\dot{V}CO_2$) respiratory exchange ratio (R) and ventilatory equivalent ($\dot{V}_E/\dot{V}O_2$) were also calculated. At the end of the third trial, the subjects were asked to rate, on a scale of 0-100, the relative effort which they perceived during each trial, where 100 was the value assigned to the most strenuous.

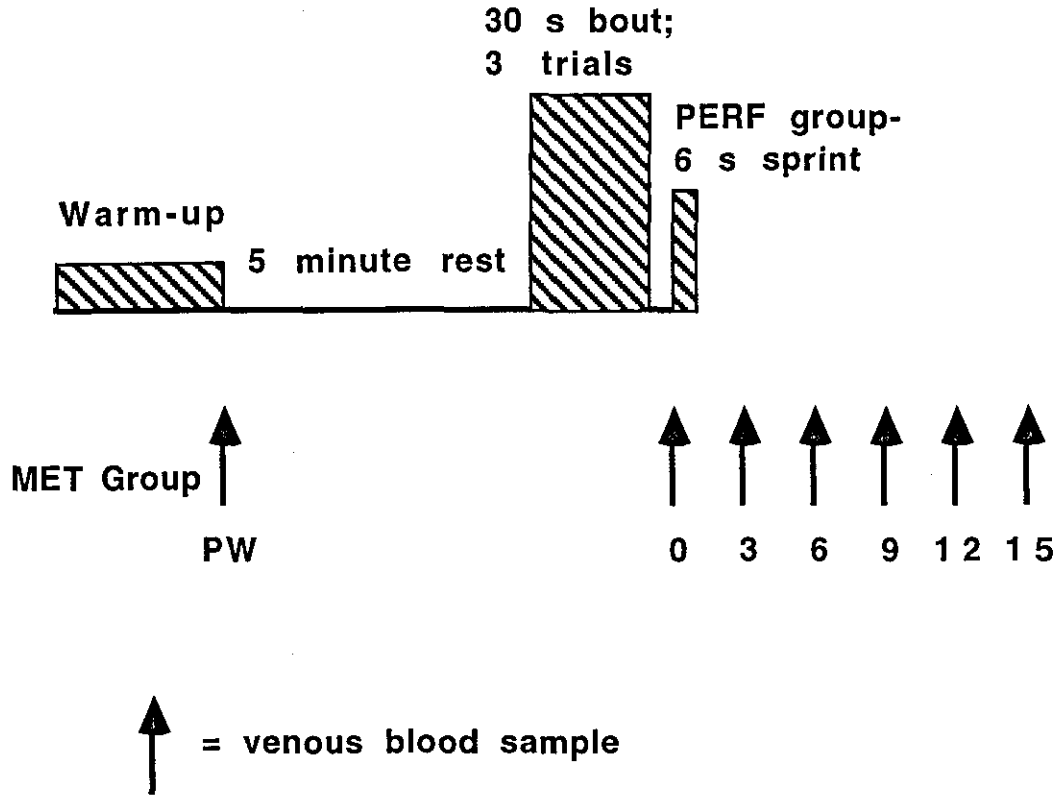

Fig. 4.3. Schematic representation of protocol. Subjects were assigned to either a metabolic (MET) group or a performance (PERF) group. All subjects performed a 2 minute warm-up followed by 5 minutes rest, then a 30 s bout of exercise. At the end of the 30 s bout of exercise, subjects in the PERF group were asked to perform a 6 s sprint. Venous blood samples were taken from subjects in the MET group at post-warm-up (PW), and t = 0, 3, 6, 9, 12 and 15 minutes after the 30 s bout of exercise.

Statistical analysis: Results are presented as mean±SE unless otherwise stated. Two-way analyses of variance for repeated measures were used where appropriate to compare trial 1 with, separately, trials 2 and 3 (factors being time, trial and time/trial interaction). If any significant differences were found with the ANOVA then a post-hoc Tukey test was performed to ascertain exactly where they were to be found. Where single measures only were taken per trial, a students t-test was used. Significance was deemed at the $P<0.05$ level.

Results

Trial 1 (all-out): The MET and PERF groups achieved similar peak power output, peak pedal rate and total external work during the 30 s sprint. Peak power output occurred during the 2nd second in 20 of the 24 subjects, the remaining subjects achieving maximal power in either the 1st or 3rd second. Mean pedal rate was 11.1±0.3rad·s^{-1} (106±2 rev·min^{-1}). The male subjects performed better than the females on all performance variables ($P<0.01$).

Trials 2 & 3 (constant pace): In the faster of the paced trials, to replicate the external work of the all-out trial the subjects were required to pedal at a rate of 13±0.4 rad·s^{-1} (124±4 rev·min^{-1}); range 10.2-15.7 rad·s^{-1} (97-150 rev·min^{-1}). However, they were unable to maintain the set rate for the entire 30 s resulting in a mean pedal rate of 12.5±0.3 rad·s^{-1} (119±3 rev·min^{-1}) and significantly (approximately 7%) less work ($P<0.05$). In contrast, during the slower paced trial at constant average pedalling rates of 8.0±0.2 rad·s^{-1} (76±2 rev·min^{-1}); range 6.3-9.4 rad·s^{-1} (60-90 rev·min^{-1}) they were able to sustain the required rate for the entire 30s and so replicated the external work (Fig. 4.4). Figure 4.5 shows the time course of external work done for a single subject by summing power output over 5 s periods. It demonstrates that during the last 10 s of the fast paced trial the required rate could not be maintained despite strong verbal encouragement from the experimenters and the maximal effort of the subjects. As the external work was similar in only the all-out and slower paced trials, metabolic data and performance indicators for the subsequent 6 s sprint are given for these two trials.

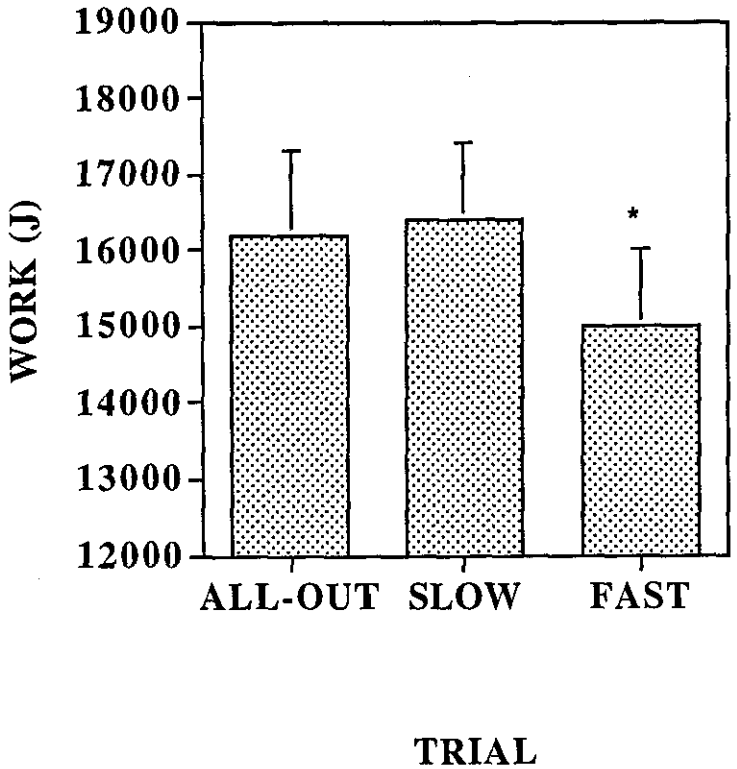

Fig. 4.4. External work production for both groups of subjects (n=24, 12M, 12F, mean±SE) during 30 s of maximal cycle exercise (ALL-OUT) and during two paced trials adopting a pedal rate of 55% (SLOW) and 90% (FAST) of the peak all-out rate. (*P<0.05 compared with all-out and slow trials)

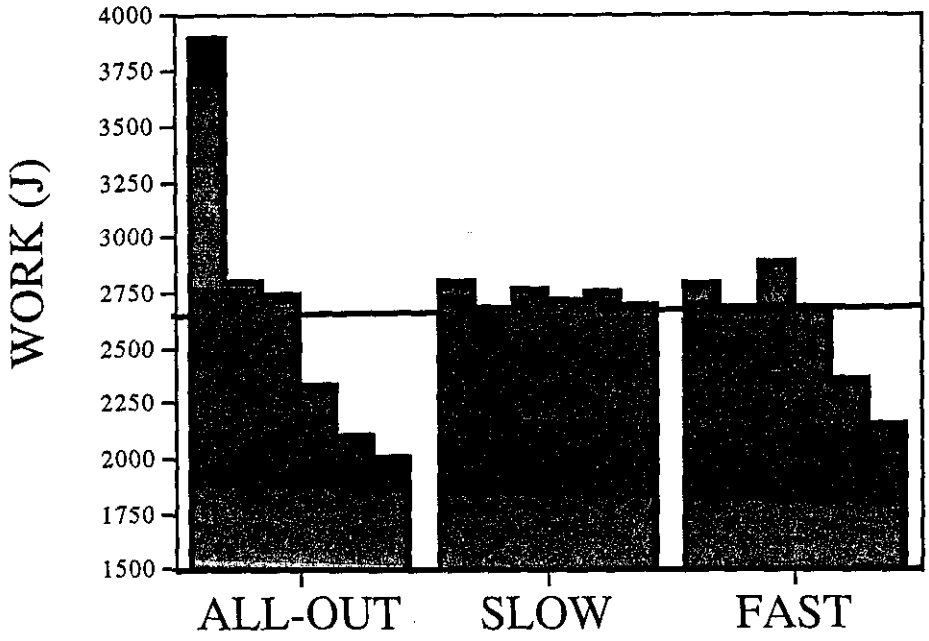

Fig. 4.5. Time course of work production for a single subject during each trial. Power output values are summed over a 5 s period so that there are 6 blocks for each 30 s bout of exercise. The horizontal line represents the work required to replicate the work of the all-out trial.

Metabolic data

Blood measurements(MET group) : Immediately following exercise, blood pH (Fig. 4.6) was reduced ($P<0.01$) to a similar extent in both the all-out and slow paced trials compared with post-warm-up (PW) values (all-out, 7.38±0.01 (PW) vs. 7.21±0.02 (t=0) : paced, 7.37±0.01 (PW) vs. 7.21±0.02(t=0)). However, at 3,6,9,12 and 15 min into recovery values were significantly higher following the paced trial than the all-out trial ($P<0.05$).

Blood lactate (Fig. 4.7) was elevated from post warm up values ($P<0.01$) as a result of exercise. During both trials, the highest values were recorded 6 min into recovery when it was higher in the all-out trial than in the paced trial, (all-out, 13.2±0.6 mM vs. paced, 10.4±0.7 mM, $P<0.01$), thereafter showing a gradual decline.

Although plasma ammonia concentration (Fig. 4.8) was elevated after exercise above post warm up values ($P<0.01$), there was no difference between trials at t=0 (all-out, 56.4±7.4μM vs. paced, 56.1±8.9μM, $P>0.05$). During recovery, plasma ammonia concentration continued to rise but at different rates and after 3 minutes of recovery was significantly higher in the all-out trial (all-out, 127.8±16.2 vs. paced, 103.7±14.2μM, $P<0.05$). The highest ammonia concentrations were observed at 3 min and 6 min post-exercise respectively in the paced and all-out trials.

Estimations of changes in plasma volume indicated that in comparison with post warm-up values, plasma volume was reduced at t=0, (all-out, -7.2±2.3% vs. paced, -8.1±2.5%, $P>0.05$), and increased at 15 minutes into recovery (all-out, +5.9±2.0 vs. paced, +5.7±3.5%, $P>0.05$).

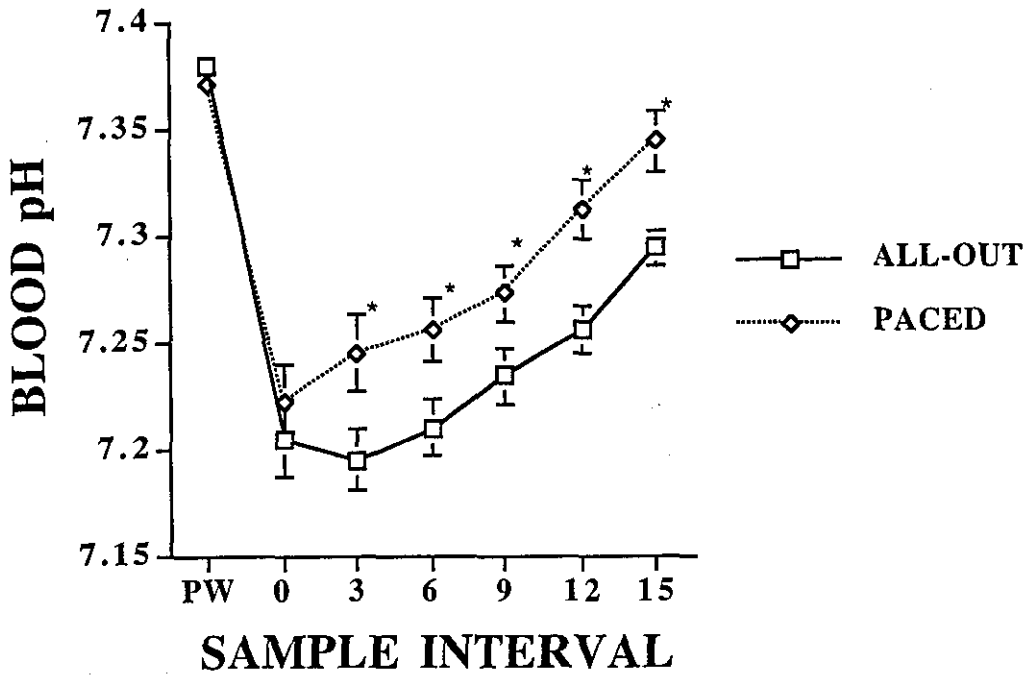

Fig. 4.6. Blood pH for the MET subjects (n=12, 6M, 6F, mean±SE) post warm-up (PW), immediately on cessation (sample interval=0) of 30 s of constant external work exercise performed under an all-out condition and at 55% (PACED) of the peak pedalling rate of the all-out trial, and during 15 min of recovery (main effect condition, $P<0.05$; main effect time, $P<0.01$; condition-time interaction, $P<0.05$; *$P<0.05$ post-hoc test, ALL-OUT vs. PACED trial).

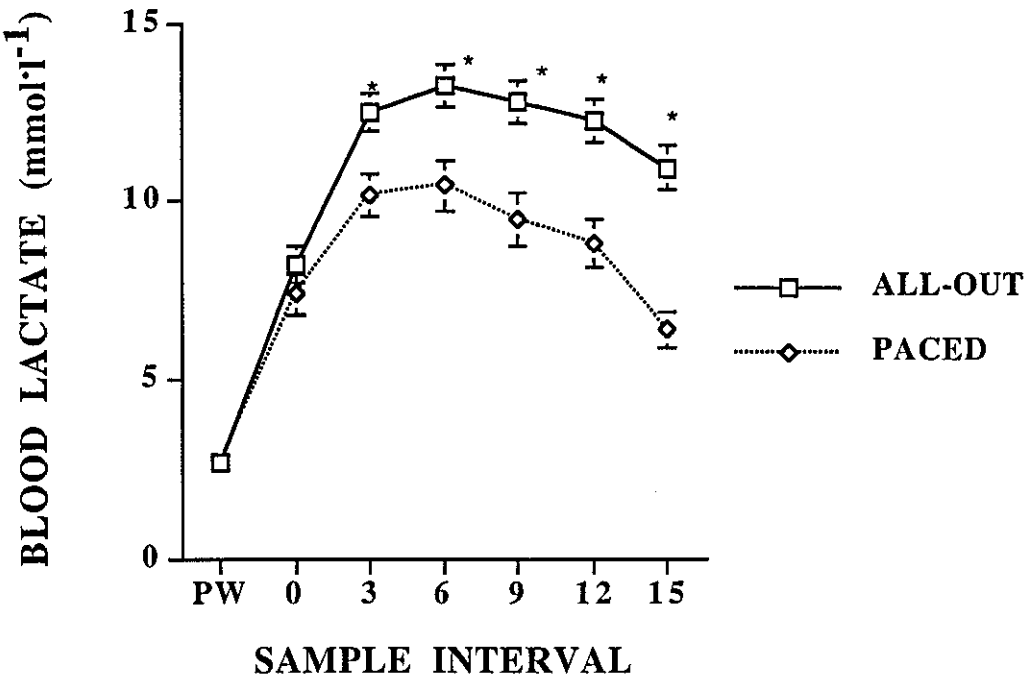

Fig. 4.7. Blood lactate concentrations for the MET subjects (n=12, 6M, 6F, mean±SE) post warm-up (PW), immediately on cessation (sample interval=0) of 30 s of constant external work exercise performed under an all-out condition and at 55% (PACED) of the peak pedalling rate of the all-out trial, and during 15 min of recovery (main effect condition, $P<0.05$; main effect time, $P<0.01$; condition-time interaction, $P<0.05$; *$P<0.05$ post-hoc test, ALL-OUT vs. PACED trial).

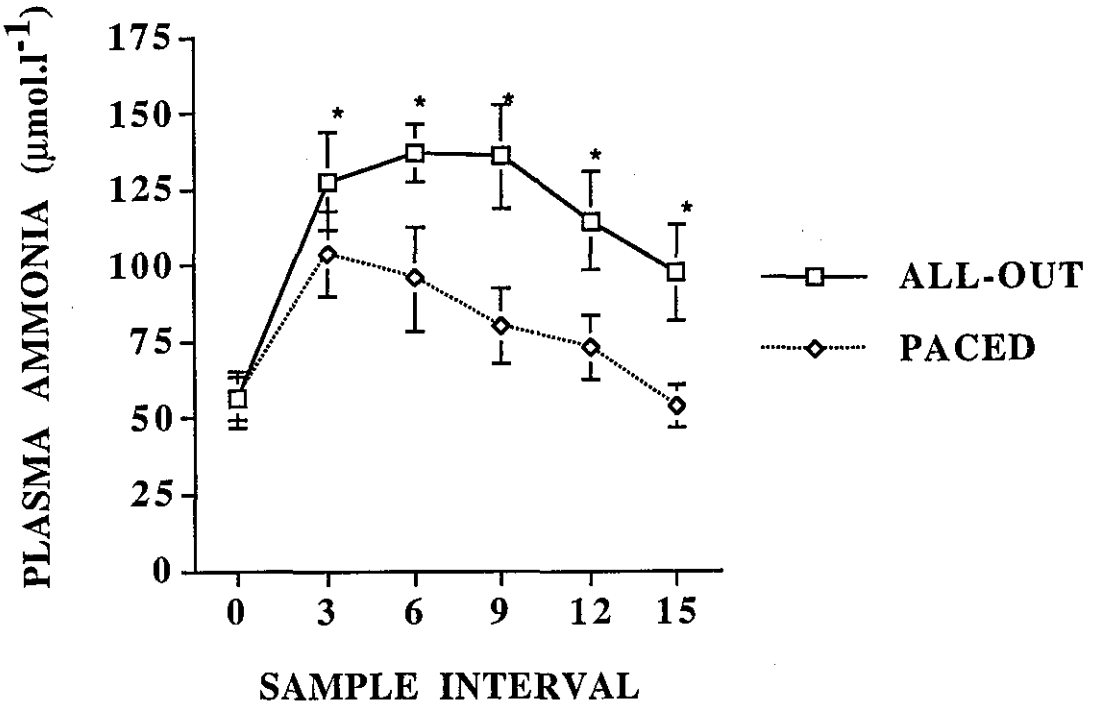

Fig. 4.8. Plasma ammonia concentrations for the MET subjects (n=12, 6M, 6F, mean±SE) post warm-up (PW), immediately on cessation (sample interval=0) of 30 s of constant external work exercise performed under an all-out condition and at 55% (PACED) of the peak pedalling rate of the all-out trial, and during 15 min of recovery (main effect condition, $P<0.05$; main effect time, $P<0.01$; condition-time interaction, $P<0.05$; *$P<0.05$ post-hoc test, ALL-OUT vs. PACED trial).

Other metabolic data (both MET and PERF groups)

Expired Air Data: Oxygen uptake was similar during the all-out and paced trial (all-out, 2.2±0.1 vs. paced, 2.1±0.1 $l \cdot min^{-1}$, P>0.05). Similarly, there were no differences in the respiratory exchange ratio between trials (all-out, 1.15±0.04 vs. paced, 1.11±0.03, P>0.05). However, $\dot{V}_E$ (all-out, 99.2±5.9 vs. paced, 76.0±4.4 $l \cdot min^{-1}$, P<0.01) and $\dot{V}_E/\dot{V}O_2$ (all-out, 44.1±2.2 vs. paced, 37.1±1.6, P<0.01) values were greater for the all-out trial.

Heart rate: Maximal heart rate was similar between trials (all-out, 178±2beats·min^{-1} vs. paced, 174±2beats·min^{-1}, P>0.05).

Perceived exertion: The all-out bout was rated the most difficult and slow paced trial easiest (all-out rating, 99±1 vs. paced rating, 73±3, P<0.01). Twenty-two of the twenty-four subjects tested accorded with this view but the two other subjects both rated the slow paced trial the most difficult.

Performance data (PERF group only)

During the second sprint of 6 s duration, completed approximately 3 s after each trial, the subjects were unable to perform as well after either the all-out or paced trial compared with the first 6 s of the 30 s all-out sprint (P<0.01). Performance in the second sprint was less impaired following the paced trial than the all-out reflected by a greater peak power output and pedal rate in the subsequent sprint (peak power output; all-out, 580±24 W vs. paced, 715±25 W : peak pedal rate; all-out, 9.3±0.2 rad·s^{-1}(89±2 rev·min^{-1}) vs paced, 11.1±0.2 rad·s^{-1}(106±2 rev·min^{-1}), P<0.05) (Fig. 4.9). For 9 of the 12 subjects peak power output occurred after 2 seconds and in the other 3 subjects after seconds 1 or 3 and was not significantly different from the time at which maximal power occurred in the previous 30 s exercise bout.

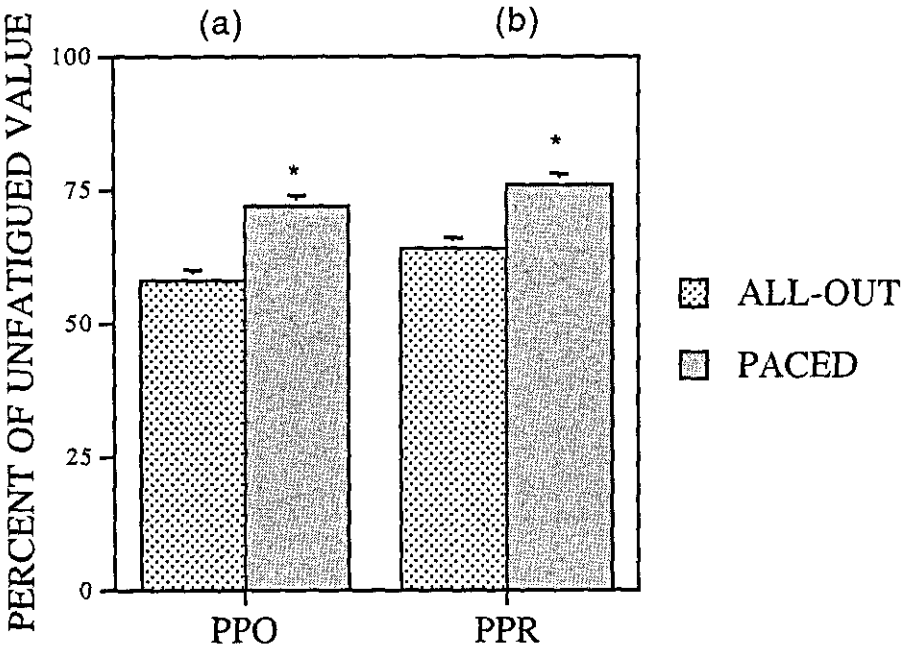

Fig. 4.9. (a) Peak power output (PPO) and (b) peak pedalling rate (PPR) achieved by the PERF group (n=12, 6M, 6F, mean±SE) in a second 6 s sprint expressed as a percentage of that achieved in the first 6 s of the all-out trial. (* P<0.05 significantly different between all-out and paced trials).

Discussion

The purpose of the present study was to describe some of the metabolic and mechanical responses to constant external work high-intensity cycle exercise of 30s duration when the pedal rate was freely chosen or imposed. It was possible to produce the same external work as that achieved during an all-out effort against an applied resistance equal to 75 $N \cdot kN^{-1}$ of the subject's bodyweight by imposing constant average pedal rates of 6.3-9.4 $rad \cdot s^{-1}$ (60-90 $rev \cdot min^{-1}$). However, at higher rates of 10.2-15.7 $rad \cdot s^{-1}$ (97-150 $rev \cdot min^{-1}$) it was not possible to maintain the required rate for the entire 30 s resulting in a significant reduction in external work. Constant external work exercise performed under paced conditions resulted in a lower level of fatigue, reflected by less of a reduction in peak power output in a subsequent sprint, and less disturbance to blood pH, lactate and ammonia post exercise, compared with an all-out strategy. This suggests that the magnitude of both the mechanical and metabolic changes were reduced during the low rate paced strategy compared with all-out.

The finding that the external work of an all-out sprint could be replicated by altering the applied resistance and adopting a constant average pedal rate conflicts with previous research which has suggested that when exercise is performed on a friction loaded cycle ergometer that an all-out policy produces the best performance for exercise lasting up to 2 minutes (Katch *et al.*, 1976; 1977). However, in that study only one constant pace strategy was used which required the subjects to pedal at 10.2 $rad \cdot s^{-1}$ (97 $rev \cdot min^{-1}$) and the authors expressed the opinion that other rates may give different results. The present study indicates that their speculation was correct. Subjects can achieve the same performance as a high speed all-out effort at rates of 6.3-9.4 $rad \cdot s^{-1}$ (60-90 $rev \cdot min^{-1}$) but are unable to do so at higher constant average rates. This is in close agreement with the findings of a study which found that an upper limit of 10.5 $rad \cdot s^{-1}$ (100 $rev \cdot min^{-1}$) exists for maximum mean power output for 30 s cycling exercise (Zoladz and Rademaker, 1994). It is important to emphasise that the results do not suggest that either policy adopted in this study is optimal for maximal power production during 30 s. The power-velocity relationship of muscle suggests a policy where pedalling rate steadily declines from an initial rate which is optimal for unfatigued muscle will produce maximal external work. This policy accords more closely with an all-out compared with an even paced strategy. However, the applied resistance in the

all-out sprint in this study was such that very high peak pedalling rates were obtained which were far from the optimal region for external power production.

The male subjects outperformed their female counterparts in respect of all performance variables in the all-out trial namely peak power output, peak pedalling rate and total work achieved. The enhanced performance in respect of peak power and work done can be attributed to the greater muscle mass of the males (Miller *et al.*, 1993). A number of factors also contributed to the higher peak pedal rates exhibited by the males and hence the higher rates imposed in the paced trials. Firstly, the cranks of the ergometer were of a fixed length and placed the shorter females at somewhat of a disadvantage. Secondly, the applied mass was calculated as a proportion of body mass and not muscle mass. Nevertheless the two groups, MET and PERF, were comprised of equal numbers of each sex and performance was matched between the two groups. In addition, there was some degree of overlap between the female and male subjects. Also, in some instances, females were able to outperform their male counterparts of similar age and body mass. Therefore the author considers that for the subjects who participated in this study, the pooling of male and female data is considered valid.

The paced trial resulted in a lower level of fatigue, reflected by less of a reduction in peak power output in a subsequent sprint, compared with the all-out trial. This suggests that the extent of the mechanical changes during 30 s of high-intensity exercise are reduced when similar external work is produced with a paced strategy compared with an all-out sprint. Previous research has also found that greater fatigue arises at high as opposed to low pedalling rates (Beelen and Sargeant, 1991: 1993) but in those studies prior exercise was at an intensity of less than maximal oxygen uptake for 6 minutes at different pedalling rates. Thus, it is possible that the extent of fatigue is influenced by the number of muscle actions which take place. However, by imposing a pedalling rate upon a friction loaded ergometer the subjects in the current study were initially working submaximally albeit at an external power output in excess of that required to elicit maximal oxygen uptake. A strict interpretation of the 'size principle' (Henneman *et al.*, 1965) would suggest that during the paced exercise some fibres at the top of the hierarchy would not be recruited resulting in a sparing of some type II fibres. However, the literature suggests that even at an intensity below maximal oxygen uptake all fibres are recruited (Greig *et al.*, 1986) and that force is modulated by adjusting the firing frequencies (rate

coding) of the active fibres (Sargeant, 1994). Thus, it is probable that the same motor unit pool was recruited throughout the all-out and paced exercise but that in the paced exercise some fibres, especially those towards the top of the hierarchy, did not have to fire at frequencies needed to elicit their maximal tension. The fibres towards the top of the hierarchy were thus working submaximally. The lower blood concentrations of lactate and ammonia together with the more rapid restoration of blood pH in the MET subjects following the paced trial compared with all-out indicates that there may have been a lower degree of activation of some fibres but in the absence of muscle metabolite data this is only speculation.

In conclusion it was possible to achieve the same external work as that produced during a high speed all-out effort by adopting a paced strategy. The paced strategy gave rise to a lower level of fatigue and less disturbance to selected blood metabolites compared with all-out. This attenuation of the metabolic responses in the paced trial was probably due to a partial sparing of muscle fibres towards the top of the hierarchy compared with all-out. The differential ability to generate peak power in the second sprint may also have been the result of the difference in the magnitude of the metabolic responses or alternatively, may have arisen because of a different number of preceding muscle actions during the paced and all-out trials.

CHAPTER 5

EFFECT OF THE NUMBER OF PRECEDING MUSCLE ACTIONS UPON THE MAGNITUDE OF THE MECHANICAL CHANGES.

The main study described in the previous Chapter suggested that the ability to generate peak power in a sprint may be influenced by the pedalling rate at which prior fatiguing exercise is performed. Greater peak power output was possible following paced exercise at a slow average pedal rate compared with a high-speed all-out effort. However, the extent to which this difference was due to changes in the total number of muscle actions, or the partial sparing of some fibres at the top of the hierarchy during the paced exercise, was not resolved. The purpose of the present Chapter is to establish which, if either, of these factors is the major one determining the short term recovery of power output following high-intensity exercise.

Introduction

The ability of muscle to generate power in short term maximal exercise will be influenced by many factors including the extent to which fatigue arises as a result of prior exercise (Sargeant, 1994). In cycle ergometry, it was established as early as 1928 that it was pedalling rate rather than the applied load which was the major factor influencing the ability of muscle to generate external power (Dickinson, 1928). Subsequently it has been demonstrated that a greater fatigue index (FI) during a single sprint arises at high compared with low pedal rates (McCartney *et al.*, 1983a). An alternative to the FI as a means of assessing the extent of fatigue is the ability to generate peak power in a sprint performed after 2-3 s of recovery from fatiguing exercise. For example, Beelen and Sargeant (1993) demonstrated that 6 minutes of cycle exercise at an external power output below that required to elicit maximal oxygen uptake performed at 12.6 $rad \cdot s^{-1}$, had a greater detrimental effect on peak power output in a subsequent sprint, compared with fatiguing exercise at the same external power output performed at 6.3 $rad \cdot s^{-1}$. Similarly, the previous Chapter indicated that peak power output in a 6 s sprint performed approximately 3 s after high-intensity exercise was greater following a strategy where the same external work was produced at a moderate average constant pedal rate compared with a high speed all-out effort.

Thus previous studies, together with the preceding Chapter, have found that a greater level of fatigue, reflected by a greater reduction in power output in a subsequent sprint, occurs when prior exercise is performed at high as opposed to low pedalling rates. Whether these differences are due to differences in the number of muscle actions *per se* or differences in the metabolic responses is unclear. It has been speculated that the greater fatigue with increasing pedalling rate is due primarily to a greater or selective fatigue of type II fibres (Lodder *et al.*, 1991; Beelen and Sargeant, 1991; Sargeant, 1994). However, whilst studies which have investigated metabolic responses at different pedalling rates have shown the concentration of possible fatigue causing metabolites in the blood to be greater post-exercise at high pedalling rates (Jones *et al.*, 1985; Chapter 4), analysis of mixed muscle metabolites has shown no difference between high, 14.7 $rad \cdot s^{-1}$ (140 $rev \cdot min^{-1}$) and low, 6.3 $rad \cdot s^{-1}$ (60 $rev \cdot min^{-1}$)rates (Jones *et al.*, 1985). At the single fibre level, research comparing muscle responses at different muscle action speeds is scarce. Beelen *et al.*, (1993) compared glycogen concentrations when two 6 minute bouts of cycle exercise at 90 % $\dot{V}O_2$ max, with 10 minutes recovery in-between, were performed at 6.3 and 12.6 $rad \cdot s^{-1}$. They found a greater depletion in the type IIb fibres at the higher rate compared with the lower rate, but this trend was also found in the other fibre types, suggesting that greater fatigue occurs in all fibres, not just the type IIb, with increasing pedalling rate.

The purpose of the present study was to consider whether the magnitude of the mechanical changes arising during high-intensity exercise was primarily affected by the number of preceding muscle actions or the pattern of external work production. Fatigue was induced by 30 s of maximal intensity exercise on a cycle ergometer where pedalling rate was manipulated by altering the applied resistance and/or the initial external power output. The extent of the mechanical changes was assessed by measuring the very short-term recovery of power output.

Methods

Subjects: Seven physically very active females volunteered to take part in the study (Age 21±1 years: Body mass 63.4±7.0 kg ; values ±SD). Subjects were informed and familiarised prior to the main trials as outlined in Chapter 3.

Equipment and Measurements: All trials took place on a modified friction-loaded cycle ergometer (Monark 864). Full details of the equipment and procedures employed to standardise measurements between trials are fully outlined in Chapter 3. In addition to peak power output (PPO), total work, peak pedalling rate (PPR) and mean pedal rate (MPR), a measure of the number of muscle actions in each 30 s was recorded. The number of muscle actions can be regarded as a function of the pedalling rate in rev·min^{-1} (each pedal revolution comprised 2 actions but exercise only lasted 30 s). All measurements are presented as mean±SE unless stated.

Protocol: Upon each visit to the laboratory, separated by at least 48 hours, the subjects were required to complete a 2 min warm-up, followed by 5 min rest, then 30 s of high intensity exercise from a rolling start of 7.3 rad·s^{-1} (70 rev·min^{-1}) and a subsequent sprint of 6 s duration. The only variation between trials concerned the 30 s block of exercise where pedalling rate was adjusted. For clarity the trials can be considered as being performed in three groups with each group providing one "TEST" value relating to the external work production in each 30 s (Fig. 5.1).

First, the subjects performed a group of high-speed all-out trials where they attempted to pedal as fast as possible against an applied resistance equal to 75 N·kN^{-1} body weight until the external work of two trials was within 1 %. The mean of these two trials was regarded as the maximal reproducible all-out work and recorded as "test 1". At the end of the study, the group of trials which were used to obtain the TEST 1 value were repeated to assess the extent of training over the course of the study.

The second group of trials required the subjects to maintain a constant average pedal rate equal to 55 % of their peak all-out rate. The first trial in this group of paced trials was against an applied resistance which enabled similar external work to TEST 1 to be produced, if the required rate was maintained for the full 30 s. During subsequent trials, the applied resistance was increased so that greater external work compared with the previous trial would be achieved if

the rate was maintained. The process continued until a maximum was obtained and the external work of this trial was recorded as "test 2".

Third, the subjects pedalled as fast as possible, against the applied resistance which gave the maximum external work in the paced trials (test 2 resistance). The external work was recorded as "test 3". This represented the external work of a slow speed all-out effort in contrast to the TEST 1 which represented the external work of a high-speed all-out effort.

This protocol resulted in two tests, where the subjects exerted a maximal all-out effort for the whole 30 s but pedalling rate was markedly different because of differences in the applied resistance, and a third paced trial where pedalling rate was imposed. Any similarities or differences in the ability to generate peak power in the subsequent 6 s sprint could indicate whether differences in the total number of muscle actions, or the pacing strategy adopted, are major determinants of the short-term recovery of power.

Effect of each strategy on subsequent peak power output: In order to assess the effect which each 30 s block of exercise had upon subsequent peak power output in every trial the subjects pedalled unresisted for 2-3 s at 7.3 $rad \cdot s^{-1}$ (70 $rev \cdot min^{-1}$) and then performed a 6 s sprint against an applied resistance of 75 $N \cdot kN^{-1}$ body weight.

Statistical Analysis: Single factor analysis of variance tests (ANOVA) with repeated measures were performed to examine the difference between mean values attained in each of the three tests. Significance (*) was accepted at the $P<0.01$ level and where the F-statistic showed a significant difference, a post-hoc test, was performed in order ascertain precisely where any differences occurred.

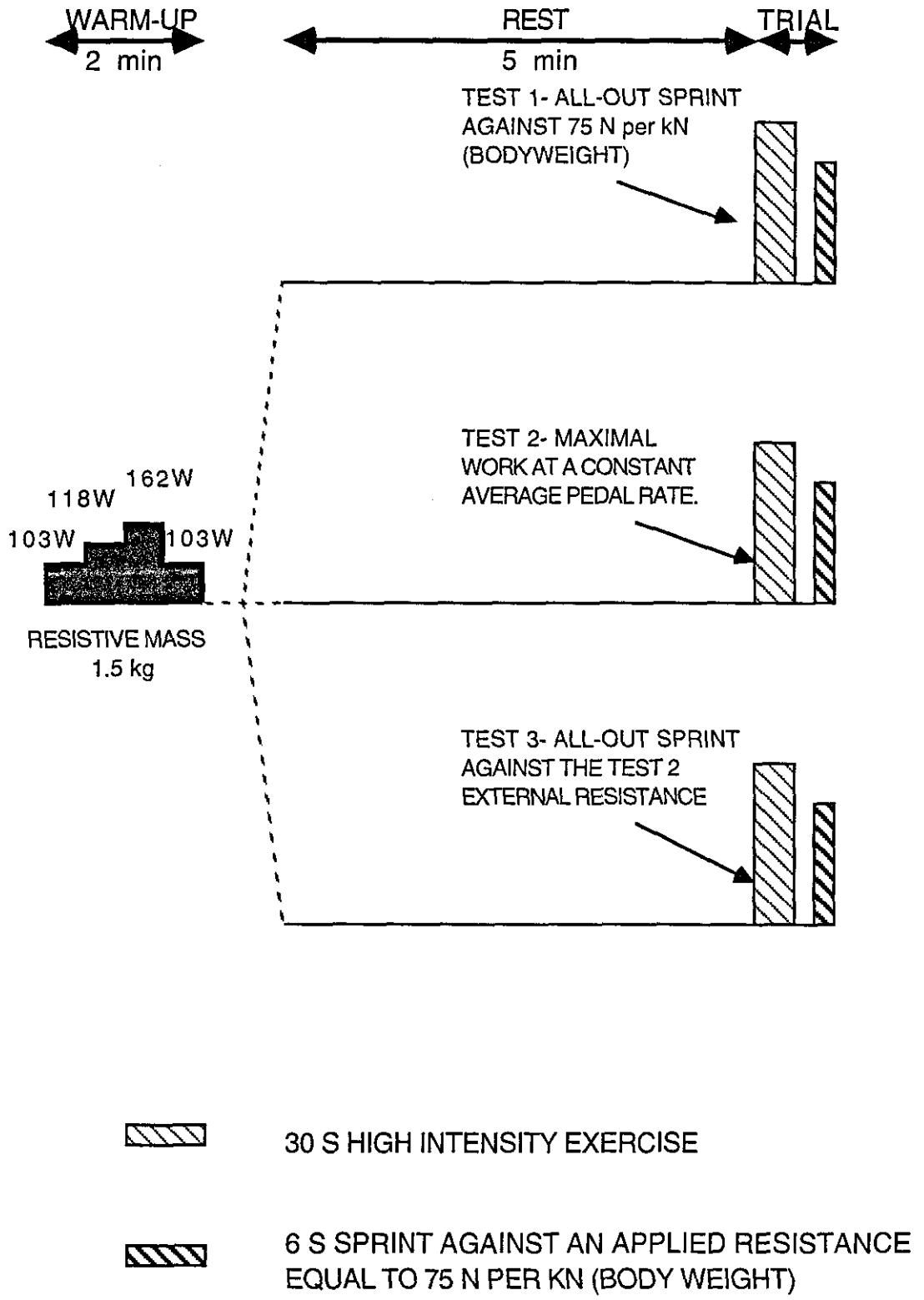

Fig. 5.1. Schematic representation of protocol.

Results

The results for the first 30 s period of exercise are shown in Table 5.1. During the batch of paced trials which were performed to obtain the test 2 value, the subjects were required to pedal at a constant average rate of 7.2±0.4 rad·s^{-1} (69±4 rev·min^{-1}). The subjects were able to maintain this rate for the entire 30 s against a resistance of 109±4 N·kN^{-1} body weight which enabled them to replicate the work of test 1. However, against a greater resistance the rate could not be sustained for a sufficient time to enable them to increase their total work ($P>0.05$, work done test 1 vs. test 2,). However, they were able to exceed their test 1 work by approximately 9 % when applying maximal effort against this resistance in test 3 ($P<0.01$, test 1 vs. test 3). There was no significant difference in the external work performed between tests 2 and 3 and tests 2 and1. Mean pedal rate and consequently the number of muscle actions was highest in test 1 ($P<0.01$, test 1 vs. tests 2 and 3). A repeat of test 1 at the end of the study revealed that the subjects had acquired no training effect.

The ability to generate peak power output in the second 6 s sprint is shown in Figure 5.2. The extent of fatigue arising from all three tests was sufficient to reduce peak power in the second sprint ($P<0.01$) compared with that achieved when sprinting against the same resistance in the unfatigued state. However, peak power was lower after the prior fatiguing exercise was produced under the test 1 condition compared with tests 2 and 3 ($P<0.01$, peak power in a 6 s sprint after test 1 vs. tests 2 and 3; $P>0.01$, test 2 vs. test 3).

Discussion

A 30 s all-out sprint against an applied resistance of 75 N·kN^{-1} body weight resulted in a greater average pedalling rate and the production of less external work compared with an all-out sprint against a heavier applied resistance of 109 N·kN^{-1} body weight. Nevertheless, despite the production of more external work, a lower level of fatigue, reflected by less of a reduction in peak power output in a subsequent 6 s sprint, arose following the sprint against the heavier resistance compared with the lighter. A similar lower level of fatigue also arose following a paced trial against the heavier resistance.

	TEST 1- HIGH SPEED ALL-OUT SPRINT	TEST 2- PACED EXERCISE	TEST 3- LOW SPEED ALL-OUT SPRINT
WORK(kJ)	13.3±0.8[b]	13.8±1.0	14.5±0.8[b]
PPO(W)	702±65	537±39a	722±70
PPR(rad·s^{-1})	13.2±0.6[a]	7.8±0.4[a]	9.9±0.7[a]
MPR(rad·s^{-1})	10.0±0.4[a]	7.2±0.4	7.8±0.3
NUMBER OF MUSCLE ACTIONS IN 30 s	96±4[a]	69±4	75±3

Table 5.1. Work, peak power output, peak pedal rate and number of muscle actions arising during 30 s of high intensity exercise produced under three contrasting conditions. (n=7; mean ± SE) ([a] significantly different, P<0.01, compared with other two tests; [b] significantly different P<0.01 between tests 1 and 3).

PPO, peak power output; PPR. peak pedalling rate; MPR, mean pedalling rate.

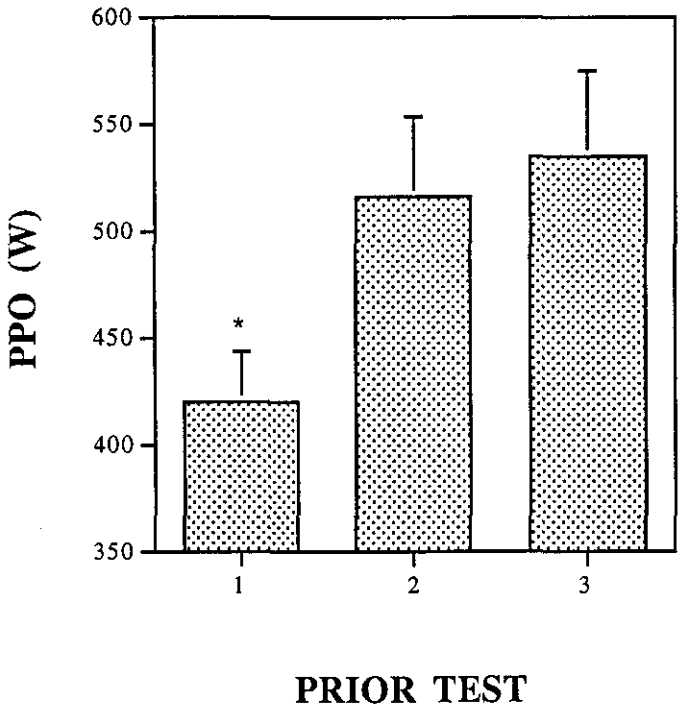

Fig. 5.2. Peak power output in a 6 s sprint performed 2-3 s after 30 s of fatiguing exercise. Prior test 1 involved a maximal all-out sprint against a resistance equal to 75N·kN^{-1}BW; test 2 was the maximal external work that could be achieved during a constant pace trial against 109N·kN^{-1}BW whilst test 3 was an all-out sprint against the greater applied resistance. Values are mean values and the vertical bars represent the SE of the mean (n=7) (*P<0.01 different peak power output following test 1 compared with test 3).

Approximately 9 % more external work was produced in 30 s in test 3, where the subjects pedalled as fast as possible against the heavy resistance, compared with test 1, where they pedalled all-out against the light resistance. This suggests that the greatest period was spent at, or very close to, the optimal region for external power production as suggested by the power-velocity relationship of muscle in test 3. For unfatigued mixed muscle, the optimal velocity for external power production lies upon a 'plateau' on the power-velocity relationship at pedalling rates of approximately 11.5 $rad \cdot s^{-1}$ (110$rev \cdot min^{-1}$) (Sargeant *et al.*, 1981), or a little higher, at about 12.5-16.8 $rad.s^{-1}$ (120-160 $rev.min^{-1}$) (McCartney *et al.*, 1983a). As exercise proceeds these rates become too fast as fatigue causes a reduction in the optimal velocity for power production (de Haan *et al.*, 1989). Thus, the optimal strategy for maximal external work production would be one where the subjects were pedalling within the optimal region at the start of exercise but pedalling rate gradually declined as the bout proceeds. This is the situation that occurs during an all-out effort where pedalling rate is freely chosen. Thus, of the two all-out trials in this study, the one against the heavy resistance was closer to the optimal than against the lighter resistance.

Greater fatigue, reflected by a lower peak power output in a subsequent sprint, arose following the high speed all-out effort against the light resistance compared with both the paced and all-out bouts against the heavier resistance. This is agreement with the findings of Kim and Chung (1995) who investigated the effects of resistance and frequency during repetitive dynamic lifting. They found that the trunk muscles fatigued faster during a light load-high frequency task compared with a heavy load-low frequency task. Three factors influence the rate of ATP utilisation and thus possibly the level of fatigue arising as a result of exercise. Firstly, the relative contribution of different fibre types to power output at different pedalling rates; secondly, the speed at which muscle actions take place; thirdly, the number of muscle actions which occur.

The contribution to power output of different fibre types at contrasting pedalling rates is one factor which may have contributed to the greater level of fatigue after test 1. This factor has been widely investigated and previous research has suggested that greater fatigue arises at high as opposed to low muscle action speeds because of a selective fatigue of type II muscle fibres (Lodder *et al.*, 1991; Beelen and Sargeant, 1991; 1993). The main findings of some previous studies are brought together in a model presented by Sargeant

(1994). In that model, muscle is regarded as being composed of only two fibre types fast and slow, with their relative ability to contribute to power output dependent upon the muscle action speed chosen. The slow fibres whose maximal velocity of shortening appears to equate to a pedalling rate of approximately 17.3 $rad \cdot s^{-1}$ (165 $rev \cdot min^{-1}$) are able to make a significant contribution to power output at rates close to their optimal velocity for power production of approximately 6.3 $rad \cdot s^{-1}$ (60 $rev \cdot min^{-1}$). However their relative contribution to power output becomes substantially less at faster pedalling rates. Conversely the relative contribution of the fast fibres to power output increases with increasing pedalling rate. The fast fibres have a greater maximal rate of ATP utilisation than their slow counterparts. Thus at fast muscle action speeds, such as that encountered during the high speed all-out effort against the light resistance, the increased contribution to power output of type II fibres may have resulted in an increase in ATP utilisation. However studies which have examined muscle metabolism at different pedal rates have not found a greater or selective fatigue of type II fibres at high pedal rates (Jones *et al.*, 1985; Beelen *et al.*, 1993). Thus, the muscle metabolite data suggest that this factor is unlikely to be a major factor influencing the differential recovery of power output found in the present study.

A further factor influencing the ATP cost of exercise is the speed at which muscle actions take place. Both peak and mean pedalling rates were higher during the all-out sprint against the light resistance compared with both trials against the heavier resistance. According to Harridge and White (1993), the major factor governing the shape of the torque-velocity curve is the time allowed for torque generation. With increasing muscle action speed there will be less time during each pedal stroke at which the lower limb, will be in position to generate effective force on the pedals. According to Bagshaw (1993) the ATP cost of a fast concentric action is less than for a slow action and thus the ATP cost per muscle action would be expected to become reduced with an increase in average pedalling rate.

A third factor to be considered in explaining the greater fatigue following test 1 is the number of muscle actions which have taken place as it will also influence ATP utilisation. A greater number of muscle actions occurred during test 1 compared with tests 2 and 3, which were similar even though pedalling rate was imposed in test 2 but freely chosen in test 3. Peak power output in the subsequent 6 s sprint was reduced to a greater extent following test 1 but the reduction was less following tests 2 and 3. These results suggest that it is the

number of muscle actions performed in prior fatiguing exercise and not the total or pattern of external work production which influence the recovery of peak power output.

During electrically induced muscle actions it has been shown that fatigue develops faster, and the ATP cost is higher, where the same amount of work is produced with many short lasting muscle actions compared with fewer longer lasting actions. The primary reason propounded for the higher ATP cost is that for a 1 s action, approximately 37% of the ATP cost is related to activation and relaxation (Bergstrom and Hultman, 1988). This is in close agreement with those studies which have assessed the individual energy requirements of the Ca^{2+} and Na^{+}-K^{+} ATP ases. Of the total ATP consumption Ruegg (1987) suggests that calcium handling requires 30 % and Clausen (1997) indicates that the Na^{+}-K^{+} pump's energy requirement is approximately 9 %.

In summary, this study considered whether the magnitude of the mechanical changes was primarily affected by the pattern of external work production or the number of preceding muscle actions. Despite producing more external work, there was a greater initial recovery of peak power following 30s of maximal all-out exercise against a heavy compared with a light resistance. Where similar external work to both all-out trials was produced under paced conditions, the recovery of peak power was similar to the all-out sprint against the heavy resistance but dissimilar to the all-out sprint against the light resistance. These results suggest that the short-term recovery of power output is unaffected by total external work production. However, the number of muscle actions in the paced trial and the all-out trial against the heavy resistance were similar suggesting that the magnitude of the mechanical changes is influenced by the number of preceding muscle actions.

CHAPTER 6

THE MAGNITUDE OF THE BIOCHEMICAL CHANGES DURING MAXIMAL HIGH-INTENSITY EXERCISE AT DIFFERENT AVERAGE PEDALLING RATES.

The previous two Chapters both indicated that the magnitude of the mechanical changes during high-intensity exercise, measured as the ability to generate peak power in a sprint performed 2-3s after such exercise, is dependent upon pedalling rate. The blood metabolite assays performed in Chapter 3 indicated that a mechanism for these mechanical differences might be found from the analysis of selected muscle metabolites, particularly those involved in the anaerobic provision of ATP. Specifically, the literature indicates that the ability to generate peak power is highly correlated with the availability of one particular metabolite, namely phosphocreatine. The literature also suggests, but no study has shown, that the magnitude of the metabolic changes, may be velocity dependent. The previous Chapter indicated that the most important factor determining the short-term recovery of power output was the number of preceding muscle actions and that a large proportion of the ATP cost will be related to processes connected with excitation and relaxation. For each action potential, there will be a movement of ions associated with excitation and relaxation. Clearly, the extent of these shifts may also be dependent upon pedalling rate as the number of muscle actions is a function of pedalling rate. The purpose of the present Chapter is, therefore, to determine if either differences in the magnitude of the changes in the concentration of muscle metabolites (with special attention focused upon muscle phosphocreatine), or ionic movements, can explain the differential ability to generate power 2-3 s after high-intensity exercise performed at different average pedalling rates.

Introduction

A number of factors, both central and peripheral, have been cited as possible causes of fatigue (Bigland-Ritchie, 1984). However, for dynamic exercise of a short duration at, or near, maximal intensity it seems that central factors can be disregarded, as the maximal voluntary force generated is similar to that attained when muscles are electrically stimulated using surface electrodes (Beelen *et al.*, 1995). The peripheral factors that are suggested as being responsible for causing fatigue are sarcolemma excitability, excitation-

contraction coupling, contractile mechanisms, metabolic energy supply and metabolite accumulation. It is now generally accepted that no single peripheral factor can be regarded as the sole contributor to the decline in force or power during high-intensity exercise. Recent research has concentrated on two biochemical factors, namely phosphocreatine availability and potassium accumulation.

A good correlation between the recovery of peak power and the extent of resynthesis of phosphocreatine after prior exercise has led some authors to suggest that phosphocreatine availability may be important in the fatigue process (Bogdanis *et al.*, 1995). Perhaps an even stronger argument implicating phosphocreatine availability is that supplementation of the diet with creatine, thereby increasing the resting phosphocreatine/creatine pool, resulted in enhanced performance in the latter bouts of repeated exercise (Greenhaff *et al.*, 1993). The recovery period between exercise bouts in these investigations has been one minute or longer and whether muscle phosphocreatine is crucial for the very short term (<5 s) recovery of power has not been investigated. Indeed, very few studies have required subjects to perform repeated bouts of exercise with a very short period of recovery (<5 s). These short term recovery studies have entailed a short maximal sprint approximately 2-3 s after exercise (Beelen and Sargeant, 1993; Previous Chapters). They have all demonstrated that the ability to generate power is significantly lower following exercise at high as opposed to moderate average pedalling rates. No study has compared muscle metabolites during maximal intensity exercise at differing muscle action speeds on friction loaded ergometers in an attempt to find a mechanism for these differences in the ability to generate power. However, it has been shown at high and low pedalling rates on an isokinetic ergometer that both the mixed muscle (Jones *et al.*, 1985) and single fibre (Beelen *et al.*, 1993) responses are similar. Clearly, if similar muscle metabolite responses are also found on a friction-loaded ergometer, this strongly suggests that the mechanism, or mechanisms, underpinning the differential recovery of power output are to be found elsewhere.

An action potential results in a net accumulation of K^+ in the extracellular space thereby increasing the extracellular K^+ concentration ($[K^+]extr$). This can result in a decrease in membrane potential and muscle force (Sjogaard, 1996). During high-intensity exercise there is a marked increase in $[K^+]extr$ and this may explain the rapid and large loss of force seen in this type of exercise (Bangsbo *et al.*, 1996). Further evidence suggesting that K^+ accumulation may

be an important factor is that the exercise induced efflux rate is determined by the number of action potentials (Hallen *et al.*, 1994; Vollestad *et al.*, 1994). With increasing pedalling rate there will be an increase in the number of muscle actions which may result in a greater K^+ efflux causing a larger shift of ions across muscle membranes. Thus ionic changes may provide an alternative explanation as to why greater fatigue arises following prior exercise with a high number of preceding muscle actions. Although measurement of $[K^+]$extr is not practical during or after dynamic exercise plasma potassium concentration is generally regarded as a good indicator of $[K^+]$extr (Vollestad *et al.*, 1996).

The purpose of the present study was to further examine the influence of manipulating average pedalling rate on a friction-loaded cycle ergometer upon the short-term recovery of power output in humans. The study also assessed the magnitude of the biochemical changes in an attempt to establish whether ionic imbalance or changes in muscle metabolite concentrations may explain how power output recovers in the first 3 s following prior exercise at different pedalling rates.

Methods

Subjects: Eight females (age 22±3 years; height 168±9 cm; bodymass 63±9 kg; mean ±SD) volunteered to take part.

Measurements and Equipment: Exercise took place on a friction loaded cycle ergometer (Monark 864) which was interfaced to a microcomputer (BBC). Full details of the equipment and the procedures for data collection and standardisation between trials can be found in Chapter 3.

Protocol: After familiarisation, each subject came to the laboratory on four separate occasions, completing one trial on each visit. In two trials, biochemical responses to a single 30s maximal all-out cycle sprint against different applied resistances were examined, whilst two further trials examined the effect which such exercise had upon the ability to generate peak power during a second sprint of short duration. All trials were preceded by a standard 2 min warm up (30 s @ 88W; 30 s @ 103W; 30 s 160W; 30 s @ 88W) and 5 minutes rest.

(a)Biochemical Trials:
On separate occasions, each subject performed a maximal 30 s sprint from a rolling start of 7.3 $rad \cdot s^{-1}$ (70 $rev \cdot min^{-1}$) against an applied resistance equal to

either 75 or 110 $N \cdot kN^{-1}$ body weight. For the first trial, the applied resistance was selected at random and 7 days later the subjects sprinted against the other resistance so that the number of muscle actions performed would be manipulated. Blood and muscle samples were obtained at selected times (Fig. 6.1).

(b)Performance Trials:
The subjects participated in two further trials also separated by 7 days. These trials, which involved no biochemical measurements, required the subjects to perform a 30 s sprint against either 75 or 110 $N \cdot kN^{-1}$ body weight as in the metabolic trials, followed by a second maximal sprint of 6 s against the lesser resistance. The period of recovery between the two sprints was 2-3 s (Fig. 6.2).

Muscle and Blood Sampling Procedures.
Muscle Samples: Two muscle samples, resting and post-exercise, were obtained during each of the two biochemical trials, resulting in each subject providing a total of four samples. During the first trial, samples were obtained from a leg chosen at random; the samples obtained during the second trial being taken from the opposite leg. Approximately 30 minutes before each trial two small incisions were made under local anaesthesia (1 % lignocaine) through the skin and fascia of the vastus lateralis muscle whilst the subject rested upon a couch. A resting muscle biopsy was obtained using the needle biopsy technique (Bergstrom, 1962) with suction applied to maximise the size of sample attained (Evans *et al.*, 1982). The post-exercise sample was obtained immediately after exercise through the second incision, whilst the subject was still seated upon the ergometer. All samples were immediately immersed in liquid nitrogen. The time lag between the end of exercise and immersion in liquid nitrogen of the post-exercise sample was recorded. Samples were stored in plastic screw-top eppendorf tubes immersed in liquid nitrogen until they were freeze dried (Edwards Modulyo 4K freeze dryer).

Analytical Methods: The freeze dried samples were stored at -70°C and at a later date approximately one third of each sample was dissected free of connective tissue and blood and homogenised using an agate pestle and mortar. The powdered muscle samples were carefully weighed using an electrical balance (Perkin Elmer AD-4 Autobalance). Metabolites were extracted from the muscle powder with 0.5 M perchloric acid/1 mM EDTA·Na_2 and neutralised with 2.1M potassium hydrogencarbonate. Phosphocreatine (PCr), adenosine 5-triphosphate (ATP), creatine (Cr), pyruvate (Pyr) and lactate (La)

were analysed enzymatically (Lowry and Passonneau, 1973; Harris *et al.*, 1974; Bogdanis, 1994). Muscle metabolite concentrations (except La) were adjusted to the mean total PCr and Cr content of the resting and post-exercise samples of each trial to correct for any remaining fat, blood or connective tissue. Muscle metabolite concentrations are expressed as mmol·kg dry muscle^{-1}.

Calculations: Anaerobic ATP turnover was calculated from the following equation (Katz *et al.*, 1986). No correction was made for aerobically produced ATP or for La efflux during exercise:-

$$\text{Anaerobic ATP turnover} = 2(-\Delta ATP) - \Delta PCr + 1.5\Delta La + 1.5\Delta Pyr$$

Blood Sampling: Venous blood samples were obtained from a cannula placed in an antecubital vein in the subject's arm. Each sample was dispensed into three separate tubes and subsequently analysed for pH, plasma ammonia, lactate (using the automated method), plasma volume change from haematocrit and haemoglobin measures and the ions K^+ and Na^+. Full details of the handling and storing procedures can be found in Chapter 3. Full details of the analytical methods can be found in Appendix B.

Statistical Analysis: The majority of the data were analysed using a two way analysis of variance. For the muscle metabolites the factors were applied resistance (75 or 110 N·kN^{-1} body weight) and time (rest and post-exercise); for the blood samples applied resistance and time (PW, 0, 2, 5, 10, and 15 min samples); for performance variables applied resistance and sprint (sprint 1 and 2). Mean pedal rate and ATP turnover rate of each trial were compared using a t-test for correlated data.

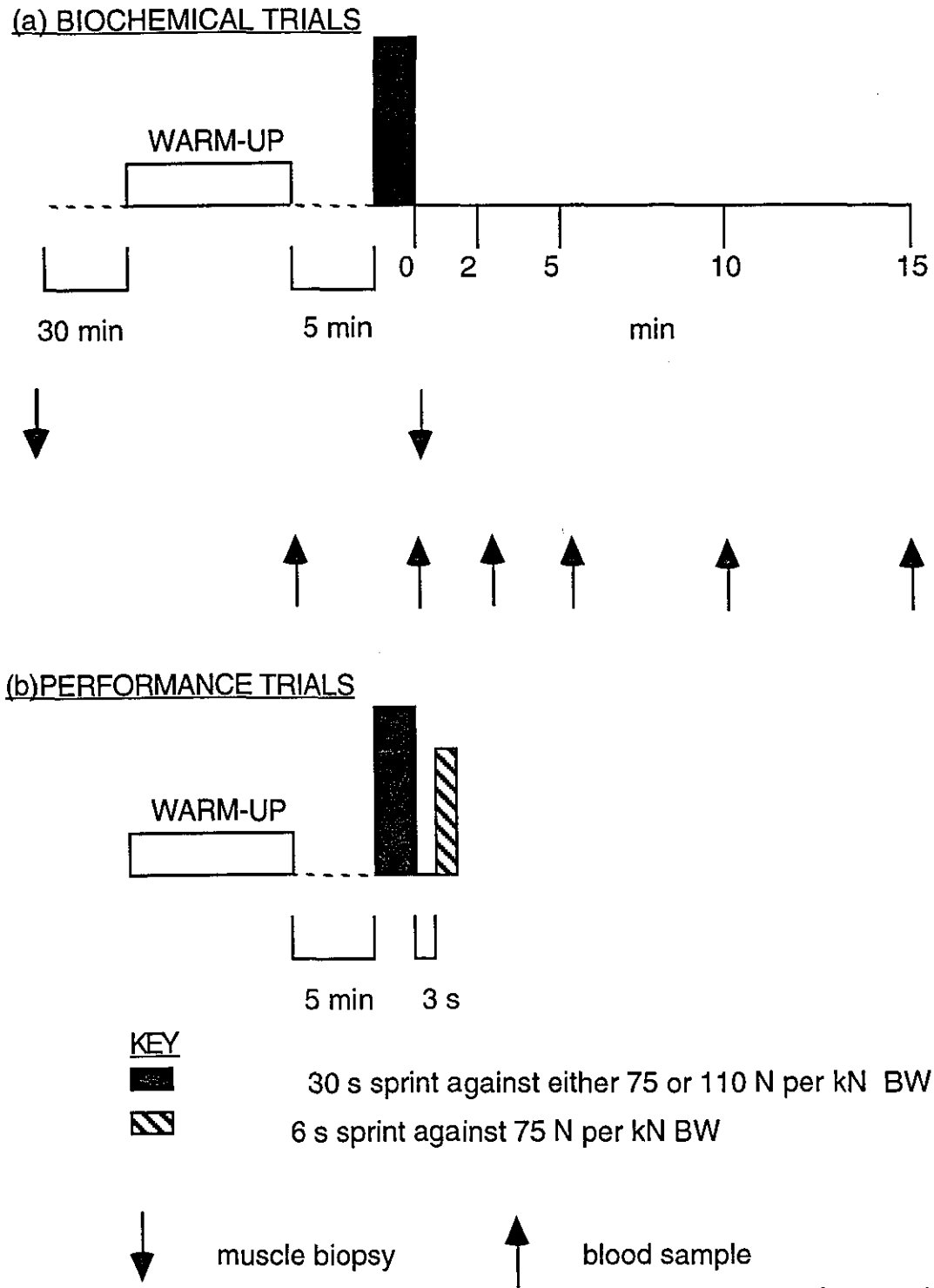

Fig. 6.1. Schematic representation of protocol. (a) 2 biochemical trials involved a single 30 s sprint against either an applied resistance equal to 75 or 110 $N \cdot kN^{-1}$ body weight, with blood and muscle samples taken at selected time intervals. (b) 2 performance trials involved a 30 s sprint against an applied resistance equal to 75 or 110 $N \cdot kN^{-1}$ body weight followed almost immediately by a further sprint of 5-6 s duration against the lesser resistance, no blood or muscle samples being taken.

Results

Biochemical Trials: The muscle sample was immersed in liquid nitrogen 5.8±0.2 s after exercise and was obtained before the first blood sample (designated as being the blood sample at time=0) which was not taken until 31±1 s after each 30 s sprint. The resting and post-exercise muscle metabolite concentrations are shown in Table 6.1. For both applied resistances the post-exercise concentration of each muscle metabolite was significantly different from the resting value (main effect, time $P<0.01$ for all metabolites). The magnitude of the changes were similar for both resistances (interaction, time and resistance, $P>0.05$). The anaerobic ATP turnover was calculated to be 218 mmol·kg drymuscle^{-1} when sprinting against the lighter and 231 mmol·kg drymuscle^{-1} against the heavier load ($P>0.05$). Blood metabolite responses for each trial are shown in Figure 6.2. The highest recorded plasma ammonia concentrations, which occurred at 5 minutes post-exercise in both trials were similar (70±11 μM against the lighter resistance and 65±5 μM against the heavier; $P>0.05$). There were no difference in plasma potassium concentrations at all blood sample intervals between the two applied resistances nor in the pattern of recovery (interaction applied resistance/time $P>0.05$; Fig. 6.2c).

Performance Trials: In the first 30 s sprint, work and peak power were similar to that attained in the metabolic trials indicating that the subjects were well practised and their performance had been unaffected by the procedures associated with muscle and blood sampling (Table 6.2). Mean pedal rate, and hence the number of muscle actions, was greater ($P<0.01$) when sprinting against 75 compared with 110 N·kN^{-1} body weight. Peak power in the second sprint was reduced compared with that achieved in the first few seconds of the first sprint ($P<0.01$ in both trials) but to a lesser extent following prior exercise against the heavier resistance (61 % of sprint 1 PPO) than following the lighter (54 % of sprint 1 PPO) ($P<0.05$).

Power in the final second of sprint 1 was similar for the two applied resistances (313±17 W against 75 N·kN^{-1} BW vs. 332±28 W against 110 N·kN^{-1} BW; $P>0.05$) but within 2-3 s had recovered rapidly (end power of sprint 1 vs. peak power of sprint 2; $P<0.05$ for both applied resistances). This recovery tended to be higher when sprint 1 was against the heavy resistance compared with the light but was not significant ($P=0.056$).

	APPLIED RESISTANCE (N per kN BODY WEIGHT)			
	75		110	
	REST	POST-EXERCISE	REST	POST-EXERCISE
PCr	79.6±1.8	25.0±3.6*	78.5±3.2	20.3±4.4*
Cr	34.9±5.9	89.4±2.5*	30.6±4.5	88.8±1.5*
Lac	6.3±8.2	110.4±1.6*	5.7±8.8	113.1±1.5*
ATP	21.3±2.1	19.4±2.1*	24.0±2.1	19.9±0.5*
Pyr	0.8±0.5	3.2±0.1*	0.4±0.4	2.9±0.1*

Table 6.1. Resting and post-exercise muscle metabolite concentrations (mmol·kg dry muscle^{-1}), prior to (REST) and after (POST-EXERCISE) the performance of a single 30 s sprint against an applied resistance equal to either 75 or 110 N·kN^{-1} body weight (n=8: main effect, rest vs. post exercise, *P<0.01 significant difference between the rest and post-exercise samples for each resistance; main effect, trial, P>0.05; interaction, P>0.05).

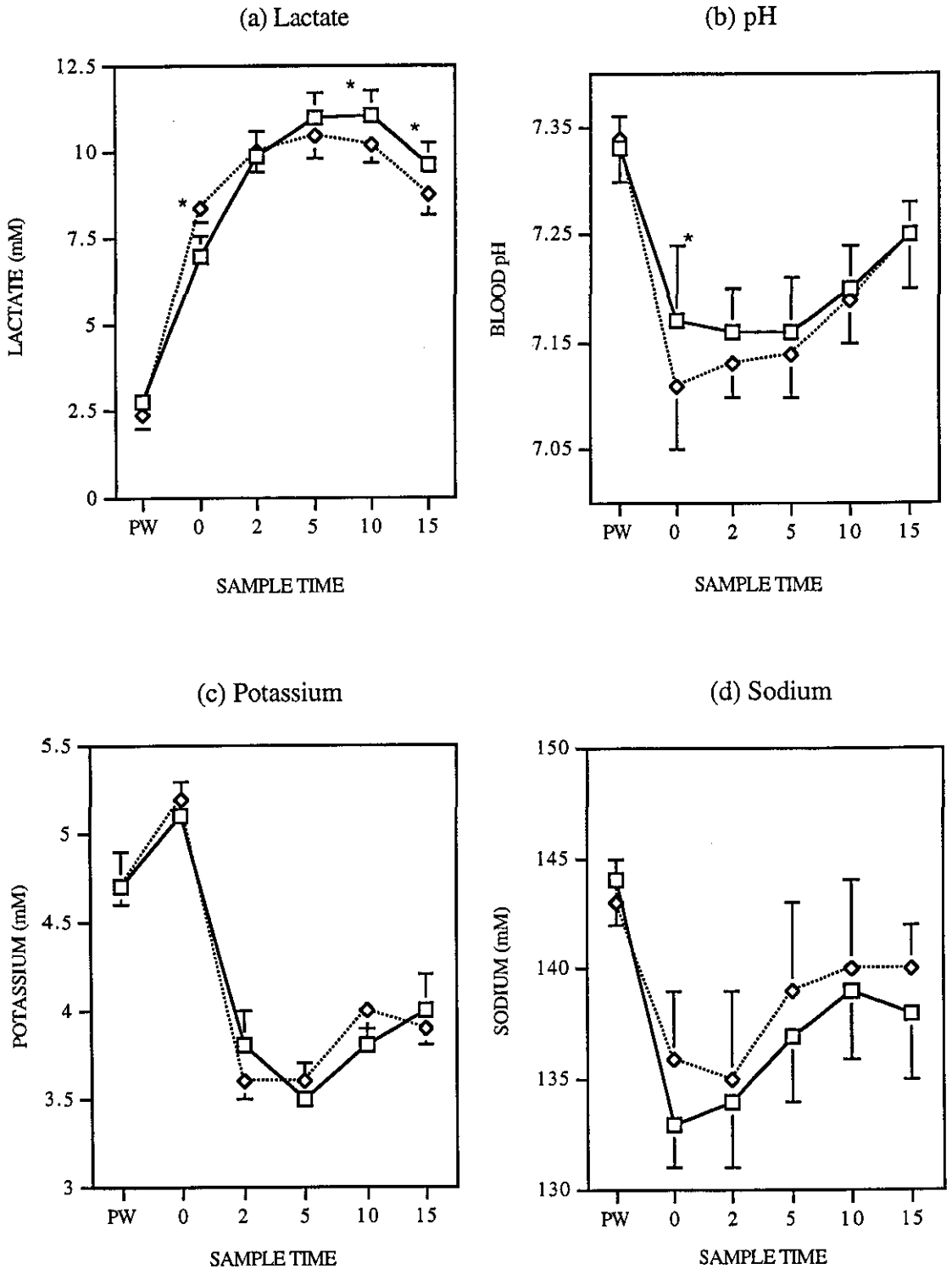

Fig. 6.2. Selected blood metabolites measured post warm-up (PW) and 31 s (0), 2, 5, 10, and 15 minutes following a 30 s sprint against an applied resistance of either 75 (square symbols) or 110 (diamond symbols) $N \cdot kN^{-1}$ body weight (main effect time = $P<0.01$ for all metabolites; Main effect applied resistance=ns; interaction $P<0.05$; post-hoc *=$P<0.05$).

	SPRINT 1 APPLIED RESISTANCE (N per kN BODY WEIGHT)							
	75				110			
	SPRINT 1			SPRINT 2	SPRINT 1			SPRINT 2
	Work (kJ)	PPO (W)	MPR ($rad.s^{-1}$)	PPO (W)	Work (kJ)	PPO (W)	MPR ($rad.s^{-1}$)	PPO (W)
Performance trial (2 sprints)	13.8±0.8	743±72	10.6±0.3	402±33†	14.7±1.6	729±86	7.7±0.4**	442±31*†
Metabolic trial (1 sprint only)	13.7±0.8	774±75	10.6±0.3	---------	14.6±1.3	720±76	7.6±0.4**	---------

Table 6.2. Performance data during a 30 s sprint against an applied resistance equal to either 75 or 110N·kN^{-1} body weight (SPRINT 1) followed by a 5-6 s sprint against 75 N·kN^{-1} body weight (SPRINT 2). (n=8; † $P<0.01$ reduction in peak power output in sprint 2 compared with that achieved in the first few seconds of sprint 1: ** $P<0.01$ difference in mean pedal rate during sprint 1 when performed against either 75 or 110 N·kN^{-1} body weight: * $P<0.05$ difference in peak power output in sprint 2 following sprint 1 against either 75 or 110 N·kN^{-1} body weight).

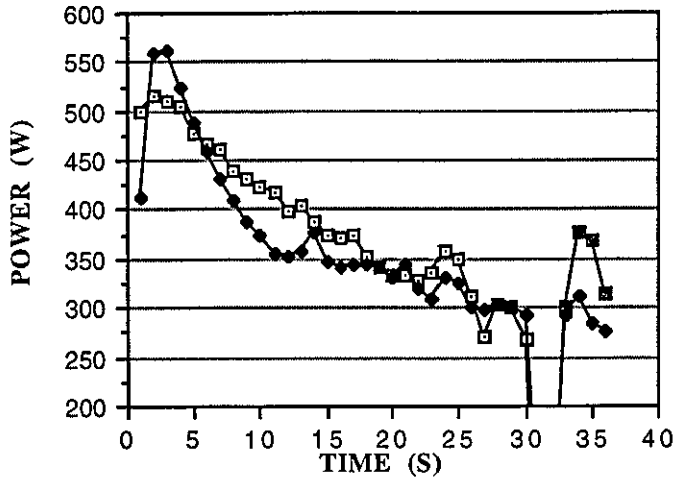

Fig. 6.3. Power profiles obtained from a single subject performing a 30 s sprint followed almost immediately by a 5-6 s sprint. Dotted symbols represent a first sprint performed against an applied resistance of 110 and filled symbols against 75 N·kN^{-1} body weight. The second sprint was always performed against 75 N·kN^{-1}body weight.

Discussion

This Chapter describes the magnitude of the mechanical and biochemical changes arising from maximal high-intensity exercise performed against different applied resistances upon a friction-loaded cycle ergometer. The mechanical changes were assessed, in common with the previous Chapters, as the ability to generate peak power approximately 2-3 s after exercise. The extent of the biochemical changes was assessed by measuring the changes in the concentration of certain mixed muscle metabolites and the pattern of recovery of selected blood metabolites. The main findings were that the mechanical changes were affected by the applied resistance and hence average pedalling rate. However, the results found that the magnitude of the biochemical changes was unaffected by changes in pedalling rate. Therefore, the biochemical data were unable to suggest a mechanism for the differences in peak power in a subsequent sprint found in this and the two preceding Chapters.

The two applied resistances in the present study resulted in average pedalling rates of approximately 7.7 and 10.6 $rad \cdot s^{-1}$ (74 and 101 $rev \cdot min^{-1}$) which is an approximate 26 % difference in the number of muscle actions between the two trials. Nevertheless, despite the different number of muscle actions the extent of the changes in the concentration of selected muscle metabolites was similar. The similarity of the muscle metabolite responses at the two different average pedalling rates is in agreement with previous findings which have analysed mixed muscle at fast 14.7 $rad \cdot s^{-1}$ (140 $rev \cdot min^{-1}$) and slow 6.3 $rad \cdot s^{-1}$ (60 $rev \cdot min^{-1}$) pedalling rates on a constant velocity ergometer (Jones *et al.*, 1985). It has frequently been suggested that the relative contribution to power output of type 1 fibres is at its highest at approximately 6.3 $rad \cdot s^{-1}$ (60 $rev \cdot min^{-1}$) but is considerably less at higher pedalling rates (Sargeant, 1994). Although single fibre analysis was not performed on the muscle samples obtained in this study, a previous single fibre study involving dynamic exercise, at 6.3 and 12.6 $rad \cdot s^{-1}$ (60 and 120 $rev \cdot min^{-1}$), showed no difference in the concentration of muscle metabolites at the two rates in a particular fibre type (Beelen *et al.*, 1993). Thus, the results of this study on a friction loaded cycle ergometer, and previous research involving dynamic exercise suggest that there are no differences in the muscle metabolite responses at pedalling rates which are typically employed on a cycle ergometer. It also suggests that there is no selective change in the concentration of muscle metabolites in some type II fibres (Beelen and Sargeant,

1993; Sargeant, 1994) at different pedalling rates which may be adopted by human athletes.

The ability to generate peak power in a further, almost immediate, sprint was influenced by the resistive mass employed during the first sprint. Peak power was greater following prior exercise against the heavier resistance giving rise to a fewer number of muscle actions. This finding is in agreement with previous studies (Beelen and Sargeant, 1993; Chapters 4 and 5). Previously it has been suggested that the recovery of peak power is dependent upon the muscle concentration of phosphocreatine (Bogdanis *et al.*, 1995). This may not be so where recovery is very short (2-3 s) as the post exercise concentration of phosphocreatine was similar, as well as a number of other metabolites involved in the anaerobic pathways, despite a difference in the ability to generate peak power. However, the muscle biopsy sample was not frozen until 6 s after sprint 1. Nevertheless, it is likely that in 6 s only limited resynthesis of phosphocreatine has taken place as the half time of recovery following the mode of exercise used in this study has been reported as approximately 57 s (Bogdanis *et al.*, 1995). It is reasonable, therefore, to assume that the 6 s sample is a good reflection of the concentration of phosphocreatine at 2-3 s. Therefore, the changes in the concentrations of muscle metabolites do not suggest a mechanism for the differences in the short-term ability to generate power.

The similarity of the plasma concentrations of K^+ and Na^+ in the blood samples following exercise against both resistances fails to indicate a mechanism for the differences in the ability to generate power in sprint 2. However, because the post-exercise muscle biopsy was required to be taken quickly and rapidly frozen, the first post-exercise blood sample was delayed until 31 s after the end of the sprint. The blood sampling procedure normally takes a total of approximately 30-60 s so the post-exercise blood sample provides only an indication of plasma potassium and sodium approximately 30-90 s post-exercise. Using catheters inserted in the femoral vein and artery it has been shown for intense exercise lasting one minute, that the peak plasma concentration of potassium is found almost immediately after exercise and declines with a half-life of 25 s (Medbo and Sejersted, 1990). Clearly, the major ionic changes in the plasma were missed in the present study and if repeated using more sophisticated methods a difference between trials may be observed.

Interestingly, both performance trials suggested that the recovery period of 2-3s was of sufficient duration to allow a significant recovery of power output.

Animal experiments have frequently demonstrated that following high-frequency stimulation, force initially recovers very rapidly (Jones and Bigland-Ritchie, 1986; Westerblad et al., 1991) and this rapid phase, linked with the re-establishment of ionic balance, has a half-time of 1 s (Lannergren and Westerblad, 1986). Therefore, an alternative possible approach to establish whether the magnitude of the ionic changes is different at different pedalling rates may be to examine the very short-term pattern of recovery of power in more detail. In this Chapter and the two previous ones, the period of recovery has been approximately 2-3 s. However, if the short-term pattern of recovery is to be assessed in more detail, any hiatus between the two sprints must be more accurately measured and sensitivity increased.

Also, it is probable that fatigue during high-intensity exercise is multi-factorial. Indeed, electrical stimulation studies have demonstrated that two kinetically distinct phases of recovery exist (Balog and Fitts, 1996). The authors of that study noted a rapid initial recovery phase which was correlated well with the recovery of the membrane potential and intracellular $[K^+]$ and $[Na^+]$ after exercise and a slower second phase linked to the recovery of metabolic factors such as inorganic phosphate and pH. Thus, whilst the existence of a rapid recovery phase may point to the importance of ionic factors, it may not be the sole cause of fatigue. Therefore, it is vital that the pattern of recovery following high-intensity exercise at different average pedalling rates is investigated over a substantial period of time.

In conclusion, this Chapter has shown that the magnitude of the biochemical changes during 30 s of maximal high-intensity exercise is unaffected by the applied resistance. However, the mechanical changes, reflected as the ability to generate peak power in a subsequent sprint are influenced by the applied resistance and hence the average pedal rate. Therefore, the biochemical data failed to suggest a mechanism for the differential rate of recovery of power output. Nevertheless, further experiments investigating in more detail the pattern of recovery of power output may a valuable insight into the aetiology of fatigue during high-intensity exercise.

CHAPTER 7

SHORT-TERM RECOVERY OF POWER OUTPUT FOLLOWING MAXIMAL HIGH-INTENSITY EXERCISE PERFORMED AT DIFFERENT AVERAGE PEDALLING RATES.

The extent of the mechanical changes have been assessed in the previous Chapters by the ability to generate peak power in a short sprint which commenced 2-3 s after 30 s of high-intensity exercise performed at different average pedalling rates. However, it is probable that many factors are involved in the fatigue process during such exercise and it is unlikely that a single period of recovery will highlight all potential causes. Also, the previous Chapter speculated that if the initial recovery phase was related to the restoration of ionic balance, then this phase would exhibit a rapid rate of recovery of power output. With this in mind, it is vital that the duration of any recovery interval is precisely known.

Introduction

The model of repeated exercise has become popular with exercise physiologists in order to further understanding of the aetiology of fatigue during high-intensity exercise (Nevill *et al.*, 1996). However, the period of recovery between bouts of exercise is usually in excess of the duration of exercise. For example, a popular model is two 30 s periods of maximal exercise separated by 4 minutes of recovery (Bogdanis *et al.*, 1995). These studies commonly find a close correlation between the recovery of power output in the second period of exercise with a particular, or more than one, muscle metabolite. However, the evidence is only circumstantial whether the two are causal (McComas, 1996) and whether the availability of a particular metabolite such as PCr is the factor limiting performance is still in question (Nevill *et al.*, 1996).

The previous studies outlined in this thesis have used the model of repeated exercise with a much shorter recovery period of approximately 2-3 s between 30 s of high-intensity exercise and a 6 s all-out sprint. All the Chapters have shown that the ability to generate peak power in the second sprint is dependent upon the average pedal rate of bout 1; peak power output being lower following exercise at high as opposed to moderate average pedalling rates. Also, in Chapter 6 it was suggested that this differential rate of recovery in the

very short term recovery of power was unlikely to be the result of metabolic factors. This view was expressed because the extent of the changes in the concentration of muscle metabolites was similar at different average pedalling rates and also the initial rate of recovery of power appeared to be too rapid to be related to metabolite resynthesis. However, there was no direct evidence to suggest that ionic factors were responsible for the differential rate of recovery of power as the changes in the plasma concentrations of sodium and potassium ions were also similar at the two rates. This similarity indicated *prima facie* that a greater ionic imbalance at the faster of the two rates was not the cause of fatigue.

However, the literature does seem to suggest that the very short term recovery of force is predominantly mediated by the restoration of ionic balance and the longer term recovery by the resynthesis of muscle metabolites (Balog and Fitts, 1996). In view of the methodological difficulties regarding the ionic measurements, important information concerning the aetiology of fatigue may be obtained from a more detailed study of the recovery of power. If the two stage hypothesis is correct, then there should be rapid recovery phase complete within a few seconds of completing prior fatiguing exercise, as ionic balance is restored, and a slower secondary phase as muscle PCr, pH etc. recovers. Moreover, if high-intensity exercise at different average pedalling rates does differentially affect the magnitude of the ionic changes then it follows that only the very short term recovery of power should be affected by the pedal rate at which prior exercise is performed. Conversely, Chapter 6 demonstrated that the extent of the changes in the concentration of muscle metabolites was unaffected by changes in pedalling rate, so the second phase of recovery, which is believed to be related to metabolite resynthesis, should be similar irrespective of the average pedalling rate.

Methods

Subjects: Six females (age 21±1 years; height 163±7 cm; bodymass 59±9kg; mean±SD) volunteered to participate in the study. Full details of the ethical considerations, familiarisation procedures and subject documents can be found in Chapter 3 and Appendix A.

Equipment: Full details of the equipment and procedures employed to standardise trials can be found in Chapter 3.

Protocol and experimental procedures: After prior familiarisation, the subjects came to the laboratory on nineteen further occasions, each visit involving a single main trial. The trials were separated by at least 8 hours. The protocol for each trial was based upon that used for the performance trials in previous Chapters, namely two sprints of 30 and 6 s duration separated by a period of recovery. All trials were preceded by a standard 2 min warm up (30 s @ 88W; 30 s @ 103W; 30 s 160W; 30 s @ 88W) and 5 minutes rest. Unlike the performance trials of previous Chapters, where a single recovery period of approximately 2-3 s was used between sprints, this study investigated the pattern of recovery of peak power at a number of intervals up to 300 s. Nine trials were performed against an applied resistance of 50 $N \cdot kN^{-1}$ body weight in sprint 1 and a further ten against 75 $N \cdot kN^{-1}$ body weight. The second sprint of 6 s was always performed against the greater resistance. The only variation for each trial concerned the period of recovery between sprints 1 and 2, which was one of nine intervals; 1, 3, 6, 9, 12, 15, 60, 120 or 300 s. A further recovery period of 0 s, i.e. the subjects performed a single maximal sprint against 75 $N \cdot kN^{-1}$ body weight for 36 s, accounted for the "extra" tenth trial where the greater resistance was applied in sprint 1.

A number of changes to the protocol, compared with the performance trials of previous Chapters, were made in order to ensure more accurate and precise measurements of the recovery period and to standardise procedures between sprints 1 and 2. Previously all sprints were performed from a rolling start but in this study every sprint was performed from a stationary start. This allowed both sprints in each trial to be performed from the same initial pedal position. A large part of the familiarisation sessions was devoted to the subjects resetting their feet as quickly as possible at the end of sprint 1. A stationary start also allowed use of the computer's internal clock to accurately record the recovery period. This was achieved by incorporating into the program the recording of the exact time of commencement of sprints 1 and 2. As a result, the recovery periods, up to 15 s of recovery, could be measured to 1/100th of a second; the longer recovery periods (60,120 and 300 s) were only recorded to the nearest second. The computer's internal clock also enabled the period of recovery to be relayed to the subjects by programming the computer to generate a loud audible bleep to signify the end of sprint 1 followed, after the appropriate period of recovery, by a second bleep to instruct the subjects to commence sprint 2.

Statistical Analysis: Data were analysed using two-way analysis of variance. The factors being the applied resistance in trial 1 and recovery interval. Significance was accepted at the $P<0.05$ level. Results are presented as mean±SEM unless otherwise stated.

Results

In sprint 1, the number of muscle actions in 30 s was manipulated by approximately 21 % as a result of employing the two applied resistances; mean pedal rate being 13.7±0.5 rad·s^{-1} (131±5 rev·min^{-1}) against 50 N·kN^{-1} body weight and 10.9±0.4 rad·s^{-1} (104±4 rev·min^{-1}) against 75 N·kN^{-1} body weight ($P<0.01$). Mean power output was greater against 75 N·kN^{-1} ($P<0.05$) compared with 50 N·kN^{-1} of body weight. The actual period of recovery between sprints 1 and 2 was typically 0.2-0.4s more than the interval between the two computer generated bleeps, but the total recovery time (time between bleeps + subject's reaction time) was similar ($P>0.05$) for both loads at all recorded time points from 1-300 s. Thus, for clarity, the periods of recovery are subsequently referred to as 1, 3 s etc. although the actual hiatus was slightly longer.

At the end of the first bout of 30 s of high-intensity exercise, power output was reduced by approximately 60 %, compared with the peak generated in the unfatigued state, against both resistances. There was however a large inter-subject variation, some subjects still achieving a high percentage (a fatigue index of approximately 35 %) of their peak power at the end of exercise, whilst others exhibited a much higher fatigue index of 70+ %.

Table 7.1 shows the absolute values of peak power output achieved in sprint 2 from 0-300 s of recovery. The same data, expressed as a percentage of the unfatigued peak power output, is shown in Figure 7.1(a). This graph clearly demonstrates that following high-intensity exercise the recovery of power output has more than one phases; an initial rapid and extensive recovery phase complete within a few seconds of exercise followed by a slower secondary phase, or phases.

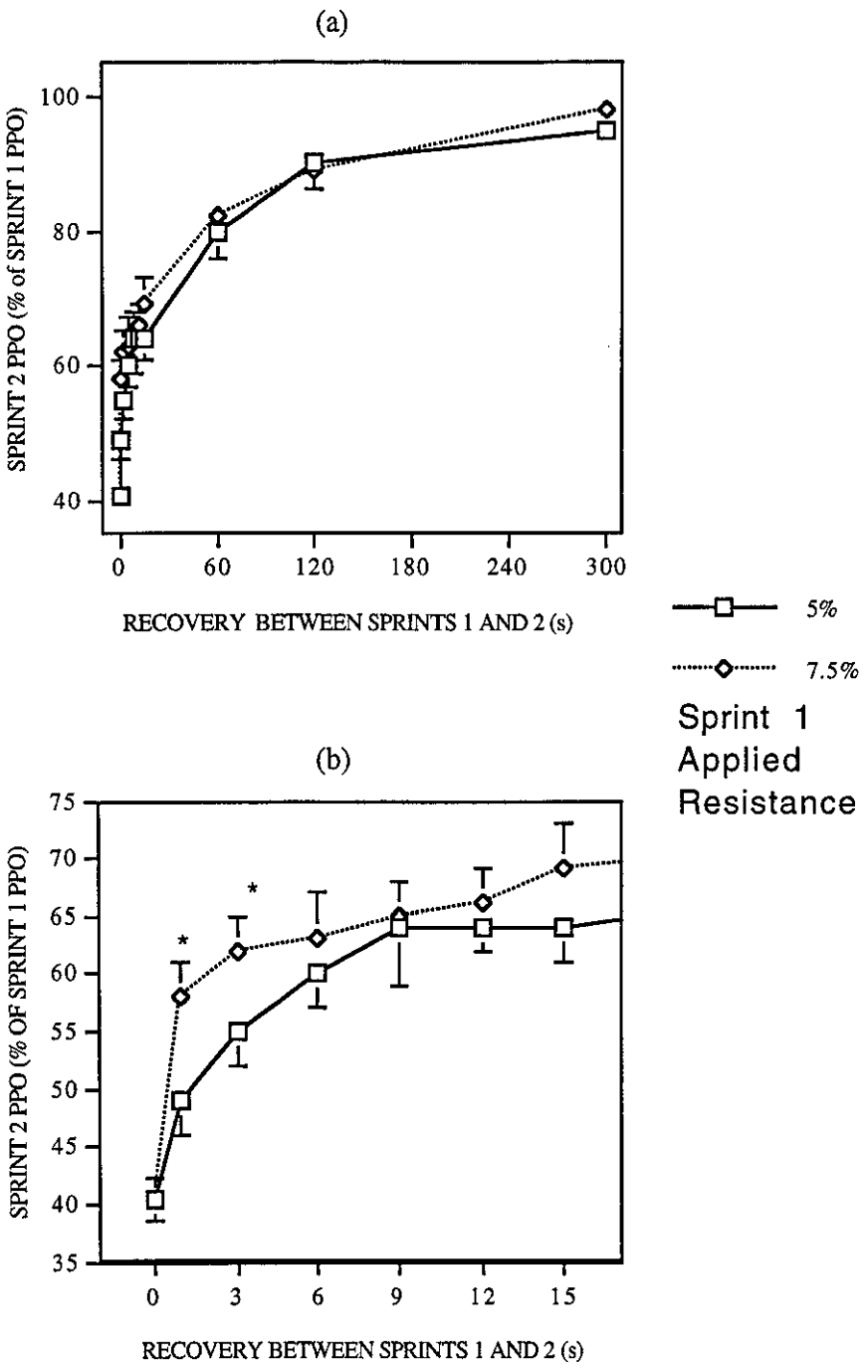

Fig. 7.1. (a) Peak power output (PPO) in a 6 s sprint, expressed as a percentage of the PPO achieved in a prior 30 s sprint. The period of recovery between the two sprints is shown on the x-axis; (b) An enlarged view of the first 15 s of recovery emphasising that the immediate recovery of power is influenced by the average pedal rate in a prior fatiguing 30 s maximal sprint (n=6, * P<0.05 significantly different PPO following exercise against applied resistances equal to 50 (5%) and 75 $N \cdot kN^{-1}$ (7.5 %) of the subject's body weight).

SPRINT 1	RECOVERY INTERVAL BETWEEN SPRINTS 1 AND 2 (S)								
($N \cdot kN^{-1}$ BW)	1*	3*	6	9	12	15	60	120	300
50	425±42	468±35	519±37	543±39	540±47	539±41	679±60	756±67	816±92
75	478±31	503±29	495±40	538±33	525±33	546±38	656±52	725±74	788±84

Table 7.1. Peak power output (W) achieved in a 6 s sprint (sprint 2) performed after a 30 s maximal all-out effort (sprint 1) against an applied resistance of either 50 or 75 $N \cdot kN^{-1}$ of the subject's body weight. (n=6, *=P<0.05 significantly different peak power attained in sprint 2 after this period of recovery from a maximal 30 s all-out sprint against 50 and 75 $N \cdot kN^{-1}$ body weight).

It is clear from Figure 7.1(a) that these two phases exist irrespective the number of muscle actions as when presented over a period of 300s, the pattern of recovery of power output seems to be unaffected by average pedalling rate. However, Figure 7.1(b) which focuses upon the first 15 s of recovery, graphically demonstrates that the rate of recovery of power output in the first 3s is dependent upon prior average pedalling rate. Although there was a initial rapid recovery of power output irrespective of pedal rate, the extent of recovery at 1 and 3 s post-exercise was greater following exercise at the slower average pedalling rate (where the heavier of the two resistances was applied) compared with the faster (where the lighter resistance was applied). At all other time points the extent of recovery of power output was unaffected by prior pedalling rate.

Discussion

The purpose of this study was to examine, in more detail, the pattern of recovery of power output following high-intensity exercise at different pedalling rates. It has found that recovery has at least two phases; an initial rapid recovery of power occurs during the first 6 seconds of recovery followed by a slower phase of recovery until 300 s had elapsed. Although an initial rapid recovery of power output occurred following exercise against both loads, the initial recovery phase was affected by the resistance applied during prior fatiguing exercise but the slower phase was independent of pedal rate.

The results from this study showed that 300 s of recovery enabled power output to recover to 96 % of the unfatigued value compared with only 40 % at the end of the first exercise bout. However, irrespective of prior pedalling rate, a very large proportion of this recovery occurred in the initial few seconds of recovery. For example, at 9 s post exercise peak power output had returned to 64 % of the unfatigued value. This demonstrates that the initial phase of recovery is very rapid. Indeed, the recovery of power output which took place in the first few seconds after sprint 1 against both loads is so rapid that it strongly suggests that this phase is unconnected with metabolite resynthesis. According to Westerblad *et al.* (1991), a very rapid recovery of force strongly suggests that fatigue is the result of a T-tubule conduction failure.

A further pointer suggesting that the initial rapid recovery may be primarily linked to ionic factors is that the power profiles bear a striking similarity to the force profiles which have been produced by electrically stimulating a muscle at

high frequencies, and following a short period of rest restimulating (Jones, 1996). This "High frequency fatigue" resulted in a loss of force during the first period of stimulation which was rapidly reversed during the recovery period. The authors of these studies have suggested that this rapid recovery cannot be related to muscle metabolite resynthesis. It is more likely that it represents the recovery of a failure which has occurred in the excitation-contraction system (Lannergren, 1992), perhaps the restoration of ionic balance across muscle membranes.

High frequency fatigue has been for many years regarded as unlikely to occur during normal human activity because the stimulation frequencies are not high enough. However, recently the features of high frequency fatigue were demonstrated during electrical stimulation at a frequency of 60 Hz (Sacco *et al.*, 1994). The intense nature of the exercise in the present study may entail high enough stimulation frequencies to give rise to such fatigue as initial firing rates of 70-90 Hz have been reported (Tanji and Kato, 1972). The cause of such fatigue is believed to be ionic as changes in extracellular Na^+ and K^+ modify the force profiles (Jones, 1996).

The present study suggests that this initial rapid recovery phase is influenced by the pattern of previous activation. A possible insight into the factors which may be causing greater ionic imbalance where prior exercise involves a high number of muscle actions, compared with fewer, may be found in the work of Clausen and colleagues (Cairns *et al.*, 1995, Harrison *et al.*, 1996). These authors have recently concentrated on the role of the Na-K pump which attempts to restore ionic balance after each action potential by pumping K^+ back into and Na^+ back out of skeletal muscle. When rat skeletal muscle is stimulated at 90 Hz there is a rapid and considerable loss of force which is rapidly reversed once stimulation is stopped. Moreover, if a large proportion of the pump capacity is inhibited by preincubation in a inhibitor of the pump, such as ouabain, then the initial rate of force recovery is much slower but by 5 minutes into recovery there is no difference in the ability to generate force (Harrison *et al.*, 1996). Also, on rat skeletal muscle at an elevated $[K^+]$extr, the same group has found that modifying the frequency and intensity of stimulation impacts upon the tetanic force. In particular, when using strong 1 ms impulses tetanic force is reduced to lesser extent than when more but weaker pulses are applied every 0.1 ms. The authors suggest that the most likely cause of these findings is an increased number of inexcitable fibres arising from the more rapid frequency regimen of stimulation (Cairns *et al.*, 1995).

The results from this study demonstrate that the secondary phase (or phases) of recovery of power output is unaffected by changes in pedalling rate. It seems most likely that the extent of recovery in this phase is determined by the extent of resynthesis of muscle metabolites. In particular recent research has suggested that the ability to generate peak power in repeated sprints is closely correlated with the availability of muscle phosphocreatine (for example Bogdanis *et al.*, 1995). The results from Chapter 6 showed that muscle phosphocreatine availability 6 s after exercise was similar, irrespective of pedalling rate, and therefore if the secondary phase is primarily governed by the availability of phosphocreatine then no differences in power output from 6-300 s of recovery would be anticipated.

Electrical stimulation studies have also demonstrated that two kinetically distinct phases of recovery exist (Balog and Fitts, 1996). The authors of that study also noted a rapid initial recovery phase which was correlated well with the recovery of the membrane potential and intracellular $[K^+]$ and $[Na^+]$ after exercise and a slower second phase linked to the recovery of metabolic factors such as inorganic phosphate and pH.

In conclusion, this study has demonstrated that the recovery from high-intensity exercise is multi-phasic. There is a very rapid and substantial recovery of power, complete within 6 s of maximal dynamic exercise which probably results from the restoration of ionic balance rather than metabolites. There is a slower secondary phase which is probably related to the resynthesis of muscle metabolites particularly phosphocreatine.

CHAPTER 8

GENERAL DISCUSSION

In this Chapter a summary of the main results is presented first. Secondly, the extent of the mechanical and biochemical changes associated with high-intensity exercise are considered and how, if at all, their magnitude is affected by changes in average pedalling rate. There is no discussion regarding the electrical changes as these were not investigated because of the methodological problems of measurement outlined in Chapter 2. Thirdly, this Chapter considers the extent to which the experiments have achieved the ultimate aim of this thesis, which was to further understanding of the causes of fatigue during high-intensity exercise. Next, the author's opinions concerning the possible factors primarily responsible for fatigue are expressed based upon the results of the experimental studies, combined with further knowledge acquired from background reading and attendance at scientific meetings during the course of completing this thesis. Finally, the Chapter concludes with some ideas for future research and the implications of the results for athletic performance and training regimens.

8.1. MAIN FINDINGS

The experiments are described in Chapters 4-7 and describe the magnitude of the mechanical and biochemical changes during 30 s of high-intensity exercise on a friction-loaded cycle ergometer at different average pedal rates. The mechanical changes were assessed by the ability to generate peak power in a subsequent 6 s sprint. In the studies described in Chapters 4, 5 and 6, a single period of recovery of 2-3 s was employed, but in the study contained in Chapter 7, a number of recovery intervals from 0-300 s were used. The biochemical changes were described in Chapters 4 and 6. In Chapter 4, the changes in the concentration of selected blood metabolites arising from exercise are reported, whilst Chapter 6 also describes the analysis of muscle biopsy samples from females.

Chapter 4. This Chapter contains details of a pilot study and a more extensive main study which describe the biochemical and mechanical changes of constant external work exercise under conditions where average pedalling rate was imposed or freely chosen. The pilot study suggested that where constant

external work was produced under conditions where pedalling rate was imposed, the extent of the metabolic changes may be less than a strategy where pedalling rate was freely chosen. The main study confirmed that it was possible to replicate the external work of a high-speed 30 s all-out sprint against an applied resistance of 75 $N \cdot kN^{-1}$ body weight by imposing moderate constant average pedal rates of 6.3-9.4 $rad \cdot s^{-1}$ (60-90 $rev \cdot min^{-1}$) but not at higher rates of 10.2-15.7 $rad \cdot s^{-1}$ (97-150 $rev \cdot min^{-1}$). Where the same external work was produced under paced conditions, the ability to generate peak power in a subsequent sprint was greater compared with all-out. Similarly, during 15 minutes of recovery, the concentration of selected blood metabolites returned more rapidly towards their unfatigued values in the paced condition compared with all-out.

Chapter 5. The study described in this Chapter attempted to resolve whether it was the total number of muscle actions, or if it was the pacing strategy (all-out vs. paced), which was the major factor influencing the differential short-term recovery of peak power output. A 30 s all-out sprint against an applied resistance of 109 $N \cdot kN^{-1}$ body weight resulted in approximately 9 % more external work and 21 fewer muscle actions compared with an all-out sprint against 75 $N \cdot kN^{-1}$ body weight. Peak power output in a further sprint, performed after a recovery of 2-3 s, was 20-25 % greater following both the 30 s sprint against the heavier applied resistance and 30 s of paced exercise at a similar average pedalling rates, compared with a high speed all-out sprint against the lighter resistance. The results suggest that it is the number of muscle actions and not the pacing strategy which seems to be the major determinant of the differential rate of recovery of power output following exercise at different average pedalling rates.

Chapter 6. The study described in this Chapter compared the extent of the biochemical and mechanical changes of 30 s of maximal intensity exercise (i.e. where the subjects always adopted an all-out effort) against two different applied resistances. In agreement with the previous Chapter, the results from this study found that peak power output in a 6 s sprint, performed approximately 2-3 s after fatiguing high-intensity exercise, is dependent upon the number of preceding muscle actions; greater fatigue arose following exercise against the lighter of the two applied resistances. However, the biochemical data, which included the analysis of a number of metabolites involved in the anaerobic pathways of ATP provision and changes in plasma electrolytes, did not suggest a mechanism for these differences.

Chapter 7. The study described in this Chapter investigated, in more detail, the pattern of recovery of peak power output following 30 s of maximal high-intensity exercise against different applied resistances. The results showed that recovery has at least two phases; an initial rapid recovery of power which was most evident during the first 6 s of recovery and a slower recovery phase until 300 s had elapsed. The initial recovery phase was rapid irrespective of pedalling rate although it's rate during the first 3s of recovery was dependent upon the prior pedalling rate. The recovery of power output in this phase was so rapid that it was unlikely to be associated with metabolite resynthesis. Peak power was 13 and 8 % higher at recovery intervals of 1 and 3 s following prior exercise against the heavier of the two applied resistances which may have benn a consequence of differences in ionic balance between the two applied resistances. However, at recovery intervals between 6 and 300 s, the pattern of recovery of power output was independent of average pedal rate. The secondary phase was probably reflective of metabolite resynthesis after exercise.

8.2. INFLUENCE OF CHANGES IN AVERAGE PEDALLING RATE UPON THE MAGNITUDE OF THE MECHANICAL AND BIOCHEMICAL CHANGES.

8.2.1. Mechanical Changes

The rapid and substantial decline in power output exhibited during high-intensity cycle exercise suggests that the extent of the mechanical changes is extensive. Previously, the extent of these changes has been assessed using either a single bout of exercise, or the model of repeated exercise bouts where the recovery period is equal or longer than the duration of exercise. The experiments described in this thesis used the model of repeated exercise but, unlike other studies which have used this model, concentrated on the very short-term recovery of power output. Indeed, the recovery interval predominantly used in this thesis of 2-3 s, represented only about one-tenth of the duration of prior exercise. However, this was always sufficient time to enable a fixed resistance of 75 $N \cdot kN^{-1}$ body weight to be applied to the belt surrounding the flywheel of a friction-loaded cycle ergometer.

With hindsight, a better experimental design would have been to require the subjects to perform three bouts of exercise when considering the recovery of

power output. The first and third being 6s sprints against a fixed resistance. In the second bout, 30s of exercise where pedalling rate was manipulated should have been performed. Peak power in the third bout (fatigued) could then have been compared with that achieved during the first bout which involved an unfatigued sprint of the same duration and against the same applied resistance.

A consistent finding of the studies was that the recovery of power exhibited within the first 2-3 s after fatiguing high-intensity exercise was dependent upon the prior average pedalling rate. All the main studies demonstrated that prior exercise at high average pedalling rates resulted in a slower initial recovery of power compared with low or moderate rates.

The greater initial recovery of peak power at low, as opposed to high, average pedalling rates agrees with previous studies which have investigated the extent of the mechanical changes during a single sprint by calculation of a fatigue index (FI) (the method of calculation is described in Chapter 2). However, if the FI values obtained from previous studies at different pedalling rates are compared with the results of the studies described herein, it is apparent that the magnitude of the differences is dissimilar. On a constant-velocity ergometer, it has been shown that the FI for peak power during a 30 s sprint was 23.6 % at 6.3 rad·s^{-1} (60 rev·min^{-1}) and increased to 45.7 % at 10.5 rad·s^{-1} (100 rev·min^{-1}) (McCartney *et al.*, 1983a). Using data from Chapter 6 (Table 6.2), and substituting "peak power in sprint 1" for "initial power" and "peak power in sprint 2" for "final power" in the FI equation, gives FI's of 39 and 46 % at 7.7 rad·s^{-1} and 10.6 rad·s^{-1} (74 and 101 rev·min^{-1}) respectively. Although these pedalling rates are not exactly the same as those of McCartney and co-workers, both methods show that the extent of the mechanical changes is dependent upon the pedalling rate. However the difference between the two figures is much greater using the single sprint method (22 %) compared with the 2-3 s recovery data (7 %).

The calculation of the FI during a single sprint at different average pedalling rates must be treated with caution. This is because of the reduction in the velocity for peak power production as fatigue arises will tend to exaggerate the difference in FI between low and high rates. For example, a velocity which the literature suggests is optimal for power production as described by the power-velocity relationship of muscle in the unfatigued state (approximately 11.5 rad·s^{-1} (110 rev·min^{-1})), will become too fast as fatigue ensues. Conversely, slower rates will be far from optimal at the start of exercise but will lie nearer

the optimum at the end of exercise. To some extent, the problem of calculation in a single sprint was overcome in the experiments described herein by assessing the magnitude of the mechanical changes with a subsequent sprint against a fixed resistance. However, even this protocol is not without flaws. Specifically, it is probable that the repeated exercise model with a 2-3s recovery period under-estimated the FI. Previously it has been suggested that within 2-3 s there is no recovery of power output (Buttelli *et al.*, 1996). However, the results of Chapter 7, when the pattern of recovery of peak power was studied in more detail, demonstrated that even within 2-3 s of high-intensity exercise, there is a significant recovery of power.

It was suggested in the discussion in Chapter 7, that the rapid decline and recovery of power output exhibited during and immediately following high-intensity exercise, may indicate that the subjects were experiencing high frequency fatigue (HFF). It must be emphasised that the rapid recovery of power output was a feature following high-intensity exercise, irrespective of the pedalling rate at which it is performed (Fig. 7.1(b)). One of the features of HFF is a loss of force after stimulating at high frequencies which is reversed rapidly by reducing the frequency (Jones, 1996), or restimulating after a short period of rest (Lannergren, 1992). This rapid recovery of force is thought to be reflective of the reversal of changes which have taken place during stimulation in the excitation-contraction system, rather than in the contractile apparatus of the muscle. The critical factor determining whether the features of HFF arise seems to be the frequency of muscle stimulation. In mouse soleus muscle, a rapid rise in force occurs after 50 s of stimulation at 100 Hz when the frequency is reduced to 20 Hz but not so following 50 s of stimulation at 50 Hz (Jones *et al.*, 1979). In human tibialis anterior muscle, there is a large, almost immediate, recovery of force following stimulation at 60 Hz but not so at 15 and 30 Hz (Sacco *et al.*, 1994). The literature suggests that human subjects are only able to very briefly achieve firing rates of 60 Hz and above and that the maximum mean sustainable rates are of the order of 25 Hz (Chapter 2). Therefore, it seems unlikely that human subjects are able to voluntarily sustain firing frequencies at a sufficiently high rate to result in HFF. However, when human tibialis anterior muscle was shortened from its optimal length for force production, the features of HFF, which at the optimum length had only been found at a frequency of 60+ Hz, were demonstrated at a stimulation frequency of 30 Hz (Sacco *et al.*, 1994). During cycle exercise, where the vast majority of the force is generated concentrically (Chapter 3), it is possible in highly motivated subjects that a mean firing rate of approximately 30 Hz is sustainable. It is, therefore, likely

that a failure in the excitation-contraction coupling system plays a role in the fatigue process during high-intensity exercise.

The mechanism which underlines the loss of activation seen during HFF, namely that of a slow inactivation of Na^+ channels, was explored in Chapter 2. Although there was a rapid recovery of power output irrespective of the pedalling rate at which exercise was performed (Chapter 7), all the studies showed that the rate of recovery in the first 2-3s post-exercise was dependent upon the prior pedal rate. Assuming that the Na^+-K^+ pump is similarly activated following exercise at different pedalling rates, then the differential initial rate of recovery of power is possibly suggestive of a 'threshold potential' at which fibres become excitable once again following exercise. Clearly the slower initial rate of recovery in the first 2-3s following exercise at high pedal rates suggest that the depolarization during exercise was greater than at slow speeds. If so, this would support the hypothesis that ionic factors play a role in the fatigue process during high-intensity exercise.

However, Jones (1996) makes the point that the only difficulty with slow inactivation as an explanation of HFF is the slowness of the process, since the inactivation appears to develop over the course of minutes, whilst HFF can occur within seconds (and the recovery of peak power following high-intensity exercise) if the frequency is high enough. Thus a fully satisfactory explanation of the rapid recovery of force following high frequency stimulation is still awaited. A possible alternative explanation of the differences in the initial recovery of power output could be that they arose due to central factors. Although for exercise of 45-60s duration the literature suggests that central factors are not important where the subjects are highly trained and motivated (Chapter 2), it must be acknowledged that central factors cannot be entirely ignored. In particular, the methods employed in this study manipulated pedal rate by varying the applied resistance during the first 30s bout of exercise, but always used the same fixed resistance during the second sprint. This meant that during 'low' speed trials the subjects were performing the second sprint against a lighter applied resistance compared with the first. Conversely, in the faster trials subjects generally went from a 'light' applied resistance in sprint 1 to a heavier resistance in sprint 2. Specifically it could be argued that the Golgi tendon organs, which are the receptors which discharge in response to the tension in the contracting muscle, may have inhibited the muscle to a greater extent when encountering a heavier resistance in sprint 2, compared with sprint 1, compared with two sprints against the same applied resistance. However, it

is difficult to reason why this should be the case. Although any central effects may be most pronounced in the first few seconds after exercise, it is difficult to understand why, if central factors are the reason for the differential initial rate of recovery, why a differential ability to generate power does not persist throughout recovery.

It is also evident from the measurements of the recovery of power output from 0-300 s that at least two stages of recovery are present. The initial rapid recovery phase is relatively short lasting, although it is difficult to establish exactly where a slower phase (or phases) of recovery commences (Fig. 7.1(a)). The fact that there are at least two kinetically distinct phases of recovery suggests that there are at least two distinct mechanisms of contractile impairment (Fitts and Balog, 1996). A bi-phasic pattern of recovery from fatigue has been demonstrated previously using muscle preparations (Chapter 2). These studies have typically shown a rapid phase that is complete within 2 minutes and slower phase lasting 30-60 minutes to the unfatigued level of tension. However, in Chapter 7 peak power in sprint 2 had returned to the unfatigued level within 5 minutes of recovery. It would have been useful to have data which followed the recovery of peak power over a similar time scale to those used during the muscle preparation experiments to make a valid comparison of the two different experimental models. It would also have been interesting to establish whether mean power in 30 s recovered in a manner similar to peak power, or not. Nevertheless, the existence of more than one phase in the recovery of peak power is strongly suggestive that fatigue during high-intensity exercise is multi-factorial.

8.2.2. Biochemical Changes

It was anticipated that the biochemical measurements taken in Chapters 4 and 6 would suggest a mechanism, or mechanisms, to explain the differences observed in the recovery of peak power in the initial 3s of recovery. The blood data obtained in Chapter 4 did show differences between a paced and all-out strategy but the blood and muscle data obtained in Chapter 6 were unaffected by changes in average pedalling rate. Thus, a biochemical mechanism for the differences in the mechanical changes was not found.

(i) Metabolic Changes

Two methods were used to manipulate average pedalling rate in this thesis. The first was to impose a pre-set constant average pedalling rate ("paced") so that external work production was evenly produced. The second was to require the subjects always to try to pedal as fast as possible ("all-out"), but vary the applied resistance.

In Chapter 4, selected blood metabolite responses to constant external work exercise produced under two separate strategies, a paced strategy at a moderate average pedal rate and a high speed all-out effort, were examined. The magnitude of the changes in the concentration of key metabolites such as lactate and ammonia was found to be less as a result of the paced exercise compared with all-out. Although both strategies probably involved the recruitment of all motor units throughout exercise, the recruited units would not have to fire at their optimal frequency for maximal power production throughout the paced bout because of the initial sub-maximal power production. Thus, the extent of the metabolic changes may have been less under the paced condition because of a sub-maximal activation in some units. However, the difference may also have been due to a difference in the total number of muscle actions between the two strategies.

In Chapter 6 it was shown that where an all-out strategy is adopted, differences in average pedalling rate have no effect on the muscle metabolite responses. For example, the anaerobic ATP turnover rates of 218 and 231 mmol·kg dry muscle^{-1} for the eight female subjects against different applied resistances were similar. However, both values were a little lower than the 255 mmol·kg dry muscle^{-1} previously reported for eight males performing similar exercise (Bogdanis *et al.*, 1995). Also. the reduction in the concentration of phosphocreatine of 55 (-69 %) and 58 (-74 %) mmol·kg dry muscle^{-1} was unaffected by pedalling rate. The similar muscle and blood data at different pedalling rates, where subjects always adopted an all-out strategy reveal that the metabolic differences of Chapter 4 were predominantly due to the sub-maximal activation of some motor units and not to differences in the number of muscle actions. The biochemical data, therefore, do not provide a satisfactory mechanism to explain the velocity dependent initial recovery of power.

However, the similarity in the muscle responses at different average pedalling rates combined with the fact that the recovery of power output from 6-300 s post-exercise is unaffected by pedalling rate (Chapter 7), suggests that the secondary phase of recovery of power output may be connected with

resynthesis of muscle metabolites. In particular, the similar post-exercise concentration of muscle phosphocreatine following exercise at different pedal rates is considered an important aspect of this slower phase of recovery. The results of other studies also support such a hypothesis. The resynthesis of phosphocreatine following 8-9 minutes of isokinetic cycling at 6.3 rad·s^{-1} (60 rev·min^{-1}) is bi-phasic; an initial phase with a half-time of approximately 21s and a slower secondary phase at 173s (Harris *et al.*, 1976). For 30 s of high-intensity cycle exercise on a friction-loaded cycle ergometer, the recovery of phosphocreatine has been modelled which indicates the half-time to be approximately 57s (Bogdanis *et al.*, 1995; Nevill *et al.*, 1997). Therefore, it seems unlikely that any significant resynthesis of phosphocreatine takes place in 2-3s and so the similar concentrations measured 6s after exercise can be regarded as very close to the concentrations at 2-3s after exercise. Thus the similar post-exercise phosphocreatine concentration cannot explain the differences in power output in the rapid recovery phase but may be a major factor determining the recovery of power thereafter.

It has been speculated that the differential rate of recovery of power output may be due to a selective fatigue of some type II fibres (for example Sargeant, 1994). In view of the similarity of the mixed muscle responses in Chapter 6 and those of a previous study by Jones *et al.* (1985), it seems very unlikely that analysis of muscle metabolites in single fibres would have suggested a metabolic mechanism for the differential recovery of power output in the rapid initial phase.

(ii) Ionic Changes

In was anticipated that the magnitude of the changes in the plasma concentration of the electrolytes, K^+ and Na^+, might provide a mechanism for the initial rapid recovery of power and the differential pattern of recovery of power output, 2-3 s after exercise. The fact that recovery was so rapid suggests that it is due to a failure somewhere in the excitation-contraction system. The discussion in Chapter 7 suggested that restoration of ionic balance across muscle membranes was a likely candidate. Specifically, the extent of K^+ fluxes is dependent upon (i) the number of active fibres in the muscle, (ii) the muscle blood flow and (iii) the action potential frequency (Sjogaard, 1996). High-intensity exercise at different average pedalling rates is unlikely to have major effects on (i) and (ii) but may markedly affect (iii). According to Hallen (1996),

assuming a firing frequency of 30 Hz, pedalling at 1 Hz (i.e. 6.3 rad·s^{-1}) will result in six action potentials per second, as the muscle is only active for approximately 20 % of each pedal cycle. A doubling of pedalling rate to 2 Hz (12.6 rad·s^{-1}) will, therefore, probably increase the number of action potentials per second. The extent of K^+ efflux, which is of the order of 2 μmol kg^{-1} action potential^{-1} will thus be greater at the higher rate. It was suggested in Chapter 2, that an accumulation of potassium, particularly in the depths of the t-tubules where the concentration of Na^+-K^+ pumps is low, may be a cause of fatigue. At high pedalling rates, the accumulation of potassium may be greater compared with low rates because of the increase in K^+ efflux.

The post-warm up plasma [K^+] of 4.7 mM (Chapter 6) is a little higher than resting values of approximately 4 mM reported elsewhere. Therefore, it is likely that the warm-up, although of a fairly low intensity, resulted in a net efflux of K^+. Irrespective of the applied resistance, post-exercise measurements of [K^+] (and [Na^+]) were similar. This indicated that the magnitude of the ionic changes was independent of the pedalling rate and hence the number of preceding muscle actions. However, the highest recorded values of 5.2 mM were much lower than the 8.3 mM previously reported for similar exercise where the measurement were made using indwelling catheters (Medbo and Sejersted, 1990). The major source of the difference was that the first blood post-exercise blood sample was delayed because of the muscle sampling and as K^+ recovers with a half-time of 25 s, the peak value and major changes were missed. In addition, the concentrations were corrected for the estimated changes in plasma volume which took place, although this would only account for a minor part of the difference. Clearly, if plasma K^+ is to be measured, it is vital that the sample is taken as quickly as possible after exercise. Plasma [Na^+] also showed no differences between the two applied resistances. However, it is doubtful whether the plasma changes can be accurately recorded in the first 20 s of recovery even if indwelling cannulae are used (Medbo and Sejersted, 1990). Also, the literature suggests that it must be doubtful whether the plasma changes are a truly accurate measure of the changes taking place in the muscle (Dorup *et al.*, 1988b; Dorup, 1996; Sjogaard, 1996). For example, long-term treatment of patients with diuretics has been shown to lead to potassium (and magnesium) deficiencies in the muscle which are not detectable using the standard methods of serum analysis (Dorup *et al.*, 1988b). According to Sjogaard (1996), neither the exact magnitude or the time history of changes in the concentration of muscular K^+ can be deduced from plasma K^+.

Thus, in conclusion neither the muscle metabolite nor the plasma ion data could provide a biochemical mechanism to explain the differential rate of recovery of power output in the first few seconds after exercise at different average pedalling rates. However, methodological problems may explain why any differences in the ionic changes were not found.

8.3. THE AETIOLOGY OF FATIGUE DURING HIGH-INTENSITY EXERCISE

The primary objective of this thesis was to further understanding of the aetiology of fatigue during high-intensity exercise. Although the model of repeated bouts of exercise has been widely used by other research groups, the recovery period, or periods, have usually been relatively long (>30s). The experiments described in this thesis have predominantly used a much shorter duration of recovery (often 2-3s). The biochemical changes arising from maximal intensity exercise have also been previously studied at different muscle action speeds. However, these experiments have used an exercise regimen where the external angular velocity is imposed, for example by use of an isokinetic cycle ergometer, which does not truly resemble the situation in sporting events where the limbs are free to accelerate.

All five experiments showed that the recovery of peak power in the first few seconds of recovery was dependent upon the average pedalling rate at which prior fatiguing exercise was performed. The muscle metabolite data were unable to suggest a mechanism for the differential initial rate of recovery. Also, the initial phase of recovery of power output was so rapid that it was unlikely to be connected with the resynthesis of muscle metabolites. These findings strongly suggest that factors other than the availability of muscle metabolites are involved in the fatigue process.

In the author's opinion, this other factor, in addition to metabolite availability, which may be involved in the fatigue process may be within the excitation-contraction coupling system. Specifically, high-intensity exercise requires the muscles to fire at high stimulation frequencies involving many action potentials, each of which causes an efflux of K^+ and influx of Na^+. The extent of these changes may be so great that a substantial depolarisation may arise which may result in some myofibrils becoming inexcitable. The initial rapid recovery phase could represent the reversal, at least in part, of these changes thorough the action of the Na^+-K^+ pumps. The extent of the ionic changes would be

greater with increases in average pedalling rate which, in turn, would be reflected by a slower recovery of peak power. However, no biochemical data could be presented to support the author's opinion that fatigue in high-intensity exercise is due, at least in part, to a failure in the E-C coupling system.

However, it must be emphasised that the recovery of power output from 6-300s of recovery was independent of average pedalling rate, as were the changes in the concentration of muscle metabolites. These findings suggest that energy supply, and in particular muscle phosphocreatine availability, is a vital part of the fatigue process. Assuming that the initial recovery phase is reflective of the restoration of ionic balance, it could be argued that the inability of the Na^+-K^+ pump to restore ionic balance is also due to problems relating to energy supply as the pump is energy dependent. However, Clausen and co-workers have conducted many experiments which have manipulated the number of active Na^+-K^+ pumps. They have consistently found that force is enhanced when pump activity is increased and substantially reduced in the presence of a inhibitor such as ouabain. These findings suggest that it is the number of pumps which is the critical factor affecting the ability of the muscle to generate force, and not whether there is sufficient ATP available so that the Na^+-K^+ pumps can function maximally.

8.4. AREAS FOR FUTURE RESEARCH

In the review of literature (Chapter 2), it was suggested that with increases in pedalling rate there may be a greater fatigue of fast fatiguing fibres. Also, it was indicated that the extent of the ionic shifts associated with high-intensity exercise may be velocity dependent. The results of the experiments described herein have proved inconclusive. However, this thesis has demonstrated that some of the features of high frequency fatigue can be exhibited in well trained athletes performing high-intensity cycle exercise.

In humans performing dynamic exercise, biochemical evidence suggesting that a selective fatigue of a small group of type II fibres with increasing pedalling rate arises, is still awaited. The mixed muscle data obtained in Chapter 6 strongly indicate that analysis of single fibres will not suggest a metabolic mechanism to explain this effect. However, a study which has actually analysed metabolites in single fibres has not been undertaken.

It may be the case that if a greater fatigue of fast fatiguing fibres does occur at high pedalling rates, it may not be reflected in the changes in the concentration of muscle metabolites. Moreover, if the extent of ionic movements does play a part in the fatigue process, the literature suggests that there are fibre type differences which may provide a mechanism. Although there are more Na^+-K^+ pumps in type II fibres compared with type I, the ionic movements are substantially greater per action potential in type II fibres (Clausen *et al.*, 1997). As a result, the extent of ionic disturbance is greater in type II fibres compared with type I; the magnitude of this fibre type difference would be expected to increase with an increase in the frequency of action potentials.

Unfortunately, methodological problems will no doubt hinder the development of further understanding of ionic movements, especially if the model of dynamic exercise is used. Ionic movements occur very rapidly during exercise and the results from this thesis suggest that they may be reversed in a rapid manner. Surface EMG measurements would, at first glance seem the obvious tool to assess whether any depolarisation is dependent upon average pedalling rate. However, the literature suggests the technique is not sensitive enough to record any differences. Also, it is not merely the number of Na^+-K^+ pumps that may dictate the extent of this recovery but how many are located in the depths of the t-tubules. Nevertheless, as a start, it is now possible by means of a simple and rapid needle biopsy procedure to detect alterations not only of electrolytes but also of Na^+-K^+ pumps (Dorup *et al.*, 1988a). However, such experiments would need to be carefully designed as the biopsy procedure takes about 5-10 s to complete and many of the ionic changes may be largely reversed within 6 s of exercise.

Energy supply is still the most frequently cited cause of fatigue during high-intensity exercise. For example, some studies have shown an enhancement of performance following a period of creatine supplementation but no mechanism has yet been put forward which satisfactorily explains how such supplementation works. What is certain from the literature is that creatine supplementation results in a significant increase in body weight, believed to be as a result of the retention of water. Interestingly, it is known that the reverse effect, i.e. treatment with diuretics, dramatically reduces the number of pumps in skeletal muscle and contractile force (Dorup *et al.*, 1988b). Clearly, any mechanism capable of enhancing water retention, such as creatine supplementation, may have the reverse effect. A study combining creatine

supplementation and the measurement of the number of electrolyte and pump status may be useful.

The experiments described in this thesis only followed the recovery of peak power generated in a second bout of exercise of 6 s duration; the first bout always being 30 s. It would have been very useful to have followed the recovery of mean as well as peak power following exercise at different average pedalling rates. This would involve two sprints of equal duration separated by a period of recovery. Whether subjects could perform two 30 s sprints with only a very short recovery (2-3 s) should be investigated before such a study is embarked upon. It would also have been helpful to establish whether recovery has two, or perhaps more phases, by extending the range of recovery periods from 1 second up to 1 hour.

In closing, if ionic balance is a major influence on the fatigue process during high-intensity exercise, what are the implications for athletic training. The athlete should be adopting practices which ensure that the electrolyte and Na^+-K^+ pump stores in skeletal muscle are not depleted and enhanced where possible. The status of both has been shown to be affected by the K^+ and interestingly, the Mg^{2+}, content of the diet. Also, athletes should always ensure they are well hydrated as patients treated with diuretics show a dramatic loss of pump function and force (Dorup *et al.*, 1988b). Pump function is activated and deactivated by a number of factors and these were summarised in Fig. 2.17. However, the most potent regulator of pump activity is the degree of activation of the muscle. Hard training will therefore reap rewards!

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APPENDIX A

Subject Documents

A.1. Health History Questionnaire

LOUGHBOROUGH UNIVERSITY OF TECHNOLOGY
Department of Physical Education, Sports Science and Recreation Management

CONFIDENTIAL

HEALTH HISTORY QUESTIONNAIRE

DATE _____ AGE _____

NAME DATE OF BIRTH _____ / _____ / _____

ADDRESS TELEPHONE

_____ _____

_____ MARITAL STATUS

_____ _____

_____

_____

OCCUPATION

_____

DOCTOR

_____

PAST HISTORY (please tick yes or no) FAMILY HISTORY (please tick yes or no)

Have you ever had ? Have any of your family ever had ?

	Yes	No		Yes	No
Rheumatic fever / heart murmur	[ ]	[ ]	Heart disease	[ ]	[ ]
High blood pressure	[ ]	[ ]	High blood pressure	[ ]	[ ]
Any heart trouble	[ ]	[ ]	High cholesterol	[ ]	[ ]
Disease of arteries	[ ]	[ ]	Stroke	[ ]	[ ]
Varicose veins	[ ]	[ ]	Diabetes	[ ]	[ ]
Lung disease	[ ]	[ ]	Congenital heart disease	[ ]	[ ]
Asthma	[ ]	[ ]	Heart operations	[ ]	[ ]
Kidney disease	[ ]	[ ]	Any blood clotting disorders	[ ]	[ ]

Liver disease	[ ]	[ ]	Early death	[ ]	[ ]
Diabetes	[ ]	[ ]	Other family illnesses		
			_____		
Epilepsy	[ ]	[ ]	_____		
			____		
Thyroid disease	[ ]	[ ]	_____		
			____		
Any blood clotting disorders	[ ]	[ ]	_____		
			____		
Any abnormal bruising	[ ]	[ ]	_____		
			____		
Any form of depressive illness	[ ]	[ ]	_____		
			____		

HOSPITALISATIONS

Year _____ Reason _____

Year _____ Reason _____

Any other medical problems ?

_____

_____

_____

_____

PRESENT SYMPTOMS

(Have you recently had ?)

	Yes	No
Chest pain / discomfort during exercise	[ ]	[ ]
Shortness of breath during exercise	[ ]	[ ]
Heart palpitations	[ ]	[ ]
Skipped heart beats	[ ]	[ ]
Cough on exertion	[ ]	[ ]
Coughing of blood	[ ]	[ ]
Dizzy spells	[ ]	[ ]
Frequent headaches	[ ]	[ ]
Frequent colds	[ ]	[ ]

Recurrent sore throat	[ ]	[ ]
Back pain	[ ]	[ ]
Aching or swollen joints	[ ]	[ ]
Orthopaedic problems	[ ]	[ ]
Unexplained weight loss (>5 lbs)	[ ]	[ ]
Any form of depressive illness	[ ]	[ ]

*Are you currently taking any prescription or non-prescription medications ?

Medication	Reason for taking	For how long?	Dosage

*Do you currently smoke ? Yes _____ No _____

If so, what ? _____ How much ? _____

Have you ever quit smoking ? _____ For how long did you smoke ? _____

*How much alcohol do you consume in one week ?

What type ?	Beer (pints)	Wine (glass)	Other
	_____	_____	_____

*How much caffeinated beverage do you consume per day ?

What type ?	Coffee (cups)	Tea (cups)	Other
	_____	_____	_____

*Do you take any dietary supplements (carbohydrate drinks, vitamins, creatine, others) ?

If so what ? _____ Dosage _____

*How would you describe your state of well-being at this time (please tick one) ?

Very, very good	[ ]	Poor	[ ]
Very good	[ ]	Very poor	[ ]
Good	[ ]	Very, very poor	[ ]
Neither good nor poor	[ ]		

Female Subjects Only:-	Yes	No
Are you currently pregnant ?	[ ]	[ ]
Are you trying to become pregnant ?	[ ]	[ ]
Have you recently given birth ?	[ ]	[ ]

If the answer is yes to any of the above please give details:-

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A.2. Consent Form

LOUGHBOROUGH UNIVERSITY, DEPARTMENT OF PHYSICAL EDUCATION, SPORTS SCIENCE AND RECREATION MANAGEMENT

High-Intensity Cycle Exercise Study

Title Of Study Inserted Here

Statement of Informed Consent:

Your permission to take part in this study is voluntary. You are free to deny consent or to withdraw from the study at any point and without explanation, if you so desire.

I have read the information regarding this study and had the opportunity to ask questions of the investigators. I understand the procedures involved and consent to participate in this study.

Signature of subject: Date:

Signature of witness:

APPENDIX B

Blood Assays

Haemoglobin (The Cyanmethaemoglobin Method)

Principle

Haemoglobin + Cyanide + Ferricyanide ———> Cyanmethaemoglobin

(Van Kampan and Zijlstra, 1961)

Reaction Mixture

The reaction mixture ('Drabkins') was made from a kit (Boehringer Mannheim) which contains phosphate buffer, potassium cyanide, potassium ferricyanide and detergent.

Procedure

1. In duplicate, 20 µl of whole blood was added to 5 ml of the reaction mixture and mixed well in a glass tube.
2. The tube was left to stand for at least 5 minutes; the exact time varied from one run to another.
3. The absorbance of the sample was measured using a spectrophotometer (Cecil Instruments) at a wavelength of 546 nm.
4. The relative absorbance (A) (absorbance of sample - absorbance of reaction mixture) was calculated. The haemoglobin concentration was calculated from the following equation:-

$$\text{haemoglobin concentration } (g \cdot 100ml^{-1}) = (37.2 \times A) + 0.06$$

(Wintrobe, 1956)

Heamatocrit (% Cell Volume)

Procedure

1. Three haematocrit tubes (Scientific Instruments) were approximately three-quarters filled with blood.
2. The tubes were sealed at one end with plasticine, placed in a microcentrifuge (Hawksley) and centrifuged for 14 minutes.
3. The percentage cell volume was calculated by placing each tube in a reader (Hawksley) where the bottom line was aligned with the base of the sample, the middle line intersected the top of the red cells and the top line intersected the top of the plasma.

Lactate

Principle:-

The fluorimetric assay for lactate is based upon that described by Maughan (1982).

$$\text{Lactate} \xrightarrow[\text{lactate dehydrogenase}]{\text{NAD------>NADH}} \text{Pyruvate}$$

The lactate concentrations reported in Chapters 3 were obtained using a non-automated analyser protocol whilst an automated analyser (Cobas) was employed in the study described in Chapter 5. Essentially, the two methods are the same but both procedures are explained in full.

Non-Automated Analyser Protocol

Reaction Mixture

For each ml of hydrazine buffer pH of approximately 9.0 the following were added:-

2.0 mg NAD
10.0 µl LDH (5500 $U \cdot ml^{-1}$)

Standards
In addition to a blank of 2.5 % perchloric acid, working standards of 1, 2.5, 5, 10, 15 and 20 mM were prepared from 1 M stock standard (Boehringer).

Procedure (each sample was analysed in duplicate)
1. After being allowed to thaw, samples were mixed (Whirlimix) and centrifuged for 3 minutes at 13000 rev·min^{-1} (Sanyo).
2. 20 µl of either standard or supernatant was pipetted into a glass fluorimeter tube and 200 µl of reaction mixture was added.
3. The tubes were mixed thoroughly (Whirlimix), covered, and left to incubate for approximately 30 minutes.
4. The fluorescence of the blanks, standards and samples were measured (Locarte).
5. A linear regression plot of the standards was made and the lactate concentration of the samples calculated.

<u>Automated Analyser Protocol</u>

Reaction Mixture
For each 1 ml of hydrazine buffer pH of approximately 9.0 the following was added:-

1.7 mg NAD
7 µl LDH (5500 U·ml^{-1})

Standards The Cobas automated analyser automatically blanks itself. The reagent tray can only accommodate three standards and standards with concentrations across appropriate ranges were used. In addition, a commercially available quality control (Sigma) at a concentration of 2.1 mM was run with each batch of samples. Both standards and quality control were diluted in 2.5 % perchloric acid in the same ratio as the samples.

Procedure (each sample was performed in duplicate)
1. The automated analyser (Cobas) power switch was depressed and the self-check was completed.
2. On the keyboard, the test code LACTATE was selected.
3. The standards, quality control and samples were mixed (Whirlimix) and centrifuged for 3 minutes at 13000 rev·min^{-1} (Sanyo).

4. The reagent tray was filled with standards (usually 5, 10 and 15 mM for studies involving a single bout of high-intensity exercise) and reaction mixture. The appropriate button was then depressed to run the assay. When the assay was complete, the values of the standards in units as well as the percentage of theoretical vs. actual was printed. The process was continued until satisfactory results (in the range 99-101 % for all standards) were obtained.
5. Approximately 100 µl of sample was placed into a one of the 25 cups comprising the sample disc. During each run, 100 µl of quality control was pipetted into two cups. When the sample disc was full the sample cups were pressed firmly into position and the disc placed on the turntable.
6. The reagent tray was re-filled with reaction mixture, if required, and the assay was run. The concentration of each sample and quality control was printed in mM.

Ammonia

Principle:
The spectrophotometric assay is based on a method described by Da Fonseca-Wollheim (1973).

$$\text{a-oxoglutarate} + NH_4^+ + \text{NADPH} \xrightarrow{\text{GLDH}} \text{L-glutamate} + NADP^+ + H_2O$$

Reaction Mixture
The reagents are contained in three bottles (numbers 1, 1a and 2) found in a commercially available kit (Boehringer Mannheim). Bottle 1 contains NADPH, bottle 1a buffer and bottle 2 GLDH. A reagent solution was made by adding 2.5 ml of bottle 1a to the contents of bottle 1. An enzyme solution was made by adding 0.5 ml of bottle 1a to the contents of bottle 2. This resulted in the following concentrations:-
Reagent solution
triethanolamine buffer at a pH of approximately 8.6 0.15 M
NADPH 0.12 mM
a-oxoglutarate 15 mM
ADP 1.5 mM
Enzyme solution
GLDH at an activity of not less than 755 $U \cdot ml^{-1}$

Standards

A set of Preciset® ammonia standards obtainable in kit form from Boehringer Mannheim was used which contained ammonia concentrations of 58.8, 117.6 and 176.5 µM.

Procedure (each sample was analysed in triplicate)

1. Once thawed, 100 µl of sample plasma or standard was pipetted into a 1.5 ml disposable plastic cuvette. Each cuvette was capped with a plastic tight fitting lid wrapped in Nescofilm®.
2. 500 µl of the reagent solution was added to the sample cuvette, agitated gently and the lid replaced. During each run, a blank comprising 600 µl of reagent was pipetted into a cuvette. The sample (or standard) and blank was left for 15 minutes.
3. The spectrophotometer (Cecil) was set to zero with the blank and the absorbance of the sample/standard recorded at 340 nm (A_1).
4. Using a positive displacement pipette, 4 µl of the enzyme solution was added to the sample/standard and blank cuvettes, agitated gently, left for 15 minutes and the absorbance read (A_2).
5. A further 4 µl aliquot of the enzyme solution was added to the cuvettes containing sample, blank and standards. The cuvettes were left for 15 minutes and the absorbance read again (A_3).
6. The ammonia concentration was calculated using the following equations:-

$$\Delta A = (A_1 - A_2) - (A_2 - A_3)$$

$$[\text{ammonia}]\ (\mu M) = 959 \times (\Delta A_{(\text{sample/standard})} - \Delta A_{(\text{blank})})$$

Glucose (Pilot Study In Chapter 4 Only)

Principle:
A colourimetric method based on the following principles (Werner *et al.*, 1970):

$$\text{Glucose} + O_2 + H_2O \xrightarrow{GOD} \text{Gluconate} + H_2O_2$$

$$H_2O_2 + \text{ABTS}^{®} \xrightarrow{POD} \text{Coloured complex} + H_2O$$

(ABTS® is a kit reagent)

Reaction Mixture
The reagents are contained in a kit (Boehringer Mannheim). The contents of a bottle (Catalogue number 124 036) was dissolved in 1000 ml of water. This resulted in a reaction mixture with the following reagent concentrations:-

phosphate buffer at a pH of approximately 7.0 100 mM
POD 0.8 $U \cdot ml^{-1}$
GOD 10 $U \cdot ml^{-1}$
ABTS® 1 $mg \cdot ml^{-1}$
(di-ammonium 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonate)

Standards
The kit also contains a glucose standard at a concentration of 0.505 mM

Procedure
1. Once thawed, the samples were mixed thoroughly (Whirlimix) and centrifuged at 13000 $rev \cdot min^{-1}$ (Eppendorf) for 5 minutes.
2. 20 µl of supernatant or standard was added to 1 ml of reaction mixture and left to incubate at room temperature for 20-30 minutes.
3. The absorbance (A) of the samples and standard was measured at a wavelength of 610 nm on a spectrophotometer (Cecil).
4. The glucose concentration (c) of each sample was calculated using the following equation:-

$$c = 5.55 \times (A_{sample}/A_{standard}) \ (mM)$$

APPENDIX C

Muscle Assays

Full details of the recipes for the reagents used in the muscle assays can be found in Appendix D of G. Bogdanis' thesis (1994). This Appendix describes the spectrophotometric procedures used to validate standards and fluorometric methods to analyse samples. They are amended versions of those also to be found in Appendix D of the aforementioned thesis.

AC.1. Spectrophotometric Assays Used To Validate Standards

Most enzymatic reactions are linked to the interconversion of NAD/NADH or NADP/NADPH. Reduction of adenine nucleotides is accompanied by an increase in absorbance at 340 nm. Beer's Law can be written as:-

$$A=E.b.c \qquad \text{(Equation: AC.1)}$$

Where A is absorbance, E is the molar extinction coefficient, b is the pathlength and c is concentration. Knowing that the milli-molar extinction coefficient of NAD/NADPH is 6.22 mol^{-1} cm^{-1} at 340 nm and by keeping the pathlength fixed at 1 cm, the concentration of a substance will be proportional to the change in absorbance and can be thus calculated without the need for external standards. Assays to validate standards were performed in plastic semi-micro cuvettes and the concentration of the standard calculated by rearranging Beer's Law and modifying. Rearranging Beer's Law for concentration gives c=A/(E.b) but b can be ignored as the cuvettes used are 1 cm, so c=A/E. During the spectrophotometric validation assays, the change (delta) in absorbance is recorded as the contents of the cuvette was increased by adding standard and then enzyme to 1 ml of reaction mixture. Thus, the rearranged equation can be modified to:-

$$[\text{standard}]\ \text{mM} = \frac{\text{final cuvette volume (ml)} \times \Delta A}{\text{volume of standard added (ml)} \times 6.22}$$

(Equation: AC.2)

Spectrophotometric assays were used to validate standards for the muscle assays for lactate, ATP, PCr and Cr. For the remaining muscle assay, pyruvate, reference was made to external standards prepared by chemical manufacturers. All enzymes which are marked * were diluted with tris-HCl 20 mM, pH of approximately 8.1 with 0.02 % bovine serum albumin (BSA); where the asterisk is omitted the enzyme was un-diluted.

Adenosine Triphosphate (ATP)

Principle:-

$$ATP + glucose \xrightarrow{hexokinase} ADP + G\text{-}6\text{-}P$$

$$G\text{-}6\text{-}P \xrightarrow{NADP \text{ ----> } NADPH/G6P\text{-}DH} 6\text{-}P\text{-}gluconolacton$$

Reaction Mixture

For each 1 ml of Tris-HCl 50 mM buffer, at a pH of approximately 8.1 with added bovine serum albumin (BSA), the following reagents were added:-

15 µl NADP 50 mM	10 µl ADP 10 mM
10 µl glucose 100 mM	10 µl Dithiothreitol (DTT) 50 mM
50 µl $MgCl_2$ 100 mM	15 µl G6P-DH* 14 $U \cdot ml^{-1}$

Procedure

1. 1 ml of reaction mixture was added to 20 µl of 2mM ATP standard in a plastic 1 cm cuvette and the absorbance read.
2. 10 µl of HK* (28 $U \cdot ml^{-1}$) was added to the cuvette, the contents of which were mixed well, and the reaction followed to completion (approximately 10 minutes). The absorbance reading at completion was recorded.
3. A further 10 µl aliquot of HK* was added to the contents of the cuvette and the absorbance was read for the third time after thorough mixing.
4. The ΔA value was calculated and substituted into the rearranged version of Beer's Law (Equation AC.2).

Phosphocreatine (PCr).

Principle:

$$PCr + ADP \xrightarrow{\text{creatine kinase}} Cr + ATP$$

$$ATP + \text{glucose} \xrightarrow{\text{hexokinase}} ADP + \text{G-6-P}$$

$$\text{G-6-P} \xrightarrow{NADP \rightarrow NADPH/\text{G6P-DH}} \text{6-P-gluconolactone}$$

Reaction Mixture

For each 1 ml of Tris-HCl 50 mM buffer, at a pH of approximately 8.1 with added bovine serum albumin (BSA), the following reagents were added:-

15 µl NADP 50 mM
10 µl glucose 100 mM
50 µl $MgCl_2$ 100 mM
15 µl HK* 28 $U \cdot ml^{-1}$
10 µl ADP 50 mM
10 µl Dithiothreitol (DTT) 50 mM
15 µl G6P-DH* 14 $U \cdot ml^{-1}$

Procedure

1. 1 ml of reaction mixture was added to 20 µl of 2mM PCr standard in a plastic 1 cm cuvette and the absorbance read.
2. 15 µl of CK* (1290 $U \cdot ml^{-1}$) was added to the cuvette, the contents of which were mixed well, and the reaction followed to completion (approximately 10 minutes). The absorbance reading at completion was recorded.
3. A further 15 µl aliquot of CK* was added to the contents of the cuvette and the absorbance was read for the third time after thorough mixing.
4. The ΔA value was calculated and substituted into the rearranged version of Beer's Law (Equation AC.2).

Creatine (Cr)

Principle:

$$Cr + ATP \xrightarrow{\text{creatine kinase}} Cr + ADP$$

$$ADP + \text{P-pyruvate} \xrightarrow{\text{pyruvate kinase}} ATP + \text{Pyruvate}$$

$$\text{Pyruvate} \xrightarrow{\text{NADH ---> NAD/lactate dehydrogenase}} \text{Lactate}$$

Reaction Mixture

For each 1 ml of Imidazole-HCl 50 mM buffer at pH of approximately 7.5 the following reagents were added:-

20 µl NADH 5 mM	1 µl LDH* 240 $U \cdot ml^{-1}$
10 µl ATP 100 mM	7 µl phosphoenol pyruvate (PEP) 30 mM
50 µl $MgCl_2$ 100 mM	10 µl KCl 3 M
12.5 µl PK* 75 $U \cdot ml^{-1}$	

Procedure

1. 1 ml of reaction mixture was added to 20 µl of 2mM Cr standard in a plastic 1 cm cuvette and the absorbance read.
2. 15 µl of CK* (1290 $U \cdot ml^{-1}$) was added to the cuvette, the contents of which were mixed well, and the reaction followed to completion (approximately 10 minutes). The absorbance reading at completion was recorded.
3. A further 15 µl aliquot of CK* was added to the contents of the cuvette and the absorbance was read for the third time after thorough mixing.
4. The ΔA value was calculated and substituted into the rearranged version of Beer's Law (Equation AC.2).

Lactate

Principle:

Lactate $\xrightarrow{\text{NAD-->NADH/Lactate dehydrogenase}}$ Pyruvate

Reaction Mixture

For each 1 ml of hydrazine 1.1 M buffer at a pH of approximately 9.0 with 1 M EDTA·Na_2, the following was added:-

40 µl NAD 50 mM

Procedure

1. 1 ml of reaction mixture was added to 20 µl of 2mM Cr standard in a plastic 1 cm cuvette and the absorbance read.
2. 5 µl of LDH (5500 $U \cdot ml^{-1}$) was added to the cuvette, the contents of which were mixed well, and the reaction followed to completion (approximately 20 minutes). The absorbance reading at completion was recorded.
3. A further 5 µl aliquot of LDH was added to the contents of the cuvette and the absorbance was read for the third time after thorough mixing.
4. The ΔA value was calculated and substituted into the rearranged version of Beer's Law (Equation AC.2).

AC.2. Mixed Muscle Metabolite Fluorometric Assays

The mixed muscle metabolite assays are based on methods described by Lowry and Passoneau (1973) and Harris *et al.* (1975). In order to make the assays run in the muscle metabolism laboratory at Loughborough University, a number of minor adaptations have been made. The revised methods were described in full recently (Bogdanis, 1994) but some further adjustments in the concentrations of certain reagents were required to obtain the muscle metabolite data in Chapter 5. Therefore, the assays are again described in full. In order to obtain satisfactory results, assays were firstly run on rat and unwanted human muscle. The coefficients of variation for each assay can be found in Chapter 3.

Adenosine Triphosphate (ATP) and Phosphocreatine (PCr).

Principle:

The phosphorylation of ADP by the creatine kinase reaction results in the formation of an equivalent amount of ATP (Reaction 1). The reaction of ATP with glucose in the presence of hexokinase produces equimolar amounts of ADP and glucose-6-phosphate (Reaction 2). The change in fluorescence as NADP is reduced to NADPH (Reaction 3) is thus proportional to [PCr] in the presence of CK and HK and to [ATP] where the CK is omitted.

Reaction 1. PCr + ADP $\xrightarrow{\text{creatine kinase}}$ Cr + ATP

Reaction 2. ATP + glucose $\xrightarrow{\text{hexokinase}}$ ADP + G-6-P

Reaction 3. G-6-P $\xrightarrow{\text{NADP---> NADPH/ G6P-DH}}$ 6-P-gluconolactone

Reaction Mixture

For each 1 ml of Tris-HCl 50 mM buffer, at a pH of approximately 8.1 with added bovine serum albumin (BSA), the following reagents were added:-

15 µl NADP 5 mM	10 µl ADP 10 mM
10 µl glucose 10 mM	10 µl Dithiothreitol (DTT) 50 mM
50 µl $MgCl_2$ 100 mM	10 µl G6P-DH* 14 $U \cdot ml^{-1}$

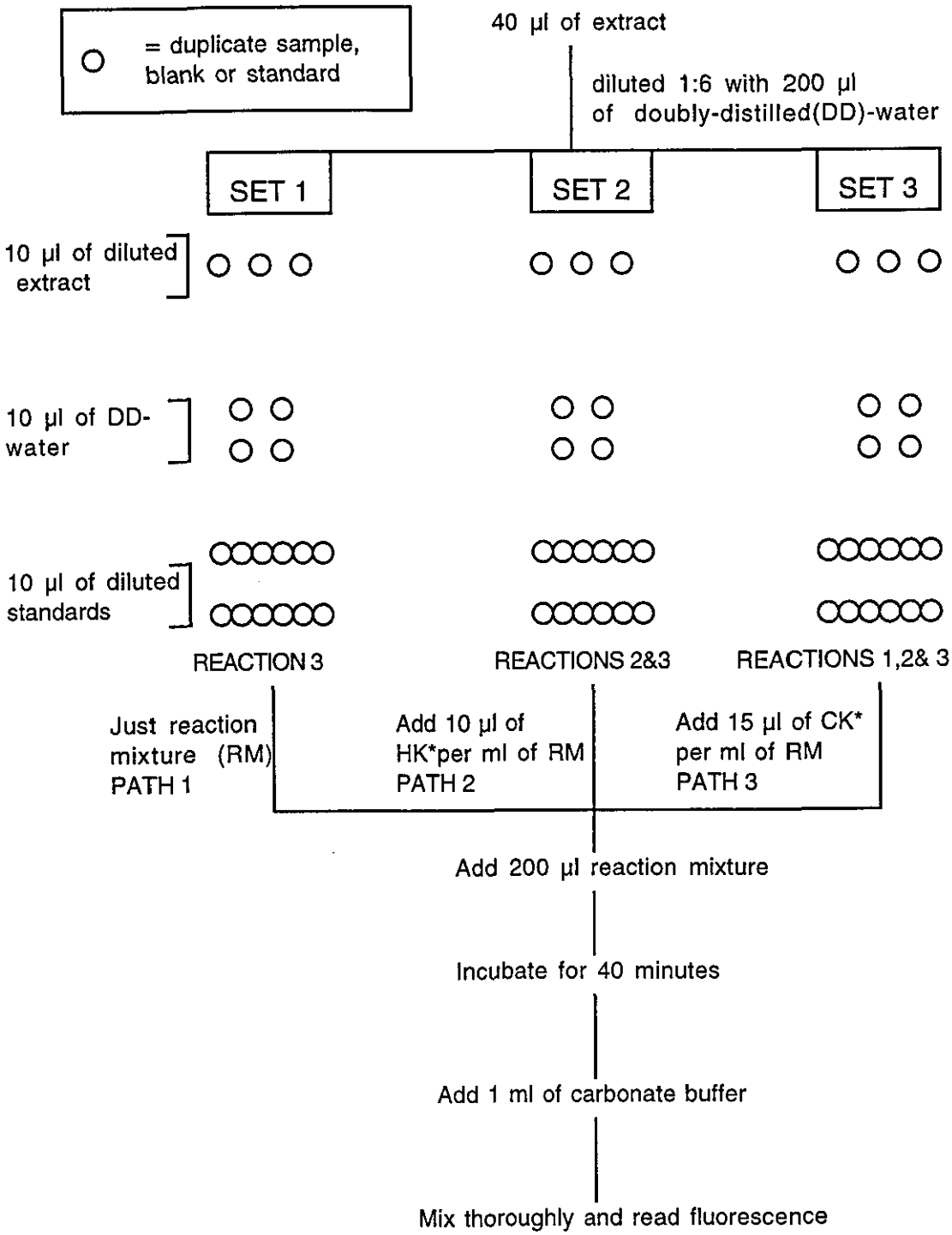
Standards

In addition to blanks containing doubly-distilled water, working standards were prepared with [ATP] of approximately 25, 50, 100, 150 and 200 µM and a [PCr] of approximately 200 µM from stock standards (2 mM for both ATP and PCr).

Procedure

The reaction mixture was prepared immediately prior to analysis for 3 sets of duplicate samples, quadruplicate double distilled water blanks and a set of quadruplicate standards. The procedure is outlined in figure AC1.

Figure AC.1:PCr and ATP



For ATP determination subtract the relative fluorescence of PATH 1 from PATH 2.

For PCr determination subtract the relative fluorescence of PATH 2 from PATH 3.

The concentration was determined from linear regression of the standards then correcting for dilutions during the extraction procedure and the dilution during the assay.

Creatine (Cr)

Principle:

An equimolar amount of pyruvate can be formed from creatine by two reactions catalysed by creatine kinase (Reaction 1) and pyruvate kinase (Reaction 2). When pyruvate is converted to lactate, there is a change in fluorescence as NADH is oxidised to NAD (Reaction 3).

Reaction 1. Cr + ATP $\xrightarrow{\text{creatine kinase}}$ PCr + ADP

Reaction 2. ADP + P-pyruvate $\xrightarrow{\text{pyruvate kinase}}$ ATP + Pyruvate

Reaction 3. Pyruvate $\xrightarrow{\text{NADH---> NAD/lactate dehydrogenase}}$ Lactate

Reaction Mixture

For each 1 ml of Imidazole-HCl 50 mM buffer at pH of approximately 7.5, the following reagents were added:-

60 µl NADH 1 mM	3 µl LDH* 240 $U \cdot ml^{-1}$
20 µl ATP 10 mM	25 µl phosphoenol pyruvate (PEP) 2 mM
50 µl $MgCl_2$ 100 mM	10 µl KCl 3 M
1 µl EDTA·Na_2 100 mM	15 µl PK* 75 $U \cdot ml^{-1}$

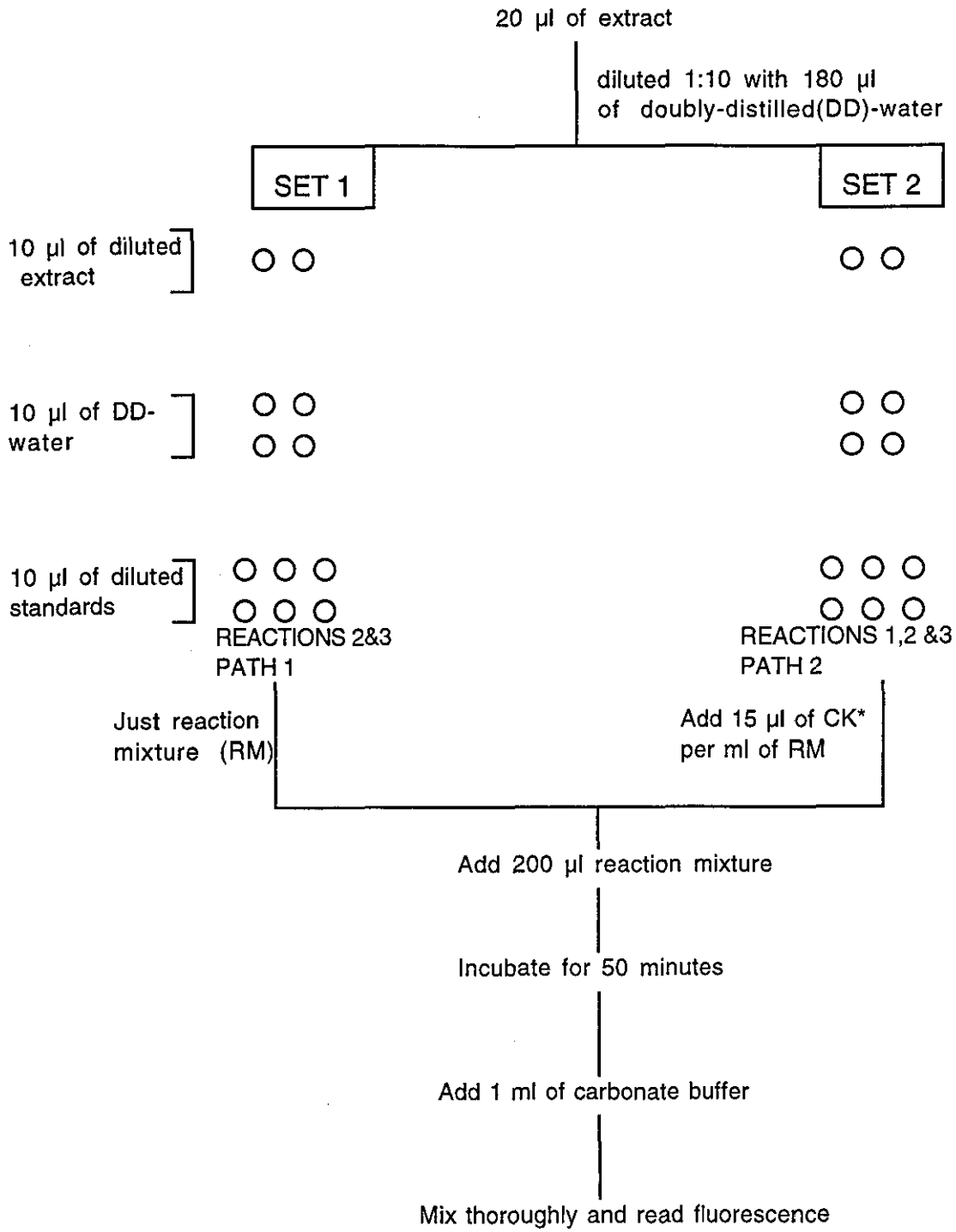
Standards

In addition to blanks containing doubly-distilled water, working standards with [Cr] of approximately 50, 100 and 150 µM were made from 2 mM stock standard.

Procedure

The reaction mixture was prepared immediately prior to analysis for 2 sets of duplicate samples, quadruplicate double distilled water blanks and a set of quadruplicate standards. The procedure is outlined in figure AC.2.

Figure AC.2: Creatine (Cr)



For Cr determination subtract the relative fluorescence of PATH 1 from PATH 2.

The creatine concentration was calculated from linear regression of the standards and were corrected for dilutions during the extraction procedure and the dilution during the assay

Pyruvate

Principle:
When pyruvate is converted to lactate, there is a change in fluorescence as NADH is oxidised to NAD.

Pyruvate $\xrightarrow{\text{NADH--> NAD/lactate dehydrogenase}}$ Lactate

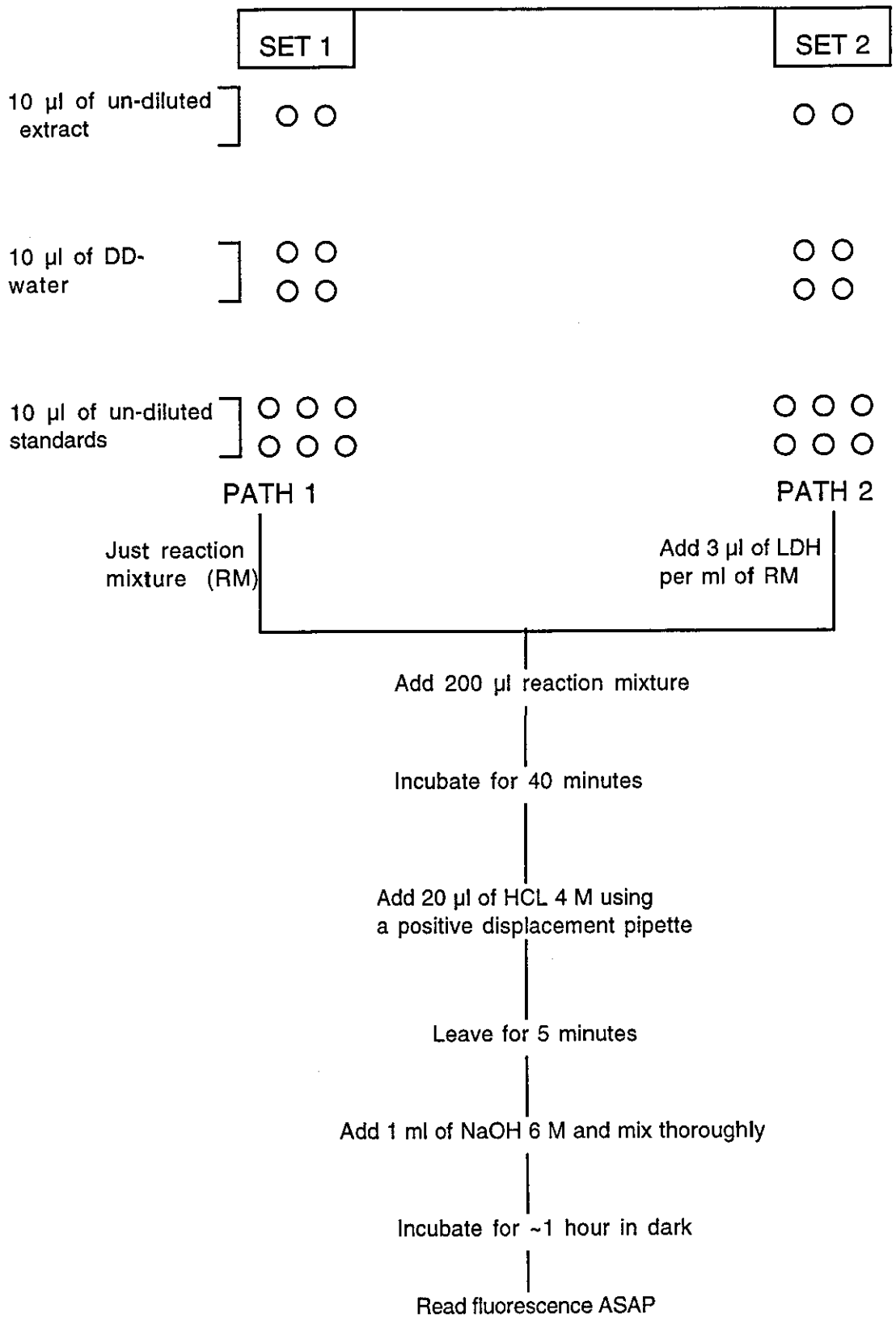
Reaction Mixture
For each 1 ml of Imidazole-HCl 50 mM buffer at pH of approximately 7.0, the following was added:-

3 µl NADH 5 mM
10 µl EDTA·Na_2 100 mM

Standards
In addition to blanks containing doubly-distilled water, working standards of [Pyruvate] approximately 10, 30, 50 and 100 µM were made from 2 mM stock standard.

Procedure
The reaction mixture was prepared immediately prior to analysis for 2 sets of duplicate samples, quadruplicate double distilled water blanks and a set of quadruplicate standards. The procedure is outlined in figure AC.3.

Figure AC.3:Pyruvate



For pyruvate determination the relative fluorescence of the blanks, PATH 1, was subtracted from PATH 2

Lactate

Principle:

As lactate is converted to pyruvate, there is a change in fluorescence as NAD is reduced to NADH.

$$\text{Lactate} \xrightarrow{\text{NAD---> NADH/lactate dehydrogenase}} \text{Pyruvate}$$

Reaction Mixture

For each 1 ml of hydrazine 1.1 M buffer at pH of approximately 9.0 with 1 M EDTA·Na_2 the following was added:-

10 µl NAD 50 mM
10 µl LDH 5500 $U \cdot ml^{-1}$

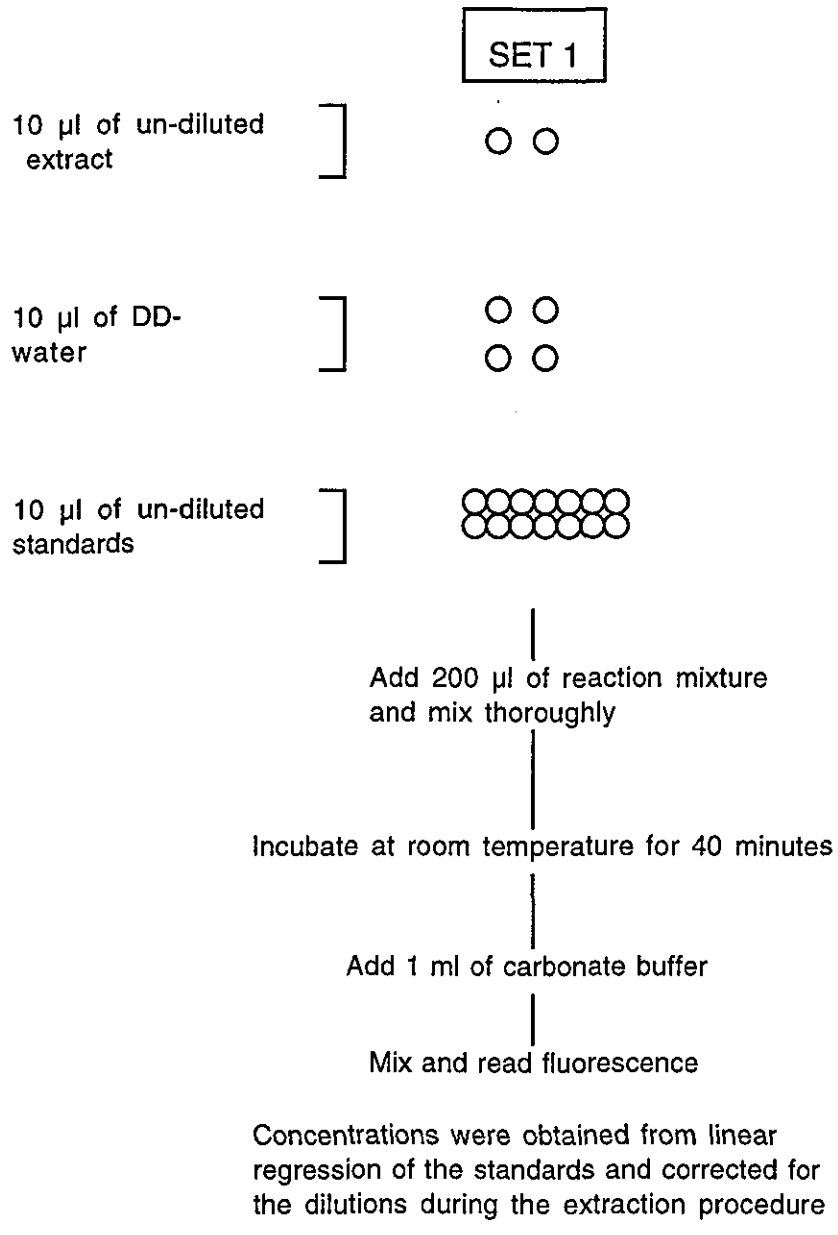
Standards

In addition to blanks containing doubly-distilled water, working standards with [Lactate] of approximately 50, 75, 100, 150, 400, 750 and 1200 µM were made from 2 mM stock standard.

Procedure

The reaction mixture was prepared immediately prior to analysis for 2 sets of duplicate samples, quadruplicate double distilled water blanks and a set of quadruplicate standards. The procedure is outlined in figure AC.4.

Figure AC.4:Lactate



Concentrations were obtained from linear regression of the standards and corrected for the dilutions during the extraction procedure

APPENDIX D

Terms, Definitions And Formulae

The Twentieth Century has seen the rapid expansion of scientific research and the introduction of a wide range of methodologies. No longer is research the preserve of the individual but is practised primarily in educational establishments, corporations and governments, by research groups, where substantial funding is frequently available. Moreover, the growth of communications has meant that scientific work can now be relayed world-wide. As a consequence, standardisation of terms and definitions has been required. Unfortunately, this process has taken time and is still not complete. By way of example, atmospheric pressure has been quoted at various times in terms of millimetres of mercury, Pascal's, atmospheres and the bar. The result is that papers contain a plethora of terms and definitions, adopting the fashion prevailing at the time of writing.

Below are the current Système International d'Unités (SI) units, as at 1995, for a variety of measurements, together with an brief explanation of a number of terms used frequently in this project:-

Current SI Units

Mass	kilogram	(kg)
Distance	metre	(m)
Time	second	(s)
Force	newton	(N)
Work	joule	(J)
Power	watt	(W)
Velocity	metres per second	$(m \cdot s^{-1})$
Torque	newton metre	$(N \cdot m)$
Acceleration	metres per second^2	$(m \cdot s^{-2})$
Angle	radian	(rad)
Angular velocity	radians per second	$(rad \cdot s^{-1})$
Volume	litre	(l)
Stiffness	newton metre	$(N \cdot m^{-1})$
Compliance	metre per newton	$(m \cdot N^{-1})$

Action potential: A brief, all-or-none reversal of the membrane potential polarity.

Depolarization: When the membrane potential becomes less negative (or reverses polarity and becomes positive) than the resting membrane potential.

Energy:The capability of producing force, performing work or generating heat. The current SI unit is, like for work, the joule, which is equal to 0.238846 cal

Fatigue: The definition of fatigue commonly adopted by physiologists is that of "a reduction in the required or expected force" (Edwards, 1981). With regard to sporting arena, the athlete is concerned with the maintenance of as high an exercise intensity as possible and with this in mind the above definition was extended to include "failure to maintain the required or expected power output" (Edwards, 1983).

Force:Application of a force will alter, or at least tend to alter, the state of rest or motion in matter. It is the product of mass multiplied by acceleration. Frequently, it is gravity that provides the acceleration and where this is the case, is regarded as 9.81 $m \cdot s^{-2}$. The current SI unit of force is the newton although other terms have been frequently used by physiologists and the conversion factors are:-

$$1N=7.233 \text{ poundal}=10^5 \text{ dyne}$$

Hyperpolarization: When the membrane potential becomes more negative than the resting membrane potential.

Load:The resistance opposing tension generated within a muscle.

Mean power output (MPO): is the average power output per second achieved during the work period.

Membrane potential: The voltage difference between the inside and outside of a cell.

Peak power output (PPO): is the highest power output achieved during one second of the work period.

Pennation: In many muscles the fibres are not aligned with the long axis of the muscle. Instead, they run obliquely between the broad intramuscular expanses of the tendon. The obliquity of the muscle fibres is called the angle of pennation. Pennation is significant because it attenuates the transmission of muscle forces to tendons. Also, pennation determines the magnitude of the changes in fibre length which accompany changes in muscle length and therefore the force which the muscle fibres can produce at any given muscle length or velocity.

Power:Power is representative of the rate of doing work and is the product of force multiplied by velocity. Currently the SI unit is the watt.

In order to clarify the distinction between work and power a useful analogy is to consider a journey made frequently such as from home to work. When in good time a gentle walk may suffice but when time is short a jog may be required for part or all the way. In each case, the total work is the same as the distance from home to work and body mass is the same each day. However, the journey time is shorter when jogging so the rate of performing work is faster and hence power is greater.

Resting membrane potential: The membrane potential of a cell not producing an electric signal

Tension:The force exerted by muscle on a load. The terms force and tension when referring to muscle are used interchangeably

Torque:The effectiveness of a force to overcome the rotational inertia of an object. It is the product of force and the perpendicular distance from the line of action of the force to the axis of rotation. As there is a rotational element to the definition the force applied to the cranks of a cycle is referred to as torque. The units, are N·m.

Work:Work is achieved when a force moves through a distance. The current SI unit is the joule. One joule is equivalent to the product of one newton multiplied by one metre. (i.e. 1N·m = 1J)

Current SI Prefixes

Prefix	Value	Symbol
giga-	10^9	G

mega-	10^{6}	M
kilo-	10^{3}	k
deci-	10^{-1}	d
centi-	10^{-2}	c
milli-	10^{-3}	m
micro-	10^{-6}	μ
nano-	10^{-9}	n
pico-	10^{-12}	p