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METABOLIC CORRELATES OF FATIGUE FROM DIFFERENT TYPES OF EXERCISE IN MAN

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ABSTRACT

It is well established that muscle fatigue, defined as a decline in maximal force generating capacity, is a common response to muscular activity. To what extent metabolic factors contribute to the reduced muscle function is still debated. Metabolic effects can affect muscle through different processes, either through a reduced ATP supply or by effects on EC-coupling or crossbridge dynamics. Observations from in vitro experiments are often extrapolated to interpret fatigue mechanisms from measurements performed in vivo, without recognizing that the biochemical reactions involved can be quite different depending upon such factors as activation pattern, mode and duration of exercise. During repeated submaximal contractions, there is a negligible accumulation of H^+ and inorganic phosphate, and hence fatigue must be ascribed to other factors. Substrate depletion might contribute to exhaustion, but cannot explain the gradual loss of maximal force. Curiously, the energetic cost of contraction increases progressively during repeated isometric but not during concentric contractions. With contractions involving high-force or high power output, fatigue is better related to $H_2PO_4^-$ than to pH, but still other factors seem to play a role.

INTRODUCTION

This chapter describes the metabolic responses during different types of exercise and relates them to fatigue. Metabolic and biochemical aspects of fatigue involve energy release and utilization, and the consequences of substrate degradation and electrolyte shifts. While all of these factors may be important in fatigue, only the first two factors will be examined in this chapter. The effects of pH and electrolyte balance are covered elsewhere (Allen et al., Chapter 3; Sjøgaard & McComas, Chapter 4).

To generate force and power, ATP is hydrolyzed by myosin ATPase. In addition, ATP is utilized in the re-establishment of electrolyte homeostasis after ion fluxes associated with propagation of the action potential and Ca^{2+} release from sarcoplasmic reticulum. Adequate resynthesis of ATP, therefore, is important in the maintenance of cellular function. During

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prolonged exercise, oxidative phosphorylation of carbohydrates and free fatty acids are the main sources for energy. Although the products of this process are not known to affect force, the availability of substrates or the deterioration of mitochondrial function may contribute to muscle fatigue. With an inadequate oxygen supply, such as during sustained high-force contractions, glycogen and phosphocreatine (PCr) are rapidly broken down, and lactic acid and inorganic phosphate (Pi) accumulate. The possible role of these metabolites and substrate supply in the development of fatigue is still debated, and I will review some of the available data with emphasis upon how they relate to fatigue from voluntary activation in humans.

FATIGUE AND EXHAUSTION

The term *muscle fatigue* is used to denote a decline in the maximal contractile force of the muscle. Defined this way, fatigue may be assessed from brief maximal voluntary contractions or tetanically stimulated contractions. Most importantly, fatigue may occur during exercise at submaximal levels without being reflected in the performance. Fatigue is different to exhaustion, where the latter is defined as the moment in time when the expected force level cannot be maintained.

Muscle fatigue can occur with all types of muscular activity (i.e., isometric, shortening and lengthening contractions), and with both short-lasting intense and prolonged exercise. The various modes and intensities of exercise involve different metabolic processes, and the importance of biochemical changes to the decline in force may vary accordingly. To illustrate these possibilities, this chapter focuses on metabolic responses to repeated submaximal contractions and to exercise involving high-intensity force or power. These examples indicate that it is difficult to extrapolate the biochemical changes seen under one condition to other exercise protocols.

ANALYTICAL TECHNIQUES

Several different analytical methods have been used to study the effects of biochemical factors on performance. While some early data were obtained by manipulating substrate levels or metabolic pathways (Lundsgaard, 1930), these approaches were limited and initial biochemical analysis could only be carried out on stimulated animal muscles. With the introduction of needle biopsies in the sixties (Bergström, 1962), a more detailed picture of the metabolic changes in human muscle during various types of exercise was obtained. Substrate degradation and accumulation of metabolic products could readily be determined. The biopsy technique, however, has several limitations. First, only a limited number of samples can be obtained from each muscle. Second, the time resolution is poor. And thirdly, there is evidence that biochemical processes occur in the muscle when the sample is cut (Söderlund & Hultman, 1986). Hence, interpretation of small metabolic changes measured with the biopsy technique requires caution.

These disadvantages are not present when metabolites are assessed using ^{31}P nuclear magnetic resonance spectroscopy (^{31}P-MRS). Relative changes in PCr, Pi and ATP can be determined from the spectra, and from these ADP, $H_2PO_4^-$ and pH can be calculated (see Bertocci, Chapter 15). This nonivasive technique is based on phosphorous spectra recorded from muscles by surface coils and usable spectra may be obtained in a few seconds (Degroot et al., 1993). The greatest disadvantage of this method is that it has been applied almost exclusively to the study of phosphorous compounds in relation to fatigue. In the future, further developments in MRS may expand to analyze other aspects of metabolism. Another disadvantage of MRS is that it restricts the experimental design to the performance of a task

within a strong magnetic field. Hence, the exercise-induced changes in metabolites have mainly been studied during isometric contractions, or dynamic contractions of smaller muscle groups. A third disadvantage of the ^{31}P-MRS is that it is not possible to calibrate the quantification of metabolites determined from the recorded spectra. To compensate for this, high-energy phosphates are most often expressed in relation to total phosphate. A fourth disadvantage is that the observed changes is concentrations will always be an average of the muscle fibers within the field of view. Details about cellular changes cannot be obtained when only a small portion of the muscle fibers are active.

In a recent study, Bangsbo and coworkers (1993) compared changes in metabolites during fatigue in parallel studies using ^{31}P-MRS and biochemical analyses of muscle biopsies. Similar changes in PCr were observed with the two methods, but a somewhat larger decline in ATP was determined biochemically after high-force contractions. The increase in ADP was much more pronounced when determined by ^{31}P-MRS than biochemically. This discrepancy was caused by the fact that the biochemical analysis measures total ADP whereas free ADP is determined from ^{31}P-MRS spectra. Hence, changes in PCr and for most purposes ATP can be reliably assessed with either method. However, only ^{31}P-MRS is sensitive for changes in free ADP.

The knowledge gained over the last three decades using different experimental models and analytical techniques has shown that the biochemical changes associated with fatigue vary over a wide range. Most investigators concur that several different mechanisms may play a role depending on the type and intensity of exercise and the fiber-type composition of the involved muscles.

PROLONGED EXERCISE - REPETITIVE SHORTENING OR ISOMETRIC CONTRACTIONS

During prolonged repeated submaximal contractions, the maximal force generating capacity declines continually (Bigland-Ritchie et al., 1986b; Vøllestad et al., 1988). At the onset of submaximal exercise only a fraction of the muscle fibers in the working muscles are activated. This has been shown by biopsy studies from several laboratories (Gollnick et al., 1973; Gollnick et al., 1974; Vøllestad et al., 1984; Vøllestad & Blom, 1985; Bigland-Ritchie et al., 1986a). When performing histochemical staining for the glycogen concentration in the fibers, it has been shown that only type I fibers are recruited from onset of bicycle exercise at about 40% of the maximal oxygen uptake (VO_2max) (Vøllestad & Blom, 1985). With increasing intensity, a larger fraction of the muscle fibers are recruited at the beginning. Interestingly, we observed a linear rise in the number of active muscle fibers as the exercise intensity increased (Vøllestad & Blom, 1985). If submaximal exercise is prolonged for an hour or more, exhaustion of the glycogen stores may occur in some fibers (Gollnick et al., 1973; Vøllestad et al., 1984). Due to the lack of glycogen, these fibers will therefore have a decreased capacity to release ATP. As exercise approaches exhaustion, an increasing number of fibers show total glycogen depletion.

One appealing hypothesis is thus that exhaustion is caused by an insufficient rate of ATP release in the muscle fibers. Several lines of evidence, however, argue against such a causative relationship. Maximal force generating capacity is determined by brief MVCs or tetanically stimulated contractions. The ATP hydrolysis in these contractions may amount to 1-2 mmol/kg ww (Edwards, 1976; Katz et al., 1986), indicating that only a minor fraction of the PCr and ATP stores are utilized under the test contractions. In keeping with this, several reports based on ^{31}P-MRS show that only small, if any, changes occur in ATP during short lasting high-force contractions (Miller et al., 1988; Quistorff et al., 1992). It might be argued

that the fall in ATP concentration is much larger close to the crossbridges than in other parts of the cell. Hence, the ATP concentration determined as an average for the cell or muscle does not reflect the true conditions at the force-generating sites. However, as recently reviewed by Fitts (1994), it is unlikely that even in localized parts of the cell the ATP concentration should drop to levels below 50 μM which is shown to be sufficient for maximal isometric tension in rabbit psoas fibers (Cooke & Bialek, 1979). It may thus be concluded that ATP availability is not the main cause of fatigue under these conditions.

It is well established that lowered pH or elevated concentrations of inorganic phosphate (Pi) reduces the force generated by the crossbridges (Cooke & Pate, 1985; Donaldson & Hermansen, 1978). The effects of these factors during prolonged submaximal exercise are probably marginal because only a small fraction of the energy is released by anaerobic pathways. Hence, the accumulation of lactic acid or Pi is less than that required to affect force generation significantly (Vøllestad et al., 1988).

More recently, different research groups have studied fatigue using repeated submaximal isometric contractions. In several series of experiments, we have examined changes during quadriceps contractions held at 30% MVC for 6 s with 4 s rest between. The MVC force fell gradually during the entire exercise period reaching 40-60% of control at exhaustion, whereas serial muscle biopsies revealed only marginal changes in muscle glycogen, PCr, ATP and lactic acid for the first 30 min (Vøllestad et al., 1988). At exhaustion, however, almost total depletion of the PCr store was observed, while glycogen was reduced by about 30%. These observations indicate that also with repetitive isometric contractions the gradual decline in maximal force generating capacity during repeated shortening or isometric contractions must be explained by other factors than those connected to substrate depletion or accumulation of Pi and H^+.

Further evidence for the lack of a uniform relationship between fatigue and metabolite changes have been provided by experiments using ^{31}P-MRS. Fig. 1 shows results obtained from two subjects during repeated isometric contractions at 40% MVC with the quadriceps muscle. In one subject (dotted line), PCr fell by almost 90% within 10 min from the start of exercise, which was continued for more than 20 min with depleted PCr stores. Even though he was totally depleted of PCr, only marginal changes were seen in pH. In the other subject (solid line), PCr fell continually until exhaustion, and no changes were seen in pH. Fatigue developed gradually and by an equal amount for the two subjects. In addition to emphasizing that fatigue can develop without large changes in PCr, Pi and pH, these results show that constant force contractions can be carried out for a long time almost in the absence of PCr. This is at variance with our earlier data from repeated isometric contractions, in which exhaustion seemed to be clearly related to PCr depletion (Vøllestad et al., 1988). An example of the metabolic changes in one of these subjects is given in Fig. 2, illustrating that PCr was maintained at almost resting levels for 101 min, but totally depleted at exhaustion which was reached after 104 min. Also ATP fell rapidly by 2.3 mmol/kg ww in the last 3 min of exercise. The discrepancy between the data obtained in the two series of experiments is not understood, but could be related to differences in the angle of the knee (and thus muscle length) or to circulatory effects due to differences in body position (supine vs. sitting).

Interestingly, there is one distinct difference in the metabolic response to cycling or running compared with repeated isometric contractions. When this type of dynamic exercise is kept at a constant intensity, the oxygen consumption (VO_2) rises initially, and reaches a new steady level within a minute or two. Hence, the energy cost of contraction appears to remain constant with time. In contrast to this, repeated submaximal isometric contractions at 30% MVC with the quadriceps muscles results in an initial rise in VO_2 followed by a continual further rise until exhaustion (Vøllestad et al., 1990). Similar results have been reported by Sahlin and coworkers (1992), who also showed that the oxygen cost of isometric contractions remained elevated for an hour after end of exercise.

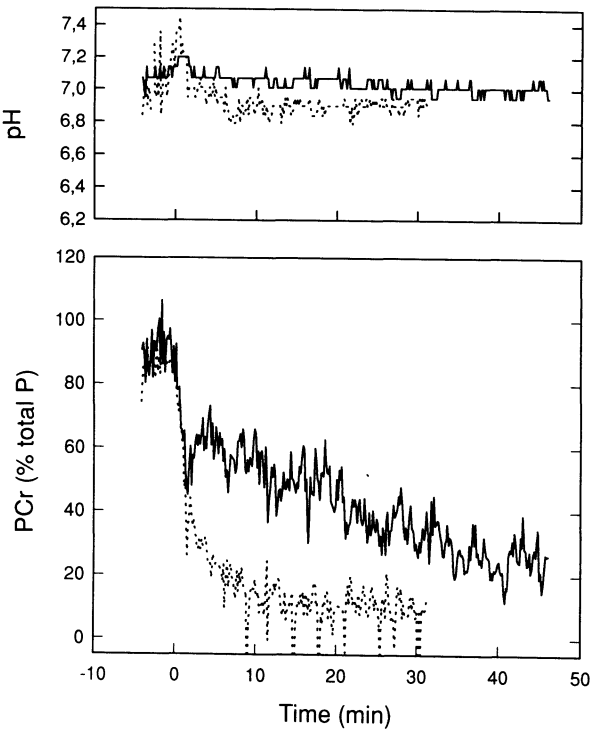

Figure 1. Changes in pH and PCr determined in the quadriceps muscle every 9 s by ^{31}P-MRS during intermittent isometric contractions at 40% MVC. The exercise was carried out with the subjects in a supine position and with each contraction held for 6 s with 4 s rest between. Data for two subjects shown by dotted and solid lines. Some of the deflections are due to maximal test contractions carried out regularly.

The causes for the gradual rise in VO_2 during repeated isometric contractions are not clarified. There are two alternative explanations. Either the ATP demand (ATP turnover:force ratio) increases, or the mitochondrial ATP production becomes less efficient (decreased P:O ratio). The latter mechanism would predict that oxygen consumption during any type of exercise involving the fatigued muscles would be elevated. Sahlin and coworkers (1992) tested this by comparing the VO_2 during bicycle exercise before and after fatigue from repeated isometric contractions. They observed no change in VO_2 during bicycling, indicating that the mitochondrial respiration was not altered. The most probable explanation for the increased VO_2 thus appears to be that the ATP turnover:force ratio increases. Two lines of

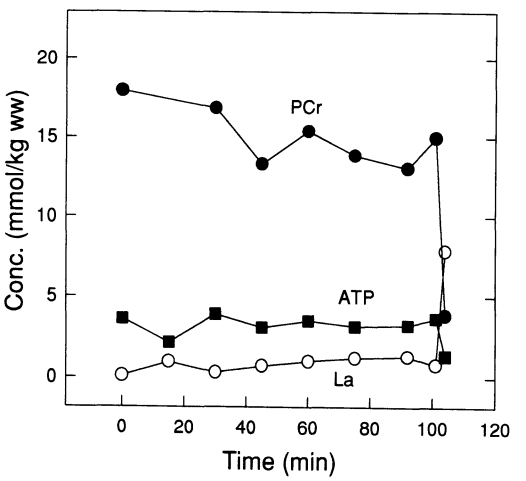

Figure 2. Changes in muscle concentrations of PCr, ATP and lactate (La) determined from biopsies taken from the quadriceps muscle of one subject. Intermittent isometric knee extension contractions at 30% MVC were carried out until exhaustion (104 min). The subject was seated in a chair and each contraction lasted 6 s with 4 s rest between.

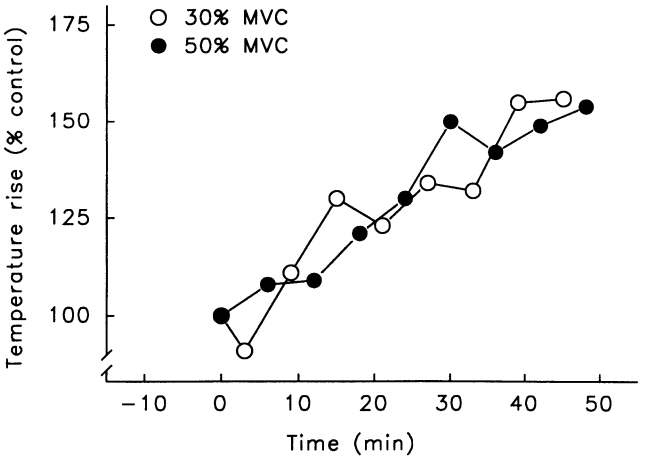

Figure 3. Temperature rise in the vastus lateralis muscle during isometric test contractions performed every 4th min of an intermittent isometric knee extension protocol (6 s contraction and 4 s rest) held at 30% and 50% MVC for 6 s with 4 s rest between. The parallel increase in the rate of temperature rise during the test contractions at the two force levels (with more type II fibers active at 50% MVC) indicate that the recruitment pattern plays a minor role for the increased energy utilization (reproduced with permission from Sejersted & Vøllestad, 1993).

evidence from recent experiments in our laboratory support this conclusion. In a separate set of experiments, we examined the rate of heat accumulation during constant force test contractions for 10-15 s carried out at regular intervals during 60 min of repeated 6 s isometric contractions. As shown in Fig. 3, the rate of temperature rise increased gradually as fatigue developed. Because the O_2 stored in the muscle is insufficient to support 10-15 s contractions at 30-50% MVC, heat accumulation after the first few seconds probably reflects the rate of anaerobic energy release and thus should be unrelated to mitochondrial respiration. In another series of experiments, we examined the changes in relaxation rate, because this parameter seems to be closely related to the ATP turnover (Edwards et al., 1975). Despite profound fatigue (MVC fell by 30-40%), the rate of relaxation rose gradually (Vøllestad et al., 1995). During repeated 30% MVC contractions carried out till exhaustion, the half-relaxation time of twitches and tetanic contractions decreased by 27 %. The exercise-induced increase in rates of heat accumulation and relaxation are in keeping with an increased ATP utilization. It must be emphasized, however, that there is no direct evidence for this conclusion. More research is needed to identify the mechanisms behind the increased metabolic rate during repeated isometric contractions. In addition, it should be sought out why the metabolic response to repeated concentric and isometric contractions differs.

HIGH-INTENSITY DYNAMIC EXERCISE AND HIGH-FORCE ISOMETRIC CONTRACTIONS

In contrast to prolonged submaximal exercise, exhaustive short lasting exercise requires extensive activation of most fibers in the working muscles, and fatigue develops rapidly. During sustained maximal voluntary contractions, the MVC force will decline by 50% within 1-3 min (Miller et al., 1988; Wilson et al., 1988; Degroot et al., 1993). Using an isokinetic ergometer or a nonmotorized treadmill, it has been shown that maximal power

falls by about 50% during 30-45 s of concentric contractions (Thorstensson & Karlsson, 1976; McCartney et al., 1983; Cheetham et al., 1986).

Under these conditions, the ATP released from aerobic processes is insufficient to match the energy demand. Hence, force and power output must rely heavily on ATP from glycolysis and PCr degradation. Accordingly, a number of studies have shown that lactic acid increases by 20-30 mmol/kg ww and PCr is reduced by more than 80% (Hermansen & Vaage, 1977; Katz et al., 1986; Cheetham et al., 1986; Miller et al., 1988; Wilson et al., 1988). Inorganic phosphate increases as a mirror image of PCr, whereas only small and insignificant changes are reported for ATP (Miller et al., 1988; Quistorff et al., 1992). Because MRS has a relatively good time resolution, it has been possible to compare the temporal changes in maximal force generating capacity and metabolite concentration during sustained contractions. Several recent studies have shown that the fall in PCr and rise in Pi parallels the reduction in maximal force over the first 1-2 min. Thereafter, the levels of PCr and Pi almost stabilizes while force continues to decline (Wilson et al., 1988; Degroot et al., 1993). In contrast, pH increases initially, before a steady decline begins (Miller et al., 1988; Wilson et al., 1988; Degroot et al., 1993). An illustration of these changes are shown in Fig. 4.

Studies of skinned fibers have shown that both low pH and increased Pi may reduce maximal force by 50% (Donaldson & Hermansen, 1978; Cooke & Pate, 1985). Accordingly, both of these factors have been assumed to be responsible for fatigue. Several lines of evidence, however, indicate that a low pH is not necessarily an important mechanism of muscle fatigue. By comparing the relationship between muscle force and metabolite levels during maximal concentric contractions of different speeds, Wilson and coworkers (1988) found that there were no uniform relation between pH and force, whereas the relationship between $H_2PO_4^-$ and force remained unaffected by the duty cycle. Hence, they concluded that fatigue was attributed more to high levels of $H_2PO_4^-$ and less to a low pH. Other studies with different contraction protocols have arrived at the same conclusion (Miller et al., 1988; Le Rumeur et al., 1990; Degroot et al., 1993). Furthermore, the recovery of muscle force is

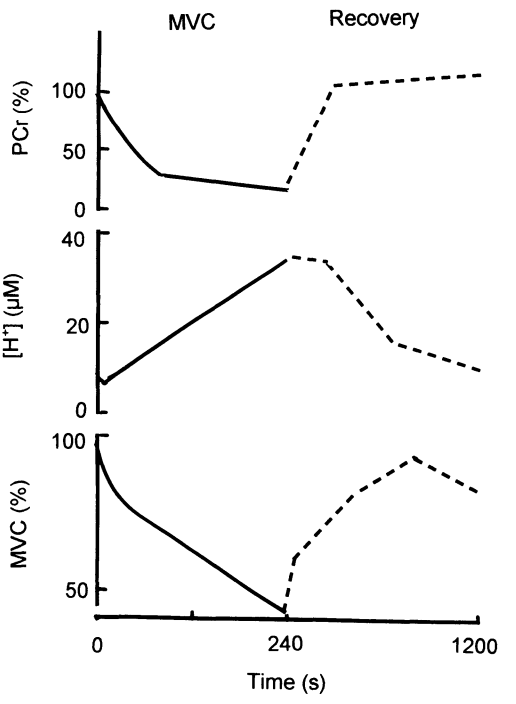

Figure 4. Changes in concentrations of PCr and H^+ of calf muscle during and after 4 min of sustained MVC (adapted from Degroot et al., 1993).

more closely related to $H_2PO_4^-$ than to pH because MVC force increases immediately after the end of exercise, while pH remains depressed or continues to decline for a minute or two before slowly returning to pre-exercise levels (Miller et al., 1988; Degroot et al., 1993).

Based on the differences in glycolytic capacity between type I (slow twitch) and II (fast twitch) fibers, it is often assumed that anaerobic energy release occurs predominantly in type II fibers. Accordingly, one would expect pH and Pi to influence the force generating capacity in type I less than in type II. From muscle biopsies obtained before and after short lasting bicycle exercise of various intensities we calculated the glycogenolytic rate of muscle fibers of various types (Vøllestad et al., 1992). At three different intensities (120-200% VO_2max), the glycogen breakdown in type I fibers was about 30% lower than in type II fibers. During exercise at the highest intensity (200% VO_2max), the glycogenolytic rate in type I fibers was twice the rate observed in type II fibers during exercise at 120% VO_2max. This observation suggests that in human muscle marked reductions in pH may occur in both fiber types, in keeping with single fiber analysis of lactate content (Essén & Häggmark, 1975) Because PCr is reported to decline by 80% or more in the quadriceps muscle during intense exercise, PCr depletion must be substantial in type I fibers. This is shown for electrically stimulated contractions (Söderlund et al., 1992). Hence, it appears that fatigue in both fiber types can potentially be due to low pH or PCr depletion. In contrast to the fast glycogenolytic rate that may be observed in type I fibers during voluntary activation, electrically stimulated contractions of the quadriceps muscle fails to stimulate glycogenolysis markedly in type I fibers (Söderlund et al., 1992). The causes for the different metabolic response to voluntary and electrically stimulated contractions are unresolved. However, these data emphasize that metabolic regulation may be task dependent.

CONCLUSION

The available data shows that fatigue related changes vary markedly between different tasks. Furthermore, correlations between fatigue and known metabolic changes all indicate that the metabolic factors are insufficient to fully explain the changes in maximal force. There is now substantial evidence that other factors also contribute to fatigue. In particular, during high-force contractions or exercise at high power output, electrolyte shifts over the sarcolemma and between sarcoplasmic reticulum and cytosol can be dramatically altered (Sejersted, 1992). Future studies should examine the interaction between electrolyte balance and metabolic changes.

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REFERENCES

Bangsbo J, Johansen L, Quistorff B & Saltin B (1993). NMR and analytic biochemical evaluation of CrP and nucleotides in the human calf during muscle contraction. *Journal of Applied Physiology* **74**, 2034-2039.

Bergström J (1962). Muscle electrolytes in man. *Scandinavian Journal of Clinical and Laboratory Investigation Supplement (Oslo)* **14**, 9-88.

Bigland-Ritchie B, Cafarelli E & Vøllestad NK (1986a). Fatigue of submaximal static contractions. *Acta Physiologica Scandinavica* **128**, 137-148.

Bigland-Ritchie B, Furbush F & Woods JJ (1986b). Fatigue of intermittent submaximal voluntary contractions: central and peripheral factors. *Journal of Applied Physiology* **61**, 421-429.

Cheetham ME, Boobis LH, Brooks S & Williams C (1986). Human muscle metabolism during sprint running. *Journal of Applied Physiology* **61**, 54-60.

Cooke R & Bialek W (1979). Contraction of glycerinated muscle fibers as a function of the ATP concentration. *Biophysical Journal* **28**, 241-258.

Cooke R & Pate E (1985). The effects of ADP and phosphate on the contraction of muscle fibers. *Biophysical Journal* **48**, 789-798.

Degroot M, Massie B, Boska M, Gober J, Miller RG & Weiner MW (1993). Dissociation of [H^+] from fatigue in human muscle detected by high time resolution ^{31}P-NMR. *Muscle &Nerve* **16**, 91-98.

Donaldson SKB & Hermansen L (1978). Differential, direct effects of H^+ on Ca^{2+}-activated force of skinned fibers from the soleus, cardiac and adductor magnus muscles of rabbits. *Pflügers Archiv* **376**, 55-65.

Edwards RHT (1976). Metabolic changes during isometric contractions of the quadriceps muscle. In: Jokl E (ed.), *Medicine and Sport*, vol. 9, pp. 114-131. Basel: Karger.

Edwards RHT, Hill DH & Jones DA (1975). Metabolic changes associated with the slowing of relaxation in fatigued mouse muscle. *Journal of Physiology (London)* **251**, 287-301.

Essén B & Häggmark T (1975). Lactate concentration in type I and II muscle fibres during muscle contraction in man. *Acta Physiologica Scandinavica* **95**, 344-346.

Fitts RH (1994). Cellular mechanisms of muscle fatigue. *Physiological Reviews* **74**, 49-94.

Gollnick PD, Armstrong RB, Saubert CW, IV, Sembrowich WL, Sherpherd RE & Saltin B (1973). Glycogen depletion patterns in human skeletal muscle fibers during prolonged work. *Pflügers Archiv* **344**, 1-12.

Gollnick PD, Piehl K & Saltin B (1974). Selective glycogen depletion pattern in human muscle fibers after exercise of varying intensity and at varying pedalling rates. *Journal of Physiology (London)* **241**, 45-57.

Hermansen L & Vaage O (1977). Lactate disappearance and glycogen synthesis in human muscle after maximal exercise. *American Journal of Physiology* **233**, E422-E429.

Katz A, Sahlin K & Henriksson J (1986). Muscle ATP turnover rate during isometric contraction in humans. *Journal of Applied Physiology* **60**, 1839-1842.

Le Rumeur E, Le Moyec L, Toulouse P, Le Bars R & de Certaines JD (1990). Muscle fatigue unrelated to phosphocreatine and pH: an "in vivo" 31-P NMR spectroscopy study. *Muscle & Nerve* **13**, 438-444.

Lundsgaard E (1930). Untersuchungen über muskelkontraktionen ohne Milchsäurebildung. *Biochemische Zeitshrift* **217**, 162-175.

McCartney N, Heigenhauser GJF, Sargeant AJ & Jones NL (1983). A constant-velocity cycle ergometer for the study of dynamic muscle function. *Journal of Applied Physiology* **55**, 212-217.

Miller RG, Boska MD, Moussavi RS, Carson PJ & Weiner MW (1988). 31P nuclear magnetic resonance studies of high energy phosphates and pH in human muscle fatigue. *Journal of Clinincal Investigation* **81**, 1190-1196.

Quistorff B, Johansen L & Sahlin K (1992). Absence of phosphocreatine resynthesis in human calf muscle during ischaemic recovery. *Biochemical Journal* **291**, 681-686.

Sahlin K, Cizinsky S, Warholm M & Höberg J (1992). Repetitive static muscle contractions in humans - a trigger of metabolic and oxidative stress? *European Journal of Applied Physiology and Occupational Physiology* **64**, 228-236.

Sejersted OM (1992). Electrolyte imbalance in body fluids as a mechanism of fatigue. In: Lamb DR, Gisolfi CV (eds.), *Energy Metabolism in Exercise and Sport (Perspectives in Exercise Science and Sports Medicine)*, pp 149-207. Carmel, IN: Brown & Benchmark.

Sejersted OM & Vøllestad NK (1993). Physiology of muscle fatigue and associated pain. In: Vaerøy H, Merskey H (eds.), *Progress in fibromyalgia and myofascial pain*, pp. 41-51. Amsterdam: Elsevier Science Publications.

Söderlund K, Greenhaff PL & Hultman E (1992). Energy metabolism in type I and type II human muscle fibres during short term electrical stimulation at different frequencies. *Acta Physiologica Scandinavica* **144**, 15-22.

Söderlund K & Hultman E (1986). Effects of delayed freezing on content of phosphagens in human skeletal muscle biopsy samples. *Journal of Applied Physiology* **61**, 832-835.

Thorstensson A & Karlsson J (1976). Fatiguability and fibre composition of human skeletal muscle. *Acta Physiologica Scandinavica* **98**, 318-322.

Vøllestad NK & Blom PCS (1985). Effect of varying exercise intensity on glycogen depletion in human muscle fibres. *Acta Physiologica Scandinavica* **125**, 395-405.

Vøllestad NK, Sejersted I, Saugen E (1995) Increased relaxation rates during and following intermittent submaximal isometric contractions. *Clinical Physiology* In press.

Vøllestad NK, Sejersted OM, Bahr R, Woods JJ & Bigland-Ritchie B (1988). Motor drive and metabolic responses during repeated submaximal contractions in man. *Journal of Applied Physiology* **64**, 1421-1427.

Vøllestad NK, Tabata I & Medbø JI (1992). Glycogen breakdown in different human muscle fibre types during exhaustive exercise of short duration. *Acta Physiologica Scandinavica* **144**, 135-141.

Vøllestad NK, Vaage O & Hermansen L (1984). Muscle glycogen depletion patterns in type I and subgroups of type II fibres during prolonged severe exercise in man. *Acta Physiologica Scandinavica* **122**, 433-441.

Vøllestad NK, Wesche J & Sejersted OM (1990). Gradual increase in leg oxygen uptake during repeated submaximal contractions in humans. *Journal of Applied Physiology* **68**, 1150-1156.

Wilson JR, McCully KK, Mancini DM, Boden B & Chance B (1988). Relationship of muscular fatigue to pH and diprotonated P_i in humans: a ^{31}P-NMR study. *Journal of Applied Physiology* **63**, 2333-2339.