Effect of oral creatine supplementation on respiratory gas exchange and blood lactate accumulation during steady-state incremental treadmill exercise and recovery in man

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INTRODUCTION

Recently published data show that a regimen of dietary creatinine (Cr) ingestion in man can elevate whole-muscle total Cr stores by approximately 20%, one-third of which is in the form of phosphocreatine (PCr) [1]. This has been shown to improve performance during bouts of repetitive maximal isokinetic knee extension [2] and maximal cycling exercise [3, 4], an effect which has been suggested to be due mainly to better maintained muscle ATP and PCr resynthesis [2]. It is also possible that an increase in intramuscular buffering capacity resulting from greater PCr hydrolysis may have contributed to the observed improvements in repetitive high-intensity muscle performance [2]. These findings suggest that athletes performing short-duration, power-related events would benefit from Cr supplementation, which has recently been confirmed [5]. There have also been some anecdotal reports that athletes involved in lower intensity, endurance events can derive benefit from Cr supplementation. Based on the knowledge that cystolic Cr is known to be an acceptor of ATP generated by mitochondria [6, 7], and that Cr availability has been shown to increase PCr resynthesis during recovery from exercise [8], it is plausible to suggest that intracellular Cr availability will have an influence on mitochondrial ATP production, and thereby increase oxygen consumption, during exercise and recovery. Any such effect might also be expected to produce a reciprocal decrease in muscle lactate production. A study was therefore performed to examine the influence of oral Cr supplementation on energy metabolism during steady-state incremental submaximal exercise and recovery.

METHODS

Subjects

Eight physically active men (Table 1) gave their written consent to participate in the study, which was approved by the Army Personnel Research Establishment Ethics Committee.

Key words: creatine, energy metabolism, exercise, phosphocreatine.
Abbreviations: Cr, creatine; PCr, phosphocreatine; RER, respiratory exchange ratio.
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Experimental protocol

The study was performed over a 3-week period. During week 1 the subjects performed an incremental exercise test running on a treadmill at 10 km/h with gradients increased from 0% in 2% increments, to the point of exhaustion. Exercise periods were of 4 min duration at gradients up to 10% and 3 min thereafter, and subjects rested for 4 min between each exercise level. Expired air was collected using Douglas bags for the final 60 s of exercise. During week 1 the subjects performed an incremental exercise test running on a treadmill at 10 km/h with gradients increased from 0% in 2% increments, to the point of exhaustion. Exercise periods were of 4 min duration at gradients up to 10% and 3 min thereafter, and subjects rested for 4 min between each exercise level. Expired air was collected using Douglas bags for the final 60 s of exercise, and these samples were then analysed for oxygen and carbon dioxide (Servomex 1400 series) contents. Expired gas volume was measured using a spirometer (Collins). Two days following this test, subjects repeated their two highest workloads to confirm that maximal oxygen uptake had been achieved. Experiments were performed at the same time of day for each of the subjects who had eaten only a light breakfast at least 2 h previously.

In week 2, subjects performed a continuous incremental exercise test, running on a treadmill at 10 km/h for 6 min exercise periods with workloads set at 50%, 60%, 65%, 70%, 75%, 80% and 90% VO₂max. Respiratory gas exchange was measured as previously on samples collected for the final 30 s at each workload, and on samples collected over 5 min periods during the first 15 min of recovery. Blood samples for lactate analysis were obtained via an indwelling 21-G forearm venous cannula, which was kept patent by intermittent flushing with saline. Samples were taken during the final 30 s at each workload and at 5, 10 and 15 min during recovery. On collection, blood samples were rapidly mixed with lithium heparin, after which 100-μl aliquots were immediately deproteinized in 1 ml of 1 mol/l ice-cold perchloric acid. This was then centrifuged and frozen at -20°C for subsequent analysis of lactate concentration [9].

Commencing 2 days after the tests in week 2, each subject consumed four doses of 5 g of Cr dissolved in a hot drink at approximately 4-h intervals. This regimen was repeated for a further 4 days to give a total Cr dose of 20 g each day for 5 days, which has previously been shown to result in an approximately 20% increase in muscle total Cr concentration [1]. Two days after completion of the creatine supplementation, subjects repeated, at exactly the same workloads, the incremental exercise test performed in week 2. Throughout the period of the study subjects were asked to maintain their dietary intake and physical activity patterns as close to normal as possible.

Data analyses

Mean and standard errors of the mean (SEM) for each parameter measured were calculated before and after Cr supplementation and were plotted for each exercise intensity and over the three 5-min periods of recovery. Analysis of variance for repeated measures was used to compare data before and after Cr supplementation. Student’s paired t-test was used to compare the body weight changes before and after Cr. On all occasions values shown in the text, table and figure represent means (SEM), and any difference between treatments was deemed to be significant at the P < 0.05 level.

RESULTS

Subjects were of moderate to high aerobic fitness, and details of ages, weights before and after Cr ingestion and maximal aerobic capacity are shown in Table 1. All subjects showed a small gain in weight with mean values rising from 75.9 (SEM 2.2) kg to 76.9 (SEM 2.3) kg (P < 0.05). As shown in Fig. 1, the ingestion of Cr had no significant effects on oxygen consumption, blood lactate concentration or respiratory exchange ratio. Running on a flat treadmill at 10 km/h corresponded to exercise intensities of greater than 50–65% VO₂ in some of the subjects, and one subject could not sustain an exercise intensity of 90% VO₂max for 6 min. Fig. 1 therefore represents varying numbers of subjects at different levels of exercise, and this is illustrated on each part of the figure. Cr ingestion also had no significant effects on carbon dioxide production, expired gas volume or heart rate.

DISCUSSION

The results from this experiment showed that Cr ingestion had no effect on respiratory gas exchange or blood lactate accumulation during incremental steady-state exercise and recovery. This is perhaps surprising in view of recent data showing that Cr supplementation leads to an increased rate of muscle PCr resynthesis, and presumably therefore an increase in oxygen consumption, during recovery from maximal contraction [8], and those studies which support the existence of the PCr/Cr shuttle [10, 11]. Greenhaff et al. [8] postulated that the greater PCr resynthesis observed during recovery from maximal exercise after Cr ingestion was...
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attributable to the concentration of muscle Cr being maintained above the Michaelis constant ($K_m$) concentration of creatine kinase for Cr (20 mmol/l, ref. 12), thereby sustaining a high flux rate through the creatine kinase reaction in favour of PCr resynthesis. With this in mind, and accepting that the free Cr concentration of muscle ranges from ~13 mmol/l intracellular water at rest [1] to 40 mmol/l intracellular water following short-term maximal exercise [13], the design of the present study was intended to produce changes in muscle Cr concentration which would fall on either side of the potentially important $K_m$ concentration of 20 mmol/l. This being the case, the clear lack of an effect of Cr ingestion on the measured variables is difficult to explain. This apparent discrepancy could be related to a number of factors. Firstly, it is possible that the expired gas and blood lactate measurements of the present study may have been too insensitive to reflect accurately the quantitatively small changes in muscle metabolism which may have occurred during exercise and recovery following Cr supplementation. Secondly, it is possible that, even before Cr ingestion, the muscle Cr concentration during exercise and recovery was always greater than 20 mmol/l, and therefore Cr availability was not a limiting factor to flux through the creatine kinase reaction. A more invasive experimental protocol involving tissue oxygen consumption and metabolite measurements would hopefully clarify these points. Finally, it is unclear whether the reported $K_m$ of creatine kinase for Cr determined in vitro [12] accurately reflects that in existence in vivo.

The lack of any effect of Cr ingestion on the measured variables in the present experiment could also clearly be interpreted to mean that Cr ingestion has no effect on metabolism during steady-state submaximal exercise, which is partly in agreement with recent data showing that dietary Cr supplementation had no influence on performance during a submaximal 6-km all-terrain run in man [14]. This interpretation is also supported by evidence in the literature indicating that there is no functional coupling between mitochondrial ATP resynthesis and mitochondrial creatine kinase [15, 16]. Using animal models, these studies demonstrated that a marked reduction in muscle Cr stores, achieved by feeding the Cr analogue $\beta$-guanidinopropionic acid, had no effect on muscle function or aerobic metabolism during submaximal steady-state contraction, and, as a consequence, it was proposed that Cr is not an important regulator of substrate utilization during steady-state contraction. Interestingly, in the study of Meyer et al. [16], in addition to the authors presenting evidence relating to submaximal steady-state contraction, it was also demonstrated that force production during very intense contraction is markedly dependent on Cr availability, which is in agreement with studies demonstrating the positive effect of Cr ingestion on performance during maximal short-term exercise in man [2–5].

The data from the present experiment must of course be interpreted in the light of the experimental weaknesses (i.e. no placebo control or cross-over design). However, the body weight gains observed in subjects following Cr supplementation were similar to those previously reported [4, 8, 14] and support the assumption that Cr supplementation was effective in raising intramuscular Cr and PCr concentrations. The lack of a cross-over design, while not desirable, was in some ways unavoidable because the Cr washout time of skeletal muscle is currently unknown; however, washout is likely to
be achieved over a period of months rather than days [17].

In summary, the results show that Cr supplementation, which has previously been shown to enhance performance during maximal short-duration exercise, has no effect on respiratory gas exchange or blood lactate accumulation during incremental steady-state exercise and recovery in man.

REFERENCES