Caffeine counteracts the ergogenic action of muscle creatine loading

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Vandenberghé, K., N. Gillis, M. Van Leemputte, P. Van Hecke, F. Vanstampel, and P. Hespel. Caffeine counteracts the ergogenic action of muscle creatine loading. J. Appl. Physiol. 80(2): 452–457, 1996.—This study aimed to compare the effects of oral creatine (Cr) supplementation with creatine supplementation in combination with caffeine (Cr+C) on muscle phosphocreatine (PCr) level and performance in healthy male volunteers (n = 9). Before and after 6 days of placebo, Cr (0.5 g·kg−1·day−1), or Cr (0.5 g·kg−1·day−1) + C (5 mg·kg−1·day−1) supplementation, 31P-nuclear magnetic resonance spectroscopy of the gastrocnemius muscle and a maximal intermittent exercise fatigue test of the knee extensors consisted of three consecutive maximal isometric contractions and three interval series of 90, 80, and 50 maximal voluntary contractions performed with a rest interval of 2 min between the series. Muscle ATP concentration remained constant over the three experimental conditions Cr and Cr+C increased (P < 0.05) muscle PCr concentration by 4–6%. Dynamic torque production, however, was increased by 10–23% (P < 0.05) by Cr but was not changed by Cr+C. Torque improvement during Cr was most prominent immediately after the 2-min rest between the exercise bouts. The data show that Cr supplementation elevates muscle PCr concentration and markedly improves performance during intense intermittent exercise. This ergogenic effect, however, is completely eliminated by caffeine intake.

exercise; phosphorus-31-nuclear magnetic resonance; phosphocreatine; adrenergic stimulation; diet

THE CREATINE KINASE reaction that catalyzes the reversible exchange of high-energy phosphate between ATP and phosphocreatine (PCr) plays a key role in energy metabolism in contracting muscle. First, PCr is the only fuel available to precipitously regenerate ATP during episodes of rapidly fluctuating energy demand. Second, PCr breakdown functions as a buffer of protons originating from anaerobic glycogen breakdown. Third, during contractile activity, the sarcoplasmic ATP/ADP ratio is maintained at a high level at subcellular sites where creatine kinase is functionally coupled to ATP-consuming processes, the kinetics of which favor rapid rephosphorylation of ADP to ATP. This, in turn, also contributes to preserve the muscle adenine nucleotide pool. Finally, the creatine kinase isoenzyme system possibly provides an efficient way of transferring mitochondrialy produced high-energy phosphate to the sites of ATP consumption in the cytoplasm (5, 27). Thus procedures capable of improving the efficiency of the PCr system are expected to yield a significant "ergogenic" benefit. Some recent studies, indeed, have indicated dietary creatine supplementation to raise creatine and PCr levels in skeletal muscle (14, 18). Moreover, this was accompanied by delayed occurrence of fatigue during high-intensity intermittent exercise (1, 6, 15, 19). This ergogenic effect may be partly explained by facilitation of PCr resynthesis during episodes of muscle relaxation (14).

Creatine entry into muscle cells is facilitated by specific creatine transporters present in the sarcolemma (12, 13, 16, 25). Because the Michaelis constant of creatine uptake is situated in the physiological range of plasma creatine concentrations (13, 21), oral creatine intake is expected to stimulate creatine uptake by muscle cells by enhanced membrane creatine transport. Furthermore, increased creatine uptake appears to result in intracellular creatine trapping, in its free as well as in its phosphorylated form (14, 18). On the basis of the above data, it is reasonable to hypothesize that intramuscular creatine and PCr levels might be even further elevated by adding a direct stimulus to the membrane creatine transport system during oral creatine administration. In this respect, sarcolemmic creatine transport is known to be critically dependent on extracellular Na⁺ (12, 16, 21). Thus adrenergic stimulation might facilitate the uptake of exogenous creatine by muscle tissue via increasing Na⁺-K⁺-adenosinetriphosphatase (Na⁺-K⁺-ATPase) pump activity, thereby increasing the Na⁺-sarcolemmic gradient (8, 9). The aim of the present study, therefore, was to investigate the ability of caffeine to amplify the effects of oral creatine supplementation on the muscle PCr pool as well as on the capacity to perform high-intensity exercise. Caffeine, indeed, acts in vivo as a potent sympathomimetic agent by elevating the circulating epinephrine level. The data demonstrate that caffeine does not improve the efficiency of oral creatine either to raise muscle PCr or to improve performance. In contrast, the ergogenic action of muscle creatine loading is fully abolished by caffeine administration during the period of creatine supplementation.

METHODS

Subjects. Nine healthy male subjects ranging in age from 20 to 23 yr gave their informed written consent to take part in the study. The study protocol was approved by the local Ethics Committee. All subjects were physically active in some form of recreational exercise but none of them was highly trained. They were informed on the experimental procedures to be undertaken and were asked to abstain from any medication during the period of the study and to avoid changes in their level of physical activity.
Study protocol. A double-blind study was performed, whereby the subjects were assigned in random order to three experimental protocols (protocols A, B, and C), each lasting 8 days and separated by a washout period of 3 wk. The subjects started abstaining from trimethylxanthine-containing beverages and foods for at least 4 days before each experimental period during which they received identical standardized meals (3,000 kcal/day, 50% carbohydrates, 32% fat, 18% proteins). During protocol A, the subjects were administered creatine supplements for 6 days (days 2–7). They ingested a total of 0.5 g creatine monohydrate (Sigma Biochemicals) per kilogram body weight per day, divided over eight similar doses taken at regular intervals. The creatine monohydrate supplements were administered with a maltodextrine and partame powder mixture (97:3), so as to match placebo taste and were rapidly dissolved in hot water immediately before intake. The last dose of creatine preceded the measurements on day 8 by at least 12 h. Protocol B was identical to protocol A, with the exception that the subjects received placebo supplements (0.5 g glucose·kg body wt$^{-1}$·day$^{-1}$) instead of creatine. Protocol C was similar to protocol A. However, on days 5, 6, and 7, in addition to the creatine supplements, the subjects were administered a single dose of 5 mg caffeine·kg body wt$^{-1}$·day$^{-1}$. Caffeine capsules were ingested after breakfast. So as to simultaneously induce a marked increase in plasma creatine and caffeine levels, a double dose of caffeine was administered at the same time as the caffeine intake. One hour later, another caffeine supplement was taken. The last dose of caffeine preceded the measurements on day 8 by at least 20 h. During protocols A and B, placebo (glucose) instead of caffeine capsules was administered to the subjects.

On the first (day 1) and the last day (day 8) of each period, the subjects reported to the laboratory after a light standardized meal. Their body weight was first measured, and thereafter $^{31}$P-nuclear magnetic resonance ($^{31}$P-NMR) spectroscopy of the gastrocnemius muscle of the nondominant leg was performed. Finally, the subjects performed an intermittent exercise test with the same leg on an isokinetic dynamometer to evaluate the isometric and dynamic strength and fatigability of the knee extensors. Over the different experimental conditions, the subjects were evaluated on the same day of the week and at the same time of the day. The results of the measurements were disclosed neither to the subjects nor to the investigators until completion of the entire study.

Determination of muscle PCr concentration. $^{31}$P-NMR measurements from the calf muscles were performed in a 4.7 T superconducting magnet with 30-cm diameter horizontal bore ( Biospec, Bruker, Karlsruhe). Before the leg was inserted in the magnet, the calf was marked at the level of its largest circumference. The mark was then positioned in the center of a 50-mm diameter surface coil and mounted in a wooden mold, with an adjustable holder allowing accurate and reproducible positioning of each of the subjects' legs during the different NMR sessions. The coil was tuned to either the proton or the phosphorus frequency. Axial proton NMR images (200 MHz) were acquired with the surface coil to verify the position of the muscle on the coil (Flash sequence with repetition time/echo time/angle = 100 ms/10 ms/40°; three slices, 10 mm thick, 10 mm gap). $^{31}$P-NMR (81.1 MHz) signals were acquired with a 160-μs excitation pulse (corresponding to an angle of ~120° in the middle of the coil) and accumulated to improve the signal-to-noise ratio (64 acquisitions, every 5 s). The broad resonance from phosphorus nuclei in less mobile molecules was removed by multiplication of the time signal with an exponential decay corresponding to a line broadening of 1,000 Hz and subtraction of the result from the original signal. The deconvoluted time-domain signal was exponentially filtered (5 Hz line broadening) to further improve the signal-to-noise ratio and was Fourier transformed for spectral analysis. Finally, the PCr and ATP peaks were manually integrated. Integral values were corrected for partial saturation due to incomplete relaxation during the 5-s repetition period (correction factor 1.36 for PCr and 1.08 for β-ATP). Correction factors were determined from a separate measurement on a single subject by comparing the partially relaxed spectrum (5-s repetition period) with a fully relaxed spectrum (50-s repetition period). The area of the water proton peak was used as an internal reference to correct for possible drifts of the spectrometer amplifier gains between the various experiments. It has been demonstrated that ATP concentration estimated by NMR spectroscopy is similar to ATP concentration measured biochemically (4). Because β-ATP peak areas were unchanged over the different treatments, the mean area for β-ATP calculated from all repeated measurements performed in the total group of subjects was set equivalent to a concentration of 5.5 mmol/kg wet muscle (17). Thereafter, individual β-ATP areas were referred to this mean ATP value, and ATP concentrations were expressed in millimoles per kilogram wet muscle. PCr concentrations were calculated by multiplying the ratio of the saturation corrected peak areas of PCr to ATP by the ATP concentration.

Determination of knee extension torque. The exercise test consisted of unilateral knee extensions performed with the nondominant leg in a sitting position on an isokinetic dynamometer that was calibrated before each experiment. After a standardized 5-min warmup, the subjects performed consecutively and separated by 2-min rest intervals the following exercises: 1) three static maximal voluntary contractions (MVCs) of the knee-extension muscles, 2) three bouts of 30 dynamic MVCs, 3) four bouts of 20 dynamic MVCs, and, finally, 4) five bouts of 10 dynamic MVCs. The bouts of 30, 20, and 10 MVCs were each separated by 60-, 40-, and 20-s rest periods, respectively. Knee-extension torque was measured during each contraction and digitized (250 Hz) by an on-line computer. Maximal isometric torque was derived as the smoothed curve during the static contractions (5 s) at a knee angle of, consecutively, 95, 120, and 145°. The dynamic torque was calculated as the mean torque during maximal knee extension at a constant velocity of 180°/s, starting from 90° knee flexion to full knee extension. After each contraction, the leg was returned (180°/s) passively to the starting position from which the next contraction was immediately initiated. Torque production was registered as the mean of five successive contractions.

Statistics. The results are expressed as means ± SE (n = 9). Statistical evaluation of body weight and NMR spectroscopy data were performed by using two-tailed paired t-tests. Comparison of knee-extension torque data was performed by using repeated-measures two-way analysis of variance (SAS Institute, general linear models procedure), and Scheffé's tests were applied for post hoc multiple comparison when appropriate. The relationship between variables was evaluated using Pearson's correlation coefficient. The level of statistical significance was set at P < 0.05.

RESULTS

Body weight, identification, and side effects of the treatment. Body weight was on average 80 ± 4 kg at the start of the study and did not significantly change over the different experimental conditions.

At the end of each protocol, the subjects were asked as to whether they had a notion of the past treatment or had experienced adverse side effects.
After creatine ingestion, eight subjects reported they were unsure about the treatment, whereas one subject answered incorrectly (placebo). After creatine + caffeine as well as after placebo treatments, one subject's identification of treatment was correct, whereas eight subjects expressed no opinion. Three subjects reported some minor gastrointestinal distress: two during the first 2 days of creatine ingestion and one over the 3 days of creatine + caffeine supplementation.

**Muscle ATP and PCr concentrations.** NMR PCr/ATP peak area ratios and the calculated muscle ATP and PCr concentrations are presented in Table 1. ATP concentrations remained constant over the three experimental conditions, before as well as after the respective treatment regimens. Furthermore, muscle PCr/ATP ratio and PCr concentration were similar at the start of the placebo, creatine, and creatine + caffeine conditions. Compared with placebo, both creatine and creatine + caffeine increased ($P < 0.05$) muscle PCr concentration. The relative increase in muscle PCr produced by creatine and creatine + caffeine amounted to 4 and 6% of the initial level, respectively. The effects of creatine and creatine + caffeine supplementation on muscle PCr concentration were not different.

**Static knee-extension torque.** Isometric torque production by the knee extensor muscles was measured at knee angles of 95, 120, and 145°, respectively, in this order. As shown in Table 2, pretreatment values were similar for the three experimental conditions and placebo did not affect isometric torque. Compared with placebo, neither creatine nor creatine + caffeine changed static knee-extension torque.

**Dynamic knee-extension torque and fatigue.** Two minutes after the static torque measurements, three series of dynamic knee-extension MVCs were performed, separated by a rest interval of 2 min: series 1 (S1) consisted of three bouts of 30 MVCs, series 2 (S2) of four bouts of 20 MVCs, and series 3 (S3) of five bouts of 10 MVCs, performed with intervals of 60, 40, and 20 s, respectively. knee-extension torques and the pattern of fatigue typical to the exercise test used are shown in Fig. 1. The data points represent the average torque value of each five consecutive contractions before and after placebo, creatine, and creatine + caffeine. Within each series, torque peaked at the start of the first bout, whereafter it progressively decreased. Compared with the peak torque observed in S1, initial torque was 2–9% lower in S2 and 5–12% lower in S3. Torque produced at the end of S1, S2, and S3 represented 55–72, 58–74, and 70–76% of the peak torque registered at the start of the exercise test, respectively. Pretreatment values were not significantly different for the three experimental conditions. Furthermore, dynamic torque production was not affected by placebo.

As indicated above, dynamic knee-extension torque progressively decreased from the start to the end of each exercise bout. Because neither creatine nor creatine + caffeine significantly altered the slope of this decrease, mean torque productions per bout were considered for further analyses of the treatment effects. Figure 2 shows the change in knee-extension torque (post minus pre ingestion) produced by placebo, creatine, and creatine + caffeine supplementation. Compared with placebo, torque generation during the successive bouts of knee-extensions was larger ($P < 0.05$) after creatine. This ergogenic effect was most pronounced during the first bout of each series performed following the 2-min rest intervals after the preceding exercise series. Thus knee-extension torque was improved ($P < 0.05$) by 23, 16, and 14% during the first exercise bout of S1, S2, and S3, respectively. During the last bout of each series, however, torque gain had diminished to ~10% (not significant). The change in muscle torque during bout 1 correlated positively ($r = 0.65, P = 0.05$) with the rise in muscle PCr concentration. The improvement of intermittent knee-extension torque seen after creatine was completely absent after creatine + caffeine. Torque production in the creatine + caffeine condition was similar to performance during placebo, from the start to the end of the exercise test.

**DISCUSSION**

Creatine loading is rapidly becoming a popular ergogenic aid in sports. This evolution ensues a recent report by Harris and co-workers (18), demonstrating that prolonged high-dose oral creatine intake elevates the muscle creatine and PCr pool. Moreover, creatine supplementation has shown significant ergogenic potency (1, 6, 15, 19), predominantly during high-intensity intermittent exercise. The present studies were set up in an attempt to contribute to a better understanding of the mechanisms underlying the improvement of physical performance caused by creatine.
CREATINE INTAKE AND PERFORMANCE

Table 2. Static torque production of knee extensor muscles before and after ingestion of placebo, creatine, and creatine in combination with caffeine

<table>
<thead>
<tr>
<th>Knee Angle</th>
<th>95°</th>
<th>120°</th>
<th>145°</th>
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<tbody>
<tr>
<td>Placebo</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pre</td>
<td>139 ± 11</td>
<td>193 ± 15</td>
<td>213 ± 10</td>
</tr>
<tr>
<td>Post</td>
<td>135 ± 15</td>
<td>197 ± 20</td>
<td>214 ± 18</td>
</tr>
<tr>
<td>Creatine</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pre</td>
<td>137 ± 15</td>
<td>175 ± 17</td>
<td>203 ± 21</td>
</tr>
<tr>
<td>Post</td>
<td>138 ± 14</td>
<td>197 ± 16</td>
<td>221 ± 15</td>
</tr>
<tr>
<td>Creatine + caffeine</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pre</td>
<td>134 ± 11</td>
<td>181 ± 15</td>
<td>197 ± 17</td>
</tr>
<tr>
<td>Post</td>
<td>136 ± 10</td>
<td>194 ± 14</td>
<td>220 ± 18</td>
</tr>
</tbody>
</table>

Values (Nm) are means ± SE of 9 observations. Maximal isometric knee-extension torque was measured consecutively at a knee angle of 90, 120, and 145° before (Pre) and after (Post) 6 days of placebo, creatine (0.5 g·kg⁻¹·day⁻¹) and creatine (0.5 g·kg⁻¹·day⁻¹) plus caffeine (5 mg·kg⁻¹·day⁻¹) administration, using an isokinetic dynamometer. See METHODS for further details. Post values were not significantly different from Pre values.

loading and thereby to possibly improve the efficiency of creatine supplementation procedures. We hypothesized that oral creatine intake, combined with a physiological degree of adrenergic stimulation by caffeine, might facilitate muscle creatine trapping. This, in turn, might enhance performance even further than simple creatine supplementation. The data demonstrate that caffeine does not improve the efficiency of oral creatine to either raise muscle PCr or enhance performance. Unexpectedly, our findings indicate that caffeine counteracts the ergogenic action of muscle creatine loading.

Creatine transfer from the extracellular space to the sarcoplasm is facilitated by specific carrier proteins residing in the plasma membrane (12, 13, 16, 25). Given the critical role of the sarcolemmal Na⁺ gradient in driving muscle creatine transport (12, 16, 21), it is reasonable to assume that direct stimulation of Na⁺-K⁺-ATPase activity associated with creatine ingestion may promote muscle creatine uptake. Following this line of reasoning, we compared the effect on muscle PCr of simple creatine intake with creatine intake in combination with caffeine. Caffeine, indeed, has been demonstrated to directly stimulate muscle Na⁺,K⁺-pump activity (8, 25). In addition, the drug is well known to increase plasma epinephrine (3, 20, 26), another direct stimulus to muscle Na⁺-K⁺-ATPase (8, 9). However, as shown in Table 1, 6 days of creatine supplementation (0.5 g·kg⁻¹·day⁻¹) expanded the muscle PCr pool by ~5% above the initial level, irrespective of simultaneous caffeine administration. Based on these findings, it would, nevertheless, be premature to conclude that caffeine does not possess the in vivo potential to enhance creatine uptake by skeletal muscle. Because caffeine might increase the pace of creatine and PCr trapping by muscle during the initial stage of creatine supplementation rather than elevate the presumed plateau level (18) achieved at the end of a prolonged

Fig. 1. Dynamic knee-extension torque during intermittent exercise test before and after placebo, creatine, and creatine + caffeine treatment. Values are means ± SE of 9 observations. Data points represent means of 5 consecutive contractions (con) before (○) and after (■) 6 days of placebo (A), creatine (0.5 g·kg⁻¹·day⁻¹) (B), and creatine (0.5 g·kg⁻¹·day⁻¹) + caffeine (5 mg·kg⁻¹·day⁻¹) (C) administration. with the use of an isokinetic dynamometer, 3 series of dynamic knee extensions were performed with a rest interval of 2 min between series. Series 1 consisted of 3 bouts of 30 contractions, series 2 of 4 bouts of 20 contractions, and series 3 of 5 bouts of 10 contractions, interrupted by rest intervals of 60, 40, and 20 s, respectively. See METHODS for further details.
supplementation period. Whether caffeine indeed accelerates the efficiency of creatine supplementation at the onset of treatment with respect to increasing the muscle PCr pool needs to be addressed in further studies.

Creatine loading has recently demonstrated significant ergogenic potency with regard to strenuous intermittent exercise (1, 6, 15, 19). Single short-term maximal exercise performance (11, 23) or endurance-exercise capacity (2), on the other hand, may not benefit from creatine supplementation. Similarly, the present experiments again clearly demonstrate that prolonged high-dose creatine intake efficiently improves high-intensity intermittent exercise performance. As shown in Fig. 2, knee-extension torque generated during repeated maximal isokinetic contractions was increased by up to 25% after a 6-day period of creatine ingestion. This ergogenic effect most prominently appeared during the exercise bouts immediately following the 2-min rest intervals separating the knee-extension series. Shorter rest episodes (≤60 s) clearly resulted in a reduced performance gain. Furthermore, oral creatine supplementation did not delay the rate of development of fatigue in the course of the exercise bouts, since the fall in knee-extension torque within bouts was similar after creatine and placebo administration. This indicates that creatine loading facilitates recovery of muscle dynamic force during rest following prior fatigue, rather than delays the process of fatigue during continued high-intensity contractile activity. Alternatively, one may argue that the subjects performed more work during the exercise bouts in the creatine condition than during placebo and, therefore, in fact fatigued more slowly. Support for creatine loading facilitating recovery of isokinetic force following fatigue comes from different recent studies. On the one hand, Bogdanis and co-workers (7) have demonstrated PCr resynthesis during recovery from intense muscular activity to be very critical to the reformation of muscle power at the onset of a next bout of maximal exercise. On the other hand, Greenhaff et al. (14) showed that a diet-induced increase in muscle total creatine content facilitates PCr resynthesis during the second minute of recovery from strenuous contractile activity. Accordingly, muscle creatine loading has been reported not to enhance performance during a single bout of short maximal exercise (11, 23).

As pointed out before, the idea supporting the present studies was that caffeine might enhance the ergogenic potency of creatine loading. Surprisingly, however, caffeine was found to counteract the benefits of oral creatine intake with respect to intermittent exercise performance (see Fig. 2). Data available to date do not provide a sound explanation of this phenomenon. Some tentative conclusions are, however, possible. Thus our findings suggest that caffeine directly interferes with the physiological mechanism causing the ergogenic action of creatine loading. This mechanism does not appear to be confined to increased muscle PCr availability per se. Creatine and creatine + caffeine supplementation, indeed, increased muscle PCr level to the same extent (see Table 1), whereas only the former treatment was beneficial to performance. In keeping with such a concept is a recent report by Greenhaff et al. (14) showing the muscle PCr resynthesis rather than an elevated muscle PCr pool per se to be critical to the ergogenic action of creatine loading. It is also worthwhile mentioning that the presently observed effects of caffeine on performance are unlikely to be due to acute effects of the drug on muscle energetics (see Refs. 10, 22 for reviews). The last caffeine dose, indeed, preceded the exercise test by at least 20 h. Given a plasma half-life of 3–5 h (10), it is reasonable to assume that the drug was fully eliminated at the time of the exercise test. Further studies will have to elucidate the interaction of creatine loading and caffeine with respect to regulation on muscle energetics and performance during intermittent exercise. Moreover, the weight of evidence today indicates that caffeine has no direct effects on high-intensity exercise performance. Finally, from a practical point of view, our findings indicate that trimethylxanthine-containing beverages are an inappropriate vehicle for ingestion of creatine supplements.
In conclusion, the present study demonstrates creatine loading to increase PCr concentration in skeletal muscle. This is accompanied by accelerated recovery of muscle contractile capacity during rest episodes of high-intensity intermittent exercise. The ergogenic action of creatine loading is counteracted by caffeine.

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REFERENCES


