



# Effects of creatine hydrochloride supplementation on physical performance and hormonal changes in soldiers

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## Abstract

**Purpose:** The aim of this study was to examine the effects of 2 weeks of creatine hydrochloride (CrHCl) supplementation on physical performance and hormonal changes in army soldiers. **Materials:** Eighteen male army soldiers were randomly assigned in a double-blind fashion to either a CrHCl (n=9) or placebo (Pl) (n=9) groups. CrHCl group costumed 3g of creatine per day, whereas Pl group was given dextrose. Before and after supplementation period, the subjects performed a battery of performance tests including one repetition maximum (1RM) of bench press and back squat, vertical jump (VJ) and Wingate anaerobic test. In addition, blood samples were obtained to determine changes in testosterone and cortisol concentrations per and post supplementation. **Results:** There were significant increases in VJ, peak and mean power, and 1RM back squat test following the 2 weeks of CrHCl supplementation ( $p < 0.05$ ) without any significant change for the Pl group. In addition significant changes were observed in testosterone and cortisol concentrations from before to after supplementation in CrHCl group ( $p < 0.05$ ) and compared with Pl group ( $p=0.001$ ). **Conclusions:** In conclusion, the present study indicates that CrHCl supplementation can improve VJ, power performance, 1RM back squat and hormonal changes in army soldiers.

**Keywords:** creatine, performance, HCl, hormones

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## INTRODUCTION

It has been well documented that the physiological stress of sustained military operations has significant effects to decrement in body mass, strength and power in soldiers [1,2]. In consideration that sustained combat operations or highly intense military training may result in energy deficit, the use of a nutrient intervention to preserve lean mass and physical performance may be desirable [1]. Creatine (Cr) has been demonstrated to enhance recovery and attenuate muscle damage resulting from high intensity exercise. Cr is a popular dietary supplement that is used by athletes to increase muscle mass and strength and especially to improve sports performance [3, 4]. Cr is an organic compound synthesized mainly in the liver and kidneys from the amino acids glycine, arginine and methionine [5]. Cr plays an important role in rapid energy provision during muscle contraction involving the transfer of N-phosphoryl group from phosphorylcreatine to ADP to regenerate ATP through a reversible reaction catalyzed by phosphorylcreatine kinase (PCK) [5]. Phosphocreatine (PCr) plays a key role in energy provision to muscle cell. Dietary supplementation of Cr has been shown to increase muscle levels of both Cr and PCr by 20–50% [6,7].

The main reports from Cr use were on weight gain and water retention [6] with some studies reporting gastrointestinal stress [7,8] and one of these studies reporting a strong correlation between diarrhea and the Cr monohydrate (CrM) doses ingested [8]. These adverse effects of CrM are probably related to the mechanism of action of the substance and the dose used to ensure the efficacy of the supplement. In order to minimize these negative effects, Creatine Hydrochloride (CrHCl) was introduced in the market by Dash et al. [9], which is a molecule that is supposed to be 41 times more soluble in water than CrM. Moreover, it appears that its permeability in the intestinal tract is also greater than CrM [10]. According to Gufford et al. [11] the amount of water to dilute 5 to 10 g of CrM is around 400 to 600 ml while CrHCl would, for the same amount, need 21 ml of water. The authors then propose that greater solubility and permeability could decrease the amount of creatine needed to fill the muscle. That would mean more absorption, less creatine excretion, and less gastrointestinal discomfort. McDonough et al. [12] reported that supplementation with 4 g of CrHCl for 7 days in healthy, resistance trained men significantly increased the number of repetitions performed during intermittent bouts of submaximal bench press and power performance. In addition, de Franca et al. [13] examined the effects of CrHCl supplementation on physical performance and found improvements in strength in recreational weightlifters.

Regarding greater solubility and permeability of CrHCl, it seems that supplementation of this creatine could play an important role in rapid energy provision during muscle contraction in military training in soldiers; however, the data about this subject is limited. In addition, few studies have also examined a CrHCl supplementation on performance and hormonal changes. Therefore, the purpose of this study was to examine the effects of 2 weeks of CrHCl supplementation on physical performance and hormonal changes in male army soldiers.

## MATERIAL AND METHODS

### *Participants*

Eighteen male military soldiers volunteered to participate in this study and were randomly divided into a creatine (CrHCl, n=9, age 21.5±1.1 years, height 174.5±3.7 cm, weight 66.5±4.8 kg) or placebo (Pl, n=9, age 22.3±2.1 years, height 176.5±4.5 cm, weight 68.7±2.9 kg) groups. The sample size was calculated based on a previous study by de Franca et al. [13] with an alpha level of 0.05, and an actual power (1- $\beta$ ) of 0.80. The a priori power analysis was computed using G × Power (Version 3.1.9.2, University of Kiel, Germany) and the t test family. The analysis revealed that a total sample size of N = 9 for each group would be sufficient to find effects of Cr supplementation on hormonal and performance changes. The subjects were free of Cr supplementation at least one year prior to beginning the study which completed with blood assessment. All subjects were required to read and complete a medical history form to ensure that eligibility criteria were met. All subjects were informed of the purpose and possible risks involved in the investigation and were required to read and sign an

informed consent prior to participation. All procedures were approved by the Human Investigation Committees (AUMS, 1398).

### *Procedure*

In a randomized-controlled-placebo, short-term design (i.e., 2 weeks). The subjects reported to the laboratory on 5 days including; 1 session for the familiarization (to eliminate any learning effects on test performance) with testing procedures, 2 sessions for pre-test (i.e., day 1; blood sampling, vertical jump, and Wingate test, day 2; one repetition maximum for the back squat and bench press) and 2 sessions for post-test. There was 24 hours interval between sessions. All tests were performed at 10 AM with 8 to 10 hours of sleep and fasting.

### *Anthropometric measurements*

Height was measured to the nearest 0.1 cm with the use of a wall-mounted stadiometer (Seca 222, Terre Haute, IN). Weight was measured to the nearest 0.1 kg using a medial scale (Camry, EF921, China).

### *Performance measurements*

#### *One repetition maximum*

The one repetition maximum (1RM) for the bench press, and back squat were used for assessing maximal strength which described in detail by Kraemer and Fry [14]. Subjects performed a warm-up a set of 8-10 repetitions with a light weight (approximately 50% of 1RM). A second warm-up consisting of set 3-5 repetitions with a moderate weight (approximately 75% of 1RM), and third warm-up including 1-3 repetitions by a heavy weight (approximately 90% of 1RM). After the warm-up, subjects performed 1RM strength by enhancing the load on consecutive trials until the subject was unable to properly perform a good lift, complete range of motion and correct technique. Three to 5 minutes rest between attempts was provided for each subject.

#### *Power performance*

Wingate anaerobic test (WAnT) was performed in a cycle-ergometric test (Monark E983, Sweden) that lasted 30 sec. Maximal load is accomplished by a built-in air resistance system on the wheels. Load registration was in real time, using a computer with a module for measuring the number of wheel turns. Software backup was provided by a program for graphic registration of load during 30 sec with data memory. This provided a follow-up of the whole test and a quick analysis of the basic variables of anaerobic performance. Also, software provided data related to the quantitative anaerobic power values for every second or period of time during 30 sec of the test. The following variables were determined: peak power (PP), mean power (MP), and also fatigue index (FI) [15]. The subjects performed vertical jump (VJ) test after 20 min of rest following WAnT. This test involves measuring the difference between a person's standing reach and the height recorded from a jump and reach. The difference between the standing height and the jump height is the vertical jump value. Subjects were instructed to perform two-foot vertical jump and peak vertical jump value was recorded in cm [16].

#### *Blood sampling and analysis*

Blood samples were obtained via venipuncture, after five minutes in a supine position, from an antecubital vein by using a 20-gauge needle and vacutainer tubes for the determination of serum testosterone and cortisol concentrations. Blood samples were obtained, pre and after 2 weeks of supplementation before any performance tests, in the morning hours (9 to 10 A.M), and after an 8 to 10 hour overnight fast and sleep and occurred during a standardized time of day for each subject in order to minimize the effects of diurnal variations and circadian rhythm on hormonal changes. The blood was processed and centrifuged, and the resultant serum was stored at  $-80^{\circ}\text{C}$  until analyzed. Total serum testosterone and cortisol were determined in duplicate by using standard radioimmunoassay procedures and were assayed via kits (Monobind kit, USA).

### Supplementation

Cr group consumed 3g CrHCl (Creatine HCl-HydroCrea, GT USA; Resistant Starch, Orion Pharmacy) as well as Pl group consumed 3g dextrose for 2 weeks. The subjects were instructed to mix the 3g CrHCl or dextrose with 200 ml of juice one time per day [13]. The supplement was distributed premixed in resalable baggies containing instructions for consumption for the week. All subjects returned empty baggies on the day of post-testing and verbally confirmed consuming the supplement according to instructions. Subjects were informed not to make any significant changes to their diet during the testing or training program over the course of the study. Subjects were also asked to maintain their normal level of daily activity.

### Statistical analysis

All data are presented as mean  $\pm$  SD. Statistical analysis was performed using SPSS version 21.0 software. A 2 (group)  $\times$  2 (time) analysis of variance (ANOVA) was used to determine significant differences between the groups at pre and post-test. The level of significance was set at  $p \leq 0.05$ .

## RESULTS

There were significant increases in VJ, PP, MP, and 1RM back squat test following 2 weeks of CrHCl supplementation ( $p < 0.05$ ) without any significant change for the Pl group. In addition no significant differences were observed between the groups ( $p > 0.05$ ). No group or group by time interactions ( $p > 0.05$ ) was seen in FI and 1RM bench press following 2 weeks of supplementation period. Significant changes were observed in testosterone and cortisol concentrations from before to after the supplementation in CrHCl group ( $p < 0.05$ ). In addition, CrHCl group indicated significant increases in testosterone and decreases in cortisol concentrations compared with Pl group after supplementation period ( $p = 0.001$ ) (Table 1).

Table 1. Pre to post changes in performance variables and hormones in CrHCl and Pl groups.

Variables	Time	Groups	
		CrHCl	Pl
Vertical jump [cm]	Pre	51.8 $\pm$ 7.5	50.3 $\pm$ 8.7
	Post	55.7 $\pm$ 8.5*	52.3 $\pm$ 13.1
PP [w]	Pre	642.8 $\pm$ 79.4	700.8 $\pm$ 112
	Post	790.3 $\pm$ 75.2*	747.1 $\pm$ 93.6
MP [w]	Pre	293.8 $\pm$ 92.6	308.5 $\pm$ 130
	Post	371.2 $\pm$ 76.1*	335.8 $\pm$ 108
FI [scale]	Pre	56.3 $\pm$ 18.9	55.4 $\pm$ 18.7
	Post	56.6 $\pm$ 7.1	55.3 $\pm$ 12.1
1RM bench press [kg]	Pre	78.6 $\pm$ 8.4	76.6 $\pm$ 13.4
	Post	81.3 $\pm$ 8.6	80.6 $\pm$ 11.4
1RM back squat [kg]	Pre	95.8 $\pm$ 10.2	105.1 $\pm$ 15.9
	Post	104.9 $\pm$ 9.4*	102.7 $\pm$ 11.6
Testosterone [ng/ml]	Pre	0.30 $\pm$ 0.06	0.26 $\pm$ 0.1
	Post	0.36 $\pm$ 0.07*†	0.20 $\pm$ 0.06
Cortisol [ng/ml]	Pre	620.8 $\pm$ 244.7	532.6 $\pm$ 179.1
	Post	396 $\pm$ 72.6*†	614 $\pm$ 135.3

\*significant differences compared with pre value, † significant differences compared with Pl

## DISCUSSION

To our knowledge, this investigation was the first study to examine the effects of CrHCl loading on functional performance and hormonal changes in military soldiers. We found that 2 weeks CrHCl supplementation induced significant increases in VJ, PP, MP, and 1RM back squat test. In addition, testosterone and cortisol concentrations improved from before to after supplementation in CrHCl group, and in comparison to Pl group.

In relation to VJ and power performance (i.e., PP and MP), these findings are in line with previous studies that explored the effects of Cr loading and found increases in power performance [17,18]. Vandenberghe et al [19] compared knee extension torque production during maximal intermittent tests conducted after Cr supplementation, and reported that Cr loading increased muscle phosphocreatine (PCr) content and significantly improved torque production by 5 to 13%. This increase can be availability of PCr into the muscle fibers before the jump and power performance [6]. Moreover, ATP-CP system is major component for realizing energy during power test, that CrHCl loading can enhance these potential [20]. The basics of performance enhancement are such that Cr supplementation increases the storage of muscle Cr, resulting in the production of intramuscular PCr and adenosine triphosphate [21]. Naturally, this process will eventually lead to prolonged vigorous physical activity and improved performance [22]. According to previous reports, the Cr has a buffering effect on muscle acidity. On the other hand, one of the main reasons for discontinuing exercise is the increased generation of lactic acid that is produced following the glycolysis [3,6]. The benefits of increasing the mentioned buffer capacity with Cr usage is that the Cr provides a substrate to the muscle with increased buffer capacity, which can fulfill its work before getting pH-dependent fatigue, although the accumulation of lactic acid is higher the soldiers could perform power exercises with higher ability and tolerance [15,23].

In strength performance, only 1RM back squat increased post supplementation period in the CrHCl group. Rossouw et al. [17] reported significant improvements in 1RM dead lifts after Cr supplementation. These findings are in agreement with the previous study by Volek et al. [23] which found increases in back squat in Cr group than placebo subjects. In contrast, Also, Stevenson and Dudley [17] suggested that Cr loading does not augment unilateral strength or multi-set resistance exercise performance for knee extensions compared with placebo loading. The findings of present study are agreement with Law et al. [24] who found no statistically significant difference in 1RM bench press strength after Cr compared with baseline. But, they found significant increases in 1RM back squat following Cr loading. In the present study, we found that 2 weeks CrHCl loading is required to achieve an increase in muscle strength in military soldiers. It seems that 2 weeks CrHCl supplementation could increase content of muscle PCr, resulting in muscular strength performance enhancements in army soldiers.

The present study examined the effects 2 weeks CrHCl supplementation on hormonal concentrations and found increase in testosterone concentration and decrease in cortisol concentration. Also, CrHCl supplementation could induce significant changes in hormonal variables compared to Pl group. A significant elevation in resting testosterone was observed in young men. The results of current investigation are in agreement with Faraji et al. [25] that examined the effects of Cr loading on testosterone, and cortisol concentrations. In contrast, Volek et al. [26] and/or Op't Eijnde and Hespel [27] found no measurable alteration in endocrine status after Cr supplementation. The possible explanations for this discrepancy could be due to type of Cr consumption, supplementation period, and fitness level of subjects. Testosterone is an important mediator of the adaptations to exercise and plays an important role to the synthesis of muscle contractile proteins and recovery from the training [21]. On the other hand, increases in testosterone following CrHCl supplementation could help the soldiers to training in military exercise and recover faster than others [9]. In fact, enhanced testosterone concentrations in the CrHCl group suggest that Cr supplementation provides a superior anabolic milieu and promote recover period from the training.

In contrast, cortisol is a catabolic hormone and plays an important role in recover period from the exercise. In this study, CrHCl supplementation decreased cortisol levels after 2 weeks which indicate anti-catabolic effects of CrHCl for the soldiers. Indeed, the CrHCl ingestion could prevent from increasing cortisol concentration. Therefore, Cr supplementation for 2 weeks is sufficient for

increasing testosterone and decreasing cortisol level. These changes appear to results in greater increases in anabolic hormone and decreases in catabolic hormone for this supplementation in soldiers to increase recovery time following military training.

## CONCLUSIONS

In summary, the present study indicated that CrHCl supplementation could result in significant improvements in vertical jump, power performance, and 1RM back squat. However, Cr loading was not sufficient for improving 1RM bench press and FI. In addition, 2 weeks CrHCl supplementation induced significant changes in testosterone and cortisol concentrations and compared with Pl group after supplementation period. We recommend that, military soldiers use 2 weeks CrHCl supplementation for improving strength, power performance and hormonal variables to improve practices in military events and better quality with greater recovery in military training.

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## CONFLICT OF INTEREST

There are no conflicts of interest.

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