Low-Dose Creatine Combined with Protein during Resistance Training in Older Men

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[1]Faculty of Kinesiology and Health Studies, University of Regina, Regina, Saskatchewan, CANADA; [2]College of Kinesiology, [3]College of Pharmacy and Nutrition, and [4]Department of Psychiatry, Neuropsychiatry Research Unit, University of Saskatchewan, Saskatoon, Saskatchewan, CANADA

ABSTRACT

CANDOW, D. G., J. P. LITTLE, P. D. CHILIBECK, S. ABEYSEKARA, G. A. ZELLO, M. KAZACHKOV, S. M. CORNISH, and P. H. YU. Low-Dose Creatine Combined with Protein during Resistance Training in Older Men. *Med. Sci. Sports Exerc.*, Vol. 40, No. 9, pp. 1645–1652, 2008. **Purpose:** To determine whether low-dose creatine and protein supplementation during resistance training (RT; 3 d·wk^{-1}; 10 wk) in older men (59–77 yr) is effective for improving strength and muscle mass without producing potentially cytotoxic metabolites (formaldehyde). **Methods:** Older men were randomized (double-blind) to receive 0.1 g·kg^{-1} creatine + 0.3 g·kg^{-1} protein (CP; n = 10), creatine (C; n = 13), or placebo (PLA; n = 12) on training days. Measurements before and after RT included lean tissue mass (air-displacement plethysmography), muscle thickness (ultrasound) of elbow, knee, and ankle flexors and extensors, leg and bench press strength, and urinary indicators of cytotoxicity (formaldehyde), myofibrillar protein degradation [3-methylhistidine (3-MH)], and bone resorption [cross-linked N-telopeptides of type I collagen (NTx)]. **Results:** Subjects in C and CP groups combined experienced greater increases in body mass and total muscle thickness than PLA ($P < 0.05$). Subjects who received CP increased lean tissue mass (+5.6%) more than C (+2.2%) or PLA (+1.0%; $P < 0.05$) and increased bench press strength (+25%) to a greater extent than C and PLA combined (+12.5%; $P < 0.05$). CP and C did not differ from PLA for changes in formaldehyde production (+24% each). Subjects receiving creatine (C and CP) experienced a decrease in 3-MH by 40% compared with an increase of 29% for PLA ($P < 0.05$) and a reduction in NTx (−27%) versus PLA (+13%; P = 0.05). **Conclusions:** Low-dose creatine combined with protein supplementation increases lean tissue mass and results in a greater relative increase in bench press but not leg press strength. Low-dose creatine reduces muscle protein degradation and bone resorption without increasing formaldehyde production. **Key Words:** AGE, EXERCISE, LEAN TISSUE MASS, MUSCLE, STRENGTH, BONE

BASIC SCIENCES

Aging is associated with a decrease in muscle mass and strength (7). Some (3,12,31) but not all (2,14) studies have shown that creatine supplementation during resistance training (RT) increases muscle mass and strength to a greater extent than RT alone in older individuals. Despite its potential benefits, high-dose (21 g·d^{-1}) creatine supplementation in young adults results in an approximately 4.5-fold increase in formaldehyde production (26), which may increase cytotoxicity (41). Cytotoxicity refers to a destructive action on cells. It is hypothesized that formaldehyde may have a destructive effect especially on endothelial cells, which may lead to tissue damage in blood vessels (41). We have previously shown in young adults that low-dose creatine supplementation (i.e., approximately 8 g·d^{-1}) is effective for increasing muscle mass and strength (6), especially when combined with protein supplementation (4). The primary purpose of this study was to determine the effect of low-dose creatine supplementation combined with protein on muscle mass, strength, and formaldehyde production in older individuals. On the basis of the results in younger individuals (4,19), we hypothesized that low-dose creatine (i.e., approximately 8 g·d^{-1}) combined with protein would be effective for increasing muscle mass and strength. We also hypothesized that low-dose creatine supplementation would not result in the same formaldehyde production as has been shown with higher doses of creatine (26).

A secondary purpose of this study was to determine the effects of creatine on markers of myofibrillar protein degradation and bone resorption in older individuals. Creatine decreases protein degradation in younger men (24) and attenuates bone resorption in young boys experiencing Duchenne dystrophy (21,30). However, no study has determined the effects of creatine alone or in combination with protein on these variables in older individuals, who are susceptible to increased muscle protein loss and bone turnover. We hypothesized that creatine supplementation

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would decrease myofibrillar protein degradation and bone resorption in older men.

METHODS

Subjects. Forty healthy older men (59–77 yr), with no previous history of creatine supplementation, volunteered for the study. Subjects were required to fill out a Physical Activity Readiness Questionnaire, which screens for health problems that present a risk with performance of physical activity (32). Subjects that indicated a health problem and all subjects over 69 yr were required to have medical approval before participating in the study. The study was approved by the University of Saskatchewan Biomedical Research Ethics Board for research in human subjects. Subjects were informed of the risks and purposes of the study before written consent was obtained.

Randomization and supplementation. The study used a double-blind repeated measures design in which every subject participated in supervised RT three times per week and were randomized to supplement with creatine and protein (CP) [0.1 g creatine·kg^{-1} contained in 0.45-g·kg^{-1} SyntheVol™ 2 HP; combined with 0.3 g protein·kg^{-1} contained in 0.55-g·kg^{-1} Myoplex®; Experimental and Applied Sciences, Inc., Golden, CO; see Table 1 for complete ingredients; and 0.2 g·kg^{-1} of a chocolate and cherry-flavored sucrose powder], creatine (C) [0.1 g creatine·kg^{-1} contained in 0.45-g·kg^{-1} SyntheVol™ 2 HP and 0.75 g·kg^{-1} of chocolate and cherry-flavored sucrose powder]; or placebo (PLA) [1.2 g·kg^{-1} of chocolate and cherry-flavored sucrose powder], distributed in three equal doses throughout each training day (i.e., 0.4 g·kg^{-1} of supplement powder dissolved in water immediately before their training session, 0.4 g·kg^{-1} of supplement powder dissolved in water immediately after their training session, and 0.4 g·kg^{-1} of supplement powder dissolved in water before going to bed). The supplements were mixed with the chocolate- and cherry-flavored sucrose powder to ensure that supplements and placebo were similar in energy content, taste, texture, and appearance; however, the CP supplement was more nitrogenous because it contained protein. The creatine dose of 0.1 g·kg^{-1} was chosen because we have shown it to be effective when combined with protein (4) or when given on its own in young subjects (6). The protein dose of 0.3 g·kg^{-1} was chosen because it is an approximate amount shown to increase muscle mass during RT (4). Creatine supplementation occurred only on training days because we have previously found this to be effective for increasing lean tissue mass, muscle thickness, and strength in young individuals (11) and daily creatine supplementation in older men (i.e., 0.3 g·kg^{-1}·d^{-1} creatine for 5 d and 0.07 g·kg^{-1}·d^{-1} thereafter for 12 wk) resulted in minor adverse events (i.e., loose stools, muscle cramping, and muscle pulls/strains) (12). All supplementation was double blind. To achieve the double blind, an individual who was not involved in the study was responsible for mixing the supplements into bags and for labeling the bags with subjects' names, according to the randomization list. Compliance with the supplementation protocol was monitored by having subjects return empty supplement bags when picking up additional supplements. Subjects fasted for 3 h before each training session. The supplement was provided immediately before (approximately 5 min) and immediately after (approximately 5 min) each training session, because it has been shown that the timing of either protein and/or creatine ingestion is crucial for creating an anabolic environment for muscle growth, with ingestion immediately before (13,34) and immediately after RT (11,13,15) appearing optimal.

TABLE 1. Ingredients in Myoplex® and SyntheVol™ 2 HP supplements (g·kg^{-1}·d^{-1}).

Ingredient	Myoplex®	Ingredient	SyntheVol™ 2 HP
Protein	3.0×10^{-1}	Protein	4.0×10^{-2}
Carbohydrates	1.7×10^{-1}	Carbohydrates	2.7×10^{-1}
Fat	1.8×10^{-2}	Aspartame	1.5×10^{-3}
Cholesterol	1.0×10^{-4}	Creatine monohydrate	1.0×10^{-1}
Sodium	2.9×10^{-3}	Glutamine peptides	6.0×10^{-2}
Calcium	3.6×10^{-3}	Inzitol® (D-Pinitol)	8.0×10^{-4}
Iron	1.0×10^{-5}		
Vitamin A	2.1×10^{-6}		

Experimental design. Before the first visit to the laboratory for initial testing and data collection, all subjects were instructed to refrain from physical activity for 48 h and not change their diet before or during the RT program. The dependent variables measured before and after the 10 wk of supplementation and training were the following: lean tissue mass; muscle thickness of flexors and extensors of the elbow, knee, and ankle; strength [leg press and bench press one repetition maximum (1-RM)]; urinary 3-methylhistidine excretion (3-MH; an indicator of myofibrillar protein degradation); urinary cross-linked N-telopeptides of type I collagen (NTx, an indicator of bone resorption); and urinary formaldehyde (an indicator of cytotoxicity). In addition, subjects completed dietary records for 3 d during the first and final week of RT and supplementation to assess energy and macronutrient differences between groups. At the end of the study, a treatment identification questionnaire was administered as a test of our blinding success to determine whether subjects perceived they were on creatine, placebo, or were unsure what supplement they consumed.

Detailed procedures for the RT program (8), lean tissue mass (8), muscle thickness (7), muscle strength (12), myofibrillar protein degradation and bone resorption (25), and diet (8) were previously described; therefore, only a brief description is provided.

RT program. Before each training session, but after the supplement drink was consumed, each subject warmed up on a stationary bicycle for 5–10 min and completed light stretching. Subjects trained 3 d·wk^{-1} for three sets of 10 repetitions with 2-min rest between sets. An initial intensity corresponding to approximately 70% 1-RM was used for the leg press and bench press, and a weight corresponding to individual 10-repetition maximum was used for other

exercises. Resistance was increased once an individual was able to complete the required number of repetitions for an exercise with good form. Resistance exercises performed in order were bench press, leg press, lat pull down, leg (knee) extension, shoulder press, leg curl, biceps curl, calf press, and triceps extension. Subjects maintained daily training logs where average training volume per session (weight × sets × repetitions) was determined for each subject.

Lean tissue mass. To estimate lean tissue mass, air-displacement plethysmography (Bod Pod S/L; Life Measurement, Inc., Concord, CA) was used. Reproducibility was assessed by testing 16 subjects 1 wk apart. The coefficient of variation (CV) was 0.87%, the intraclass correlation coefficient (ICC) was 0.99, and the SEM was 0.84 kg.

Muscle thickness. Elbow, knee, and ankle flexor and extensor muscle thickness was measured using B-mode ultrasound (Aloka SSD-500, Tokyo, Japan). Reproducibility of muscle thickness measurements was determined by testing 16 subjects 1 wk apart and for each site was as follows: elbow flexors (CV = 2.6%; ICC = 0.96; SEM = 0.15 cm), elbow extensors (CV = 2.1%; ICC = 0.88; SEM = 0.18 cm), knee flexors (CV = 2.3%; ICC = 0.99; SEM = 0.14 cm), knee extensors (CV = 2.1%; ICC = 0.99; SEM = 0.15 cm), ankle plantar flexors (CV = 3.1%; ICC = 0.98; SEM = 0.21 cm), and ankle dorsi flexors (CV = 4.0%; ICC = 0.87; SEM = 0.18 cm).

Muscle strength. Leg press and bench press strength were assessed using a 1-repetition maximum standard testing procedure before and after supplementation and RT. Reproducibility of the strength measures was assessed on 10 subjects, 1 wk apart. The leg press had a CV of 3.8%, ICC of 0.99, and SEM of 6.0 kg, and bench press had a CV of 3.1%, ICC of 0.99, and SEM of 3.2 kg.

Myofibrillar degradation and bone resorption. Myofibrillar protein degradation was assessed by 3-MH, and bone resorption was assessed from NTx from 24-h urine samples collected before and immediately after the 10 wk of training and supplementation. Urine collection was preceded by a 3-d meat-free diet. Meat consumption affects 3-MH levels, and at least 3 d of a meat-free diet are required to return urine concentrations of 3-MH to baseline levels (22). The concentration of 3-MH was determined by high-performance liquid chromatography (3-mm Chromsep ODS-2 column; Varian, Inc., Mississauga, Ontario, Canada; flow rate 1.0 $mL \cdot min^{-1}$) and 2475 multiwavelength fluorescence detection (Waters, Mississauga, Ontario, Canada) using the methods of Wassner et al. (36), with modification for sample volumes. The concentration of 3-MH in urine samples was corrected for creatinine content [modified Jaffe reaction; see Burke et al. (5)] and multiplied by 24-h urine volume to produce a value for daily 3-MH excretion. The intra-assay CV for duplicate samples was 5%, the ICC was 0.99, and the SEM was 0.9 $\mu mol \cdot L^{-1}/mmol \cdot L^{-1}$ creatinine.

The concentration of NTx was determined using a competitive-inhibition enzyme-linked immunosorbent assay (ELISA, Osteomark NTx test; Ostex International, Inc., Seattle, WA). Samples were analyzed in duplicate within a single assay. The concentration of NTx in urine samples [expressed as bone collagen equivalents (BCE)] was corrected for urinary creatinine and multiplied by 24-h urine volume to produce a value for daily NTx excretion. The intra-assay CV was 9%, the ICC was 0.99, and the SEM was 2.3 $nmol \cdot L^{-1}$ BCE/$mmol \cdot L^{-1}$ creatinine.

Formaldehyde. Urinary formaldehyde concentration was assessed by high-performance liquid chromatography using previously described procedures (42). Daily formaldehyde excretion was determined by multiplying formaldehyde concentration by 24-h urine volume. The intra-assay CV was 1.3%, the ICC was 0.99, and the SEM was 6.9 μg.

Dietary intake. Dietary intake was recorded for 3 d during the first and final week of supplementation and RT to assess differences in total energy and macronutrient composition between groups over time. Dietary intakes were recorded on days that were different from the 3-d meat-free diet imposed for the 3-MH assessment. The Interactive Healthy Eating Index (Center for Nutrition Policy and Promotion, USDA) was used to analyze 3-d food records.

Statistical analyses. A 3 (CP vs C vs PLA) × 2 (pre- and posttraining) ANOVA with repeated measures on the second factor was used to assess changes in lean tissue mass, muscle thickness, strength, 3-MH, NTx, formaldehyde, and diet (energy and macronutrient contents) over time. To simplify the presentation of our results, we determined the absolute changes in dependent variables for each group as the difference between pre- and posttraining scores and the relative changes as the difference between pre- and posttraining scores divided by the pretraining score and multiplied by 100. A one-factor ANOVA was used to determine differences in change scores between groups. A one-factor ANOVA was also used to determine differences in average training volume (kg × sets × reps) per session between groups and to determine whether there were differences in baseline measurements between groups. As a follow-up to ANOVAs, we performed contrast analyses for the change scores between groups to determine whether the creatine groups combined (i.e., CP + C) differed from the placebo group and whether the CP group differed from the groups that did not receive protein (i.e., C + PLA). A least significant difference *post hoc* test was used to identify differences between means when interactions were found. All results are expressed as mean ± SE. Statistical analyses

TABLE 2. Mean ± SE subject characteristics at baseline for the CP, C, and PLA groups.

Group	Age (yr)	Mass (kg)	Height (cm)
CP (n = 10)	67.3 ± 3.1	82.5 ± 2.8	177.2 ± 2.1
C (n = 13)	65.5 ± 2.7	86.0 ± 5.4	175.4 ± 2.3
PLA (n = 12)	64.1 ± 3.1	82.9 ± 2.9	178.3 ± 1.3

There were no differences between groups at baseline.

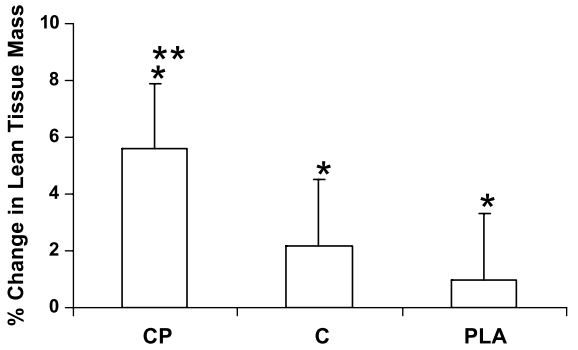

FIGURE 1—Change in lean tissue mass after 10 wk of supplementation and RT for CP (n = 10), C (n = 13), and PLA (n = 12) groups. Values are mean ± SE. *All groups increased lean tissue mass with training ($P < 0.05$). **CP increased lean tissue mass more than the C and PLA groups ($P < 0.05$).

were carried out using Statistica 7.0 (StatSoft, Tulsa, OK). Significance was set at $P \leq 0.05$.

RESULTS

Of the original 40 subjects who volunteered, 35 subjects completed the study. Two subjects from the PLA group withdrew because of shoulder and hip pain, one subject in the CP group withdrew because of gall bladder surgery, and two others in the CP group withdrew because of time constraints. Baseline characteristics of subjects who completed the study are shown in Table 2. There were no differences between groups in any of the baseline measurements. Supplementation compliance, on the basis of the number of empty supplement bags returned, was similar (CP = 94%, C = 93%, PLA = 95%) between groups.

There was a significant group × time interaction for lean tissue mass ($P < 0.05$). *Post hoc* analysis indicated that lean tissue mass significantly increased in all groups with training ($P < 0.05$; Fig. 1). The gain was greater in the CP group (3.2 ± 0.6 kg or 5.6 ± 0.9%) compared with C (2.1 ± 0.4 kg or 2.2 ± 0.8%) and PLA groups (0.6 ± 0.6 kg or 1.0 ± 1.0%; $P < 0.05$). The increase in body mass in the subjects who supplemented with creatine (i.e., C and CP groups combined) was greater (+0.6 ± 0.4 kg or +0.8 ± 0.4%) compared with PLA (−0.7 ± 0.5 kg or −0.8 ± 0.5%; $P < 0.05$).

A significant increase in muscle thickness for all muscle groups ($P < 0.05$; Table 3), except the ankle dorsi flexors, was observed for all groups with training. The increase in total muscle thickness for the six muscle groups combined was significantly greater in the subjects who supplemented with creatine (C and CP groups combined; +2.0 ± 0.3 cm) compared with PLA (+0.8 ± 0.3 cm; $P < 0.05$).

There was a significant time main effect ($P < 0.05$) for leg press and bench press strength with training (Figs. 2A and B). The increases in leg press 1-RM for the CP, C, and the PLA groups were 20 ± 6 kg or 14 ± 4%, 20 ± 5 kg or 12 ± 3%, and 21 ± 5 kg or 12 ± 3%, respectively. The increases for bench press 1-RM for the CP, C, and PLA groups were 19 ± 4 kg or 25 ± 5%, 11 ± 3 kg or 12 ± 3%, and 9 ± 4 kg or 13 ± 5%, respectively. In the contrast analysis, the relative increase in bench press strength in the CP group was greater than the other groups (C + PLA) combined ($P < 0.05$).

There were no differences over time between the three groups for changes in 3-MH, NTx, or formaldehyde (Table 4). Contrast analyses indicated the subjects who supplemented with creatine (C and CP groups combined) experienced greater relative decreases in 3-MH and NTx compared with the PLA group ($P \leq 0.05$; Figs. 3 and 4). There were no significant differences between groups for changes in urinary formaldehyde production (Fig. 5).

There were no differences in average training volume per session between the CP, C, and PLA groups. The CP group had a mean ± SE volume of 14,249 ± 1736 kg per training session; the C group had a mean ± SE volume of 14,748 ± 1298 kg per session, whereas the PLA group had a mean ± SE volume of 15,145 ± 1156 kg per session. Dietary intake did not differ significantly between groups and did not differ significantly over the course of training (Table 5).

There were no differences between groups for reports of adverse events considered related to the supplement. Two subjects in the PLA group and one subject in the C group reported increased muscle soreness and stiffness during weeks 4 through 7 of the RT program. Correct treatment

TABLE 3. Muscle thickness measurements (cm) for the flexor and extensor muscles surrounding the elbow, knee, and ankle in older men before and after 10 wk of supplementation and RT.

	CP (n = 10)			C (n = 13)			PLA (n = 12)		
Muscle Group	**Pre**	**Post**	**%**	**Pre**	**Post**	**%**	**Pre**	**Post**	**%**
Elbow flexors	2.9 ± 0.2	3.1 ± 0.2*	9.0 ± 6	2.8 ± 0.2	3.0 ± 0.2*	8.8 ± 3.7	3.0 ± 0.2	3.2 ± 0.1*	8.0 ± 5.3
Elbow extensors	4.3 ± 0.3	4.7 ± 0.3*	11.6 ± 5	4.1 ± 0.2	4.5 ± 0.2*	11.4 ± 4.3	4.7 ± 0.1	4.8 ± 0.2*	1.4 ± 3.6
Knee flexors	4.9 ± 0.3	5.2 ± 0.2*	7.0 ± 5.2	4.5 ± 0.2	4.8 ± 0.2*	9.4 ± 3.6	4.8 ± 0.2	4.9 ± 0.2*	3.2 ± 2.9
Knee extensors	3.3 ± 0.3	3.8 ± 0.3*	13.6 ± 4.7	3.3 ± 0.2	3.6 ± 0.2*	11.3 ± 3.7	3.7 ± 0.3	3.9 ± 0.2*	5.8 ± 3.6
Ankle plantar flexors	3.8 ± 0.3	4.3 ± 0.2*	14.1 ± 5.0	3.5 ± 0.2	3.9 ± 0.3*	13.8 ± 4	3.9 ± 0.2	4.2 ± 0.2*	8.0 ± 6.4
Ankle dorsi flexors	2.3 ± 0.1	2.5 ± 0.3	4.6 ± 5.1	2.1 ± 0.1	2.3 ± 0.2	7.9 ± 6.7	2.3 ± 0.2	2.4 ± 0.2	6.8 ± 5.6
Average total change			10 ± 5.2**			10.4 ± 4.3**			5.5 ± 4.6

Values are mean ± SE. %, percent change over time.
* Significantly different after training ($P < 0.05$).
** Combined CP and C groups had a greater % change versus the PLA group after training ($P < 0.05$).

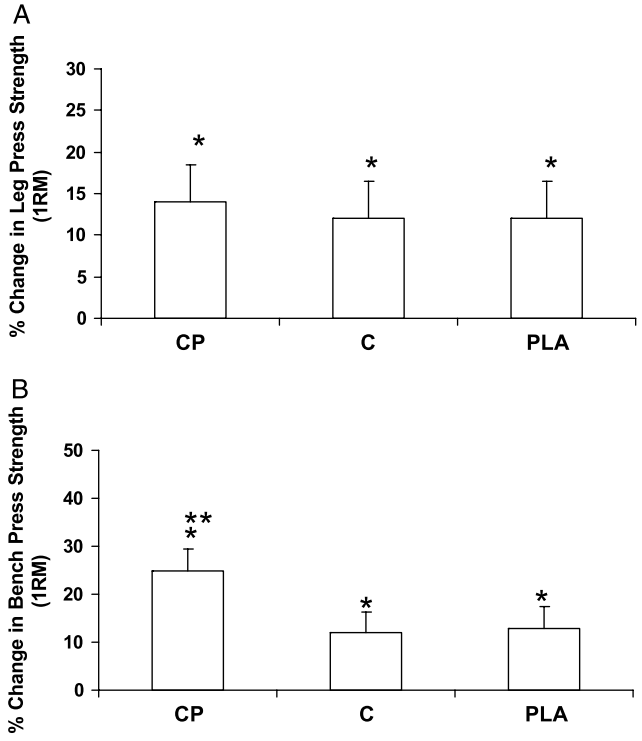

FIGURE 2—A. Change in leg press strength (1-RM) after 10 wk of supplementation and RT for CP (n = 10), C (n = 13), and PLA (n = 12) groups. Values are mean ± SE. *All groups increased strength with training (P < 0.05), with no differences between groups. B. Change in bench press strength (1-RM) after 10 wk of supplementation and RT for CP (n = 10), C (n = 11), and PLA (n = 12) groups. Values are mean ± SE. *All groups increased strength with training (P < 0.05). **CP had a greater increase in strength over C and PLA groups combined (P < 0.05).

identifications for the CP, C, and PLA groups were 10%, 8% and 17%, respectively.

DISCUSSION

To our knowledge, this is the first study to examine the effects of creatine combined with protein supplementation in healthy older men. On the basis of our previous findings in young men (4), we hypothesized that creatine and protein supplementation during RT would increase lean tissue mass and strength over creatine or placebo. Our results showed that subjects who supplemented with creatine experienced greater gains in body mass and muscle thickness, and the combination of protein and creatine increased lean tissue mass to a greater extent than creatine alone or placebo. Creatine and protein led to a greater gain in bench press strength when compared with the other two groups combined; however, there were no differences between groups for changes in leg press strength.

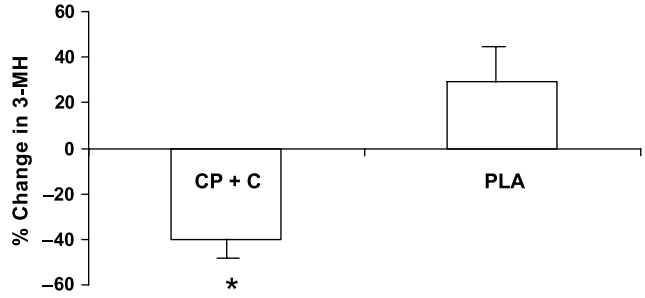

FIGURE 3—Change in 3-MH after 10 wk of supplementation and RT for CP (n = 10), C (n = 13), and PLA (n = 12) groups. Values are mean ± SE. *Creatine groups combined had a greater % decrease versus the PLA group after training (P < 0.05).

Our findings of a significant increase in lean tissue mass from creatine and protein supplementation in older men are in agreement with previous work in young men (4). Creatine has been shown to increase *in vitro* biosynthesis of muscle myosin (18), up-regulate satellite cell activity (23), and increase transcription factors that are involved in muscle hypertrophy (38). Creatine has also been shown to increase muscle fiber area (5,35) and reduce whole body protein breakdown (24). Orally administered amino acids enhance the ratio of protein synthesis to protein degradation after exercise (33). Creatine combined with protein supplementation, therefore, has potentially additive effects for muscle protein accumulation, and our results for increased lean tissue mass support this. A limitation of our study was that muscle biopsies were not collected for the above measures, and some of the increase in lean tissue mass may be caused by intracellular water retention (29).

The results of the current study in older men are similar to studies of younger men who used higher doses of creatine and protein, indicating that the adaptation of older individuals is comparable to if not greater than young. Older men who were supplemented with creatine (approximately 8 g) and protein (approximately 30 g) three times per week increased lean tissue mass by 3.2 kg or 5.6% (current study) compared with 4 kg or 6.5% experienced by young men who supplemented with a similar dose of creatine but a much higher dose of protein (approximately 96 g) everyday during 6 wk of RT (4). The increase in lean tissue mass in the older men who supplemented with creatine and protein in the present study was greater than

TABLE 4. Mean ± SE values for urinary 3-MH, NTx, and formaldehyde before and after supplementation for CP, C, and PLA.

	3-MH (μmol·L^{-1}/mmol·L^{-1} Crn)		NTx (nmol·L^{-1} BCE/mmol·L^{-1} Crn)		Formaldehyde (μg)	
Group	Pre	Post	Pre	Post	Pre	Post
CP	16.1 ± 3.2	10.1 ± 3.3	56.2 ± 9.1	35.1 ± 5.4	267 ± 41.3	306 ± 50.1
C	21.2 ± 6.2	12.2 ± 4.1	58.5 ± 8.0	45.1 ± 6.4	230.3 ± 38.2	209 ± 32.5
PLA	14.2 ± 2.4	14.4 ± 3.3	50.6 ± 7.2	47.4 ± 10.3	355 ± 62.5	327 ± 42.4

There were no differences over time between groups.
Crn, creatinine; BCE, bone collagen equivalents.

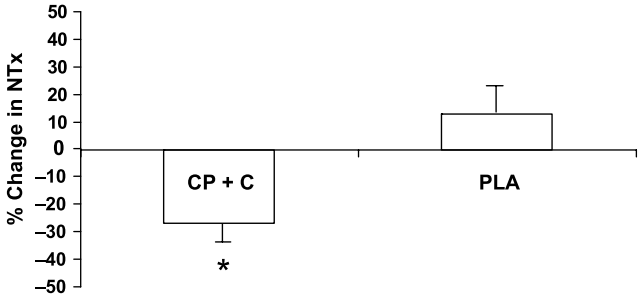

FIGURE 4—Change in NTx after 10 wk of supplementation and RT for CP (n = 7), C (n = 13), and PLA (n = 11) groups. Values are mean ± SE. *Creatine groups combined had a greater % decrease versus the PLA group after training (P = 0.05).

the increase observed in young men (2.0 kg or 3.2%) ingesting a commercial creatine (approximately 20 g) and protein (approximately 67 g) supplement [Phosphagain™; Experimental and Applied Sciences, Inc., Golden, CO; see Kreider et al. (20) for a complete list of ingredients] everyday during 4 wk of RT (20) and greater than young adults (1.6 kg or 2.8%) supplementing with creatine (3 g) and protein (74 g) everyday during 12 wk of RT (19). A unique aspect of the present study was that supplementation occurred only on training days. Justification for this protocol was based on the recent study of Chilibeck et al. (11), who found that supplementing with creatine only on training days was effective for increasing lean tissue mass and muscle thickness in young men. Also, protein requirements for the elderly are probably not increased above normal dietary levels on nontraining days (37). Therefore, it is only necessary to provide protein supplementation on training days to augment muscle protein synthesis (15). On the basis of our results, older men experience similar gains in lean tissue mass as their younger counterparts when supplementing with a lower dose of creatine and protein on training days during an RT program. This is important as compliance may be higher and costs lower when smaller, less frequent doses of supplement are consumed.

There was no greater effect from creatine supplementation by itself on lean tissue mass over placebo. This is similar to several studies that showed no effect of creatine on lean tissue mass over 30 d to 1 yr in older individuals (2,14,27,28). However, Brose et al. (3) found a significant increase in lean tissue mass (1.7 ± 1.2 kg) in older adults from creatine supplementation (5 $g \cdot d^{-1}$) during 14 wk of RT and Gotshalk et al. (17) found a significant increase in fat-free mass (2.2 kg) after 7 d of creatine supplementation (0.3 $g \cdot kg^{-1} \cdot d^{-1}$) in older men (59–77 yr). Previous work in our laboratory reported a significant increase in lean tissue mass of 3.3 ± 1.6 kg after 12 wk of creatine supplementation and RT in older men (12). These inconsistent results between studies are possibly due to methodological differences including different doses, different lengths of training, and different training programs. Creatine in the current study was only given on training days (3 $d \cdot wk^{-1}$) at a relatively low dose (approximately 8 $g \cdot d^{-1}$). This may have been insufficient for increasing lean tissue mass.

Although we were unable to detect a significant increase in muscle strength after creatine and protein supplementation when all three groups were compared, there was an increase in the CP group in contrast analysis to the other two groups combined. Our results of no greater increase in lower body strength from creatine and protein supplementation are similar to our findings in young men (4). One possible explanation may involve the complexity of the exercise involved. Complex exercises (i.e., leg press that involves movement at three joints) involve greater learning and coordination (9); therefore, the observed increases in strength may have been due to neural adaptations and learning, as all three groups increased strength over time.

The changes in 3-MH, an indicator of myofibrillar protein degradation, and NTx, an indicator of bone resorption, were similar between groups over time. Our lack of a statistically significant difference between groups may have been due to a lack of power. However, when we compared relative changes for subjects who supplemented with creatine (i.e., combined C and CP groups) to subjects who received placebo, there was a significant 40% decrease in 3-MH from creatine compared with an increase of 29% for the placebo and also a greater decrease in NTx (−27% for C group vs +13% for PLA group). Creatine supplementation has been shown to reduce NTx in young boys experiencing muscular dystrophy (21,30) and to increase lumbar bone mineral density in growing rats (1) and arm bone mineral content in older men during RT (10). In contrast, 6 months of creatine supplementation in combination with conjugated linoleic acid failed to decrease NTx or change bone mineral density in older men and women (31). Bone cells such as osteoblasts are heavily reliant on creatine kinase for energy and bone formation (1). Creatine supplementation increases the metabolic activity of rat osteoblastlike cells (16). Along with increasing bone formation, stimulating osteoblast activity would increase production of compounds (i.e., osteoprotegerin) that inhibit osteoclast activity and reduce bone resorption (39). On the basis of these findings, creatine has the potential to have a favorable effect on bone health.

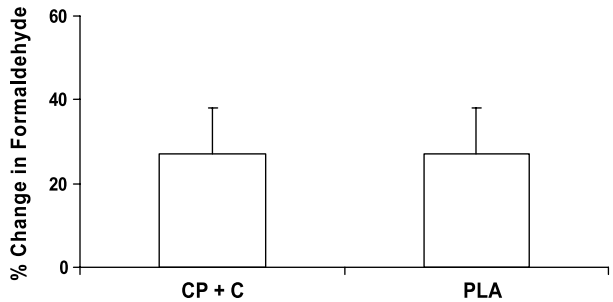

FIGURE 5—Change in formaldehyde after 10 wk of supplementation and RT for CP (n = 10), C (n = 13), and PLA (n = 12) groups. Values are mean ± SE.

TABLE 5. Mean ± SE dietary variables ($g \cdot d^{-1}$) for CP, C, and PLA groups averaged over 3 d during the first and final week of supplementation and RT.

	CP (n = 9)		C (n = 10)		PLA (n = 9)	
	Week 1	Week 10	Week 1	Week 10	Week 1	Week 10
Kilocalories per day	2543 ± 232	2368 ± 122	2558 ± 198	2513 ± 204	2162 ± 126	2195 ± 128
Carbohydrates	282 ± 33	248 ± 20	294 ± 27	275 ± 38	232 ± 18	214 ± 18
Fat	105 ± 14	102 ± 7	104 ± 13	106 ± 10	86 ± 11	96 ± 6
Protein	103 ± 9	104 ± 9	118 ± 11	120 ± 6	92 ± 6	101 ± 9

All measurements exclude the macronutrients contained in the supplements.

The metabolic degradation of creatine produces formaldehyde (41). Formaldehyde has the potential to be carcinogenic and to cross-link proteins and DNA (41). High-dose (21 $g \cdot d^{-1}$), short-term (14 d) creatine supplementation increased formaldehyde production by 348% in young adults (26); however, the increase was below the upper limit range considered safe for a healthy population. Our results show that low-dose creatine supplementation (approximately 8 $g \cdot d^{-1}$) on training days in older men did not increase formaldehyde production as urinary formaldehyde excretion did not increase significantly from before to after supplementation, and there were no differences between the creatine or placebo groups (Fig. 5). It is unknown whether exercise training or other dietary factors can affect formaldehyde levels. Formaldehyde production is affected by increased adrenaline levels (43) and by nicotine (40); therefore, stress levels and smoking may contribute to the variability in formaldehyde production. We were aware of only one subject, in the creatine group, who was a smoker in the current study; therefore, this would not have a large effect on our results.

In conclusion, older men who supplemented with creatine during a 10-wk supervised RT program experienced significant increases in body mass and muscle thickness, and the addition of protein to creatine further increased lean tissue mass. Creatine and protein were beneficial for improving bench press but not leg press strength. Creatine supplementation seems to exhibit anticatabolic properties on muscle and bone in older men during RT with no increases in formaldehyde. Future research should examine the safety and potential of long-term creatine supplementation during RT on muscle and bone health.

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