Georgia Tech Sponsored Research

Project	B-16-649
Project director	Millard-Stafford Melinda
Research unit	HLTH SCI
Title	Effect of Creatine Supplementation on Body Composition and Risk of Injury in Male and Female
Project date	8/1/1999

B-16.649

EFFECT OF CREATINE SUPPLEMENTATION ON BODY COMPOSITION AND RISK OF INJURY IN MALE AND FEMALE COLLEGIATE ATHLETES

Interim Report to the NCAA
January 4, 1998

Principal Investigator: Mindy Millard-Stafford, Ph.D.
Department of Health & Performance Sciences
750 Ferst Drive
Georgia Institute of Technology, Atlanta, GA 30332-0110
(404) 894-6274
FAX: (404) 894-9982
Email: mindy.millardstafford@sac.gatech.edu

Purpose. The purpose of this study is: 1) to assess the specific changes in body composition (i.e. changes in protein, water, and mineral fraction of the fat free mass) which occur with long term (12 weeks) Cr supplementation; and 2) to determine the relative risk of injury and alterations in blood lipids, liver and kidney function with long term (3 months) Cr supplementation in male and female athletes.

METHODS

Subjects. To date, twenty-seven healthy college student-athletes (19 males and 8 females) have served as volunteers for the study. By sport, there were six football, eight swimming, four softball, five track and field, one basketball, and three baseball athletes. Since only 25% of the Georgia Tech student population is female, our total number of female athletes is lower than many Division I-A schools and therefore, it was not possible to recruit a similar number of women (n=8) as men (n=19) for this study. All subjects were engaged in their sport-specific weight training program (3 to 4 days/ week) during the 12 weeks of the study and had never previously used Cr. This stipulation made it extremely difficult to recruit varsity athletes who had either: a) never used creatine monohydrate previously, or b) were willing to volunteer as a subject in a research study. Therefore, most subjects were incoming freshmen who had not used creatine previously in high school, and even this yielded a relatively small pool of potential subjects (i.e. most incoming athletes had either taken it in high school or wanted to start supplementation on their own -not as a part of a study). In addition, coaches in the different sports had wide-ranging opinions and willingness to allow their athletes' participation. For example, football and swimming coaches wanted all their athletes to be on Cr while the men's track coach forbid their athletes from using Cr. Therefore, in the second phase of testing we will be extending the subject population to club sport athletes at Georgia Tech in order to achieve a total population of 40 subjects. We also hope to continue monitoring several athletes from the Fall throughout the Winter quarter as funding/scheduling permits. Written informed consent was obtained from all subjects prior to their participation as stipulated by the Institutional Review Board.

Testing protocol. Physical characteristics, anthropometric measurements, and body density were measured prior to the dietary treatment and 12 weeks following the dietary treatment. Subjects reported to the laboratory after an overnight fast (8 hrs) without eating solid food and beverage except water. Subjects were instructed to ingest additional fluid the night before testing to ensure euhydration. No physical exercise was performed 12 hours before testing. No food or fluid was ingested during the morning test session.

Dietary treatment. Using a double-blind, randomized design, subjects were divided into three diet treatment groups. None of the strength coaches, athletic trainers or athletes knew which treatment had been assigned. During the 5-day loading period, the Cr supplement used was micronized creatine monohydrate (MetRx, Irvine, CA) which was mixed in warm water along with 10 grams of powdered sports drink (PowerAde, The Coca Cola Company, Atlanta, GA) and administered by a member of the research staff in order to ensure subject compliance. Using this procedure, it was virtually impossible to distinguish between a creatine or placebo supplement. During the 11-week maintenance

week period, 5 gms of powdered creatine (Perfect Creatine, Nature's Best, Hauppauge, NY) was mixed along with approximately 4 gms of sports drink powder. Subjects received each days' supplement in individual plastic bags and were instructed to consume the powder straight from the package. In only one subject, was this objectionable and so this subject was permitted to mix her daily powdered dose with water. Subjects reported to the lab each week to received the next week's supplements. The placebo group (n=8) served as a control group (PLACEBO) who consumed a sports drink solution four times daily (with no Cr) for the first 5 days and, thereafter, only 9 gms/ day of dry, powdered sports drink for the remaining 11 weeks of the study. The second group (n=9) consumed Cr (at a dosage equivalent to 0.3 gm/kg of body weight per day) four times daily in the same sports drink solution for five days (Load), and thereafter ingested the sports drink powder once a day (without Cr). The third group (n=10) followed the same procedure as the Load group for the 5-day loading phase but during the maintenance period ingested 5 gms of Cr mixed with the sports drink powder (CrL+M). Since most athletes were aware of the ergogenic potential of creatine supplementation; thus, we informed them that the intent of the study was to evaluate different dosage regimens of creatine supplementation.

RESULTS

At present, only the blood data and preliminary injury reports and body composition measures have been completed on the 27 athletes. We are still in the process of completing all the post-test DXA scans as well as the plasma purification process for total body water analysis.

Anthropometry. There were no differences in age, height, weight or % fat between the three groups prior to the intervention. Body weight increased in both Cr Load + Maintenance and Load group by 3.7 and 2.4 lbs., respectively and decreased by 0.7 lbs. for the placebo group. The % body fat (via air displacement plethysmography) was decreased by 0.5%,0.4%, and 1.1% for the Cr Load + Maintenance, Load, and Placebo groups, respectively.

Blood parameters. Creatine supplementation (either Load or Load + Maintenance) had no significant effect in the following parameters: uric acid, blood urea nitrogen, total protein, albumin, globulin, albumin/globulin ratio, bilirubin, and liver/ kidney enzymes (alkaline phosphatase, LDH, SGOT, SGPT, GGT). There was a significant group x time interaction effect for creatinine. Mean values for Cr Load + Maintenance increased from .94 to 1.2 compared to .85 to 1.07, and 1.1 to 1.2 for Load and Placebo, respectively. There were no group differences in changes in HDL, Triglyceride or Cholesterol/HDL-C ratio. However, total cholesterol and LDL were significantly higher in the Cr Load + Maintenance group both before and after supplementation compared to placebo. If this pre-test difference persists upon completion of the data set, we will use an analysis of covariance to control for pre-existing group differences.

Subjective ratings. A 10 cm Visual Analog Scale was used to quantify subjective ratings related to muscle cramping, stomach upset, joint stiffness, change in urination

frequency, and level of muscle soreness. Following the 5 day loading period, there were no differences in subjective ratings for urine frequency, stomach upset, muscle soreness or muscle cramping between the groups. However, the placebo group had significantly higher joint stiffness ratings compared to the Cr Load + Maintenance group.

There were few individual complaints during the study. However, we had one subject (pitcher) who complained about tightness in his throwing arm after the 5 day loading period and so he requested to discontinue the Cr supplementation regimen. He was retained as a member of the Load only group. We had one football player that also chronically complained of cramping in various muscle groups during both exertion and non-exertional circumstances who was in the Cr Load+ Maintenance group but he did complete his 12 week supplementation regimen. Interestingly, he was the only subject who also had blood creatinine values above the normal range. We will try to follow up with him after another 12 week wash-out period where no supplements are ingested. We also had one Placebo subject complain of stomach upset with the sports drink powder ingestion (mentioned previously), so we allowed her to mix the powder with water for ease of ingestion. This appeared to remedy the situation.

Thus, out of 27 subjects, we had only two subjects who expressed a desire to not continue with supplementation. The remaining 25 either reported ambivalence (didn't think it helped or hurt them) or that Creatine improved their training, recovery from training and /or body composition. We will continue to refine these subjective ratings with appropriate non-parametric statistical analyses.

PROJECTED TIMELINE

As originally proposed, the start date for data collection was August 30,1998 (following the football pre-participation physical) and continued through September 21 (via staggered season starting times). We will begin our second phase of testing on January 7, 1999 and conduct post-testing in April 1999. The final plasma purification processing, data analysis will be conducted for two months following the last subject testing dates, thus we project a final report submitted to NCAA early July 1999 (approximately one month ahead of schedule).

B-16-649
#2

EFFECT OF CREATINE SUPPLEMENTATION ON BODY COMPOSITION AND RISK OF INJURY IN MALE AND FEMALE COLLEGIATE ATHLETES

Final Report

Submitted to the NCAA

July 16, 1999

Principal Investigator: Mindy Millard-Stafford, Ph.D.
Department of Health & Performance Sciences
750 Ferst Drive
Georgia Institute of Technology, Atlanta, GA 30332-0110
(404) 894-6274
FAX: (404) 894-9982
Email: mindy.millardstafford@sac.gatech.edu

The purpose of this study was: 1) to assess the specific changes in body composition (i.e. changes in protein, water, and mineral fraction of the fat free mass) which occur with long term (12 weeks) creatine (Cr) supplementation; and 2) to determine the incidence of injury and alterations in blood lipids, liver and kidney function with long term (3 months) Cr supplementation in male and female athletes.

METHODS

Subjects. Forty-eight healthy college student-athletes (29 males and 19 females; 27 varsity athletes and 21 club sport/recreational weight trainers) served as volunteers for the study. Since only 25% of the Georgia Tech student population is female, our total number of female athletes is lower than many Division I-A schools and therefore, it was not possible to recruit a similar number of females as males. By varsity sport, there were six football, eight men's swimming, four softball, five women's track and field, one men's basketball, and three baseball athletes. All subjects were engaged in their sport-specific weight training program (3 to 4 days/ week) during the 12 weeks of the study and had never previously used Cr. This stipulation made it extremely difficult to recruit varsity athletes who had either: 1) never used creatine monohydrate previously, or 2) were willing to volunteer as a subject in a research study. Therefore, most subjects were incoming freshmen who had not used creatine previously in high school, and even this yielded a relatively small pool of potential subjects (i.e. most incoming athletes had either taken it in high school or wanted to start supplementation on their own -not as a part of a study). In addition, coaches in the different sports had wide-ranging opinions and willingness to allow their athletes' participation. For example, football and swimming coaches wanted all their athletes to be on Cr while the men's track coach forbid their athletes from using Cr. Therefore, with the second phase of testing we extended the subject pool to include club sport athletes and recreational weight trainers at Georgia Tech. Written informed consent was obtained from all subjects prior to their participation as stipulated by the Institutional Review Board.

There were no significant differences ($p > 0.05$) in age or height between the three groups prior to the dietary intervention, although the P group was heavier. One male subject who was randomly selected to be in the Cr Loading + Maintenance (CrL+M) dose group (baseball athlete) wanted to discontinue use of Cr after the loading phase and so was re-assigned to the Cr-Loading (CrL) only group. This explains the relatively fewer number of male subjects in the CrL+M group compared to the other groups. Mean (±SD) physical characteristics for the three groups were as follows:

	Placebo (n=16)	**Cr- Load** (n=17)	**Cr-Load + Maintain** (n=15)
Age (y)	20.8 ± 3.5	21.0 ± 3.5	20.3 ± 3.7
Height (cm)	176.4 ± 9.4	176.6 ± 10.6	175.3 ± 8.7
Weight (cm)	80.7 ± 19.9	74.4 ± 17.6	74.0 ± 20.5
#Males/females	10 / 6	11 / 6	8 / 7

Testing protocol. Physical characteristics, anthropometric measurements, and body density were measured prior to the dietary treatment and 12 weeks following the dietary treatment. In the 21 club/recreational athletes, measurements were also obtained after the 5 day loading phase in addition to baseline and 12 weeks following the intervention. Subjects reported to the laboratory after an overnight fast (8 hrs) without eating solid food and beverage except for 250 ml of water. Subjects were instructed to ingest additional fluid the night before testing to ensure euhydration. No physical exercise was performed 12 hours before testing. Subjects provided a urine specimen upon arrival in the laboratory. Urine specific gravity (USG) was obtained via a hand-held refractometer on the specimens obtained at baseline, post-loading (n=21), and post-12 week supplementation. Urine volume during the 3 h testing period was also obtained.

Dietary treatment. Using a double-blind, randomized design, subjects were divided into three diet treatment groups: Creatine Loading + Maintenance (CrL+M), Creatine Load only (Cr-L) and placebo (P). During the 5-day loading period, the Cr supplement used was micronized creatine monohydrate (MetRx, Irvine, CA) which was mixed in warm water along with 10 grams of powdered sports drink (PowerAde, The Coca Cola Company, Atlanta, GA) and administered by a member of the research staff in order to ensure subject compliance. Using this procedure, it was virtually impossible to distinguish between a creatine or placebo supplement. During the subsequent 11-week maintenance week period, 5 gms of powdered creatine (Perfect Creatine, Nature's Best, Hauppauge, NY) was mixed along with approximately 4 gms of sports drink powder. Subjects received each days' supplement in individual plastic bags and were instructed to consume the powder straight from the package. In only one subject, this was objectionable and so she was permitted to mix her daily powdered dose with water. Subjects reported to the lab each week to received the next week's supplements.

The P group consumed a sports drink solution three times daily (with no Cr) for the first 5 days and, thereafter, 9 gms/day of dry, powdered sports drink for the remaining 11 weeks of the study. The CrL group consumed Cr (at a dosage equivalent to 0.3 gm/kg of body weight per day) in three meal-time feedings each day mixed in the sports drink solution for five days (Load), and thereafter, ingested the sports drink powder once a day (without Cr). The CrL+M followed the same procedure as CrL for the 5-day loading phase but during the maintenance period ingested 5 gms of Cr mixed with 5 gm of sports drink powder. None of the strength coaches, athletic trainers or athletes knew which treatment had been assigned. Since most athletes today are aware of the ergogenic potential of creatine supplementation, we informed them that the intent of the study was to evaluate different dosage regimens of creatine supplementation.

Body Composition via DXA. Total body bone mineral content, bone mineral density and % fat were determined from whole body scans using dual-energy x-ray absorptiometry (DXA) (Lunar DPX-L , software version 1.3Z; medium mode, 3000 μA). The same technician performed and analyzed all scans. Quality control of the instrument was checked before each test session. DXA was assumed to measure bone ash, which is the total bone mineral minus volatile components that are lost in ashing, such as water of crystallization and carbon dioxide from carbonate. Bone ash is the measure of bone

mineral against which DXA has been validated. Equations for estimating body composition by use of the four-component model are based on the assumption that bone ash is measured by DXA. A correction factor of 1.27 was applied to calculate total body mineral (Brozek et al. 1963).

Total body water. Total body water was measured using deuterium oxide dilution (Davis et al. 1987). After a blood sample was obtained, subjects consumed a 100 ml solution of distilled water containing deuterium oxide (a dosage of .3 gm/kg of body weight). After a 3 hr equilibration period, another blood sample was obtained.

In the purification process, equal volumes (1.5 ml) of plasma and deionized water were incubated at 37°C for 48 hr in Conway diffusion dishes to allow deuterium equilibration. The purified water samples were then frozen until the 12 week post-test samples were obtained to allow each subject's samples to be run with the same standard curve. All samples were analyzed via mass spectrometry at the Center for Applied Isotope Studies, University of Georgia, Athens, GA. After thawing the samples, 2 microliters of sample was placed into a Pyrex ampoule with 1 gm purified zinc metal. The sample was refrozen and the ampoule evacuated to less than 10-3 Torr, and flame sealed. The water was reduced to hydrogen gas at 450° in a muffle furnace overnight. Each sample was run in duplicate on a Finnigan MAT 251 Isotope Ratio mass spectrometer. Calibration was performed using NIST standards specifically SMOW and SLAP (Standard Mean Ocean Water and Standard Light Antarctic Precipitation). Samples were reported relative to SMOW that can be equated to ppm deuterium.

Body composition calculations. In all subjects, the Siri equation (1961) was used to determine % fat (the traditional two-component model) following determination of body density via the BodPod (Life Measurements Instruments, Concord, CA). This method has been observed to be both reliable and valid compared to hydrostatic weighing (McCrory et al. 1995).

In addition to %fat obtained from DXA and air displacement plethysomography, estimates of body composition based on a four-component body composition model were calculated. Based on Lohman (1986), % fat was estimated from body density and body mineral, and body water on the basis of the four-component model (fat, water, mineral and protein). Protein content of the fat free mass was calculated by the difference from the other three components (fat mass, water mass and mineral mass).

Blood parameters. Blood was obtained via venipuncture at an antecubital vein by a trained phlebotomist. Serum was analyzed by an Olympus Simultaneous Multiple Analyzer at an external, certified laboratory (LabCorp of America, Herndon, VA) for glucose, BUN, creatinine, electrolytes, calcium, phosphorus, uric acid, total protein, albumin, globulin, total bilirubin, alkaline phosphatase, lactic dehydrogenase, SGOT, SGPT, GGT, triglycerides, cholesterol, and HDL cholesterol. LDL cholesterol was calculated based on total, HDL cholesterol and triglycerides.

Blood pressure. Systolic (SBP) and diastolic blood pressure (DBP) were measured at baseline, following the loading phase and 12 weeks of supplementation. Values were obtained via auscultation using a mercury sphygmomanometer by two trained technicians. All measurements were done in the morning following an overnight fast following 5 min of seated rest.

Injury logs/rating scale. Copies of the daily treatment report from the Georgia Tech Athletic Training room were obtained over the course of the study in the varsity athletes (n=27). In addition, the subjects completed a weekly training log and subjective rating scale describing the severity of any potential side effects. This visual analog scale consisted of verbal anchors placed on either end of a 10 cm line so that subjects could mark the appropriate response along the continuum (see example attached). The exact distance on the scale was measured from the left edge (0 cm) of each horizontal line and recorded as the rating for joint stiffness, stomach upset, muscle cramping, soreness, and increase in urination.

Statistical analysis. A repeated measures (Treatment x Time) analysis of variance (ANOVA) was used to determine differences between the diet treatment groups for body composition, subjective ratings, and blood measures. Post hoc testing, where appropriate, utilized the Tukey test. Frequency distributions for incidence of cramping and other side effects were reported for Month 1,2, and 3 of the dietary treatment period. An alpha level of 0.05 was used for determining statistical significance.

RESULTS

Body Composition. Body weight increased significantly ($p < 0.05$) over the 12 weeks but there were no dietary treatment or interaction effects among the groups. Mean changes in body weight over the 12 week supplementation period for CrL+M, CrL and P were 1.2, 0.6, and 0.7 kg, respectively. The increases in body weight after the loading phase were 5.4, 1.5, and –0.5 kg for CrL+M, CrL, and P, respectively. The relative changes in body weight after loading were 7.2%, 1.9%, and –0.5% for CrL+M, CrL, and P, respectively. When averaging both groups who loaded with Cr, the mean increase in body weight was 3.5 kg (or 4.8%), but this was not significantly different compared to P.

The % body fat (via DXA) decreased significantly over the 12 weeks by 1.4,1.8, and 0.0% for CrL+M, CrL and P, respectively (Figure 1). However, due to large variability, there were no differences among the groups in the body fat loss. Body density (and thus body fat) measured via air plethysmography did not change significantly over either the 5 day or 12 week period and was also not different among the groups.

The % body fat via the four component model significantly decreased over 12 weeks and there was a trend ($p < .06$) for a significant interaction effect but no group differences. Over the 12 weeks, % fat decreased by 0.6, 1.3, and 1.3% for CrL+M, CrL, and P, respectively. Table 1 illustrates the fat (FM) and fat-free mass (FFM), density of the fat-free mass (Dffm) and water, protein, and mineral fractions of FFM over the 12 week supplementation period. Both FM and FFM had significant time effects but no

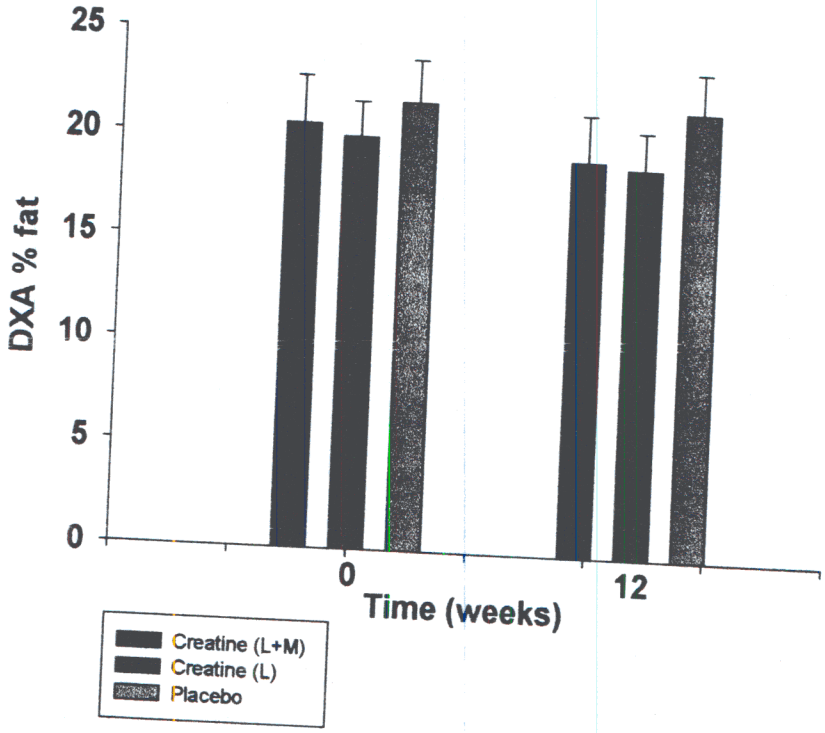


Figure 1. Percentage body fat as measured via DXA over the 12 week supplementation period in placebo (P), creatine loading group (CrL), and creatine loading and maintenance group (CrL+M).

significant group x time interaction or group effect. All groups increased FFM but did not significantly change FM ($p < .07$) over the 12 weeks. The mean (± SE) net increase in FFM over 12 weeks was 2.0, 1.9, and 2.1 kg for P, CrL, and CrL+M, respectively. The total body water increased significantly from baseline to 12 weeks but was not different among the groups. TBW increased 1.6L (3.4%), 1.8L (3.8%), and 1.4L (2.7%) for CrL+M, CrL, and P, respectively. The water and protein fractions of the FFM and density of the FFM (Dffm) did not change significantly from baseline to 12 weeks and were not different among groups. There was a small but significant decrease in the mineral fraction of the FFM over the 12 weeks but no difference among the groups.

Table 1. Body composition changes measured via a four component model (total body water [TBW], DXA and body density) before and after 12 weeks of supplementation.

	Placebo		**Cr- Load**		**Cr-L+M**	
	Pre	Post	Pre	Post	Pre	Post
FM (kg)	13.9 (2.4)	13.0 (2.1)	10.7(1.5)	9.7(1.4)	12.3 (2.1)	11.7(1.7)
FFM (kg)	62.6(4.3)	64.7 (4.7)	61.9(3.5)	63.8 (4.0)	67.2 (4.6)	69.2 (4.6)
TBW (L)	51.5 (3.6)	52.9(3.6)	47.1 (2.5)	48.9 (3.2)	47.5 (3.3)	49.1 (3.7)
Dffm	1.092(.003)	1.090(.003)	1.092(.002)	1.090(.003)	1.095(.002)	1.094(.002)
%H_2O/ FFM	75.8(0.6)	75.9(0.7)	75.7(0.7)	76.0(0.7)	75.2(0.5)	75.3 (0.4)
%mineral/FFM	6.5(0.2)	6.2(0.3)	6.6(0.1)	6.5 (0.2)	6.8(0.2)	6.7 (0.2)
%protein/FFM	17.6(0.5)	17.9(0.6)	17.6(0.7)	17.5 (0.6)	18.0(0.5)	18.0 (0.4)

After the loading phase, there was a significant increase in the water fraction of the FFM in the CrL group (5% higher) compared to either baseline or 12 weeks. CrL had higher water fractions after loading compared to P and CrL+M. When combining CrL and CrL+M groups compared to P after the loading phase, there was a significant Time effect for TBW, Dffm, and the water, mineral and protein fraction of the FFM. However, no group or time x group interaction effects. The subjects who used creatine had 0.8 L higher TBW, 0.2% lower mineral fraction, 3.3% lower protein fraction, 3.5% higher water fraction, and a lower Dffm (1.094 vs. 1.082). Mean changes for P subjects were 2.7 L lower TBW, 0.2% lower mineral fraction, 0.6% lower protein fraction, 0.8% higher water fraction, and also a lower Dffm (1.092 vs. 1.088). However, since less than half of the total subjects completed the loading phase testing using the 4 component model, these means must be interpreted with caution. On the other hand, despite the reduced statistical power resulting from smaller sample sizes, the fact that higher water fractions were found

in one of the creatine loading groups, the data do collectively suggest that gains in body weight with creatine loading are primarily due to water gain rather than protein.

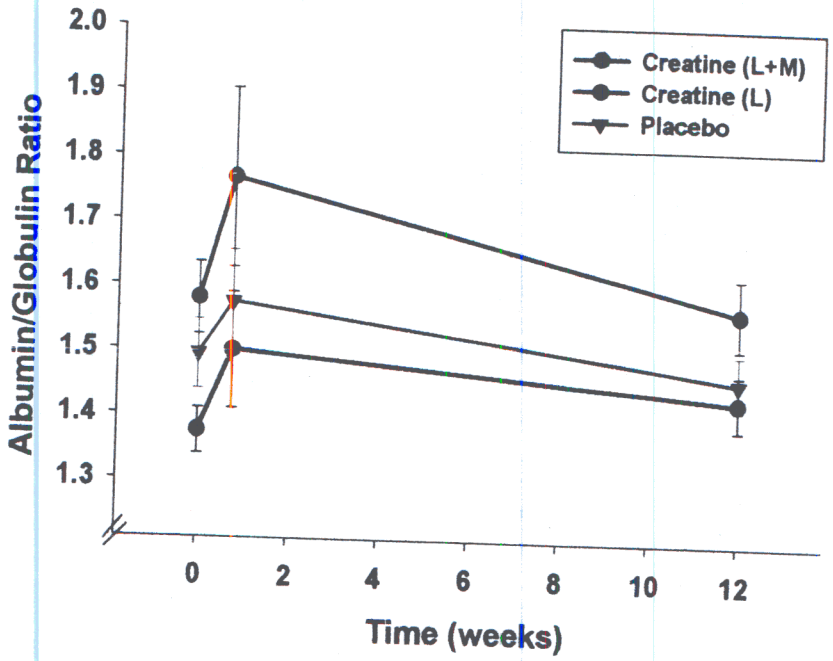
Blood parameters. Creatine supplementation had no significant effect in the following parameters: uric acid, total cholesterol, LDL cholesterol, HDL, cholesterol, total cholesterol/HDL ratio, triglycerides, sodium, chloride, phosphate, calcium, calcium/phosphate ratio, blood urea nitrogen, total protein, albumin, globulin, bilirubin, or the liver/ kidney enzymes (alkaline phosphatase, LDH, SGOT, SGPT, GGT). These values are illustrated in Figures or tables in the Appendix section.

The albumin/globulin ratio was significantly different ($p < 0.05$) between the groups (Figure 2). CrL+M group had significantly lower values than both P and CrL; however CrL was also higher than P. Lower A/G ratio is found in certain types of liver disease, inflammation or kidney disease. Thus, since the CrL group had a better profile than P and all three groups increased their values over time, there was no evidence that creatine supplementation caused adverse effects in A/G ratio.

There was a significant group x time interaction effect for creatinine (Figure 2). Mean values for CrL+M increased significantly above baseline after the loading phase (17%) and after 12 weeks (19.7%) of creatine supplementation. The CrL group also had higher serum creatinine after loading only (25%). The loading phase resulted in significantly higher creatinine levels in both the CrL+M and CrL groups ($p < 0.05$) compared to P. However, after 12 weeks, only CrL+M values were higher than the CrL group (and not different from P). Both P (6.5%) and CrL (8.2%) had non-significant increases after 12 weeks. Thus, it appears that there is a mild increase in serum creatinine values with the loading phase, which is reversible when creatine is not taken in a maintenance dose, but also is not different over long term (12 wks) compared to P.

Serum potassium was significantly different among the groups (Figure 3). CrL+M had significantly lower values after the loading and 12 wk post-supplementation compared to P despite similar baseline levels in the two groups. CrL initiated the study with significantly higher levels than P and CrL+M but decreased significantly after loading similar to CrL+M. The physiological significance of this difference is not clear since all of the values still remained within normal limits (3.5-5.3 mmol/L).

The creatine/BUN ratio (considered a marker of catabolism which is elevated in rhabdomyolysis) is illustrated in Figure 3. Creatine/BUN was not different between the groups, although there was a trend ($p < .08$) for CrL+M to increase over 12 weeks (25%) compared to changes in P (-1.4%).



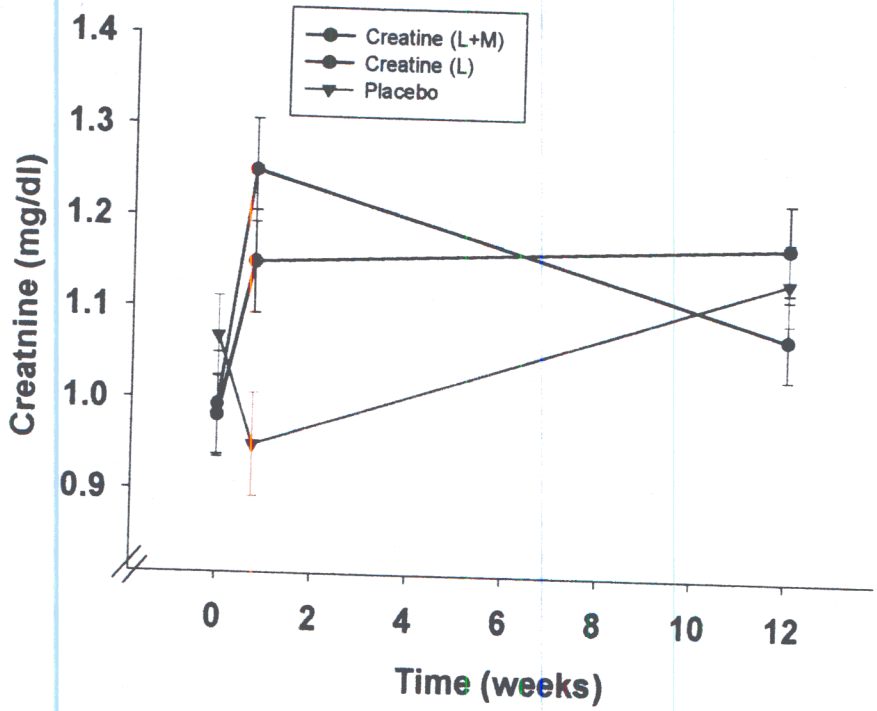
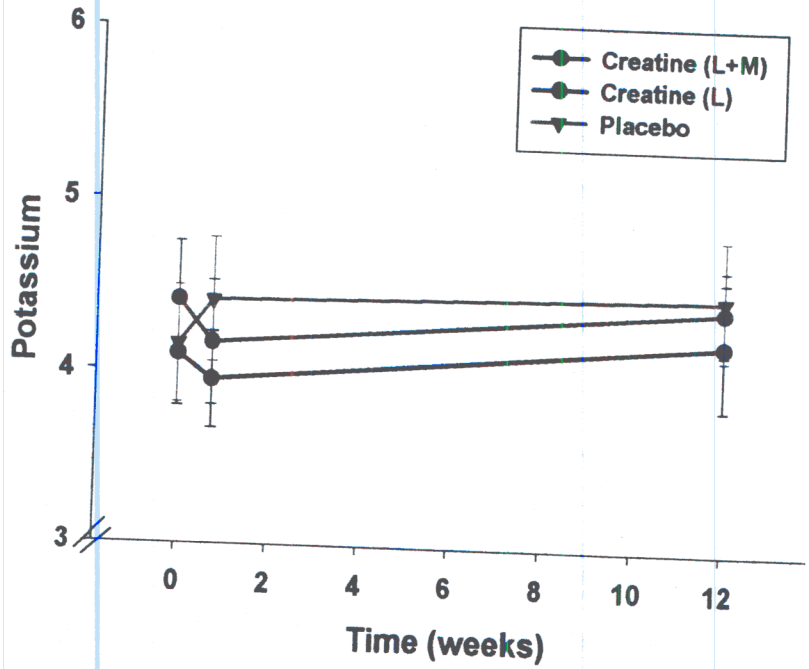


Figure 2 – Serum albumin/globulin ratio and creatinine during 12 wk supplementation period.



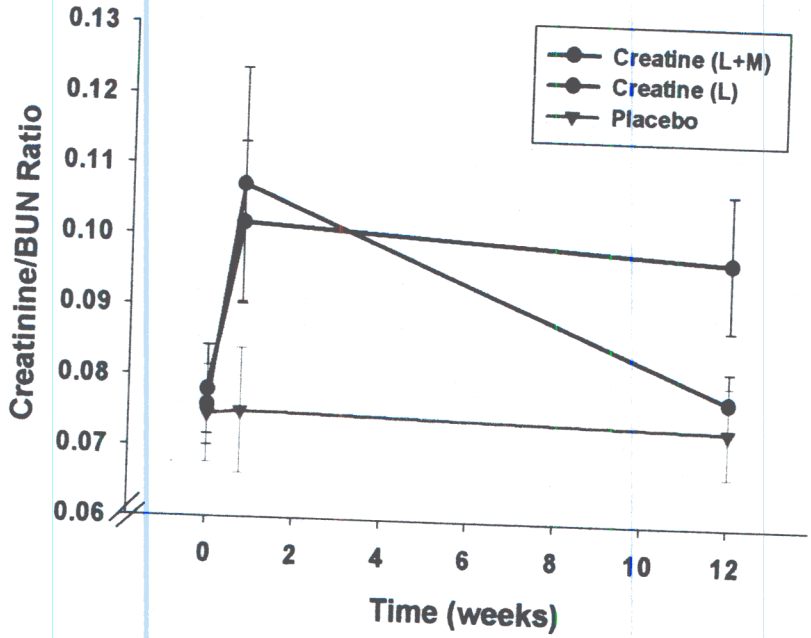
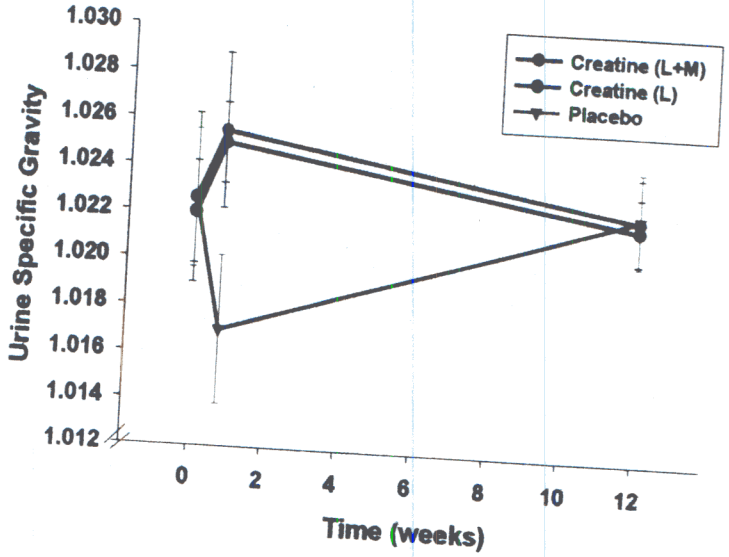


Figure 3 – Serum potassium and creatinine/BUN ratio during 12 wk supplementation period.

Blood pressure. There was no significant change in SBP over time after the 5 day loading or 12 week supplementation period or among groups. Mean (±SE) SBP values at 12 weeks were 123 ±2.5, 122± 2.1, and 124 ±3.4 for CrL+M, CrL, and P, respectively and within 3 mmHg of baseline SBP. However, there was a significant change ($p < 0.05$) in DBP over time as well as a significant interaction effect (Figure 5). Mean changes in DBP over the 12 weeks were 5.2, 1, and 0.6 mmHg for CrL+M, CrL, and P, respectively. CrL+M had significantly higher DBP following the 12 week supplementation period (no other groups had significantly different DBP at 12 weeks compared to baseline). However, the mild increase in DBP was well within the normal range (81 mmHg). In addition, there was a trend for P to have higher DBP following loading compared to CrL and CrL+M.

Urine specific gravity/volume. Usg was not significantly different over the 12 weeks or among the groups, but there was a significant interaction effect (Figure 4). P had significantly lower Usg at loading (1.017 ± .003) compared to baseline (1.023 ± .003) and 12 weeks (1.022 ± .002) while CrL and CrL+M had no difference across time. The mean values suggest the level of dehydration for all athletes was high for all groups (usg of 1.020 is defined as the limit for euhydration state by NCAA Wrestling standards). However, many athletes could not provide a urine specimen, especially in the baseline testing session.

Urine volume during the 3 hr of morning testing (during which no fluids or food was ingested) was not significantly different over the 12 weeks but there was a significant interaction effect among the three groups over time (Figure 4). Neither the CrL+M or CrL groups were different between baseline, loading or 12 weeks of supplementation; however, P had significantly greater urine volume (275 ml) after the loading phase compared to either baseline or following 12 weeks of supplementation. In addition, P had significantly higher urine volumes compared to CrL+M and CrL after the loading phase. All groups drank the additional fluids during the loading phase (and reported perceived increases in urination as mentioned in the Subjective Ratings section), but only the P group had significantly greater urine output and lower usg during the test morning following creatine loading. This suggests that subjects in the P group may either have done a better job of hydrating prior to the test session compared to the other groups or that the creatine-loaded subjects had greater fluid retention. Since the water fraction of the FFM increased only in the CrL group (not CrL+M), it is not entirely clear what effect creatine has on the body's hydration status. However, it does not appear that the hydration status is necessarily compromised when creatine is either used either in a loading dose, or in a loading plus maintenance dose.



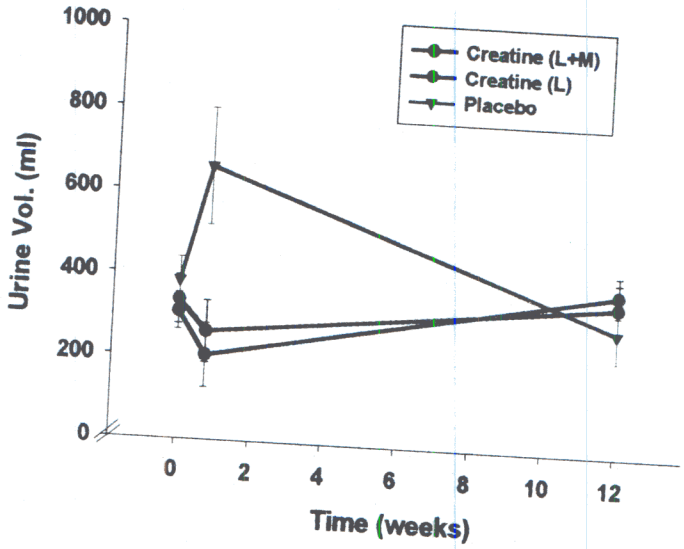


Figure 4. Mean (±SE) changes in urine specific gravity and 3 h urine volume during test mornings over the 12 week supplementation period.

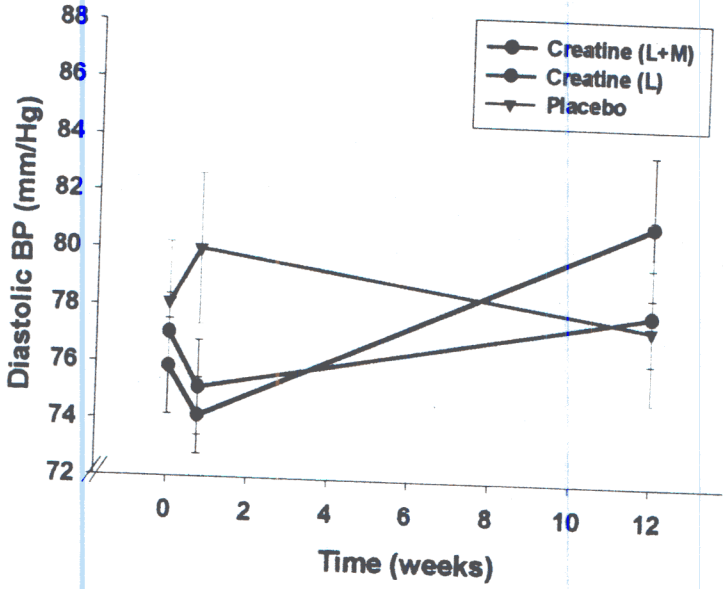


Figure 5. Mean (± SE) changes in diastolic blood pressure over the 12 week supplementation period.

Subjective ratings. A 10 cm Visual Analog Scale was used to quantify subjective ratings related to muscle cramping, stomach upset, joint stiffness, change in urination frequency, and level of muscle soreness. A score of 0 indicted no symptoms up through 10 which indicated very, very severe symptoms. Frequency distributions for the % of subject rating each variable either mild (<2) or moderate (<5) are located in the Appendix. The most severe response (on the scale of 10) is also indicated in these tables.

Over the 12 week supplementation period, there were no differences in subjective ratings for urine frequency, stomach upset, muscle soreness or muscle cramping between the groups (Figures located in Appendix). There were significant effects over time, however. Stomach upset was significantly higher following loading across all of the groups compared to the last week of the study. Perceived increases in urination were also significantly higher after loading (score of 4) for all groups compared to the rest of the weeks of the study (group scores ranging from 0.8 to 2.7). Muscle soreness ratings were higher for all groups following the loading phase (score of 5) compared to the other weeks and week 3 (which was higher than the last two ratings in the study). There were no significant differences over time in muscle cramping, however. After the loading phase, mean values for cramping in CrL+M (1.6) and CrL (0.8) were similar to P (2.0).

There was a significant interaction effect ($p < 0.01$) in joint stiffness ratings among the groups across time (Figure 6). The CrL group reported higher joint stiffness during the 3rd week (2.9 ± 0.5) of the study compared to the end of the study (1.5 ± 0.5) and CrL was higher than P (1.7 ± 0.5) during the 3rd week. However, during the last week of the study (12 weeks), the CrL+M group had significantly higher values (2.7 ± 0.9) compared to both CrL (1.5 ± 0.5) and P (0.9 ± 0.4).

The incidence of cramping was calculated by the number of times a subject indicated cramping on the weekly report forms divided by the number of forms completed (Figure 5). The mean ($\pm$ SE) percentage of time cramping occurred was not significantly different between CrL+M ($36.7 \pm 9.8\%$), CrL ($18.7 \pm 8.4\%$), or P ($22.4 \pm 6.1\%$). The number of subjects in each group who experienced at least one episode of cramping over the 12 weeks was 10/15 subjects (67%) for CrL+M, 5/16 (31%) for CrL, and 10/17 (59%) for P.

Injury logs obtained over the entire 12 week period from the training room on the 27 athletes by group indicated:

P	CrL	CrL+M
2 quadriceps strains	1 mild hamstring straing	1 severe hamstring strain
1 ankle sprain		1 mild groin strain
cramping during game		

Anecdotal reports. There were relatively few individual complaints during the study. Out of 47 subjects, we had four subjects (two male and two female) who expressed a desire to not continue with supplementation. Three subjects experienced cramping episodes that interfered with their training and all three were in the CrL+M group; however, only one of these reported not taking all of the maintenance packets as a result. One male subject (football player) complained of daily cramping in various

muscle groups during both exertion and non-exertional circumstances but he did complete his 12 week supplementation regimen. Interestingly, he was the only subject who also had blood creatinine values just barely above the normal range (1.6). The fourth individual was a baseball pitcher who after the 5th day of loading experienced arm tightness/stiffness that was not remedied with stretching, etc. This individual requested to discontinue the supplement but completed the study in the CrL group.

The remaining 43 subjects either reported ambivalence (didn't think it helped or hurt them) or that Creatine improved their training, recovery from training and /or body composition. One subject on P complained of stomach upset with the sports drink powder ingestion during the maintenance phase, so she was permitted to mix the powder with water for ease of ingestion. This appeared to remedy her discomfort. Another P subject wanted to drop out of the study after two weeks due to a perceived increased state of anxiety and skin blemishes but after consultation agreed to stick with the study protocol.

After the 3rd day of loading, three individuals (two female and one male) complained of headaches that occurred about 1 hour after consuming the creatine /sports drink solution. One of these subjects was a female ovo-lacto vegetarian. The headaches subsided following the loading phase for this female but not the other female, both of whom continued the study in the CrL+M group. The other male subject had no further headaches following the loading phase but was in the CrL group and thus did not continue a maintenance dose of Cr.

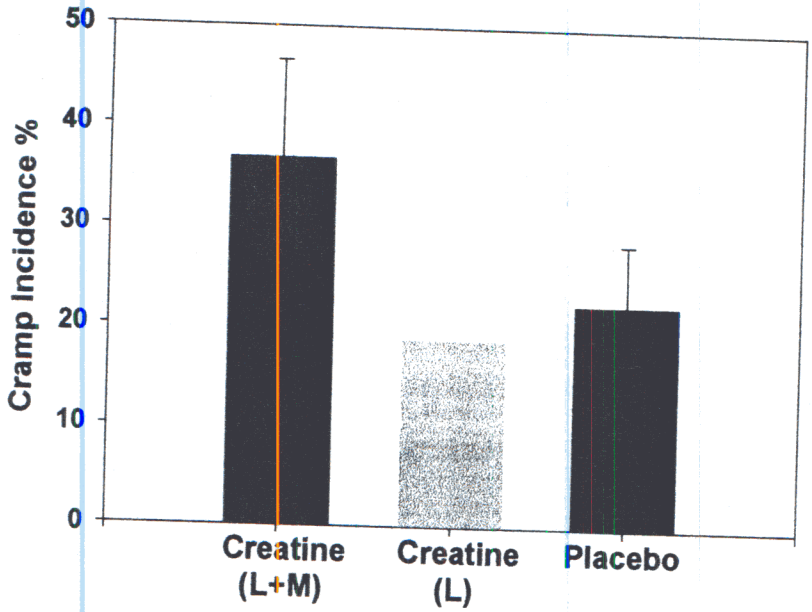


Figure 5. Mean (± SE) percentage of time that an incidence of cramping was reported on the weekly report forms throughout the 12 week supplementation period.

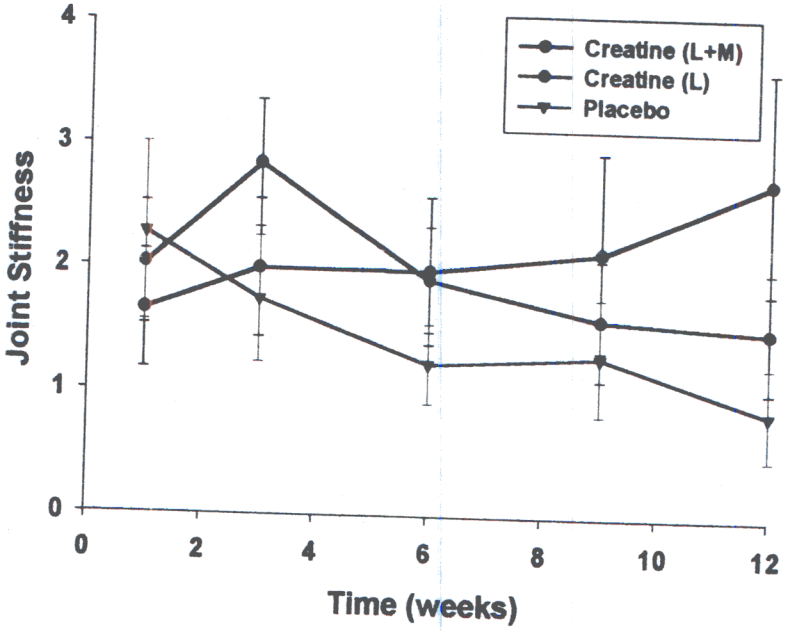


Figure 6. Mean (± SE) ratings for joint stiffness over the 12 week supplementation period.

Application of results / Importance to the NCAA. Using a double-blind, prospective research design, our data regarding adverse side effects (i.e., risk for soft tissue injury or increased muscle cramping) **do not indicate a statistically greater incidence for injury or cramping in creatine** versus non-creatine supplemented athletes over a 12 week period. However, despite no statistically significant differences, it is important to note that in some individuals (n=3) muscular cramping did occur on a regular basis (≥ 2-3 times per week) in those who ingested a loading and maintenance dose of creatine. So, there appears to be wide-ranging inter-individual differences in the cramping response to Cr. Other than some perceived arm tightness/stiffness in a male baseball pitcher during the loading phase, no other injuries occurred in subjects consuming creatine over the 12 week period. However, the VAS did indicate greater perceptions of joint stiffness in the CrL group in the 2 weeks following the loading phase, and in those that continued to ingest Cr (the CrL+M group) at the end of the 12 weeks. This finding merits additional study to verify this result and identify potential mechanisms responsible.

The amount of additional benefit that Cr (at these particular doses) provided men and women for increasing muscle mass over a 12 week period compared to a placebo was modest at best. Depending on the method used (DXA, BodPod or 4 component model), there was little to no decrease in body fatness. However, the data do suggest that most of the changes in body weight and body composition (TBW, FFM) appear to occur with the loading dose.

The indices of kidney/liver function did not provide strong evidence for problems related to creatine ingestion at the doses consumed in this study. Only creatinine was mildly elevated with Cr loading but was still within the normal range. The profile for A/G ratio changed in a similar fashion among the three groups. BUN, Creatinine/BUN ratio, uric acid, total protein, albumin, globulin total bilirubin, alkaline phosphatase, SGOT, SGPT, GGT were not different among groups.

Markers of hydration status (serum electrolytes, total body water, urine specific gravity, urine volume during 3 hr of testing) appear only marginaly affected by creatine ingestion. Of these, only usg and urine volume had group x time interaction effects during loading and only serum K^+ was lower following loading when Cr was ingested. The physiological significant of these small differences remains unclear. **Over the 12 week period, there appears to be no detriment in hydration status** among those who ingest Cr either via loading and/or combined with a maintenance dose compared to P.

Creatine has become a very popular supplement for intercollegiate athletes competing in various sports emphasizing muscular strength and power (i.e. swimming, football, basketball, baseball/softball, track and field, volleyball). The supplement is being used for prolonged periods (beyond what any research studies have investigated); thus, further research is needed to determine true "long-term" effects of the supplement at dosages that accurately represent what is being used by collegiate athletes during training. However, **for the length of time (12 weeks) and dosage consumed in the present study, significant adverse side effects were not observed.**

REFERENCES

Becque, B.D., J.D. Lochmann, and E. Melrose. Effect of creatine supplementation during strength training on 1 RM and body composition. *Med. Sci. Sports Exerc.* 29:S146, 1997.

Bosco, D. J. Tihanyi, J. Puckspk, I. Kovacs, A. Gabossy, R. Colli, G. Pulvirenti, C. Tranquillli, D. C. Foti, M. Viru, A. Viru. Effect of oral creatine supplementation on jumping and running performance. *Int. J. Sports Med.* 18:369-372, 1997

Brozek, J., F. Grande, J. T. Anderson, and A. Keys. Densitometric analysis of body composition: revision of some quantitative assumptions. *Ann. NY Acad. Sci.* 110:113-40, 1963.

Davis, J. M., D. R. Lamb, W. A. Burgess, and W. P. Bartoli. Accumulation of deuterium oxide in body fluids after ingestion of D2O-labeled beverages. *J. Appl. Physiol.*63:2060-2066, 1987.

Dempster, P. and S. Aitkens. A new air displacement method for the determination of human body composition. *Med. Sci. Sports Exerc.* 27:1692-1697, 1995.

Earnest, C. P., P.G. Snell, R. Rodriguez, A. L. Almada, and T.L. Mitchell. The effect of creatine monohydrate ingestion on anaerobic power indices, muscular strength and body composition. *Acta Physiol. Scand.* 153:207-209, 1995.

Greenhaff, P. L., K. Bodin, R.C. Harris, E. Hultman, D. A. Jones, D. B. McIntyre, K. Soderlund, and D. L. Turner. The influence of oral creatine supplementation on muscle phosphocreatine resynthesis following intense contraction in man. *J. Physiol.* 467:75P, 1993.

Greenhaff, P. L., K. Bodin, K. Soderlund, and E. Hultman. Effect of oral creatine supplementation on skeletal muscle phosphocreatine resynthesis. *Am. J. Physiol.*266: E725-E730, 1994.

Grindstaff, P.D., R. Kreider, R.Bishop, M. Wilson, L. Wood, C. Alexander, and A. Almada. Effects of creatine supplementation on repetitive sprint performance and body composition in competitive swimmers. *Int. J. Sports Nutr.* 7:330-346, 1997.

Kreider, R., M. Ferreira, M. Wilson, P. Grindstaff,. S. Plisk, J. Reinhardy, E. Cantler, and A. Almada. Effects of ingesting a supplement designed to enhance creatine uptake on body composition during training. *Med. Sci. Sports Exerc.*29:S145, 1997.

Kushner, R.F., and D. A. Schoeller. Estimation of total body water by bioelectrical impedance analysis. *Amer. J. Clin. Nutr.*44: 417-424, 1996.

Lohman, T.G. Applicability of body composition techniques and constants for children and youths. In *Exercise and Sports Science Reviews*, edited by K. Pandolf. New York:Macmillan, p. 325-357,1986.

Maughan, R. J. Creatine supplementation and exercise performance. *Int. J. Sports Nutr.* 5:94-101, 1995.

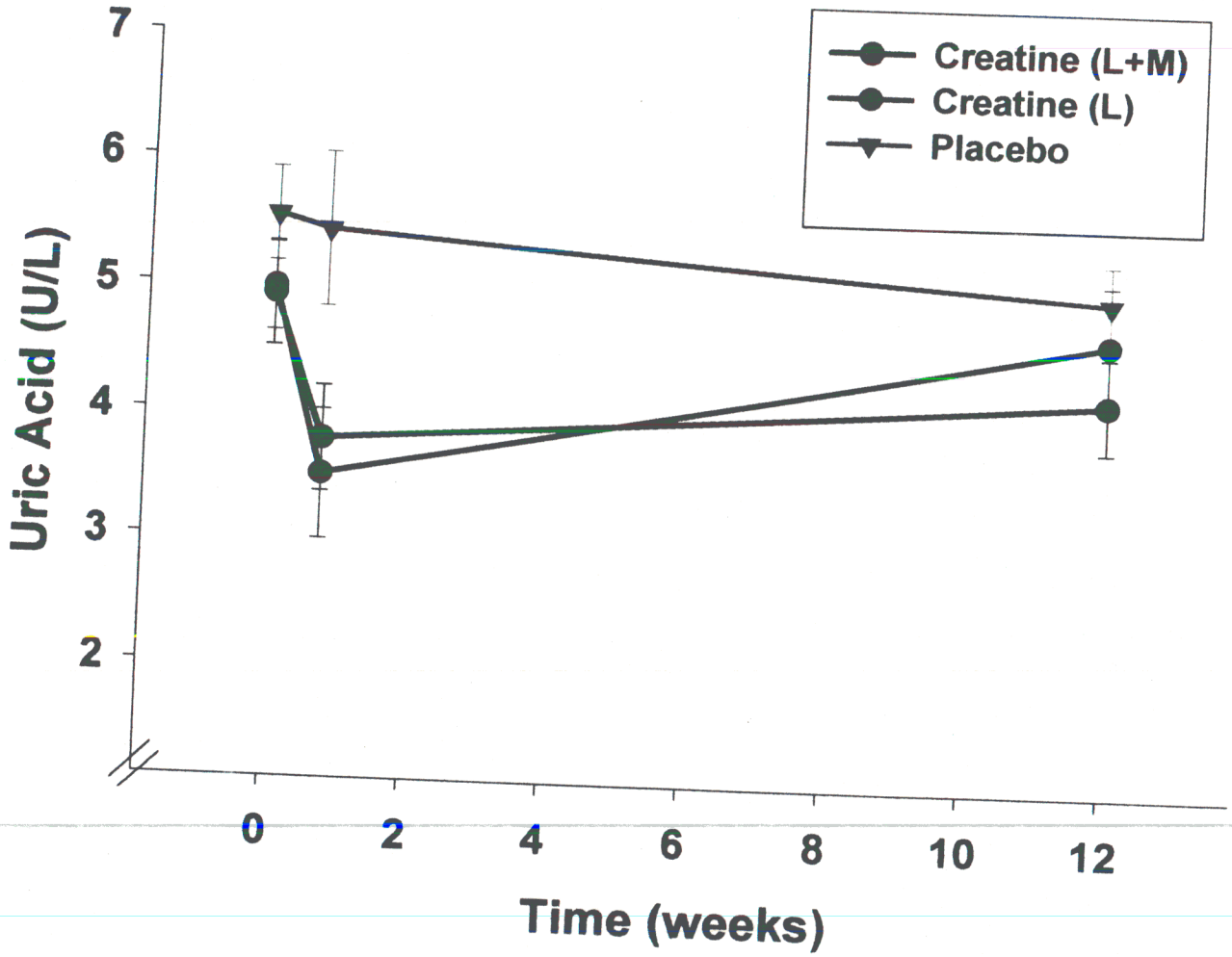
McCrory, M.A., T. D. Gomez, E. M. Bernauer, and P. A. Mole. Evaluation of a new air displacment plethysmograph for measuring human body composition. *Med. Sci. Sports Exerc.* 27:1686-1691, 1995.

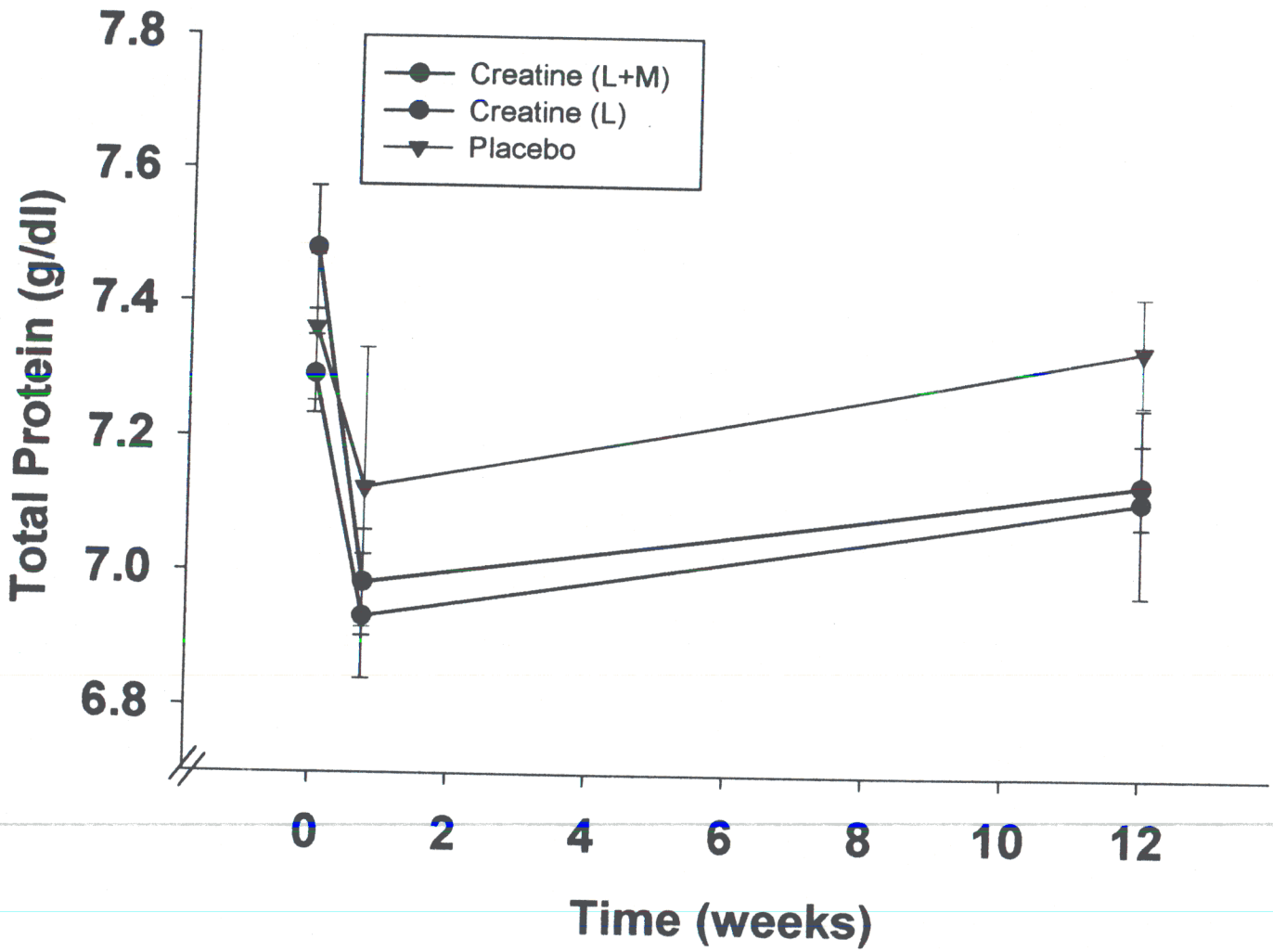
Prevost, M.C., A. G. Nelson, G.S. Morris. Creatine supplementation enhances intermittent work performance. *Res. Quart. Exerc. Sport* 68:233-240, 1997.

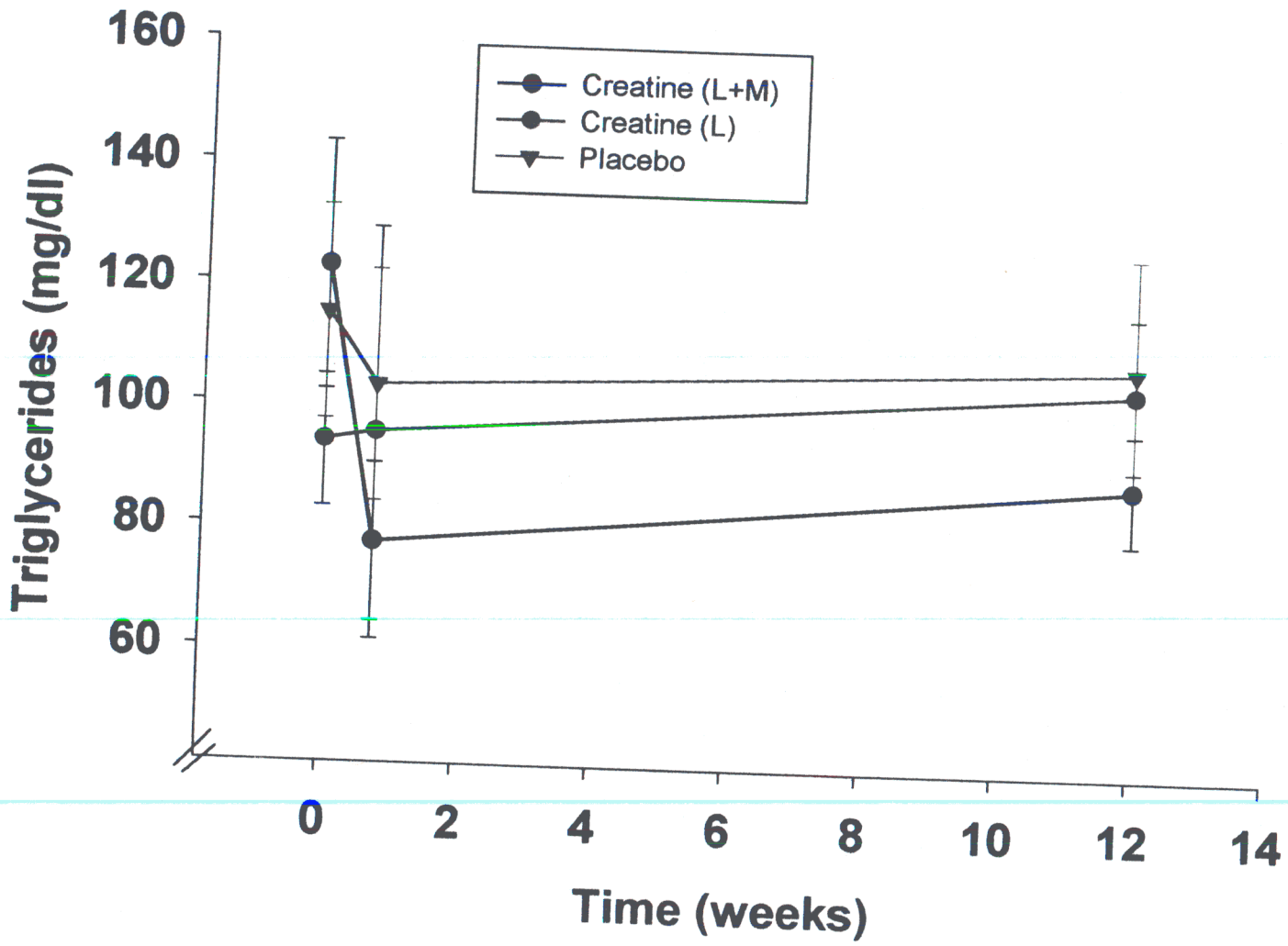
Siri, W.E. Body composition from fluid spaces and density: analysis of methods. In: *Techniques for Measuring Body Composition,* edited by J. Brozek and A. Henschel. Washington, DC:Natl. Acad. Sci.,1961, p.223-244.

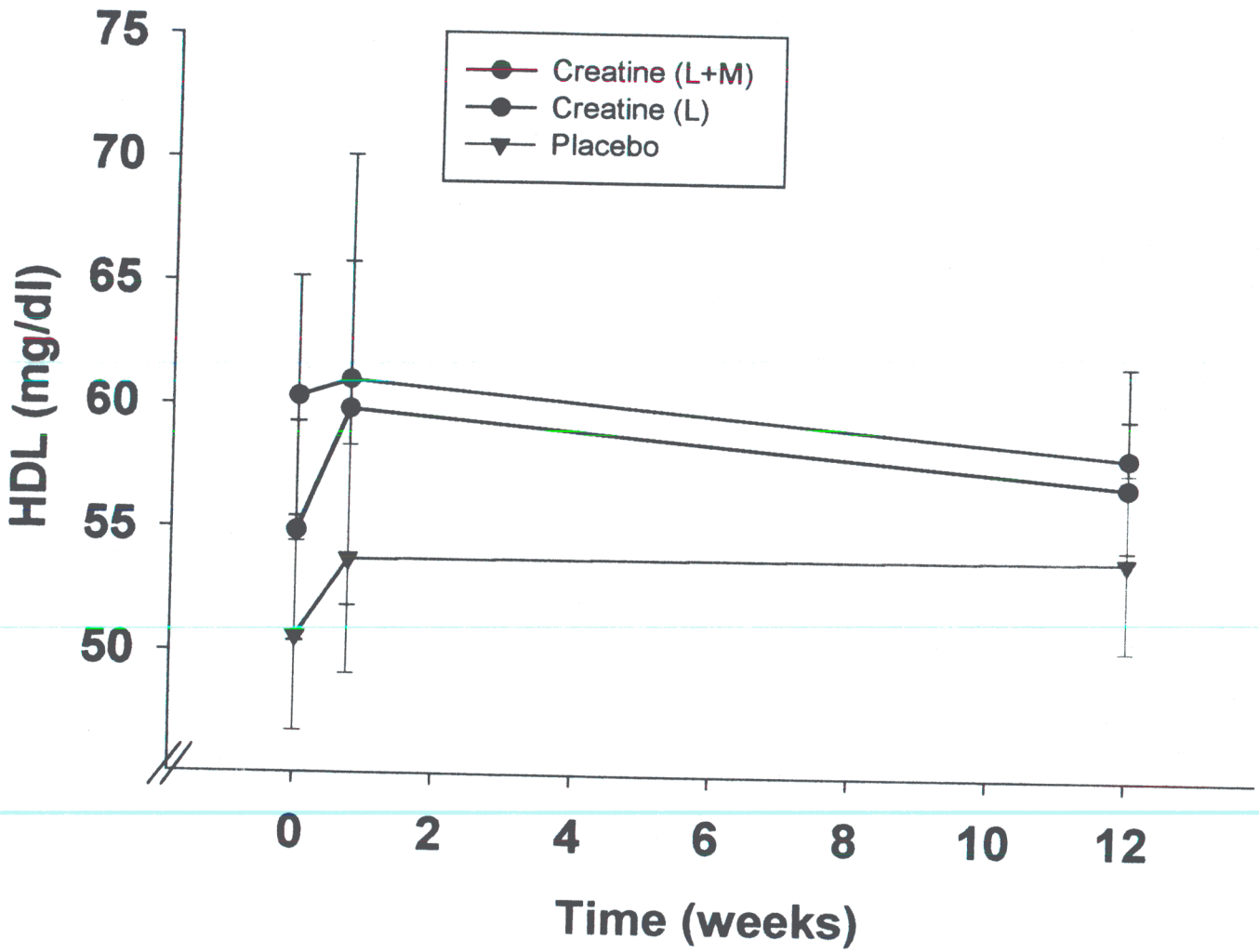
Wilson, R. L. Excessive hamstring and other muscle injuries with creatine? Gatorade Sports Science Institute Bulletin Board, July 7,1997 (http://www.gssiweb.com/).

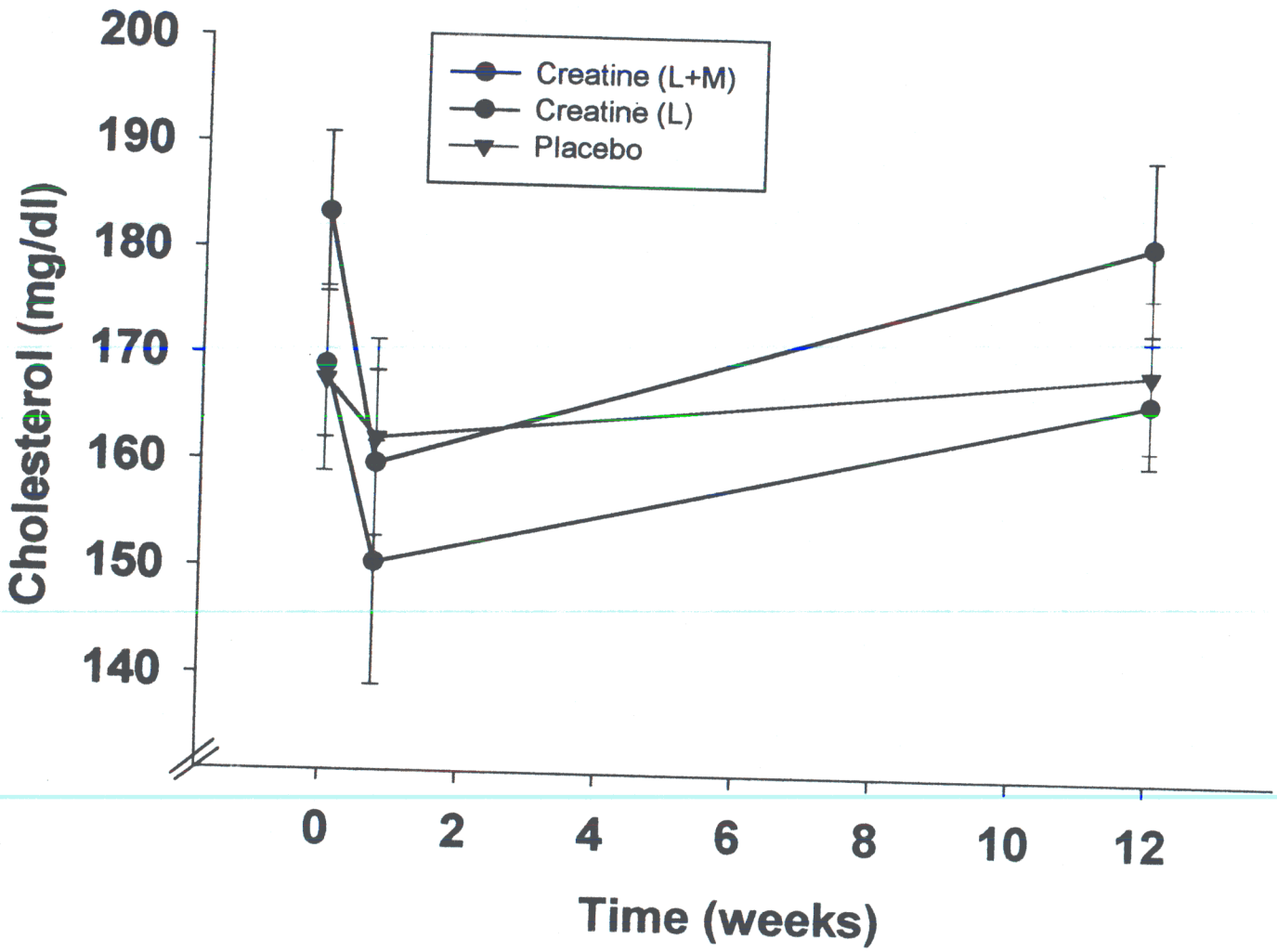
APPENDIX

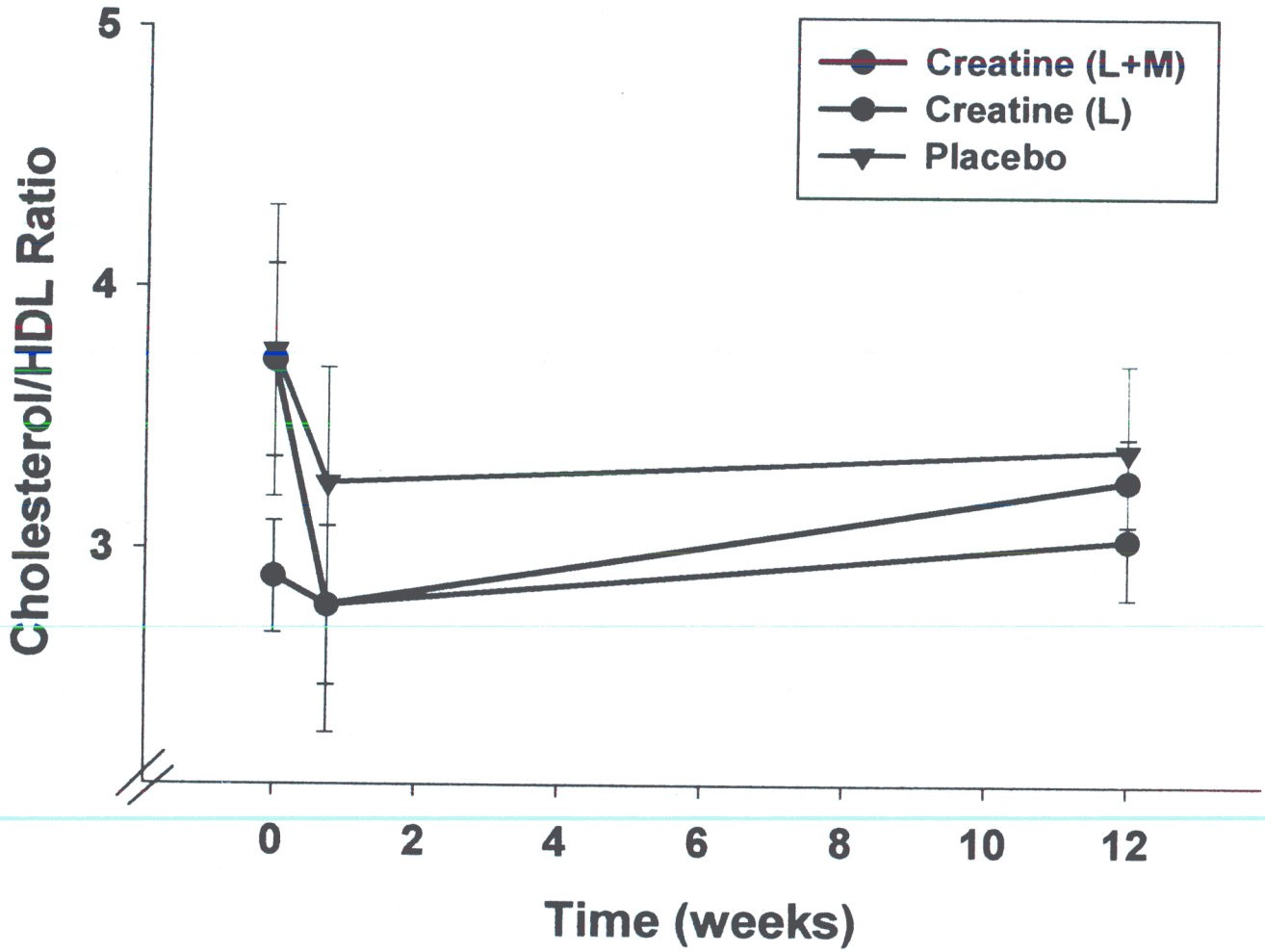

Creatine (L+M)
Creatine (L)
Placebo
Uric Acid (U/L)
7
6
5
4
3
2
0
2
4
6
8
10
12
Time (weeks)

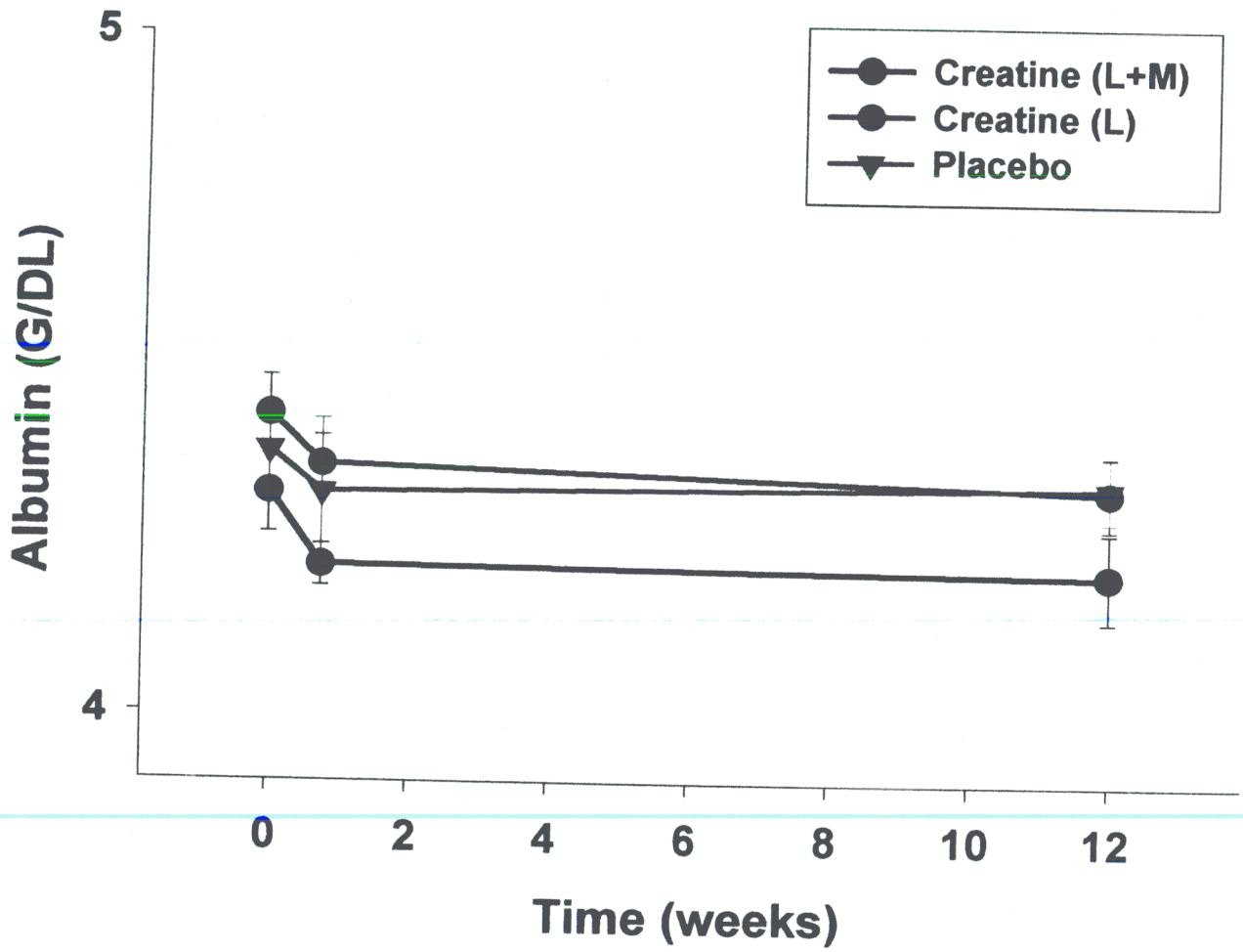

Creatine (L+M)
Creatine (L)
Placebo
Total Protein (g/dl)
7.8
7.6
7.4
7.2
7.0
6.8
0
2
4
6
8
10
12
Time (weeks)

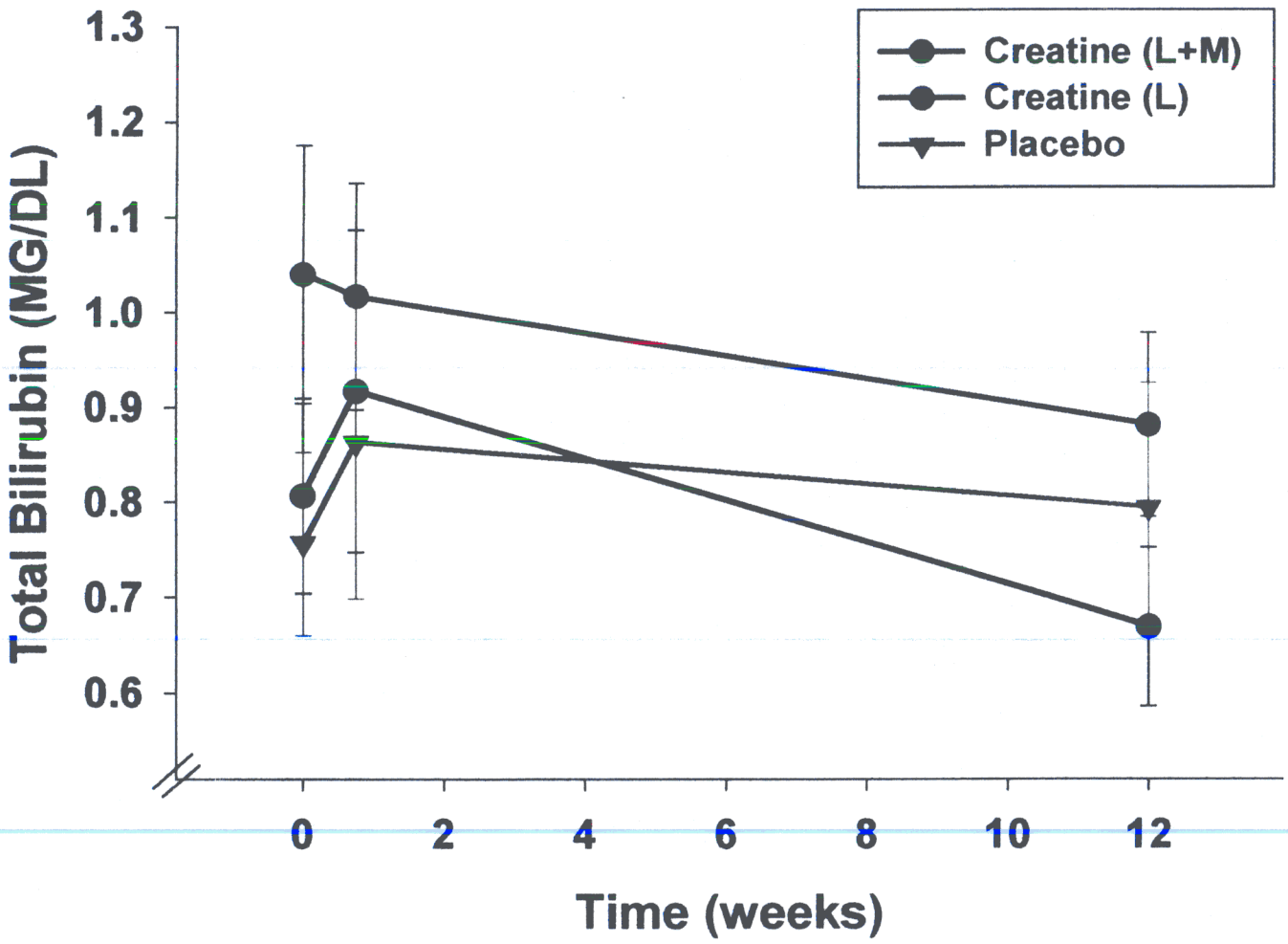

Creatine (L+M)
Creatine (L)
Placebo
Triglycerides (mg/dl)
160
140
120
100
80
60
0
2
4
6
8
10
12
14
Time (weeks)

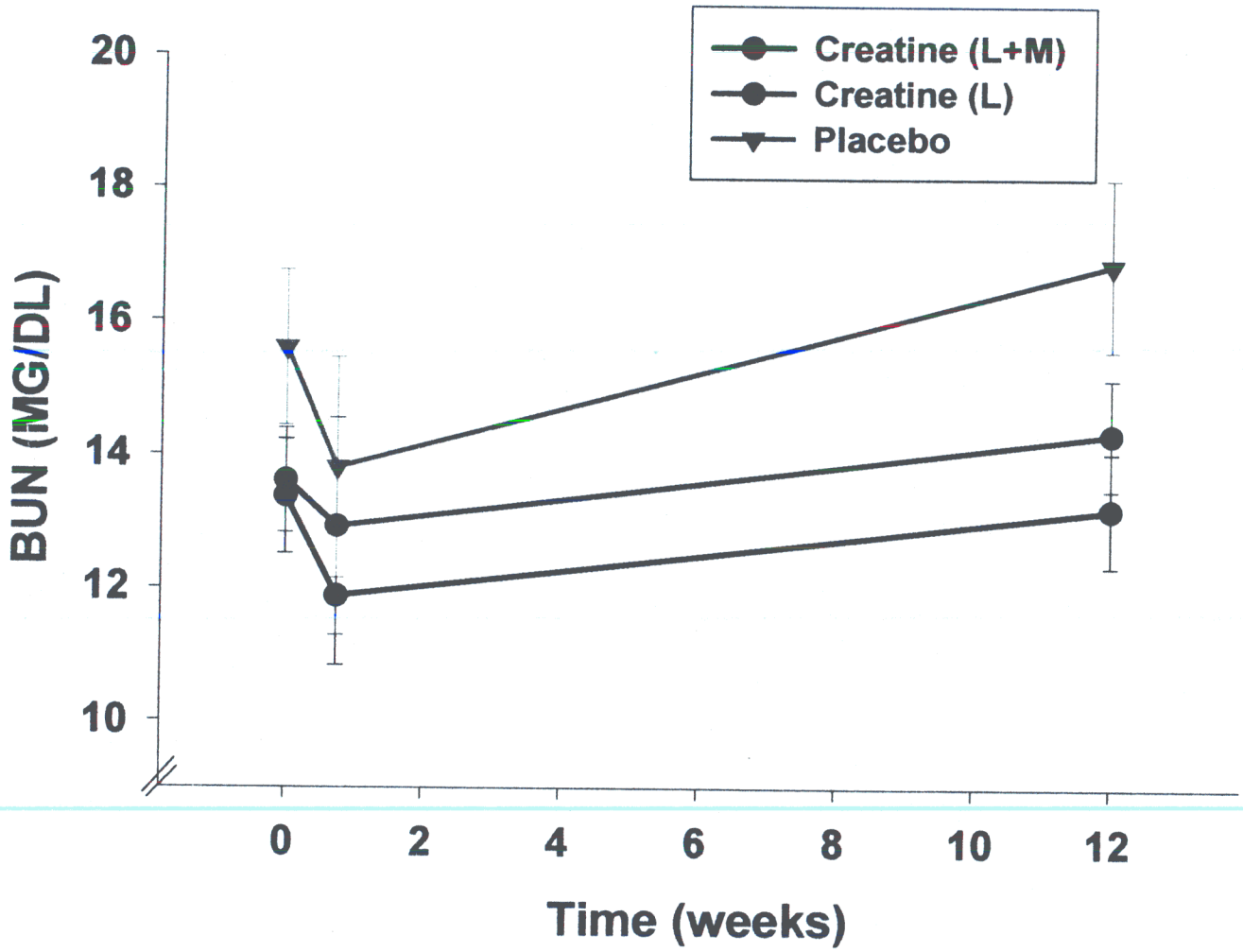

Creatine (L+M)
Creatine (L)
Placebo
HDL (mg/dl)
75
70
65
60
55
50
0
2
4
6
8
10
12
Time (weeks)

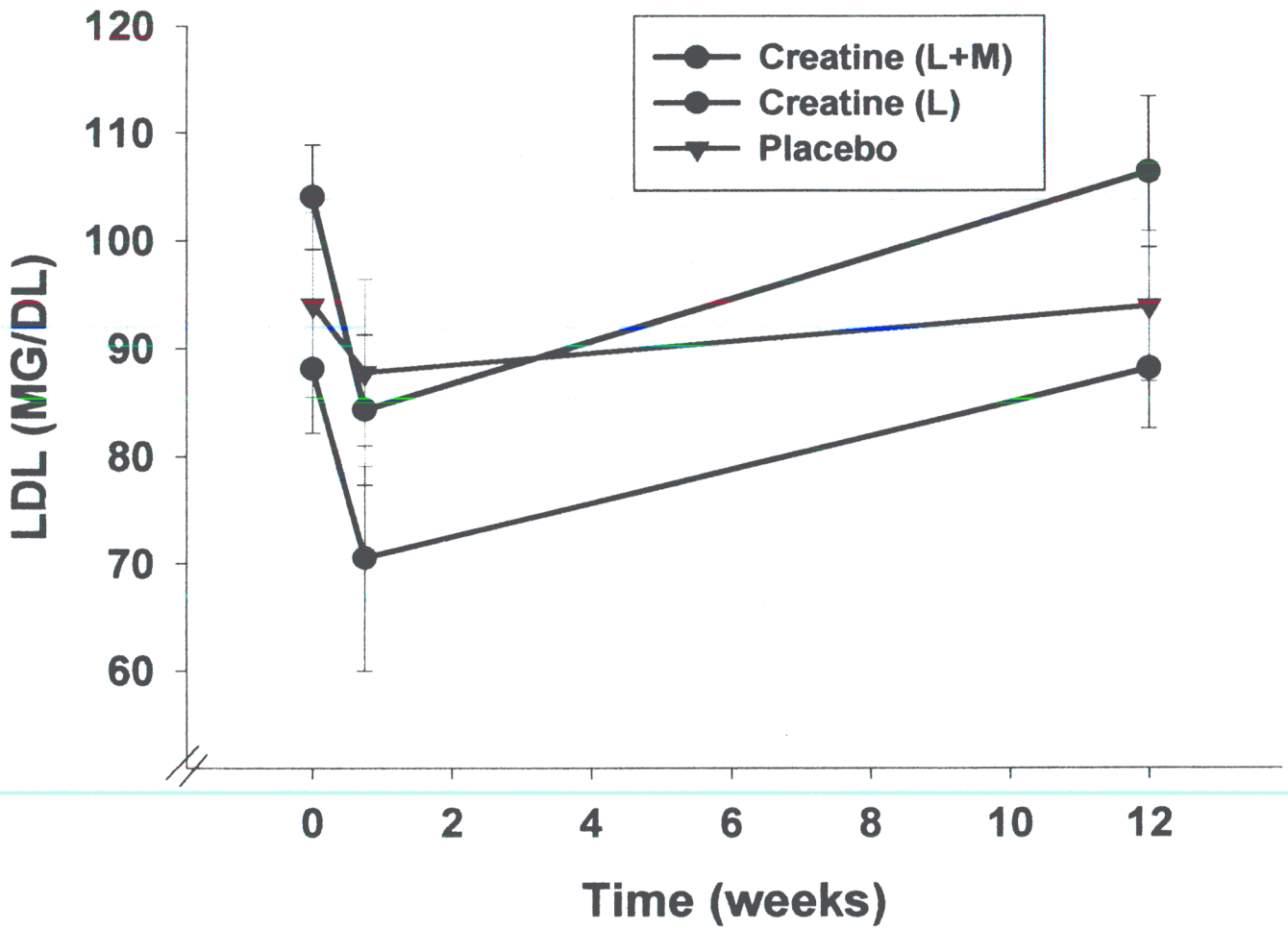

Creatine (L+M)
Creatine (L)
Placebo
Cholesterol (mg/dl)
200
190
180
170
160
150
140
0
2
4
6
8
10
12
Time (weeks)

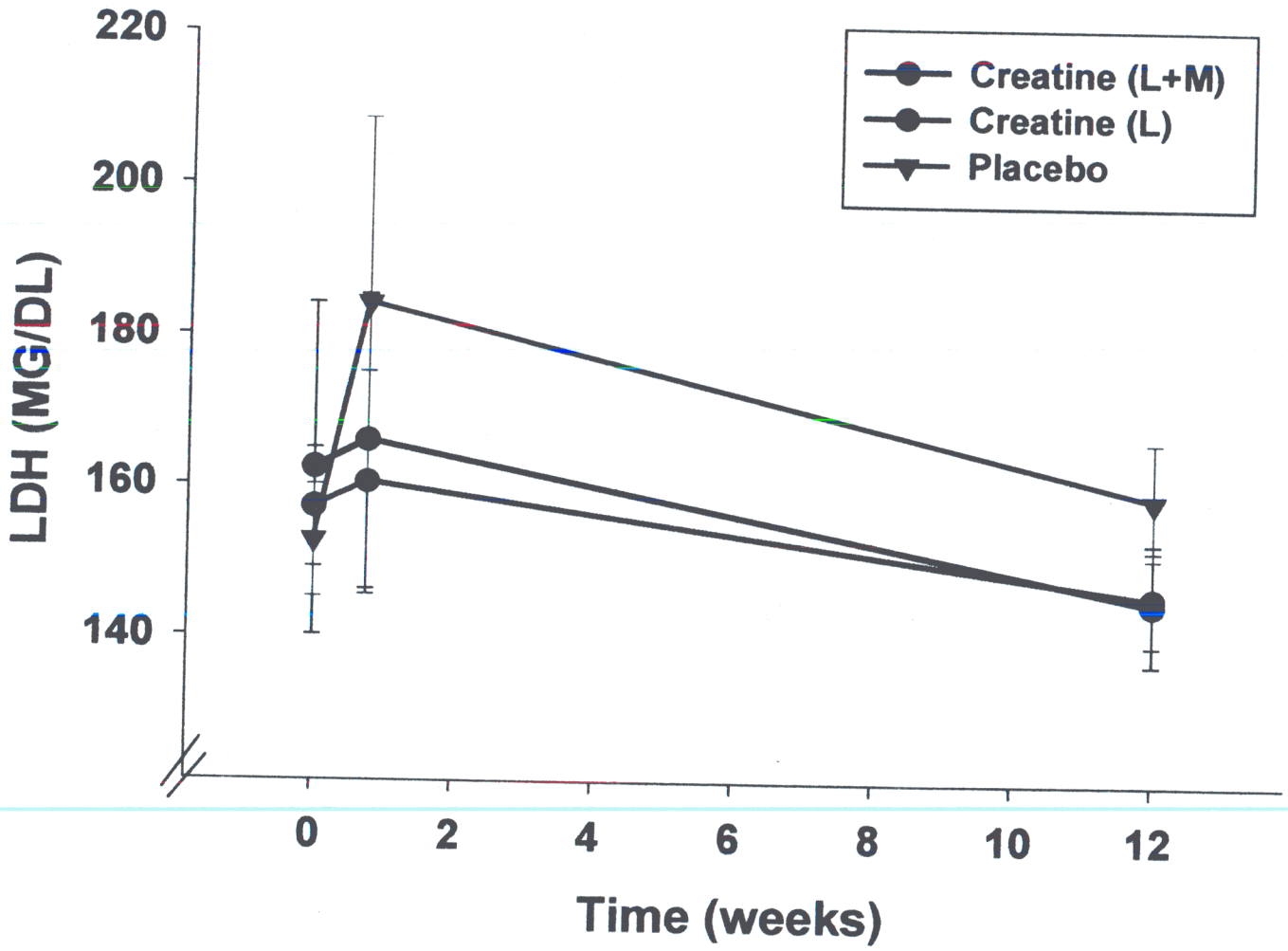

Creatine (L+M)
Creatine (L)
Placebo
Cholesterol/HDL Ratio
5
4
3
0
2
4
6
8
10
12
Time (weeks)

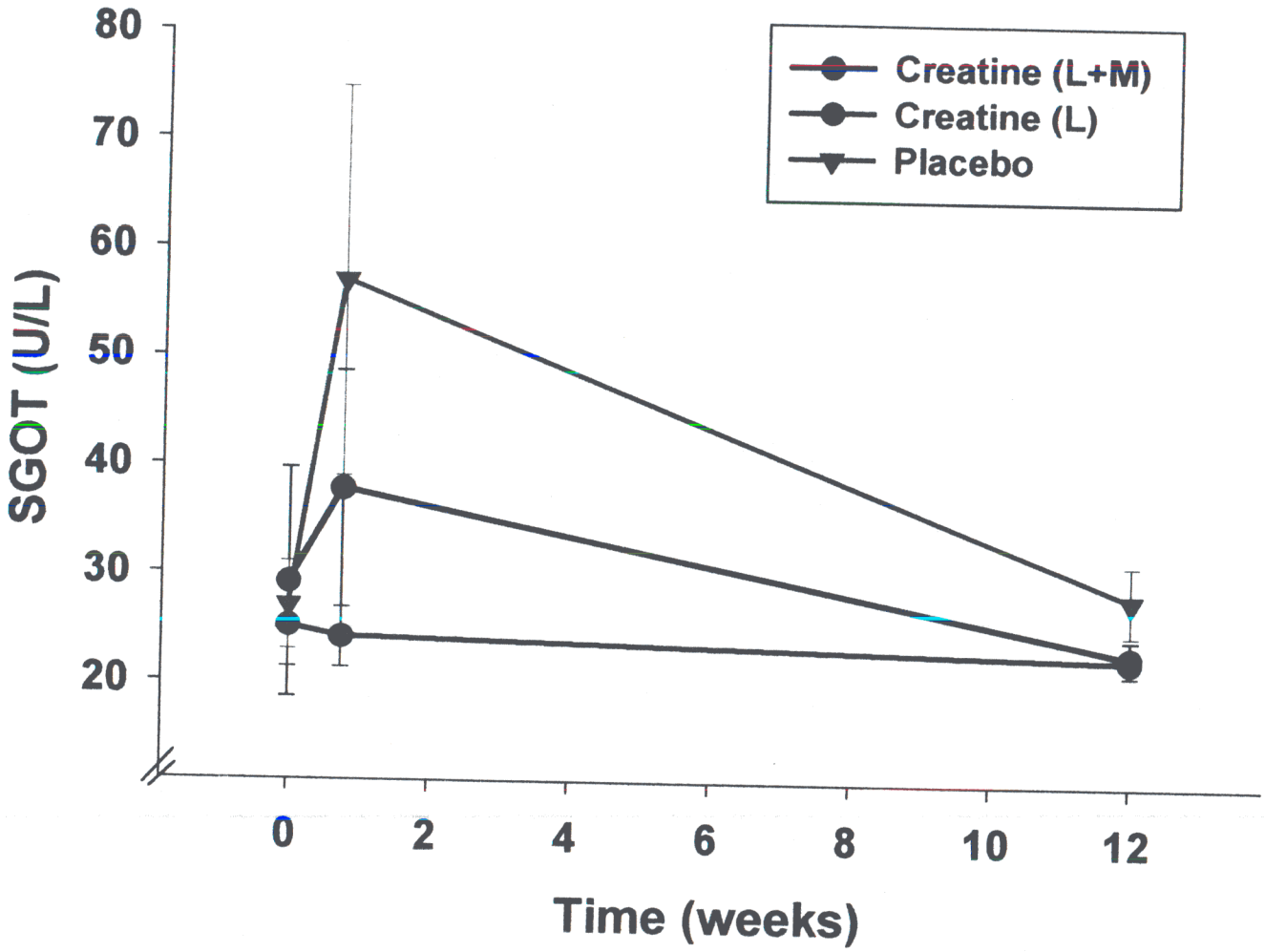

Creatine (L+M)
Creatine (L)
Placebo
Albumin (G/DL)
5
4
0
2
4
6
8
10
12
Time (weeks)

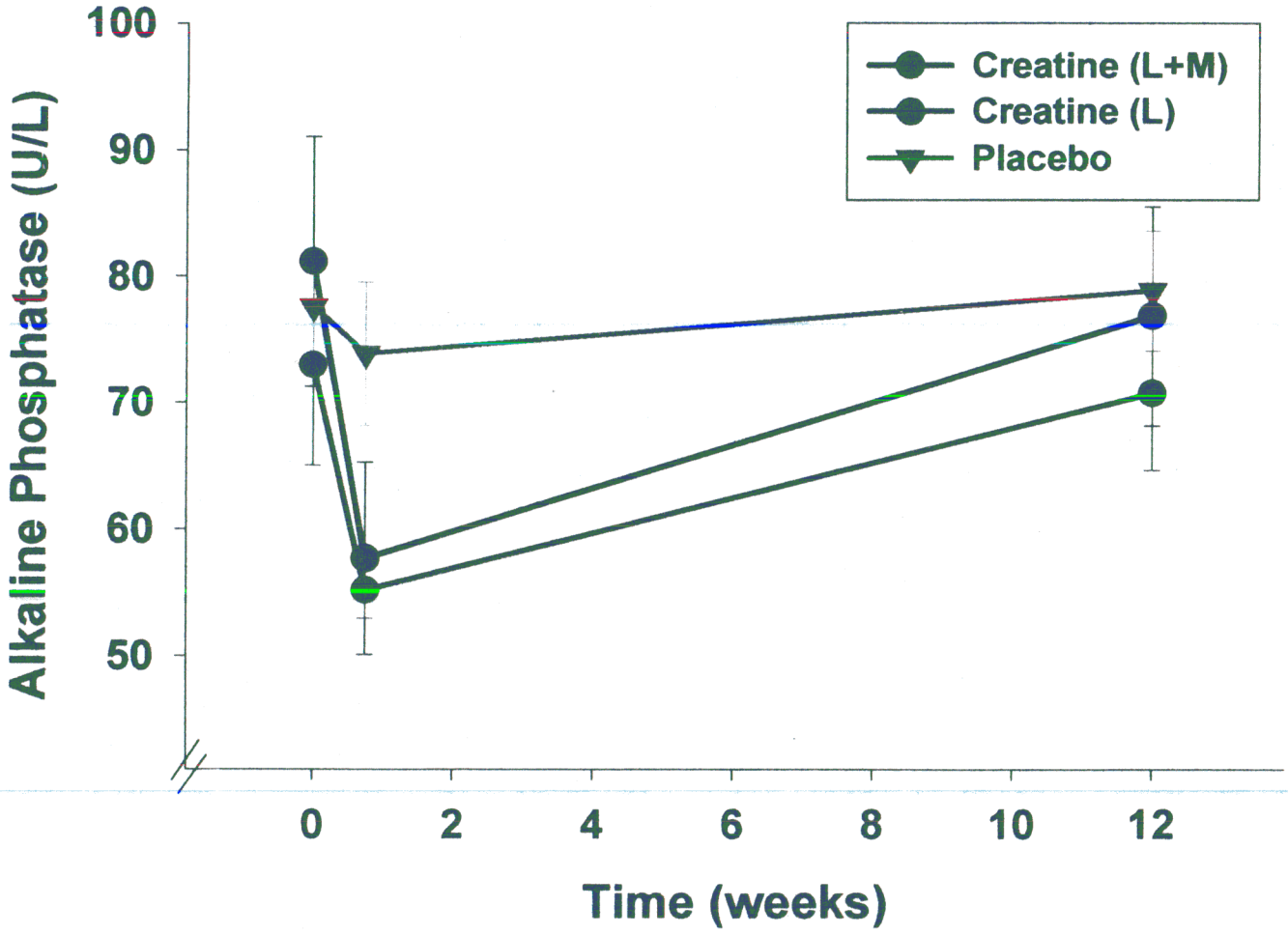

Creatine (L+M)
Creatine (L)
Placebo
Total Bilirubin (MG/DL)
1.3
1.2
1.1
1.0
0.9
0.8
0.7
0.6
0
2
4
6
8
10
12
Time (weeks)

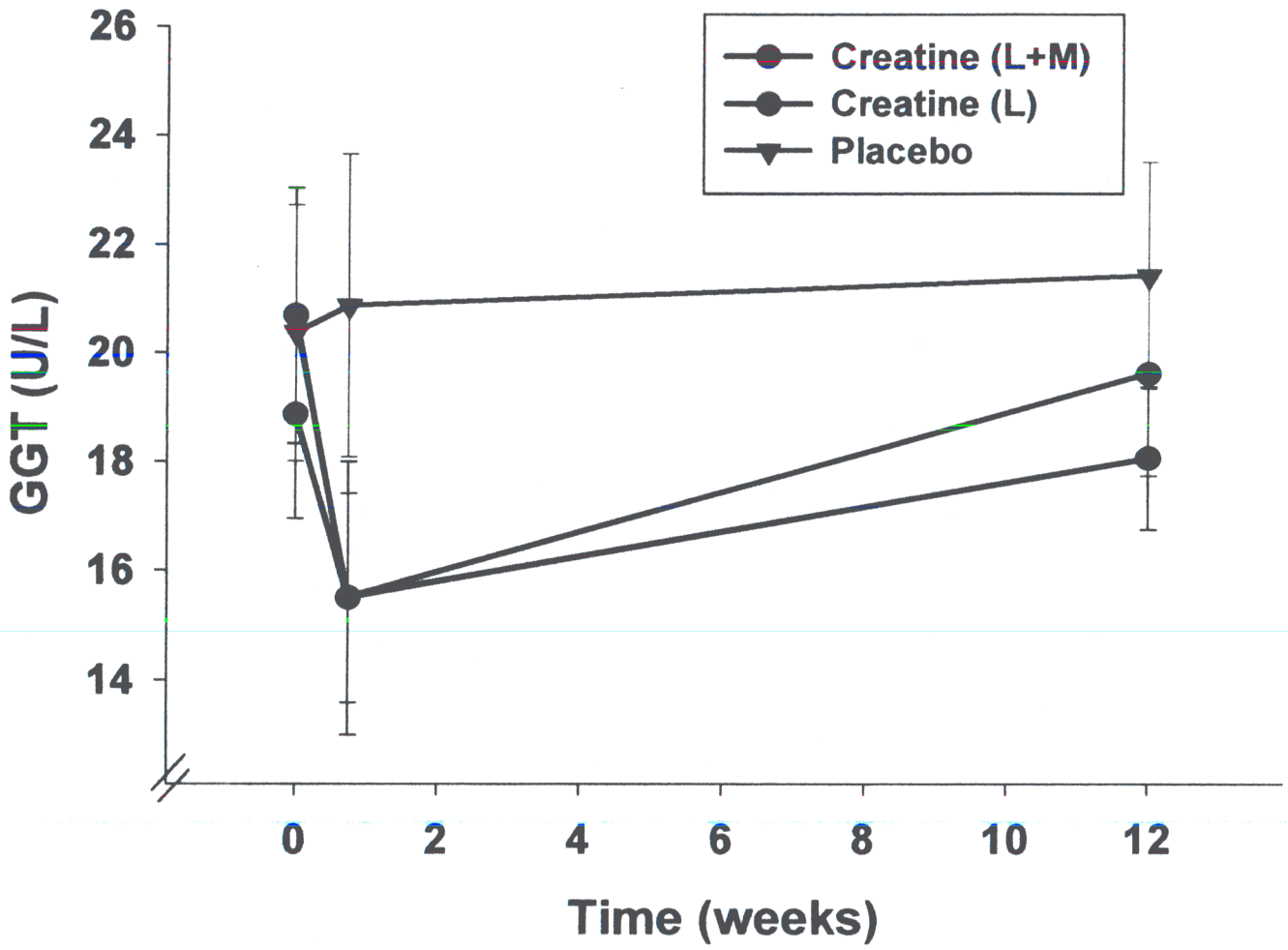

Creatine (L+M)
Creatine (L)
Placebo
BUN (MG/DL)
20
18
16
14
12
10
0
2
4
6
8
10
12
Time (weeks)

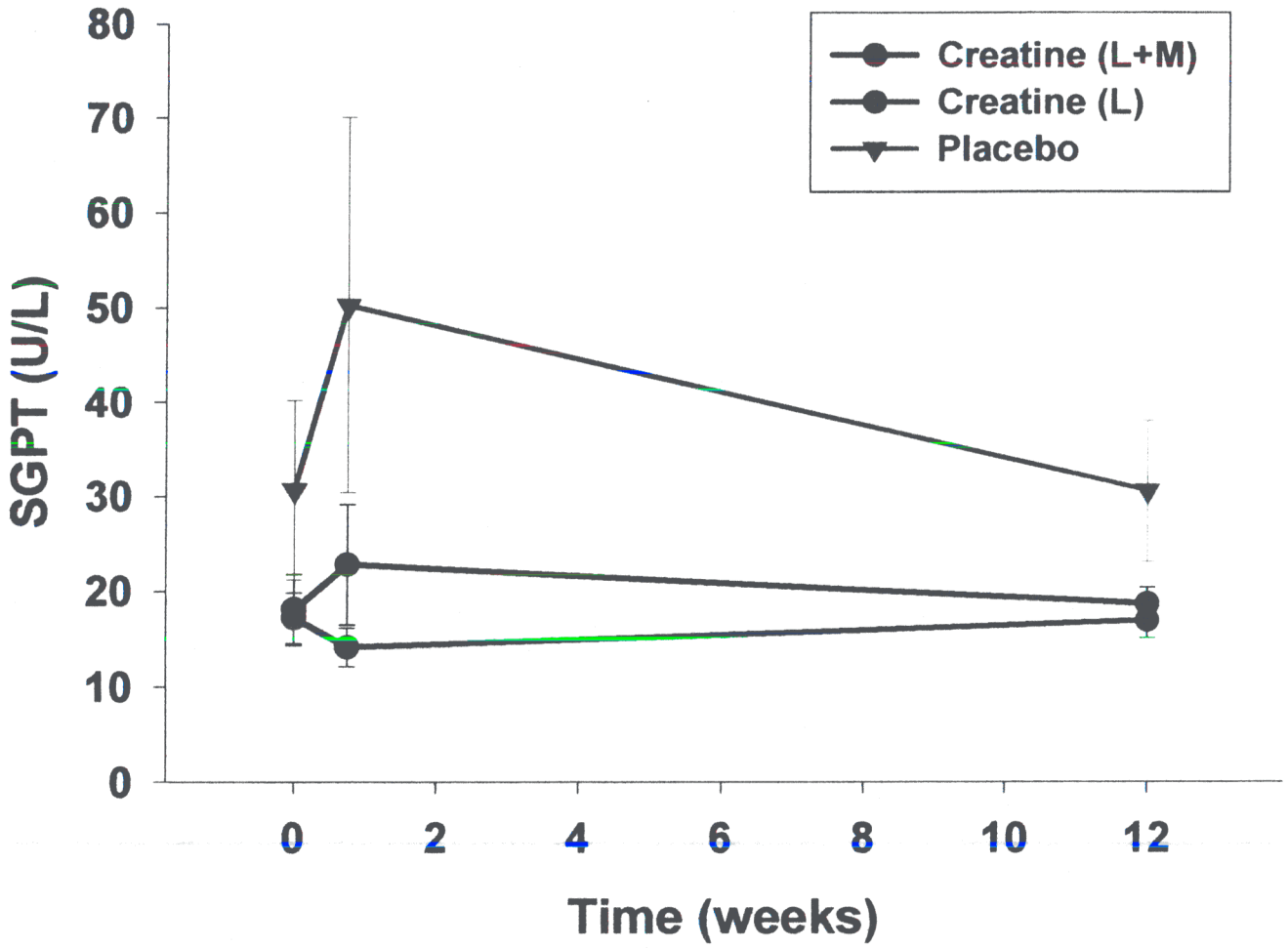

Creatine (L+M)
Creatine (L)
Placebo
LDL (MG/DL)
120
110
100
90
80
70
60
0
2
4
6
8
10
12
Time (weeks)

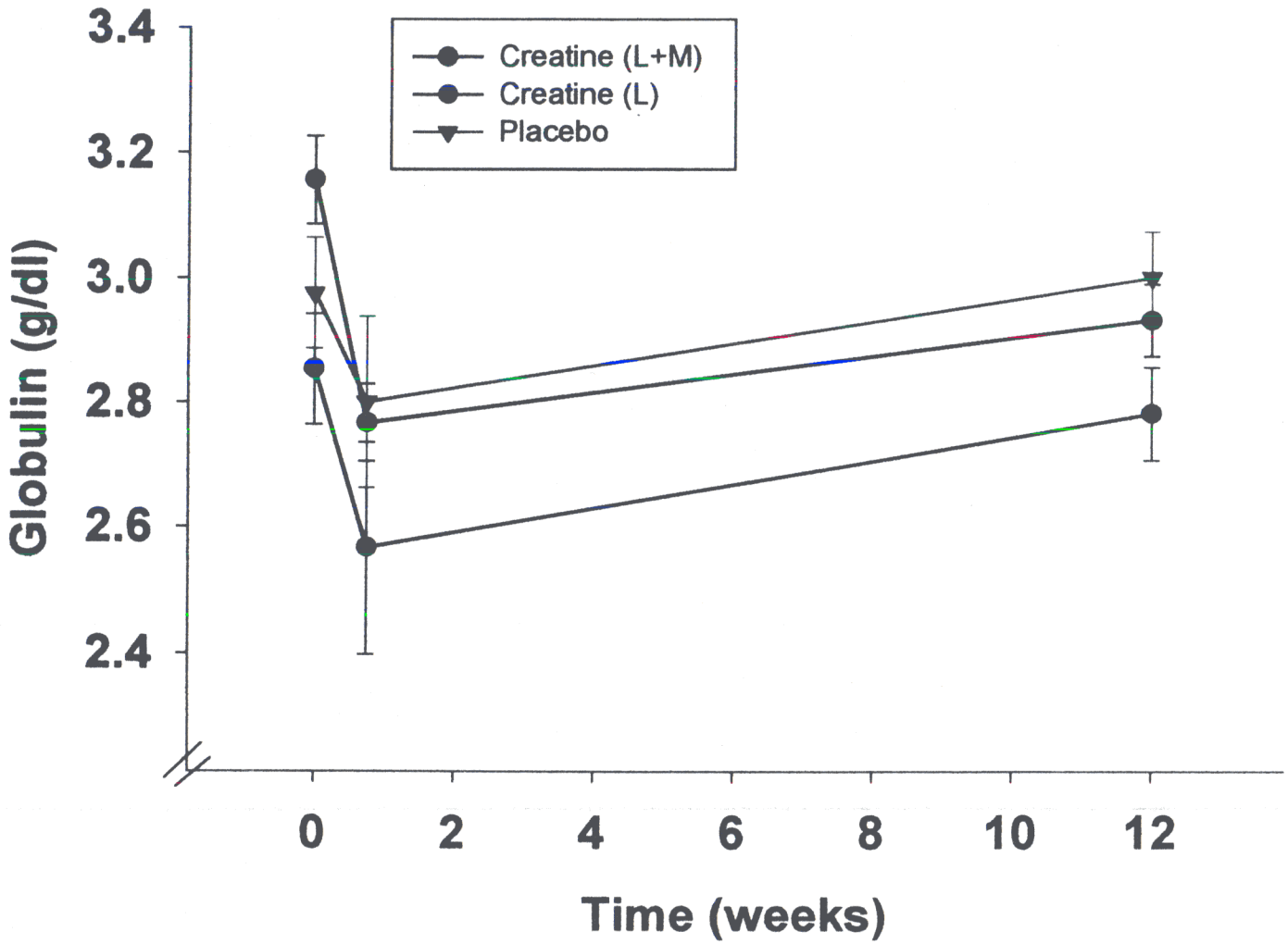

Creatine (L+M)
Creatine (L)
Placebo
LDH (MG/DL)
220
200
180
160
140
0
2
4
6
8
10
12
Time (weeks)


Creatine (L+M)
Creatine (L)
Placebo
SGOT (U/L)
80
70
60
50
40
30
20
0
2
4
6
8
10
12
Time (weeks)


Creatine (L+M)
Creatine (L)
Placebo
Alkaline Phosphatase (U/L)
100
90
80
70
60
50
0
2
4
6
8
10
12
Time (weeks)


Creatine (L+M)
Creatine (L)
Placebo
GGT (U/L)
26
24
22
20
18
16
14
0
2
4
6
8
10
12
Time (weeks)


Creatine (L+M)
Creatine (L)
Placebo
SGPT (U/L)
80
70
60
50
40
30
20
10
0
0
2
4
6
8
10
12
Time (weeks)


Creatine (L+M)
Creatine (L)
Placebo
Globulin (g/dl)
3.4
3.2
3.0
2.8
2.6
2.4
0
2
4
6
8
10
12
Time (weeks)

Group 1 = Creatine Loading and Maintenance
Group 2 = Creatine Loading
Group 3 = Placebo

Table I. Sodium Values During 12 Weeks of Supplementation

Group	Baseline	Loading	12 Weeks
1			
Mean	139.5000	141.0000	140.0625
Std. Error	.5083	.5164	.5437
2			
Mean	140.0667	141.3333	139.750
Std. Error	.5021	.6667	.6090
3			
Mean	140.2500	139.3750	140.1875
Std. Error	.3708	.3750	.5494

Table II. Chloride Values During 12 Weeks of Supplementation

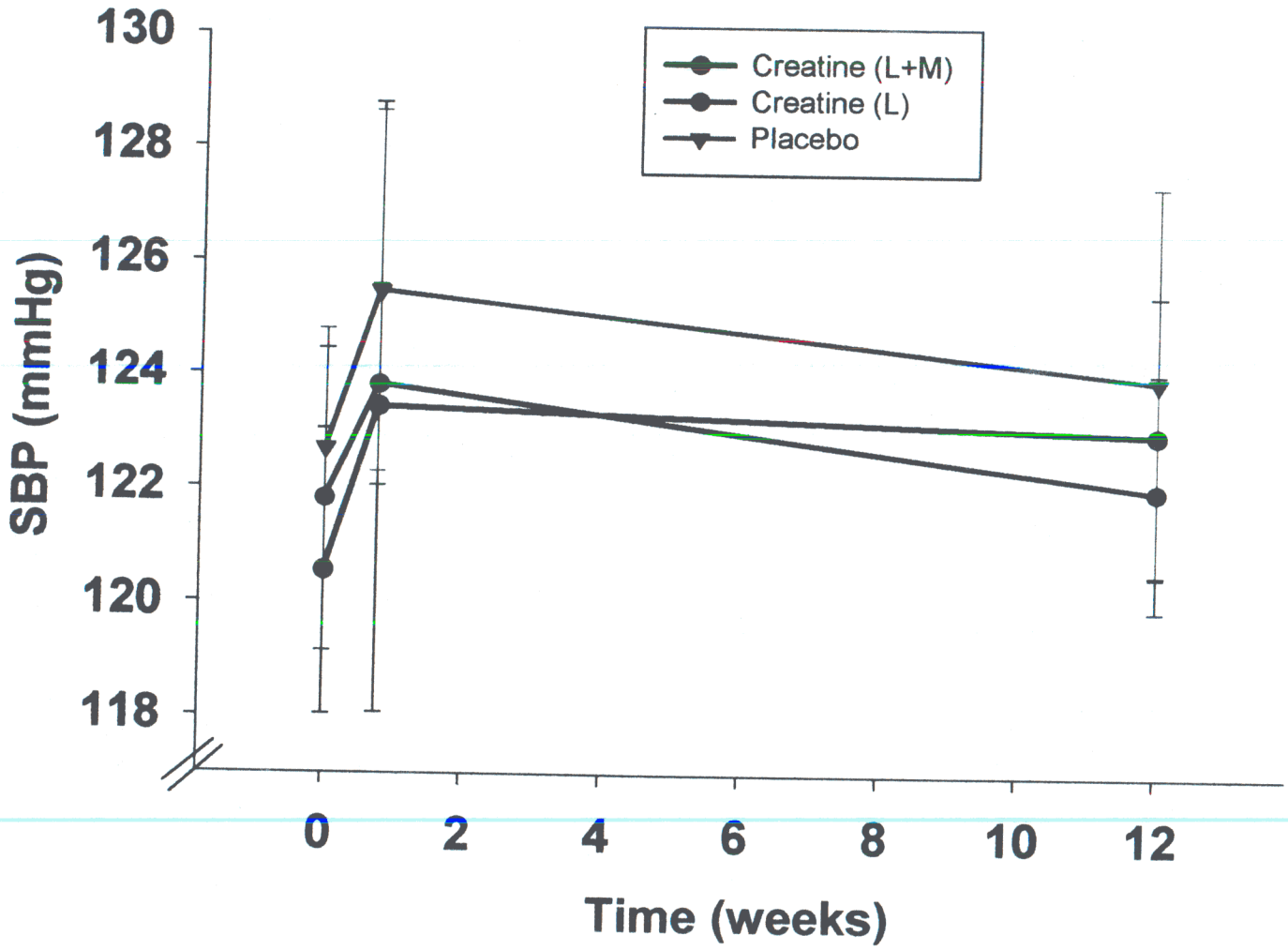
Group	Baseline	Loading	12 Weeks
1			
Mean	102.6250	101.0000	101.8125
Std. Error	.5468	.5164	.5494
2			
Mean	101.4000	101.6667	101.6250
Std. Error	.8095	.9545	.6762
3			
Mean	102.4375	101.1250	101.3750
Std. Error	.7010	1.4322	.9259

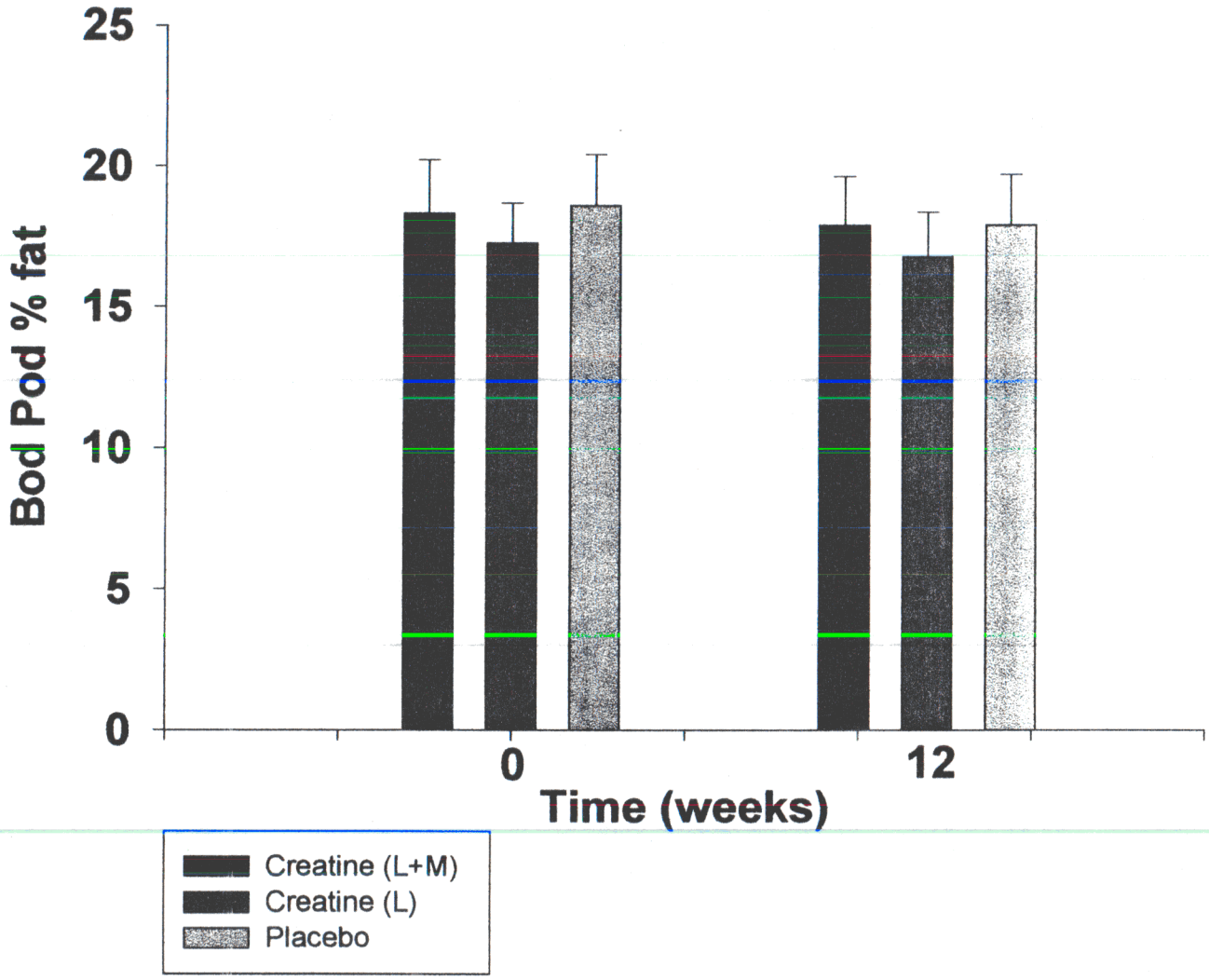
Table III. Phosphate Values During 12 Weeks of Supplementation

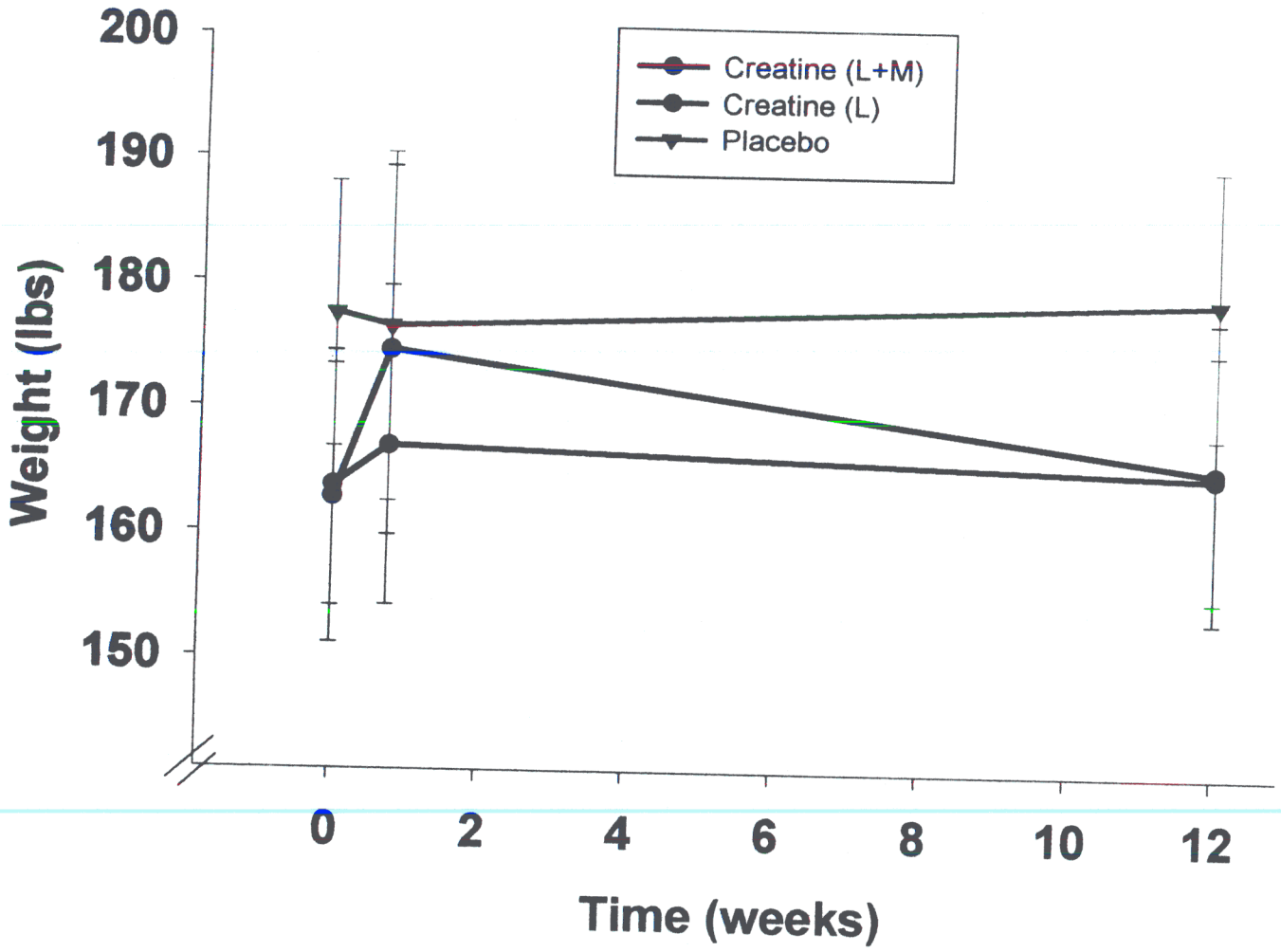
Group	Baseline	Loading	12 Weeks
1			
Mean	4.3438	4.3000	4.0813
Std. Error	.1396	.2646	.1177
2			
Mean	4.3467	3.4833	4.0563
Std. Error	.2023	.2242	.1917
3			
Mean	4.1625	3.9000	3.9438
Std. Error	.1135	.2236	.1396

Table IV. Calcium Values During 12 Weeks of Supplementation

Group	Baseline	Loading	12 Weeks
1			
Mean	9.6	9.55	9.33
Std. Error	.077	.085	.048
2			
Mean	9.7	9.6	9.6
Std. Error	.09	.09	.1917
3			
Mean	9.6	9.4	9.5
Std. Error	.08	.17	.09


Creatine (L+M)
Creatine (L)
Placebo
SBP (mmHg)
130
128
126
124
122
120
118
0
2
4
6
8
10
12
Time (weeks)


25
20
15
10
5
0
Bod Pod % fat
0
12
Time (weeks)
Creatine (L+M)
Creatine (L)
Placebo


Creatine (L+M)
Creatine (L)
Placebo
Weight (lbs)
200
190
180
170
160
150
0
2
4
6
8
10
12
Time (weeks)

Subject Ratings After Loading Phase

Symptoms	CrL+M	CrL	P
Stomach Upset			
≤ 2	68.8%	56.3%	73.3%
≤ 5	87.5%	87.5%	86.7%
*Worst Ranking	10.0 (1)	6.9 (1)	10.0 (1)
Joint Stiffness			
≤ 2	68.8%	56.3%	66.7%
≤ 5	93.8%	93.8%	80.0%
*Worst Ranking	7.1 (1)	5.5 (1)	7.9 (1)
Increase in Urination			
≤ 2	18.8%	62.5%	40.0%
≤ 5	50.0%	68.8%	60.0%
*Worst Ranking	10.0 (1)	9.7 (1)	9.7 (1)
Muscle Soreness			
≤ 2	18.8%	18.8%	26.7%
≤ 5	43.8%	62.5%	60.0%
*Worst Ranking	10.0 (1)	10.0 (1)	10.0 (1)
Cramping			
≤ 2	75.0%	87.5%	60.0%
≤ 5	87.5%	93.8%	80.0%
*Worst Ranking	9.7 (1)	5.2 (1)	6.2 (1)

*numbers indicate highest ranking and the number of subjects that reported the ranking in parentheses.

Subject Ratings After 6 Weeks

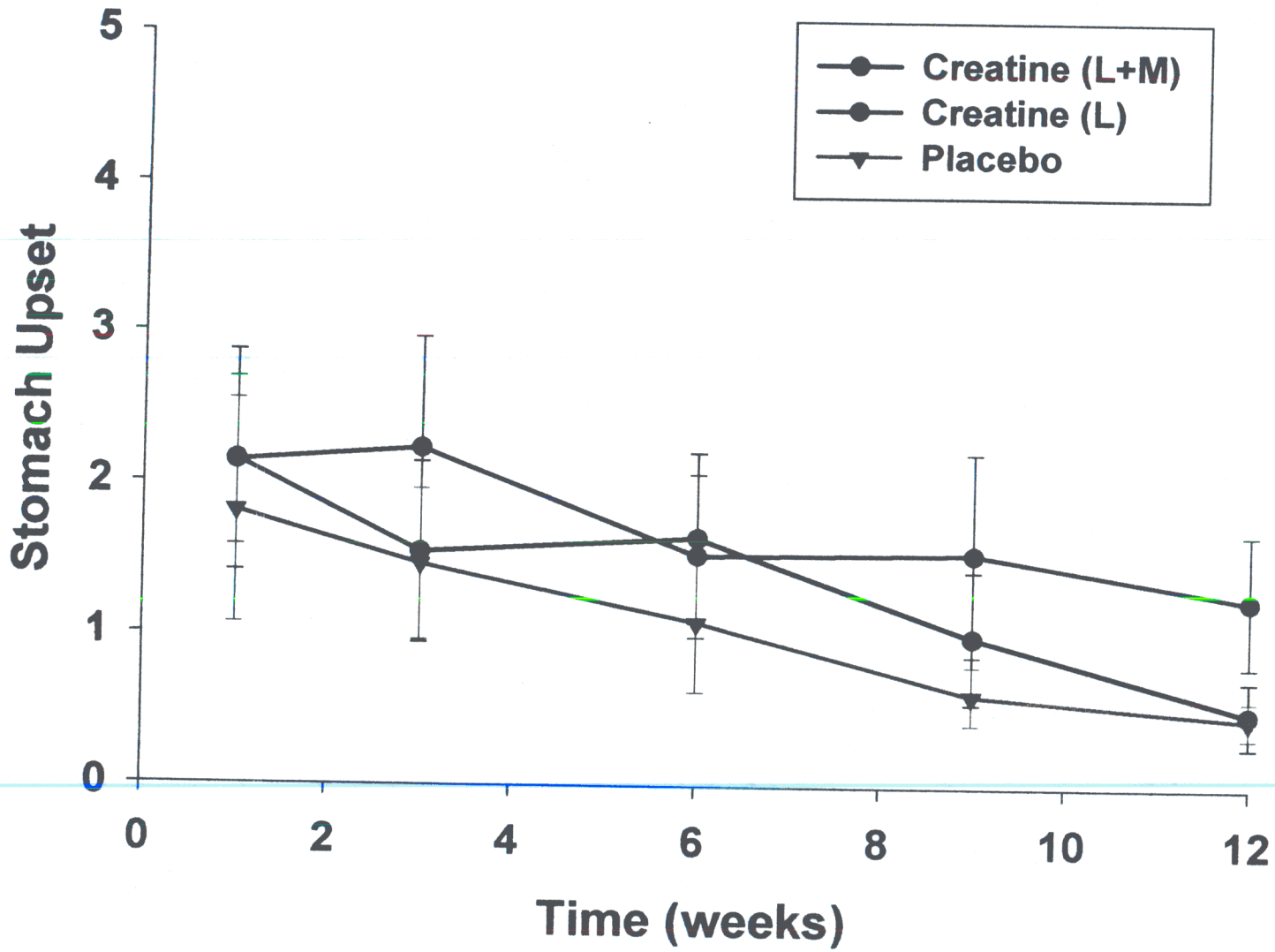
Symptoms	CrL+M	CrL	P
Stomach Upset			
≤ 2	68.8%	75.0%	85.7%
≤ 5	87.5%	93.8%	92.9%
*Worst Ranking	5.7 (1)	8.0 (1)	6.4 (1)
Joint Stiffness			
≤ 2	62.5%	56.3%	57.1%
≤ 5	93.8%	100.0%	100.0%
*Worst Ranking	8.2 (1)	4.8 (1)	2.9 (1)
Increase in Urination			
≤ 2	68.8%	81.3%	78.6%
≤ 5	87.5%	87.5%	100.0%
*Worst Ranking	9.0 (1)	7.2 (1)	4.3 (1)
Muscle Soreness			
≤ 2	50.0%	50.0%	57.1%
≤ 5	81.3%	75.0%	92.9%
*Worst Ranking	8.7 (1)	7.9 (1)	5.4 (1)
Cramping			
≤ 2	68.8%	81.3%	78.6%
≤ 5	75.0%	100.0%	92.9%
*Worst Ranking	9.4 (1)	3.7 (1)	5.1 (1)

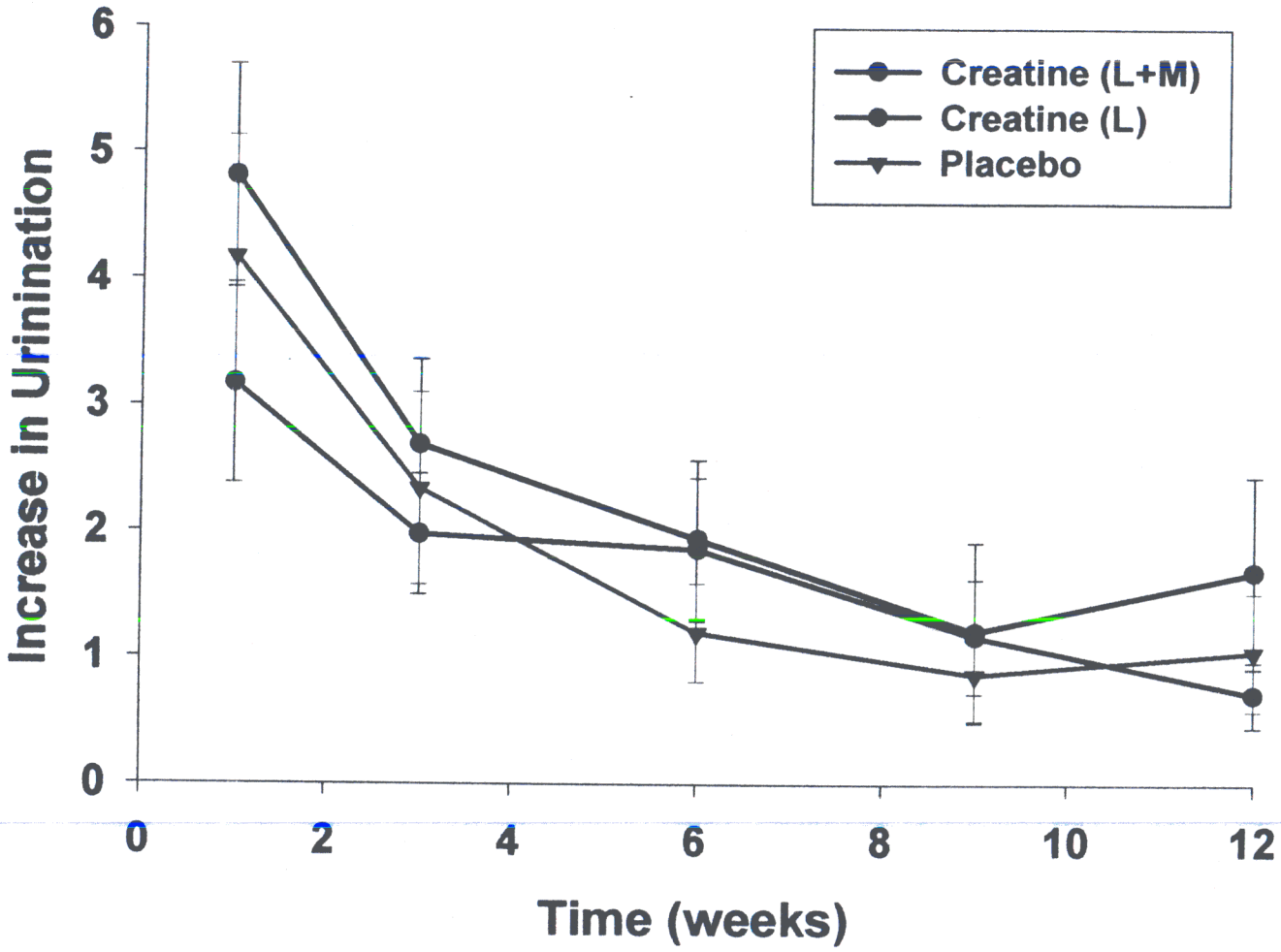
*numbers indicate highest ranking and the number of subjects that reported the ranking in parentheses.

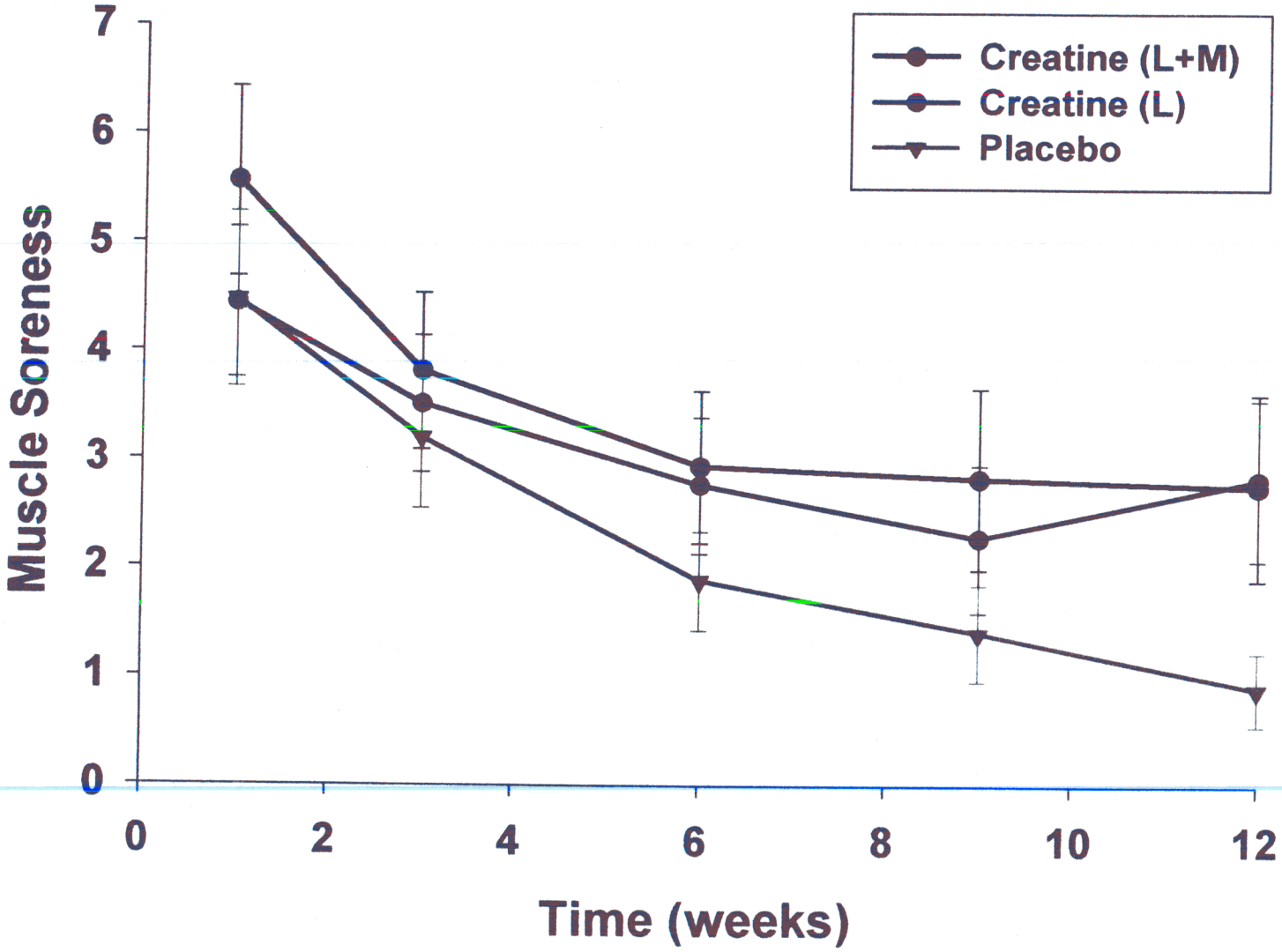
Subject Ratings After 12 Weeks

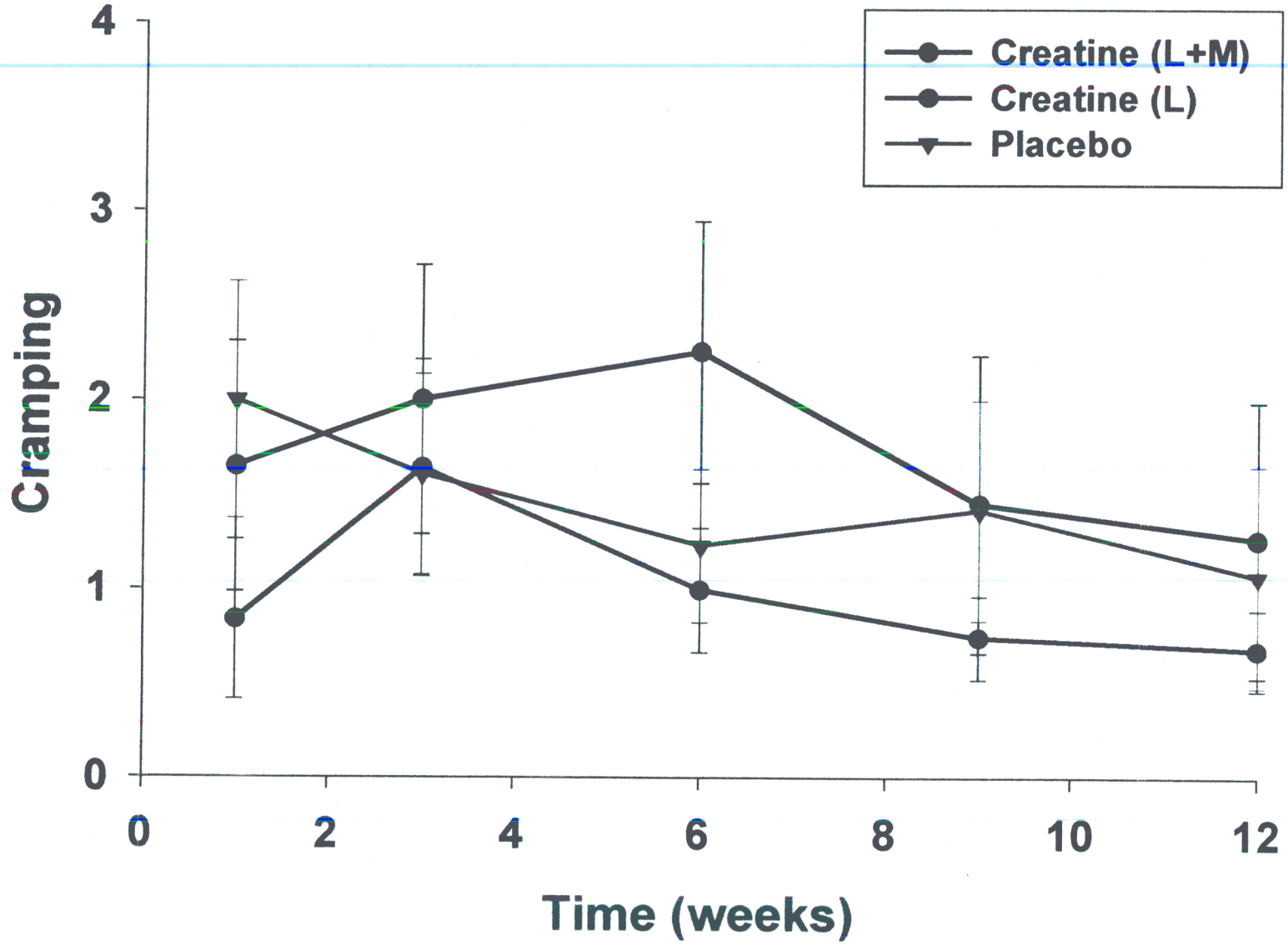
Symptoms	CrL+M	CrL	P
Stomach Upset			
≤ 2	92.3%	85.7%	100.0%
≤ 5	100.0%	92.9%	100.0%
*Worst Ranking	2.4 (1)	6.0 (1)	1.2 (1)
Joint Stiffness			
≤ 2	53.8%	64.3%	83.3%
≤ 5	76.9%	92.9%	100.0%
*Worst Ranking	9.6 (1)	6.0 (1)	3.6 (1)
Increase in Urination			
≤ 2	76.9%	92.9%	83.3%
≤ 5	92.3%	100.0%	100.0%
*Worst Ranking	9.7 (1)	3.8 (1)	4.8 (1)
Muscle Soreness			
≤ 2	53.8%	50.0%	91.7%
≤ 5	76.9%	78.6%	100.0%
*Worst Ranking	9.6 (1)	8.8 (1)	4.2 (1)
Cramping			
≤ 2	84.6%	85.7%	83.3%
≤ 5	92.3%	100.0%	91.7%
*Worst Ranking	9.6 (1)	2.6 (1)	7.0 (1)

*numbers indicate highest ranking and the number of subjects that reported the ranking in parentheses.


Creatine (L+M)
Creatine (L)
Placebo
Stomach Upset
0
1
2
3
4
5
0
2
4
6
8
10
12
Time (weeks)


Increase in Urinination
6
5
4
3
2
1
0
0
2
4
6
8
10
12
Time (weeks)
Creatine (L+M)
Creatine (L)
Placebo


Creatine (L+M)
Creatine (L)
Placebo
Muscle Soreness
0
1
2
3
4
5
6
7
0
2
4
6
8
10
12
Time (weeks)


Creatine (L+M)
Creatine (L)
Placebo
Cramping
4
3
2
1
0
0
2
4
6
8
10
12
Time (weeks)

INJURY RATING SHEET

NAME_____ SPORT_____
DATE_____

Instructions:

Rate your feelings during this week of creatine supplementation. Each horizontal line below is designed to represent various degress of a particular sensation: LEFT edge of the line represents the absence of a sensation (or 0 on a 10 point scale) and the right edge of the line represents the highest amount of that sensation that you have ever experienced (or 10 on a 10 point scale). Please make a vertical mark on each horizontal line at a point that indicates the magnitude of the sensation that you experience in response to taking the creatine supplement. We do not have a particular expected outcome in mind and our primary goal is for you to provide ratings that are as accurate and honest as possible. If you are uncertain as to the exact meaning of one of the words that constitutes the verbal anchors on the left and right of the horizontal lines please ask us to provide a definition.

1. **The amount of stomach upset I experienced this week was:**

NONE_____VERY, VERY, VERY MUCH

2. The amount of joint stiffness I experienced this week was:

NONE_____VERY, VERY, VERY MUCH

3. **Any increase in urination that I experienced this week was:**

NONE_____VERY, VERY, VERY MUCH

4. **The amount of muscle soreness I experienced this week was:**

NONE_____VERY, VERY, VERY MUCH

5. **The amount of muscle cramping I experienced this week was:**

NONE_____VERY, VERY, VERY MUCH

Please describe THE TYPE OF CRAMP and SITUATION it occurred and HOW OFTEN it occurred this week.

Did you visit the training room for treatment this week? If Yes, for what injury/ illness?

Are you taking any other medication currently (i.e. aspirin, anti-inflammatory etc.)?

Describe the amount and volume of training for the past 7 days:

	Frequency (days/wk)	Duration (min/miles each session)	Intensity (moderate, hard)
Weight training			
Sport specific			
Other			