Original Research

Effect of Chromium Supplementation and Exercise on Body Composition, Resting Metabolic Rate and Selected Biochemical Parameters in Moderately Obese Women Following an Exercise Program

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Key words: chromium, body composition, glucose tolerance, iron, obese women, zinc

Objective: To investigate the effect of chromium picolinate (CP) supplementation on body composition, resting metabolic rate (RMR), selected biochemical parameters and iron and zinc status in moderately obese women participating in a 12-week exercise program.

Methods: Forty-four women, 27 to 51 years of age, were randomly assigned to two groups based on their body mass index. Subjects received either 400 μg/day of chromium as a CP supplement or a placebo in double-blind fashion and participated in a supervised weight-training and walking program two days per week for 12 weeks. Body composition and RMR were measured at baseline, 6 and 12 weeks. Selected biochemical parameters and iron and zinc status were measured at baseline and 12 weeks.

Results: Body composition and RMR were not significantly changed by CP supplementation. No significant differences in fasting plasma glucose, serum insulin, plasma glucagon, serum C-peptide and serum lipid concentrations or in iron and zinc indices were found between the two groups over time. Serum total cholesterol concentration significantly decreased ($p = 0.0016$) over time for all subjects combined, probably as a result of the exercise training. Exercise training significantly reduced total iron binding capacity (TIBC) by 3% for all subjects combined ($p = 0.0011$).

Conclusions: Twelve weeks of 400 μg/day of chromium as a CP supplement did not significantly affect body composition, RMR, plasma glucose, serum insulin, plasma glucagon, serum C-peptide and serum lipid concentrations or iron and zinc indices in moderately obese women placed on an exercise program. The changes in serum total cholesterol levels and TIBC were a result of the exercise program.

INTRODUCTION

Chromium, an essential trace element required for normal carbohydrate, protein and fat metabolism [1], may improve impaired glucose tolerance [2–4], decrease elevated blood lipid concentrations [5–7] and result in weight loss and improved body composition [8–11] in some individuals, but results have been equivocal. As the active component of chromodulin, chromium has been suggested to potentiate the action of insulin, possibly by increasing insulin binding, insulin binding receptor number, improving insulin internalization and increasing insulin sensitivity [12,13]. Signs and symptoms of chromium deficiency in mammals include glycosuria, neuropathy, encephalopathy, decreased insulin binding and receptor number and impaired immune response [14].

While much of the research on the role of nutritional chromium has focused on its effects on blood glucose and lipid concentrations, popular interest has centered on chromium as a means to increase muscle mass and reduce body fat in obese individuals. The suggested beneficial effect of chromium on body composition was based on the rationale that chromium potentiates the functions of insulin. Insulin functions in

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transporting glucose and amino acids into muscle cells, regulating protein metabolism and synthesis. Therefore, improvements in insulin utilization should theoretically lead to increased muscle mass and reduced body fat [15]. The link between these contradictory changes in body composition may be due to chromodulin's ability to potentiate insulin's effects on converting glucose to carbon dioxide or lipid [13,16]. However, the effects of chromium picolinate (CP) supplementation on lean body mass (LBM), percent body fat and body weight are equivocal. Evans [8] showed significantly increased LBM and decreased percent body fat in young males who supplemented with 200 μg of chromium/day as CP and participated in a resistance training program. However, skinfold thickness measurements, which have a high rate of error, were used to assess body composition. In contrast, Clancy *et al.* [17] did not observe any significant changes in body composition following supplementation with 200 μg of chromium/day as CP in football players on a weight training regimen. Under water weighing, considered the gold standard for body composition analysis, was used to assess LBM and percent body fat in their study. Several studies in untrained males also found no beneficial effects of CP supplementation on body composition [18,19].

With the increasing interest in chromium supplementation for improving glucose tolerance and body composition, the adverse affects of chromium supplementation in humans has been somewhat overlooked. It has been shown *in vitro* and in animal studies *in vivo*, that chromium and iron compete for the same binding site on transferrin [20,21]. Lukaski *et al.* [19] demonstrated that supplementation with 172 μg of chromium/day as CP and 182 μg of chromium/day as chromium chloride combined with a weight training program significantly reduced urinary iron excretion compared with a placebo in male subjects. Thus, chromium supplementation may result in reducing iron status, as the body tends to conserve iron by decreasing urinary iron loss [19]. In addition, because plasma zinc and iron compete for the same binding site, chromium could potentially impair zinc metabolism.

Therefore, the purpose of this study was to examine the effects of supplementation with 400 μg of chromium/day as chromium picolinate on body composition, resting metabolic rate (RMR), plasma glucose, serum insulin, serum C-peptide, plasma glucagon and serum lipid concentrations, iron indices and plasma zinc concentrations in sedentary, moderately obese women who participated in a 12-week exercise program. Plasma, red blood cell and urinary chromium concentrations were also assessed in order to monitor changes of chromium status after CP supplementation.

MATERIALS AND METHODS

Subjects

Forty-four female subjects, with a body mass index (BMI) between 27 to 41, initially began the study. Subjects were between 27 to 51 years of age, pre-menopausal, sedentary, not taking any dietary supplements, not taking any vitamin or mineral supplements containing chromium (but could take supplements containing other vitamins and minerals), not on a weight-loss program, not taking any weight loss supplements, non-smoking and with no history of chronic diseases or recent acute illness. All subjects gave written and verbal informed consent, completed a medical history questionnaire and were required to obtain permission from their physician prior to participation. Subjects were asked not to alter their dietary habits during the course of the study, with the exception of taking the supplement or the placebo. Furthermore, subjects were asked to adhere to the supervised exercise program and not to perform any additional exercise. This study was first approved by the University of Massachusetts Human Subjects Committee.

Experimental Design

This was a double-blind, placebo-controlled study conducted for 12 weeks. Twelve weeks was chosen because other chromium supplementation studies, some with fewer subjects and lower chromium supplementation levels, were conducted for 12 weeks or less [8,9,15,19].

Forty-four free-living females were assigned, in a stratified randomized manner based on their BMI, to one of two groups: CP supplementation or placebo. The subjects received gel capsules containing either 400 μg of CP (400 μg of CP, with dicalcium phosphate as filler, and stearic acid as a lubricant) (Nutrition21, San Diego, CA) or identical looking placebo capsules (dicalcium phosphate as filler, stearic acid as a lubricant and natural pink colorant; no chromium was contained within the placebo capsules) (Nutrition21, San Diego, CA) at the beginning of the study. The chromium content of the capsules was analyzed by Nutrition21 (San Diego, CA) and contained 400 μg of chromium as chromium picolinate. Because subjects were obese, we chose to administer above the Dietary Reference Intakes (DRI) of 25 μg/day [22] and the former Estimated Safe and Daily Dietary Intake (ESADDI) of 50 to 200 μg/day [23] in order to assess if higher CP intakes would elicit significant body composition changes. The subjects were asked to take their capsules every morning during the entire 12-week study period. Capsule compliance was monitored by asking subjects how many capsules were left in their packet as well as assessment of plasma and red blood cell chromium concentrations and 24-hour urinary chromium analyses.

Measurements

Due to difficulty with scheduling subjects for data collection, all subjects were assessed for pre-test measurements within approximately two months prior to beginning the study. After stratified randomization, we staggered the subjects into groups to begin the supplementation and exercise regimen;

therefore, when we assessed them at post-test, there was, on average, two weeks' lag time from completion of the study and post-test assessment.

Body Composition

Body composition was measured at pre-, mid- and post-test by hydrostatic weighing as described by McArdle *et al.* [24]. Prior to underwater weighing, vital capacity (VC) was measured using the metabolic cart ($Cardio_2$Max metabolic cart, Medical Graphics Corporation, St. Paul, MN). Residual lung volume (RLV) was then calculated from VC as RLV = 0.28*VC [25]. Body weight in air was measured before entering the water tank on a balance-beam scale accurate to $\pm$0.5 kg while the subject was wearing a swim suit. Each subject performed the weighing procedure for at least ten trials. The three highest trials were averaged and used to compute body density, percent body fat, fat mass and LBM using Siri's equation [26]: percent body fat = (4.95/density) − 4.5. Percent coefficient of variation (%CV) for hydrostatic weighing = 7.3% to 8.3%.

Circumferences (cm) of the waist and hips were measured three times consecutively at each site. A tape measure was firmly applied to all areas without compressing the skin. The average of each site was calculated. Measurements that were greater than 2 cm different were not used and a re-trial was performed; thus, there were always three measurements averaged per site.

Resting Metabolic Rate

RMR was measured at pre-, mid- and post-test. RMR was measured after a 12-hour fast while each subject was lying comfortably in a reclining chair in a room with comfortable temperature. Prior to assessing RMR, each subject rested approximately 10 to 15 minutes. RMR (kcal/day) was measured for 30 minutes using a dilution hood connected to a metabolic cart ($Cardio_2$Max metabolic cart, Medical Graphics Corporation, St. Paul, MN) (%CV = 10.8% to 12.2%).

Blood Collection and Analyses

A total of 50 mL of blood was drawn from each subject by a registered nurse at pre- and post-test: in the fasted state (30 mL), one hour post-meal consumption (10 mL) and two hours post-meal consumption (10 mL). Subjects were given a 300 kilocalorie (kcal) meal, high in sugar, provided by the researchers in order to assess the effects of a meal high in sugar on blood chromium, zinc, glucose, insulin, glucagon and C-peptide concentrations. The meal included juice (180 mL), fruit in heavy syrup (120 mL), toasted whole wheat bread (one slice) and grape jelly (15 g) (Table 1). Monovette (Sarstedt Inc., Princeton, NJ) mineral-free tubes were used for blood collection of plasma (lithium heparin) and serum. In addition, Monovette (Sarstedt Inc., Princeton, NJ) needles were used to draw the blood.

Following the blood collection, red blood cells were separated from serum or plasma via centrifugation at 3000 rpm ($1500 \times g$) for 15 minutes. Serum blood samples were allowed to clot for one hour prior to centrifugation. Plasma samples were centrifuged immediately after each blood draw. The clear supernatant was aliquoted into properly labeled 1.5 mL Eppendorf tubes (Outpatient Services, Petaluma, CA) using disposable, mineral-free, transfer pipettes. The tubes for all determinations were stored at −80°C until required for analyses. All blood samples were analyzed, in duplicate, for the biochemical parameters described as follows:

Blood Glucose, Insulin, Glucagon and C-Peptide Analyses. Plasma glucose concentration (%CV = 1.0 to 1.5%) was measured by hexokinase phosphorylation (Hitachi 912, Japan) at University Health Services, University of Massachusetts, Amherst. Serum insulin (%CV = 3.0 to 4.5%), plasma glucagon (%CV = 3.2% to 6.5%) and serum C-peptide (%CV = 3.0% to 3.2%) concentrations were measured by radioimmunoassay (RIA) (Diagnostic Products, Corp., Los Angeles, CA). The radioactivity in each sample was determined using a gamma counter (Beckman gamma 4000 counting system, Beckman Instruments, Inc., Fullerton, CA). Serum C-peptide was measured for two reasons: it has a positive correlation with insulin concentrations, and it assesses pancreatic beta-cell secretory function [27].

Blood Lipid Analyses. Serum total cholesterol (TC) (%CV = 1.2% to 1.7%), serum low density lipoprotein cholesterol (LDL-C) (%CV = 2.4% to 3.3%), serum high density

Table 1. Detailed Composition of High Carbohydrate Meal Consumed by All Subjects Prior to Second and Third Blood Draws at Pre- and Post-Test

Food item[1]	Amount	Energy (kcal)[2]	Carbohydrates (sugars) (g)	Protein (g)	Lipids (g)	Dietary fiber (g)
Juice	180 mL	100	26 (26)	—	—	—
Fruit in heavy syrup	120 mL	90	23 (22)	—	—	1
Toasted whole bread	1 slice	60	12 (2)	3	1	2
Grape jelly	15 g	50	13 (10)	—	—	—
Total		300	74 (60)	3	1	3

[1] Juice = Capri Sun All Natural (White Plains, NY), Fruit in heavy syrup = Sweet Life Fruit Cocktail (Chaska, MN), Toasted whole wheat bread = Arnold 100% Stone Ground Whole Wheat Bread (Totowa, NJ), Grape jelly = Sweet Life Grape Jelly (Suffield, CT).

[2] kcal = kilocalories.

lipoprotein cholesterol (HDL-C) (%CV = 2.2% to 3.7%), and serum triacylglycerol (TG) (%CV = 1.4% to 3.3%) concentrations were determined by enzymatic kits (Sigma Diagnostic Co., St. Louis, MO). A spectrophotometer (Beckman Instruments, Inc., Fullerton, CA) was used for analysis at 500 nanometers for TC, LDL-C and HDL-C and 540 nanometers for TG.

Blood Iron Analyses. SmithKline Beecham Laboratory (Waltham, MA) analyzed the serum for the respective iron indices as follows: serum iron (%CV = 0.5% to 1.5%) and TIBC (%CV = 0.5% to 1.0%) were measured by spectrophotometry [28]; transferrin saturation (there is no %CV for transferrin saturation because it is a calculated value) was calculated from serum iron and TIBC [29]; serum ferritin (%CV = 6.7% to 7.0%) concentration was measured using RIA [30].

Blood Zinc Analysis. Plasma zinc concentrations (%CV = 8.4% to 19.4%) were measured at fasting and one and two hours postprandially with an atomic absorption spectrophotometer (AAS) (Perkin Elmer 2380, Norwalk, CT). Plasma samples were diluted eightfold with 5% nitric acid. Zinc standards, prepared from zinc reference solution (Fisher Scientific, Pittsburgh, PA) in 5% nitric acid, were used as an internal control.

Blood Chromium Analysis. Plasma chromium (%CV = 75% to 142%) and red blood cell (RBC) chromium (%CV = 159% to 256%) concentrations were measured at fasting and one and two hours postprandially with an AAS-Graphite Furnace (AAS-GF) (Perkin-Elmer 4100 ZL, Perkin-Elmer, Norwalk, CT). Note the exceptionally high %CV for both plasma and RBC chromium concentrations. This was a result of a combination of very low concentrations of chromium in plasma and RBC as well as a high degree of between-subject variation. Although the %CV's were high, we observed increases in plasma chromium concentration in the CP group, as expected. The Perkin-Elmer 4100 ZL AAS was equipped with a longitudinal Zeeman background correction and a pyrolytically coated transversely heated graphite atomizer with an integrated L'vov platform. All measurements were conducted at 357.9 nm resonance line. Sample aliquots of 20 μL for plasma and RBC were delivered using the Perkin-Elmer AS-71 autosampler. Argon was used as the protective gas.

The needles (Sarstedt Inc., Princeton, NJ) used to draw blood were analyzed for chromium contamination by a Spectromass 2000 inductively coupled plasma-mass spectroscopy (ICP-MS) (Spectro Analytical Instruments, Fitchburg, MA). The needles analyzed were rinsed several times with saline to represent blood. Next, the saline solution was analyzed for chromium content and compared to blanks. The background equivalent concentration was analyzed to assess the lower limits of this approach.

Urine Collection and Analyses

At the end of each test day at pre-, mid- and post-test, acid-washed urine containers were distributed to the subjects for 24-hour urine collections. The 24-hour urine collections were weighed, aliquotted into acid-washed tubes and concentrated trace-element grade hydrochloric acid (Fisher Scientific, Pittsburgh, PA) was added to each sample to prevent leaching of minerals by the container. The sample tubes were then kept in a −80°C freezer until analyses. All samples were analyzed, in duplicate, for urinary iron (%CV = 80% to 184%) and urinary chromium (%CV = 49% to 66%) concentrations. Similar to blood chromium concentrations, both urinary iron and chromium concentrations had a high %CV. This again was a result of a combination of very low concentrations of urinary iron and chromium as well as a high degree of between-subject variation. Although the %CV's were high, we observed increases in urinary chromium concentration in the CP group, as expected. Urinary sample analysis was similar to plasma and RBC analyses; however, sample aliquots of urine were delivered at 40 μL by the Perkin-Elmer AS-71 autosampler.

Urine samples were diluted 1:20 with distilled water prior to analysis. The chromium concentration was determined by AAS-GF (Perkin-Elmer 4100 ZL, Perkin-Elmer, Norwalk, CT). Twenty-four-hour urinary iron concentration was determined by AAS (Perkin-Elmer 703, Perkin-Elmer, Norwalk, CT) [31].

Dietary Intake

Nutrient intakes were assessed by three-day dietary records at pre-, mid- and post-test in order to monitor dietary patterns during the study. The dietary records included two weekdays and one weekend day. Subjects were instructed on how to report their dietary intakes properly and not alter their usual dietary habits for the duration of the study. The three-day dietary records were analyzed for total energy, carbohydrate, fat, protein, total dietary iron, absorbable iron, animal iron, zinc, calcium, vitamin C and dietary fiber using the University of Massachusetts Nutrient Data Bank (UMNDB). Dietary chromium intake was not analyzed by the UMNDB because of the lack of chromium reference data. However, estimation of dietary chromium was calculated on the basis of the estimated chromium concentration of the reported usual diet consumed in the United States, which is approximately 15 μg chromium/1000 kcal [32].

Training Protocol

All subjects participated in a 12-week supervised moderately-intense exercise program at an exercise facility on the University of Massachusetts, Amherst campus. Subjects exercised two days per week, one hour per session. This frequency and duration were chosen to increase compliance and were sufficient to elicit a training response. Subjects performed 30 minutes of weightlifting and 30 minutes of moderately-intense walking. Prior to the program, the weightlifting workout was calculated from 60% of one-repetition maximum (1-RM). Subjects' 1-RM was measured on each piece of weight-training

equipment. Instruction on appropriate weight-training techniques was given; then, a warm-up of several repetitions at very low weights was performed by each subject. One-RM was recorded as the maximum amount of weight the subject could lift just once.

The weightlifting program consisted of two sets of 8 to 12 repetitions using the following isokinetic equipment: biceps curls, triceps press, chest press, latissimus dorsi pull down, leg curls, leg extension, calf raises, leg adduction and abduction. Training logs were reviewed weekly to increase the weight load, when necessary. Once subjects performed two sets of 12 repetitions, the weight load was increased by 10 pounds for each particular piece of equipment. Therefore, this was a progressive weight training program; exercise intensity was increased based on individual progress and represented a greater percentage of 1-RM than the initial 60%. Strength, which was recorded as 1-RM, was monitored at pre- and post-test for assessing the efficacy of the weight-training program.

Following the weight-training protocol, subjects performed a 30-minute walking regimen within their specified heart rate range ([220-age] $\times$ 60% to 80%); 60% to 80% represents a moderately intense aerobic exercise program [33]. Subjects were properly trained on how to take radial and carotid pulse for 10 seconds (then multiplied by six for the actual beat per minute). Pulse was recorded before walking, at ten-minute intervals while walking and five minutes post-walking.

Statistical Analyses

Data were analyzed with the Statistical Analysis Software (SAS version 6.12; SAS Institute, Inc, Cary, NC). A power estimation was calculated to determine sample size. A total of 40 subjects (20 per group) would have been required for 80% power. We initially had 44 subjects in this study and thus met the sample size necessary to detect statistical differences. A 2×3 factorial analysis of variance (ANOVA) for treatment (placebo and chromium supplementation) and time (pre-test, mid-test and post-test) with repeated measures over time was used to analyze body composition, RMR, urinary chromium, urinary iron, urinary creatinine and dietary data. A 2×2 factorial ANOVA for treatment (placebo and chromium supplementation) and time (pre-test and post-test) with repeated measures over time was used to analyze the serum and plasma biochemical data. Tukey's post hoc test was performed when significant main effects or interactions occurred. The level of significance was set *a priori* at 0.05 for overall statistical analyses. Data are expressed as mean $\pm$ standard deviation (SD).

RESULTS

Compliance

Thirty-seven of the initial 44 subjects (84%) completed this study. Sixteen subjects reported forgetting to take their capsules and 13 subjects missed exercise training; however, no subjects reported forgetting to take the capsules more than five days (average <3 days) during the study period. Furthermore, none of these subjects missed exercise training more than four times (average <3) during the entire study. Two subjects did have minor problems with their knees or ankles, so they were unable to perform all of the specified weight training exercises (e.g., leg extension and calf raises).

Anthropometric Parameters

Age, height, body weight and BMI of the chromium and placebo groups were not significantly different at baseline (Table 2).

Body Composition

No significant main effect differences or interactions were found between the two groups over time in body weight, BMI, waist circumferences or hip circumferences, although there were non-significant decreases in circumferences over time (Table 3). Furthermore, there were no significant changes in response to the CP supplementation in percent body fat, LBM and fat mass values (Table 3). However, percent body fat and fat mass significantly decreased and LBM significantly increased at post-test compared with baseline due to the exercise training, for both groups combined. Nonetheless, the chromium group showed a tendency to have a greater decrease in percent body fat from pre- to mid- to post-test when compared to the placebo group. The mean decline of percent body fat was 1.78% with a maximum decrease of 7.54% in the chromium group, compared to a mean decline of 0.63% with a maximum decrease of 3.91% in the placebo group.

Resting Metabolic Rate

RMR values at pre-, mid- and post-test are presented in Fig. 1. At mid-point, the average RMR showed a slight, but non-significant increase in both groups. At post-test, RMR slightly decreased, but it was still greater than the average pre-test levels in both groups.

Table 2. Subject Characteristics at Pre-Test for the Chromium and Placebo Groups

	Chromium Group (n = 22)	Placebo Group (n = 22)
Age (years)	42.6 $\pm$ 6.5	42.5 $\pm$ 4.2
Height (cm)	164.6 $\pm$ 5.6	164.3 $\pm$ 5.6
Body weight (kg)	87.6 $\pm$ 11.0	89.8 $\pm$ 11.4
BMI[1] (kg/m^2)	32.5 $\pm$ 4.3	33.2 $\pm$ 4.2

Values represent means $\pm$ standard deviation (SD).

[1] BMI = body mass index.

Note: No significant differences between groups.

Table 3. Anthropometric Measurements at Pre-, Mid- and Post-Test for the Chromium and Placebo Groups

	Chromium Group			Placebo Group		
	Pre-Test (n = 22)	Mid-Test (n = 21)	Post-Test (n = 20)	Pre-Test (n = 22)	Mid-Test (n = 21)	Post-Test (n = 17)
Body weight (kg)	87.6 ± 11.0	87.7 ± 10.9	87.6 ± 10.7	89.8 ± 11.4	89.2 ± 11.6	88.0 ± 9.8
BMI[1] (kg/m^2)	32.5 ± 4.3	32.6 ± 4.6	32.7 ± 4.1	33.2 ± 4.2	33.1 ± 4.2	32.4 ± 3.7
Waist[2] (cm)	96.4 ± 8.6	92.9 ± 8.4	91.7 ± 9.3	98.4 ± 11.5	95.1 ± 10.8	92.3 ± 8.8
Hip[2] (cm)	118.0 ± 8.0	116.5 ± 7.1	114.4 ± 9.0	118.9 ± 8.4	117.2 ± 7.9	114.9 ± 6.6
Waist:Hip Ratio	0.82 ± 0.1	0.80 ± 0.1	0.80 ± 0.1	0.83 ± 0.1	0.81 ± 0.1	0.80 ± 0.1
Percent Body Fat (%)	46.15 ± 3.2	44.79 ± 3.2	44.57 ± 3.5	45.69 ± 3.7	44.84 ± 4.4	44.99 ± 3.2
LBM[3] (kg)	47.19 ± 6.5	48.54 ± 6.0	48.54 ± 5.9	48.50 ± 6.4	49.23 ± 6.3	48.22 ± 5.8
Fat Mass (kg)	40.71 ± 6.3	39.54 ± 6.4	39.28 ± 6.7	41.07 ± 7.0	40.33 ± 7.7	39.54 ± 5.6

Values represent means ± SD.

[1] BMI = body mass index.

[2] Waist and hip values are circumference measurements.

[3] LBM = lean body mass.

Note: No significant differences within or between groups over time; however, percent body fat and fat mass significantly decreased ($p < 0.05$) and LBM significantly increased ($p < 0.05$) pre- to post-test for both groups combined as a result of the exercise program.

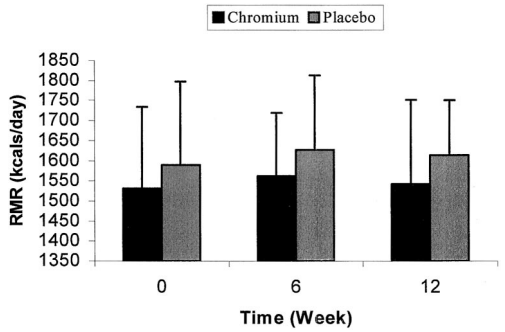

Fig. 1. Resting metabolic rate (RMR) at pre-, mid- and post-test for the chromium and placebo groups. Values represent mean ± SD. Note: No significant differences within or between groups over time.

Biochemical Parameters

Blood Glucose, Insulin, Glucagon, and C-Peptide Concentrations. Table 4 represents plasma glucose, serum insulin, plasma glucagon and serum C-peptide concentrations at pre- and post-test. There were no significant effects seen in any of these measurements in response to the chromium picolinate supplementation. Fasting plasma glucose concentrations of all subjects ranged from 52 to 138 mg/dL at the beginning of the study (only one subject was hyperglycemic [>126 mg/dL] with a blood glucose of 138 mg/dL; she was in the placebo group). At one hour post-meal, mean glucose concentration increased in both groups, but were not significantly different between the two groups. Although one subject in the chromium group had a higher than normal fasting plasma glucose concentration of 119 mg/dL, CP supplementation did not affect her levels over time. Furthermore, this same subject had a post-test fasting blood glucose level of 149 mg/dL.

Blood Lipid Concentrations. Serum lipid concentrations are presented in Table 5. No significant changes were observed between groups over time in serum LDL-C and TG concentrations; however, significant decreases ($p < 0.05$) were found pre- to post-test in serum TC and HDL-C concentrations when both groups were combined.

Iron Indices. The average serum iron concentration, TIBC, transferrin saturation and serum ferritin concentration are shown in Table 6. All values were within normal ranges for the two groups at pre- and post-test measurements. Serum iron concentrations, TIBC, transferrin saturation and serum ferritin concentrations were not significantly different at pre- and post-test between the two groups. Although serum iron concentration and transferrin saturation slightly decreased in both groups over time, the changes were not significant. However, a tendency ($p = 0.104$) was observed for serum ferritin concentration to decrease in response to exercise training in both groups. TIBC significantly decreased over time in both groups ($p = 0.0011$).

Blood Zinc Concentrations. Plasma zinc concentrations are presented in Table 7. There were no significant differences within or between groups at pre- or post-test. However, there was a significant decrease ($p < 0.01$) between the fasting state and two hours after a meal for all subjects combined.

Blood Chromium Concentrations. The needles used to draw blood were assessed for chromium, and there were no detectable levels of chromium found in the needles; thus, there was no contamination of the plasma and RBC chromium samples.

Pre- and post-test plasma chromium and RBC chromium concentrations are shown in Table 8. After 12 weeks of CP supplementation and exercise training, fasting plasma chromium concentration in the chromium group significantly increased ten-fold compared with pre-test values. At post-test, plasma chromium concentrations in the chromium group in the fasted state and two hours after a high carbohydrate meal were significantly higher than in the placebo group. However, plasma chromium concentration did not change in response to a high carbohydrate meal within either group at pre- or post-test. The RBC chromium concentration in the fasted state

Table 4. Plasma Glucose, Serum Insulin, Serum C-Peptide and Plasma Glucagon Concentrations at Pre- and Post-Test: Fasting and One and Two Hours after a High Carbohydrate Meal for the Chromium and Placebo Groups

	Fasting State	1 Hour Post-Meal	2 Hours Post-Meal	Normal Values* (Fasting)
Glucose (mg/dL)				70 to 110
Cr-Pre[1] (n = 21)	91.38 ± 12.7	105.19 ± 26.7	87.62 ± 14.0	
Cr-Post (n = 20)	89.90 ± 20.8	98.85 ± 27.5	86.50 ± 14.4	
P-Pre (n = 21)	91.14 ± 15.7	121.15 ± 42.0	87.40 ± 8.2	
P-Post (n = 17)	91.65 ± 7.3	102.37 ± 25.2	85.63 ± 7.3	
Insulin (μIU/mL)				3 to 35
Cr-Pre (n = 21)	22.39 ± 13.4	89.18 ± 72.1	35.68 ± 41.6	
Cr-Post (n = 20)	19.71 ± 14.4	92.91 ± 68.8	33.16 ± 26.0	
P-Pre (n = 21)	16.57 ± 7.6	95.67 ± 66.1	29.36 ± 20.4	
P-Post (n = 17)	18.02 ± 10.2	91.28 ± 70.3	25.58 ± 16.3	
Glucagon (pg/mL)				40 to 130
Cr-Pre (n = 21)	150.72 ± 112.0	157.21 ± 116.2	153.45 ± 107.1	
Cr-Post (n = 20)	176.17 ± 118.6	170.02 ± 121.4	165.18 ± 121.7	
P-Pre (n = 21)	116.91 ± 46.5	121.79 ± 49.5	118.57 ± 50.6	
P-Post (n = 17)	103.13 ± 29.6	108.16 ± 25.0	113.99 ± 38.1	
C-Peptide (ng/mL)				0.8 to 4.0
Cr-Pre (n = 21)	1.66 ± 1.5	5.96 ± 4.1	4.22 ± 3.2	
Cr-Post (n = 20)	1.55 ± 1.0	5.79 ± 3.1	3.61 ± 2.6	
P-Pre (n = 21)	1.52 ± 1.0	6.88 ± 3.8	3.83 ± 2.0	
P-Post (n = 17)	1.65 ± 1.3	5.43 ± 3.5	3.44 ± 2.1	

Values represent means ± SD.

[1] Cr = chromium picolinate group, P = placebo group, Pre = pre-test, Post = post-test.

Note: n = 21 for pre-test for chromium and placebo groups because we were unable to obtain sufficient blood samples from 1 subject in the chromium group and 1 subject in the placebo group.

Note: No significant differences between groups over time.

* Normal values from Radioimmunoassay Kits (Diagnostic Products, Corp., Los Angeles, CA).

Table 5. Serum Lipid Levels at Pre- and Post-Test for the Chromium and Placebo Groups

	Chromium Group		Placebo Group		Normal Values*
	Pre-Test (n = 22)	Post-Test (n = 17)	Pre-Test (n = 22)	Post-Test (n = 20)	
Total Cholesterol (mg/dL)	194.8 ± 32.3	187.0 ± 28.0	195.3 ± 45.1	187.0 ± 42.0	≤200
HDL-C[1] (mg/dL)	43.8 ± 10.0	42.5 ± 7.1	45.7 ± 8.0	43.2 ± 7.3	30 to 90
LDL-C[2] (mg/dL)	111.1 ± 25.1	109.2 ± 20.2	103.7 ± 38.5	107.7 ± 37.3	50 to 190
Triacylglycerol (mg/dL)	115.3 ± 53.5	123.0 ± 65.8	109.8 ± 67.3	115.4 ± 68.2	<160

Values represent means ± SD.

[1] HDL-C = High density lipoprotein cholesterol.

[2] LDL-C = Low density lipoprotein cholesterol.

Note: n = 21 at pre-test for serum total cholesterol and serum triacylglycerol concentrations because we were unable to obtain sufficient blood samples from 1 subject in the chromium group and 1 subject in the placebo group.

Serum total cholesterol ($p = 0.0016$) and HDL-C ($p = 0.0313$) levels were significantly decreased from pre- to post-test for both groups combined as a results of the exercise program.

* Normal values from [44].

and one and two hours after a high carbohydrate meal were not significantly different within and between groups over time.

Urinary Iron and Chromium Excretion. Daily urinary iron concentration of both groups at pre-, mid- and post-test are presented in Table 6. The urinary iron concentration was consistent within both groups throughout the study. There were no significant differences in urinary iron concentration between the two groups at each time point.

Table 6. Serum Iron Concentration, Total Iron Binding Capacity, Transferrin Saturation, Serum Ferritin Concentration and Urinary Iron Concentration at Pre-, Mid- (for Urinary Iron Concentration only) and Post-Test for the Chromium and Placebo Groups

	Chromium Group	Placebo Group	Normal Values[#]
Serum Iron (μg/dL)			20 to 52
Pre-test[1]	85.9 ± 29.6	88.4 ± 30.2	
Post-test	81.9 ± 23.3	85.5 ± 24.9	
TIBC[3] (μg/dL)			300 to 400
Pre-test	359.7 ± 47.9	352.9 ± 54.3	
Post-test	347.9 ± 57.3*	342.7 ± 49.8*	
Transferrin Saturation (%)			30% to 50%
Pre-test	24.1 ± 8.5	25.9 ± 10.3	
Post-test	24.0 ± 7.5	25.9 ± 9.6	
Serum Ferritin (ng/mL)			≥30
Pre-test	46.2 ± 34.9	45.6 ± 31.3	
Post-test	37.7 ± 25.5**	42.2 ± 31.1**	
Urinary Iron (μmol/day)			Negligible
Pre-test	2.5 ± 4.6	2.1 ± 1.5	
Mid-test	2.6 ± 1.9	2.1 ± 1.5	
Post-test	2.7 ± 1.9	1.5 ± 1.2	

Values represent means ± SD.

[1] Pre-test chromium group (n = 22), mid-test chromium group (n = 21), post-test chromium group (n = 20), pre-test placebo group (n = 22), mid-test placebo group (n = 21), post-test placebo group (n = 17).

[2] TIBC = Total iron binding capacity.

* Significantly lower than pre-test values ($p = 0.0011$).

** Lower than pre-test values ($p = 0.104$).

[#] Normal values from [27].

Table 7. Plasma Zinc Concentration at Pre- and Post-Test: Fasting and One and Two Hours after a High Carbohydrate Meal for the Chromium and Placebo Groups

	Fasting State	1 Hour Post-Meal	2 Hours Post-Meal	Normal Values*
Plasma Zinc (μg/mL)				0.7 to 1.20
Pre-Cr[1] (n = 22)	0.885 ± 0.132	0.766 ± 0.091	0.679 ± 0.057	
Post-Cr (n = 20)	0.838 ± 0.136	0.749 ± 0.135	0.704 ± 0.108	
Pre-P (n = 22)	0.861 ± 0.167	0.779 ± 0.102	0.722 ± 0.079	
Post-P (n = 16)	0.838 ± 0.150	0.825 ± 0.144	0.739 ± 0.108	

Values represent means ± SD.

[1] Cr = chromium picolinate group, P = placebo group, Pre = pre-test, Post = post-test.

Note: When both groups were combined, 2-hours post-meal zinc concentrations were lower than fasting state values ($p < 0.01$). This was due to the effect of the meal. n = 16 at post-test for the placebo group because we were unable to obtain sufficient blood samples from 1 subject in the placebo group.

* Normal values from [44].

Daily urinary chromium concentrations at pre-, mid- and post-test for both groups are presented in Fig. 2. Urinary chromium concentrations were significantly elevated at mid-test ($p < 0.05$) in the chromium group and returned to near baseline at post-test. In addition, mid-test urinary chromium concentration in the chromium group was significantly higher than in the placebo group ($p < 0.05$).

Dietary Intake

The average dietary intakes pre-, mid- and post-test are shown in Table 9. These values were an average from the three-day dietary records at each time point. There were no significant differences in total energy, carbohydrate, fat, protein, total dietary iron, absorbable iron, zinc, calcium, vitamin C and dietary fiber between the two groups at each time point. Nonetheless, iron intake was below the DRI (18 mg/day) for 35, 28 and 10 subjects for pre-test, mid-point and post-test, respectively. In addition, zinc intake was below the DRI (8 mg/day) for 14, 9 and 22 subjects for pre-test, mid-point and post-test, respectively [22].

Within each group, subjects' dietary intake was also unchanged throughout the study, except the protein intake. There were significant time and group effects and time x group interactions for total protein and percent protein intake. Total

Table 8. Plasma and Red Blood Cell (RBC) Chromium Concentrations Pre- and Post-Test: Fasting and One and Two Hours after a High Carbohydrate Meal for the Chromium and Placebo Groups

	Fasting State	1 Hour Post-Meal	2 Hours Post-Meal	Average Values
Plasma Chromium (ng/mL)				
Pre-Cr[1] (n = 22)	0.26 ± 0.33	1.01 ± 1.58	0.60 ± 1.78	0.2 to 0.5**
Post-Cr (n = 20)	2.62 ± 2.70*,[a]	1.24 ± 1.30	1.73 ± 1.28[a]	0.2 to 1.5***
Pre-P (n = 22)	0.33 ± 0.40	0.38 ± 0.45	0.12 ± 0.17	
Post-P (n = 17)	0.40 ± 0.41[b]	0.42 ± 0.68	0.50 ± 0.80[b]	
RBC Chromium (ng/mL)				No values established
Pre-Cr (n = 21)	0.63 ± 1.87	0.46 ± 0.76	0.49 ± 0.76	
Post-Cr (n = 20)	0.86 ± 2.21	0.56 ± 0.59	1.02 ± 1.73	
Pre-P (n = 22)	0.74 ± 2.15	0.41 ± 0.57	1.04 ± 1.87	
Post-P (n = 17)	0.32 ± 0.51	0.76 ± 2.10	0.58 ± 1.05	

Values represent means ± SD.

[1] Cr = chromium picolinate group, P = placebo group, Pre = pre-test, Post = post-test.

* Significantly greater than pre-test values ($p = 0.02$).

[a,b] Values in the same column with different superscript letters are significantly different from one another ($p < 0.02$).

** Average reported pre-supplementation range (from [5,36,37]).

*** Average reported post-supplementation range (from [5,37]).

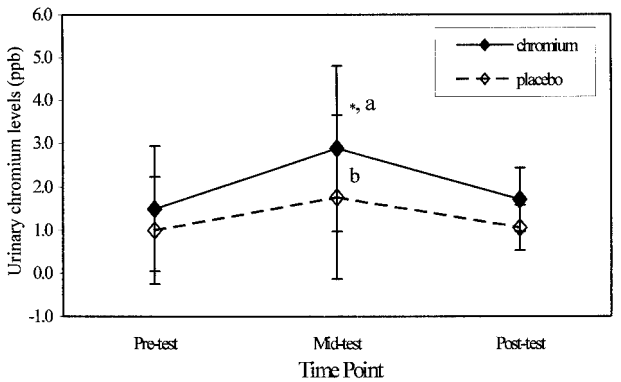

Fig. 2. Urinary chromium levels at pre-, mid- and post-test for the chromium and placebo groups. Values expressed as Mean ± SD. *Significantly different from pre-test ($p < 0.05$). [a,b]Values in the same time-point with different letters are significantly different from one another ($p < 0.05$). Note: Average reported pre-supplementation range = 0.2 to 0.8 ppb (from [4] and [45]). Average reported post-supplementation range = 0.8 to 1.5 ppb (from [4]).

protein intake was significantly lower at post-test compared to pre-test in the chromium group ($p < 0.05$). The placebo group had a significant decrease in total protein intake at post-test compared to mid-test ($p < 0.05$).

Because there is insufficient information on chromium in the database, a crude estimation of dietary chromium intake was calculated by multiplying the energy intakes at each time point by 15 μg chromium/1000 kcal on the basis of the estimated chromium concentration of the usual diet consumed in the United States [32]. The estimated dietary chromium intakes in the chromium and placebo groups ranged from 21 to 43 μg/day and 23 to 44 μg/day, respectively.

Strength Gains

The average of 1-RM values at pre- and post-test are presented in Table 10. There were no significant differences between groups; however, when all subjects were combined, there were significant increases in all strength measurements from pre- to post-test ($p < 0.05$) as a result of the weight training regimen. This demonstrates that the exercise program was effective.

DISCUSSION

From the results of the present study, we conclude that supplementation with 400 μg/day of chromium as chromium picolinate, combined with 12 weeks of a moderately-intense weight training and walking program, did not affect body composition, RMR, glucose tolerance and related hormones, serum lipid concentrations, iron status and zinc status of sedentary, moderately obese women; any changes observed were probably due to the exercise program. However, as expected, plasma and urinary chromium both increased significantly as a result of CP supplementation.

Body Composition

Although subjects in the chromium picolinate supplemented group experienced a greater loss in percent body fat than the placebo group, it was not statistically significant. However, when subjects were combined into one group, differences in

Table 9. Average Dietary Intake from Three-Day Dietary Records at Pre-, Mid- and Post-Test for the Chromium and Placebo Groups

	Chromium Group	Placebo Group
Energy (kilocalories)		
Pre-test[1]	2159 ± 585	1916 ± 434
Mid-test	2202 ± 604	2208 ± 711
Post-test	2031 ± 617	1930 ± 418
Carbohydrate (g)		
Pre-test	271 ± 72	260 ± 60
Mid-test	281 ± 92	278 ± 122
Post-test	260 ± 91	259 ± 71
Fat (g)		
Pre-test	83 ± 34	67 ± 26
Mid-test	85 ± 28	83 ± 32
Post-test	79 ± 26	70 ± 21
Protein (g)		
Pre-test	89 ± 27[a]	74 ± 17
Mid-test	83 ± 22	92 ± 27[a]
Post-test	74 ± 21[b]	74 ± 14[b]
Total Iron (mg)		
Pre-test	15.2 ± 4.6	14.6 ± 4.5
Mid-test	15.7 ± 6.4	16.7 ± 7.7
Post-test	14.3 ± 8.1	16.5 ± 7.0
Absorbable Iron (mg)		
Pre-test	1.2 ± 0.4	1.0 ± 0.3
Mid-test	1.3 ± 0.5	1.3 ± 0.6
Post-test	1.1 ± 0.4	1.2 ± 0.8
Animal Iron (mg)		
Pre-test	4.1 ± 1.9	3.2 ± 1.6
Mid-test	3.9 ± 1.6	4.4 ± 1.9
Post-test	3.5 ± 1.2	3.8 ± 5.4
Calcium (mg)		
Pre-test	926 ± 386	902 ± 244
Mid-test	901 ± 305	844 ± 450
Post-test	760 ± 413	828 ± 296
Zinc (mg)		
Pre-test	10.7 ± 4.0	8.8 ± 2.8
Mid-test	9.7 ± 3.7	11.9 ± 7.1
Post-test	9.3 ± 4.6	8.9 ± 3.2
Vitamin C (mg)		
Pre-test	139 ± 97	132 ± 73
Mid-test	146 ± 71	118 ± 96
Post-test	109 ± 66	108 ± 49
Dietary fiber (mg)		
Pre-test	14.3 ± 10.5	12.5 ± 4.3
Mid-test	14.3 ± 7.0	12.8 ± 10.1
Post-test	10.9 ± 7.3	11.7 ± 8.3

Values represent means ± SD.

[1] Pre-test chromium group (n = 22), mid-test chromium group (n = 18), post-test chromium group (n = 18), pre-test placebo group (n = 21), mid-test placebo group (n = 19), post-test placebo group (n = 17).

Note: 2 subjects in the chromium group did not complete their dietary records at mid-test or post-test, hence, n = 18.

[a,b] Values in the same column with different superscript letters are significantly different from one another ($p < 0.05$).

Note: Due to rounding of values, when calculating total kilocalories from carbohydrates, fat and protein, they do not exactly equal the total kilocalories listed in the table.

percent body fat, LBM and fat mass were significantly different from pre-test values ($p < 0.05$), which were a result of the exercise program and not the CP supplementation. This observation is consistent with others [17–19,34]. Trent and Thieding-Cancel [34] reported no beneficial effects of supplementation with 400 μg/day of chromium as CP combined with aerobic exercise training on body composition in obese Navy personnel. A number of researchers [17–19,34] do not support chromium supplementation to positively alter body composition. Although several studies have demonstrated positive body composition alterations due to chromium supplementation [8,9], their study designs have been criticized.

Body Weight

In our study, body weight did not significantly change in response to either chromium supplementation or exercise training. Other researchers have attempted to investigate the effects of chromium supplementation on weight loss [10,11]. Grant *et al.* [11] reported that 400 μg/day of chromium picolinate without exercise training was contraindicated for weight loss in young, obese women because it caused significant weight gain in these subjects. However, supplementation of 400 μg/day of chromium nicotinate combined with exercise training resulted in more beneficial effects than exercise alone in weight loss and insulin response to an oral glucose load [11].

Resting Metabolic Rate

There were no significant differences in RMR between the chromium and placebo groups in our study; however, there was a trend for RMR to increase over time in both groups combined. Because the subjects were previously sedentary, 12 weeks of exercise may have resulted in an increase in RMR. Perhaps 12 weeks of intervention was not long enough to elicit a significant increase in RMR. In a study by Kaats *et al.* [10], 30 obese subjects given a combination of chromium picolinate, L-carnitine, ascorbic acid, pyridoxine, niacin, potassium, magnesium and L-lysine had an average increase in RMR of 21%. Chromium picolinate may have contributed to the increase in RMR; however, the results were confounded by the other nutrients provided. We did not demonstrate an effect of chromium picolinate supplementation on RMR.

Glucose Tolerance

Several studies have shown that some subjects with hypoglycemia, hyperglycemia and non-insulin-dependent diabetes mellitus (NIDDM) may respond to supplemental chromium [2–5,35]. However, individuals with normal glucose tolerance, adequate dietary chromium and good chromium status do not respond to supplemental chromium [4,35–37]. Anderson *et al.* [4] showed that glucose tolerance and circulating insulin and glucagon concentrations of subjects with marginally elevated

Table 10. Averages of One-Repetition Maximum Values (in pounds) at Pre- and Post-Test for Specific Weight Training Apparatus Used During the 12-Week Study for the Chromium and Placebo Groups

	Chromium Group		Placebo Group	
	Pre-Test (n = 22)	Post-Test (n = 20)	Pre-Test (n = 22)	Post-Test (n = 17)
Chest Press	65.5 ± 15.7	80.0 ± 13.8*	70.9 ± 15.7	83.5 ± 22.9*
Lattisimus Dorsi Pull	55.5 ± 11.0	68.9 ± 13.2*	59.1 ± 12.3	75.0 ± 17.0*
Triceps Press	98.2 ± 20.3	115.0 ± 20.6*	101.4 ± 17.5	114.7 ± 15.5*
Biceps Curls	30.9 ± 11.1	50.5 ± 11.5*	31.9 ± 10.3	53.1 ± 14.5*
Leg Extension	104.6 ± 23.9	125.5 ± 26.7*	116.4 ± 23.4	140.6 ± 31.1*
Leg Curls	42.7 ± 13.5	64.0 ± 19.3*	47.3 ± 11.2	71.3 ± 22.5*
Calf Raises	128.6 ± 34.4	198.8 ± 48.3*	154.6 ± 47.9	201.3 ± 32.9*
Leg Abduction	88.6 ± 28.3	123.0 ± 23.6*	97.3 ± 23.3	128.8 ± 29.3*
Leg Adduction	79.1 ± 17.2	97.5 ± 21.0	90.5 ± 27.5	104.7 ± 30.8

Values represent means ± SD.

* Significantly different from pre-test when both groups were combined ($p < 0.05$).

blood glucose concentrations and consuming low-chromium-controlled diets were improved after they were supplemented with 200 μg/day of chromium as chromium chloride for five weeks. However, the control group with normal glucose concentration did not show any signs of chromium deficiency after the low-chromium diets and no changes after supplementation. The results presented in our study agree with these aforementioned investigations and are not surprising, because none of the moderately obese women in our study had ever been diagnosed with impaired glucose tolerance.

Serum Lipid Concentrations

We reported no significant differences in serum lipid levels as a result of the chromium picolinate supplementation. The decrease of TC concentration for all subjects may have been a result of the 12-week exercise program. Although an increase in serum HDL-C concentration would be expected as a result of the exercise training, it decreased; perhaps this was because body weight did not significantly decrease. Also, a 12-week exercise period is typically not long enough to elicit an increase in HDL-C levels. The lack of an effect of chromium supplementation on serum lipid concentrations has been shown in previous studies [2]. However, others have shown positive effects of chromium supplementation with respect to serum lipid concentrations [5–7]. The varying results of chromium's impact on blood lipid concentrations may relate to the basal level of the serum lipid concentrations and subjects' initial chromium status as well as glucose metabolism. In addition, the exercise regimen may have played a significant role on the beneficial changes in several experiments, including our study.

Iron Status

Although trivalent chromium has been determined as a potential inhibitor of iron metabolism, competing with iron by binding to transferrin (siderophilin) in rats and *in vitro* [20,21,38], the mechanism has not been determined in humans *in vivo*. In our study, iron indices were not affected by CP supplementation. Furthermore, dietary iron intakes were not significantly different between groups over time. Lukaski *et al.* [19] observed a weak tendency ($p = 0.17$) for transferrin saturation to decrease more in their CP supplemented group than in groups supplemented with chromium chloride or placebo. Furthermore, they reported a significantly lower urinary iron concentration in both chromium supplemented groups compared to the placebo group. TIBC was significantly decreased by 3% with exercise training in our study; this is in contrast to Lukaski *et al.* [19], who reported a 9% increase in TIBC, and similar to that reported by Campbell *et al.* [39], who found that hematological and iron indices were not affected by chromium supplementation in male subjects, 56 to 69 years of age, who consumed either 17.8 μmol/day of chromium picolinate or a placebo and participated in a high-intensity resistive training program for 12 weeks. Thus, depending on the dose and duration of chromium supplementation, iron metabolism may be adversely affected as a result of the body's maintaining a normal iron status by preventing urinary iron loss [19].

It is possible that the mechanical stress on red blood cells and increased rate of intravascular hemolysis induced by the activation of large muscle mass by strength training alone may lead to an increased iron requirement [40]. The decrement in serum ferritin concentration in our study was not as significant as the finding of others [40]. The difference may have been due to different training programs and gender differences.

Zinc Status

Our findings on zinc status are consistent with Lukaski *et al.* [19]. We found no significant differences between groups over time. The significant variations we found between the fasted state and the post-prandial states in both groups are similar to those reported by Lowe *et al.* [41], who found a 13% and 18% decline in plasma zinc concentration following two different types of meals. The post-prandial decline in plasma zinc levels

could be due to zinc uptake by the pancreas to facilitate the synthesis of digestive enzymes and by the liver where it is required for phosphorylation reactions [41].

Blood Chromium

Twelve weeks of CP supplementation had a significant effect on plasma chromium concentration, with an approximately tenfold increase in fasting levels in the chromium group. Our findings were similar to a three-month chromium chloride supplementation study (200 μg/day) that resulted in an approximately threefold increase in serum chromium concentration [37]. Nonetheless, the change in serum chromium concentration following chromium supplementation is suggested to be a reflection of higher chromium intake, but not a meaningful indicator of tissue chromium concentrations [14].

There are no acceptable indicators to assess chromium status because chromium concentrations in biological tissues and fluids do not reflect metabolically active chromium pools in the body [14]. In addition, there are limited reports of actual chromium concentrations in serum/plasma, whole blood, erythrocytes and urine due to methodological difficulties [29]. The best method to diagnose marginal chromium deficiency is based on an improvement in glucose tolerance after chromium supplementation [29]. As there were no changes in glucose concentration in the fasted state, and one and two hours after a high carbohydrate meal from pre- to post-test, we can assume these female subjects not only had normal glucose tolerance, but normal chromium status as well. We found no changes in serum chromium concentration 90 minutes after a glucose load compared to the fasted state in either the placebo or chromium supplementated periods. These findings are consistent with Anderson *et al.* [37].

RBC chromium was unchanged due to chromium picolinate supplementation; however, we expected it to parallel the plasma chromium level changes. As there are no published data about the changes of RBC chromium concentration after chromium supplementation, we are unable to compare our results to others. We had hoped that RBC chromium would be a better indicator than plasma chromium as an indicator of chromium supplementation, but this was not the case.

Urinary Chromium

Urinary chromium excretion decreased at post-test after a transient increase in urinary chromium excretion at mid-test in the chromium group. These changes were significant, while in the placebo group there was only a trend. The increase in chromium excretion was consistent with previous studies [17,42]. Although urinary chromium excretion is considered an indicator of excessive dietary intake [29], exercise has been suggested to induce urinary chromium excretion. Rubin *et al.* [42] demonstrated that both acute and 16 weeks of chronic resistive exercise training increased urinary chromium excretion in ten men. However, Anderson *et al.* [43] reported that trained subjects had significantly lower basal urinary chromium concentrations compared with untrained sedentary control subjects. The decline in urinary chromium excretion may be an adaptive mechanism caused by strenuous exercise [43]. People who engage in strenuous exercise tend to conserve more chromium, possibly through increased tissue storage and decreased urinary loss. Therefore, further study is required to investigate the long-term effects of resistive training and aerobic exercise on urinary chromium loss.

Dietary Intake

Dietary intake was not a confounding factor in this study, because the dietary intakes were similar in both groups according to their three-day dietary records. Compared with the DRI for chromium of 25 μg/day [22], our subjects consumed from 21 to 44 μg of chromium/day and, thus, were within or above the recommended intakes.

CONCLUSIONS

Twelve weeks of supplementation with 400 μg/day of chromium as chromium picolinate did not significantly impact body composition, RMR, plasma glucose, serum insulin, plasma glucagon, serum C-peptide and serum lipid concentrations or iron and zinc indices in moderately obese women placed on an exercise program. The changes in serum total cholesterol and TIBC were a result of the exercise program.

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