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Chromium Methionine Supplementation Decreases Obesity Indices in Rats

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Abstract

This study was conducted to determine the effects of chromium-methionine (CrMet) supplementation at various levels on obesity index, body fat, and serum glucose, insulin and leptin in rats. Forty male Sprague-Dawley rats were randomly assigned to one of four dietary groups and fed AIN-76 semi-purified basal diets supplemented with 0, 300, 600 or 1200 ppb Cr from CrMet. After 4 weeks on the respective diets, the rats were killed and serum glucose, insulin and leptin concentrations were determined. The CrMet supplementation did not affect weight gain, feed intake or feed efficiency ratio, fasting glucose, insulin or leptin levels among treatment groups. Although final body weight in all treatments were not significantly different, naso-anal length was longer in the 1200 ppb CrMet group than those of control or other groups ($p < 0.05$). The lowest obesity index and body fat were observed in the 1200 ppb dietary group ($p < 0.05$). The obesity index of the rats fed 1200 ppb supplemental CrMet was lower than in the other groups. These results suggest that CrMet supplementation results in a significant decrease in obesity index, possibly by decreasing the body fat that corresponded to increasing CrMet dosage.

Key words: chromium methionine, body fat, glucose, insulin, leptin

INTRODUCTION

Chromium (Cr) may improve impaired glucose tolerance, decrease elevated blood lipid concentrations and result in weight loss and improved body composition. However, inorganic forms of Cr are relatively unavailable for absorption. However, organic forms of Cr are usually more bio-available (1).

Several studies regarding the biological effects of organic Cr supplementation on body composition have been reported. Cr supplementation has been found to reduce body fat levels in humans and animals (2-5). McCarty (6) showed that Cr picolinate reduced body fat in rats and Evans (2) reported that Cr picolinate (200 μg/day) increased lean body mass and body fat loss in men engaged in a weight training program. In swine, Page et al. (4) found that Cr picolinate decreases body fat. Lindemann et al. (7) found linear increases in lean body mass with corresponding linear decreases in body fat in pigs supplemented with Cr picolinate. However, two human studies that used Cr as Cr nicotinate failed to observe any changes in lean body mass (8,9).

It appears that Cr supplementation is capable of altering body composition, but the ability of different forms of Cr to do this, and the dosages required, may vary; different levels of supplementation have rarely been examined. Since most of the body composition studies using Cr have evaluated the picolinate form of Cr, this study contrasted the effects of increasing amounts of dietary chromium-methionine (CrMet) upon body fat and serum glucose, insulin and leptin concentrations in the rat. Therefore, our objectives were to determine the effects of different levels of dietary Cr in the form of CrMet on obesity index, body fat and serum glucose, insulin and leptin concentrations in rats.

MATERIALS AND METHODS

Animals and dietary treatments

Forty male Sprague-Dawley rats, with original body weights of 155~156 g were divided into four treatment groups of ten each. These four groups of animals were referred to as control, CM300, CM600, and CM1200. The animals had free access to food and distilled drinking water. The animals were maintained in a controlled environment at 20°C and 40~50% humidity, with 12 h of light per 24 h period.

The formula and chemical composition of the basal diets used in this experiment are presented in Table 1. All animals received the modified AIN-76 semi-purified basal diet for 4 weeks (10). The dietary treatment consisted of the basal diet supplemented with four levels of Cr as a

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Table 1. Composition of experimental diet (g/kg diet)

Ingredients	Groups[1]			
	Control	CM 300	CM 600	CM 1200
Casein	200	200	200	200
DL-methionine	3	3	3	3
Sucrose	500	500	500	500
Corn starch	150	150	150	150
Cellulose	50	50	50	150
Corn oil	50	50	50	50
Mineral mixture[2]	35	35	35	35
Vitamin mixture[3]	10	10	10	10
Choline bitartrate	2	2	2	2
Chromium methionine	0	0.3	0.6	1.2

[1]Control: AIN-76 diet.
CM 300: AIN-76 diet with 300 ppb Cr methionine.
CM 600: AIN-76 diet with 600 ppb Cr methionine.
CM 1200: AIN-76 diet with 1200 ppb Cr methionine.
[2]AIN-76 mineral mixture.
[3]AIN-76 vitamin mixture.

CrMet were used. Treatments were supplementation of 0, 300, 600, and 1200 ppb of CrMet for rats in control, CM300, CM600, and CM1200 groups, respectively.

Body weight was measured at frequent intervals in order to estimate daily live weight gain. Food bowls were weighed daily prior to, and after each feeding, to calculate the amount of food consumed.

Obesity index

Body weight and naso-anal length were measured for obesity index at the end of the study period. We calculated Röhrer index [Body weight (g) / Naso-anal length (cm)^3 $\times$ 10^3], Lee index [Body weight (g)$^{1/3}$ / Naso-anal length (cm) $\times 10^3$] (11) and TM index [Body weight (g) / Naso-anal length (cm)$^{2.823}$ $\times 10^3$] (12) according to the respective formula. Body fat content, which is responsible for obesity, was calculated by TM index [Body fat (g/100 g body weight) = 0.581 $\times$ TM index - 22.03].

Blood glucose, insulin and leptin

At week 4, after an overnight fast, blood samples were collected by decapitation. Serum was separated by centrifugation at 4°C and kept frozen at -70°C for analysis of glucose, insulin and leptin. Glucose was analyzed by an enzymatic procedure (Boeringer Manheim, Germany). Insulin was determined by a radioimmunoassay method (Diagnostic Products Corporation, Los Angeles, CA) which has been validated for the detection of rat insulin. Serum leptin concentration was determined with a commercially available rat leptin RIA kit (Linco Research Inc., St Charles, MO, USA), in which purified recombinant rat leptin samples were used as standards.

Statistical analysis

All values are presented as the mean with standard error. Statistical analysis was performed by the GLM procedure of SAS (SAS Institute, Cary, NC USA) (13). Duncan's multiple range test was conducted to evaluate significant main effects. The differences between control and treatment groups were considered statistically significant if $p < 0.05$.

RESULTS

Body weight gain, feed intake and feed efficiency ratio

As shown in Table 2, body weight gain and feed efficiency ratio were not significantly different among groups, although there was a non-significant trend toward decreased feed efficiency ratio with increased CrMet intake. Although the feed intake of the control animals was significantly less than CrMet supplemented, there was no dose effect for CrMet on feed intake.

Body weight and naso-anal length

Changes in body weight and naso-anal lengths are shown in Table 3. After 4 weeks of feeding, the final live weight increased, reaching 325 g at the end of the study period. There were no differences among groups in final body weight, but CrMet did dose-dependently increase naso-anal length ($p < 0.05$).

Obesity index and body fat

When the obesity index was compared among the

Table 2. Effect of different levels of dietary chromium methionine on body weight gain, feed intake and feed efficiency ratio

Groups[1]	Body weight gain (g/day)	Feed intake (g/day)	Feed efficiency ratio
Control	6.20 ± 0.27[2)NS3)]	17.37 ± 0.14[b4)]	0.36 ± 0.01[NS]
CM 300	6.26 ± 0.16	18.41 ± 0.31[a]	0.34 ± 0.01
CM 600	6.44 ± 0.13	18.93 ± 0.13[a]	0.34 ± 0.01
CM 1200	6.27 ± 4.23	19.00 ± 0.15[a]	0.33 ± 0.01

[1]See the legend of Table 1.
[2]Values are mean ± SE.
[3]Not significant.
[4]Means with different letters within a column are significantly different from each other at α =0.05 as determined by Duncan's multiple range test.

Table 3. Effect of different levels of dietary chromium methionine on final body weight, and naso-anal length

Groups[1]	Final body weight (g)	Naso-anal length (cm)
Control	318.69 ± 6.08[2)NS3)]	21.50 ± 0.19[bc4)]
CM 300	327.09 ± 5.31	21.39 ± 0.11[c]
CM 600	330.18 ± 4.18	21.94 ± 0.21[ab]
CM 1200	325.81 ± 4.23	22.22 ± 0.19[a]

[1]See the legend of Table 1.
[2]Values are mean ± SE.
[3]Not significant.
[4]Means with different letters within a column are significantly different from each other at α =0.05 as determined by Duncan's multiple range test.

dietary groups, significant differences were revealed. As shown in Table 4, CrMet dose-dependently decreased obesity indices, however the decreases were only significant for the 1200 ppb supplemented group, in which all of the obesity indices were lower than the control group. This effect was consistent with reduction in the body fat content. Body fat was also significantly decreased by 1200 ppb CrMet supplementation compared to control and CM300 group ($p<0.05$). The lowest body fat content was found in CM1200 group.

Blood glucose, insulin and leptin

The concentrations of glucose, insulin and leptin in serum of rats fed CrMet are presented in Table 5. There were no significant differences among control and treatment groups. The fasting serum glucose was the lowest in rats receiving 600 ppb CrMet in the diet, although this did not appear to be significant. Serum insulin concentration was lowest in the 300 ppb CrMet group, but again the difference was not significant. Serum leptin levels were not significantly affected by supplemental CrMet levels.

DISCUSSION

We investigated the effects of CrMet supplementation on obesity index, body fat and serum glucose, insulin and leptin concentrations. CrMet supplementation resulted in a significant decrease in obesity index and body fat for rats that corresponded to increasing CrMet dosage. This is in agreement with some previous studies that have observed decreased in body fat with Cr picolinate supplementation. Cr supplementation may decrease body fat levels by increasing energy expenditure or by decreasing caloric intake. Page et al. (4) found that the decrease in body fat was accompanied by a decrease in feed intake at the higher dosages of 400 and 800 ppb Cr, but not at the lower dosages of 100 and 200 ppb Cr. In this study, however, body fat reductions occurred without a concurrent decrease in feed intake. Furthermore, body fat changes took place without any apparent differences in body weight between treatment groups, suggesting increases in lean body mass. Since these body fat reductions occurred without concurrent decreases in feed intake, it is possible that CrMet supplementation exerted a metabolic effect. More research must be done to determine the impact of supplemental CrMet source on metabolism.

The current study indicates that fasting serum glucose, insulin and leptin are not significantly affected by CrMet supplementation. This is in agreement with previous research that indicated that CrCl, Cr nicotinate, and Cr picolinate supplement did not provoke changes in blood glucose concentrations (4,14,15). In contrast, other papers have demonstrated that organic Cr decreased blood glucose values (16-18). Anderson et al. (17) reported that Cr pi-

Table 4. Effect of different levels of dietary chromium methionine on Röhrer index, Lee index, TM index and body fat content of rats

Groups[1)]	Röhrer index[2)]	Lee index[2)]	TM index[2)]	Body fat[2)] (g/100 g body weight)
Control	$33.22\pm0.65^{3)a4)}$	321.36 ± 2.10^{a}	57.17 ± 1.07^{a}	11.19 ± 0.62^{a}
CM 300	33.98 ± 0.60^{a}	323.80 ± 1.91^{a}	58.42 ± 0.99^{a}	11.91 ± 1.73^{a}
CM 600	31.85 ± 0.80^{ab}	316.81 ± 2.65^{ab}	55.00 ± 1.29^{ab}	9.92 ± 0.75^{ab}
CM 1200	30.40 ± 0.76^{b}	311.91 ± 2.59^{b}	52.61 ± 1.24^{b}	8.54 ± 0.72^{b}

[1)]See the legend of Table 1.
[2)]Röhrer index = {Body weight (g) / Naso-anal length $(cm)^3$} $\times 10^3$.
Lee index = {Body weight $(g)^{1/3}$ / Naso-anal length (cm)} $\times 10^3$.
TM index = {Body weight (g) / Naso-anal length $(cm)^{2.823}$} $\times 10^3$.
Body fat = 0.581 $\times$ TM index - 22.03.
[3)]Values are mean $\pm$ SE.
[4)]Means with different letters within a column are significantly different from each other at $\alpha = 0.05$ as determined by Duncan's multiple range test.

Table 5. Effect of different levels of dietary chromium methionine on serum glucose, insulin and leptin levels of rats

Groups[1)]	Glucose (mg/dL)	Insulin (μIU/mL)	Leptin (ng/mL)
Control	$102.67\pm6.82^{2)NS3)}$	10.14 ± 3.59^{NS}	2.47 ± 0.83^{NS}
CM 300	107.11 ± 5.69	8.88 ± 1.47	2.35 ± 0.48
CM 600	98.89 ± 4.31	15.48 ± 2.79	2.76 ± 0.34
CM 1200	104.67 ± 3.46	15.63 ± 3.40	2.69 ± 0.51

[1)]See the legend of Table 1.
[2)]Values are mean $\pm$ SE.
[3)]Not significant.

colinate supplementation decreased serum glucose concentrations. Min et al. (19) reported that 200 ppb Cr picolinate tended to increase serum glucose concentrations. Our data indicate that serum insulin concentrations were not affected in rats fed CrMet, which is consistent with other reports indicating that plasma or serum insulin concentrations are not affected by Cr picolinate, CrCl, or Cr nicotinate (15,20). Serum insulin concentrations have been reported to be decreased in pigs fed Cr picolinate (18).

In conclusion, CrMet supplementation may have in resulted in reduced obesity index due to decreased body fat. The results suggest that CrMet must be supplemented at relatively high levels for it to have positive effects on body fat.

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