



# University of Connecticut

## CLINICAL STUDY

INFLUENCES OF A DIETARY SUPPLEMENT IN COMBINATION WITH AN EXERCISE AND DIET REGIMEN, ON ADIPOCYTOKINES AND ADIPOSITY IN WOMEN WHO ARE OVERWEIGHT

Running Head: dietary supplement and weight loss

William J. Kraemer, Ph.D.  
Jeff S. Volek, Ph.D., RD  
Maren S. Fragala, MS

Human Performance Laboratory  
Department of Kinesiology  
University of Connecticut  
Storrs, CT 06269  
Phone: 860-486-6892 (6898 fax)

and

Director, Research & Development,  
Max International.  
Salt Lake City, Utah 84047

### ABSTRACT (213/300 words)

**Objective:** To examine the influence of a proprietary blend of modified cellulose and cetylated fatty acids (Max International Salt Lake City, Utah 84047) on adipocytokine and regional body composition responses to a weight loss program.

**Methods:** Twenty-two women (Supplement group (S) (n=11): age= 36.8±7.2 y; weight=87.4 ±6.1 kg; % body fat= 43.6±7.1); Placebo group (P) (n=11): age = 38.3±6.8 y; weight = 88.3±7.2 kg; % body fat=44.2±8.2) completed an 8-week placebo-controlled, double-blind study consisting of a caloric restricted diet and cardiovascular exercise. Body composition and serum insulin, leptin, and adiponectin were assessed at pre-, mid, and post- intervention.

**Results:** From pre- to post- intervention, significant decreases ( $p<0.05$ ) were observed for body weight (S: 87.4±6.1 to 78.2±3.4 kg; P: 88.3±7.2 to 82.9±4.1 kg) ( $p<0.05$  S vs P), % body fat (S: 43.6±7.1 to 36.2±4.4; P: 44.3±8.2 to 40.9±4.3) ( $p<0.05$  S vs P), leptin (S: 28.2±6.1 to 16.3±3.4 ng·ml<sup>-1</sup>; 29.3±9.1 to 19.7±3.3 ng·ml<sup>-1</sup>) ( $p<0.05$  S vs P), and insulin (S: 7.2±2.2 to 5.4±2.4 mU·L<sup>-1</sup>; P: 7.7±2.2 to 5.2±2.7 mU·L<sup>-1</sup>). Serum adiponectin increased ( $p<0.05$ ) (S: 12.2±3.1 to 26.2±3.4 µg·ml<sup>-1</sup>; 12.6±4.2 to 21.7±4.3 µg·ml<sup>-1</sup>) ( $p<0.05$  for S vs P).

**Conclusion:** Data indicated that supplementation with a proprietary blend of modified cellulose and cetylated fatty acids during an 8-week weight loss program exhibited favorable effects on adipocytokines and regional body composition.

**KEY WORDS:** Dietary supplement, weight loss, adipocytokines, diet, exercise

## INTRODUCTION (3622/5000 words)

Unsuccessful attempts to modulate human obesity have led to many research attempts to understand the neurological and hormonal pathways regulating energy intake and energy expenditure. Evidence suggests a relationship between these hypothalamic pathways and hormonal signals, from leptin and adiponectin of the adipocytes, ghrelin and polypeptides from gastrointestinal tract, and insulin from pancreas. Both ghrelin and leptin appear to modulate appetite regulatory pathways involving insulin through the neuropeptide Y pathway in the hypothalamus,<sup>15,17</sup> adiponectin appears to influence insulin sensitivity,<sup>27</sup> and insulin appears to act synergistically with leptin in the hypothalamus.<sup>28</sup> Adipocytokines appear to play an important role in the connection between the brain, adipose tissue and other peripheral organs involved in the weight regulatory pathway. Adipocytokines are signaling molecules secreted by adipose.<sup>19</sup> Adiponectin, is an adipocytokine that has been shown to have protective effects on diabetes and cardiovascular disease.<sup>19</sup> Obese individuals exhibit lower adipose tissue, adiponectin expression and hormone-sensitive lipase activity, the enzyme involved in fatty acid oxidation.<sup>5</sup> This is believed to contribute to the progression of obesity and its co-morbidities regulating hormone-sensitive lipase activity and fatty acid oxidation.<sup>5</sup> Additionally, circulating adiponectin concentrations typically increase with weight loss.<sup>33,42</sup> Furthermore, adiponectin concentrations are positively associated with insulin sensitivity<sup>14</sup> and decreased insulin resistance and blood glucose concentrations.<sup>27</sup> Moreover, weight loss has been associated with improved insulin sensitivity.<sup>41</sup>

Leptin, another adipocytokine, serves as a regulator of body fat storage through the central nervous system, by modulating satiation, appetite, glycemic control and metabolism.<sup>2</sup> Leptin is a mediator of long-term regulation of energy balance, suppressing food intake and thereby inducing weight loss.<sup>18</sup> Leptin inhibits orexigenic effects of the hormone ghrelin, which plays a role in the regulation of feeding, by centrally countering its appetite promoting effects in the hypothalamus and peripherally by attenuating gastric ghrelin secretion.<sup>37</sup> Obese individuals exhibit elevated circulating levels of leptin and appear to be leptin-resistant.<sup>18</sup> In rodents, exogenous leptin administration has demonstrated potent effects on bodyweight and adiposity.<sup>2</sup> However, a trial of leptin injection in humans was found to be unsuccessful in causing weight loss.<sup>2</sup> These findings have been attributed to possible 'leptin resistance', or failure of leptin to have the primary role in control of adipose tissue mass in humans.<sup>9</sup> In addition, some evidence suggests that leptin may restrain adipocyte adiponectin secretion.<sup>37</sup> Leptin is strongly correlated with obesity and weight loss.<sup>29</sup> Diet<sup>42</sup> and combined diet and exercise strategies<sup>38</sup> have been shown effective at decreasing body fat and plasma leptin concentrations. However, changes in leptin concentrations appear to be due to changes in body weight and body composition rather than exercise. Research demonstrates that circulating leptin concentrations did not change during nine weeks of aerobic training where aerobic fitness was improved, but body mass or body composition did not.<sup>21</sup>

While the understanding of these pathways is progressing, efforts to develop successful pharmacological or supplemental strategies to influence these pathways has been disappointing. Drug development strategies have focused on techniques to reduce intestinal fat digestion and absorption or to act centrally on blocking norepinephrine/serotonin reuptake to suppress appetite,<sup>9</sup> while lifestyle intervention programs have focused on dietary and exercise plans. Weight loss programs combining these strategies including behavior modification, medication, and meal replacements appear to be more effective than any of these strategies administered alone.<sup>40</sup> The exercise or physical activity component of weight loss strategies in combination with diet is effective in weight loss vs. diet alone.<sup>1</sup> Additionally, exercise training combined with dieting improves aerobic capacity.<sup>20</sup> The primary purpose of this investigation was to determine the effects of a proprietary blend of modified cellulose and cetylated fatty acids on a weight loss program for women.

## METHODS

### Experimental approach

Women who were overweight (BMI > 25) but otherwise healthy were recruited to participate in a supplemented 8-week, double-blinded, placebo controlled weight loss program that included dietary guidance and exercise. Participants were matched (body mass, percent fat, activity background, endurance capacity) and then each woman was randomly placed into either a group that consumed the proprietary blend of modified cellulose and cetylated fatty acids (Leptivin™) supplement (Max International, Salt Lake City, Utah 84047) or in a control group that consumed visually identical placebo capsules. Prior to participation all participants were informed of the study procedures and risks and were required to complete an informed consent document approved by the Review Board for the Use of Human Subjects. In addition, each subject was medically cleared by a physician to participate in the study.

### Participants

Twenty-two healthy women who were overweight (Supplement Group: (N= 11) Age 36.8±7.2 y; Height 163.8±9.1cm; BMI 32.12±1.34 kg/m<sup>2</sup>; Placebo group (N= 11) 38.3±6.8 y; Height 162.7±9.3 cm; BMI 32.36±1.36 kg/m<sup>2</sup>) participated in the study. No participants demonstrated any endocrine, metabolic, orthopedic, or other pathological disorders, except for being overweight. Participants were weight-stable for at least 3 months before enrollment, were not pregnant or trying to become pregnant, did not use tobacco products, nor consume more than two alcoholic beverages per day.

### Supplement

Participants consumed either three supplement (supplement group) or placebo capsules at the two largest meals each day (6 capsules total per day). Each supplement capsule contained 400 mg of Leptivin™ (a proprietary blend of modified cellulose and cetylated fatty acids). The modified cellulose is a food grade ingredient that has GRAS status. Cetylated fatty acids have no known side effects or adverse reactions. The placebo contained 400 mg of magnesium stearate per capsule. Participants logged their supplement consumption each day and returned the bottles when emptied and after the study. Additionally, participants were required to complete daily symptoms and side effect questionnaires which asked participants to cite any symptoms they experienced each day during the duration of the study whether or not they believed they were or were not associated with supplement use.

### Weight-loss program Dietetic Counseling

Using the methods described by Volek, et al.<sup>38</sup> all participants consumed a moderately caloric restricted diet of self-selected commercially available foods in a free-living environment to allow dietary modifications that could be continued with ease by participants after the duration of the study. All participants were required to attend mandatory weekly nutritional counseling meetings led by a registered dietitian. Nutritional counseling meetings focussed on techniques for behavior modification and implementation of a healthy, well-balanced restricted diet, using concepts of variety, balance and modification. Topics included measuring portion sizes, choices to make when eating out, strategies for food shopping, modifications and substitutions of recipes, self monitoring, and recent scientific research.

### Exercise Training

All participants underwent an 8-week supervised exercise program consisting of cardiovascular exercise including walking, jogging, cycling, high-lo aerobics, kick-boxing and cycling, four to five times per week in our exercise facilities. Trainers recorded the mode of exercise, duration and heart rates for all exercise sessions. Exercise duration ranged from 30-60 minutes at an intensity of 60-90% of age-predicted maximal heart rate according to procedures recommended by the American College of Sports Medicine. Each session was supervised by an exercise specialist to maintain the quality of

the workout and optimize the exercise prescription. Both the duration and the intensity progressed throughout the 8-weeks in accordance with the American College of Sports Medicine (1995) guidelines for exercise prescription.

### Diet Logs

Participants kept seven-day food and beverage diaries for the week prior to the weight loss intervention, and during weeks 1, 4, and 8. A registered dietician reviewed logs with participants to qualify completeness. Total food energy and nutrient content was analyzed with Nutritionist Pro Software (version 2.5.1). Participants also kept seven-day food analog scales during the same time points as the dietary logs week prior to the intervention and the first, fourth, and seventh weeks of the intervention. Three representative days were selected by the same registered dietitian and analyzed for total food energy and nutrient content (Nutritionist V, Version 2.1, N-Squared Computing, First Data-bank Division, The Hearst Corporation, San Bruno, CA, USA).

### Experimental Variables

Prior to the intervention, during each week, and at the completion of the intervention, body mass was measured to the nearest 0.1 kg using a calibrated clinical scale. Circumference measurements using of the abdomen, hips, and thighs on the right side of the body were also measured prior to the intervention, mid-intervention, and at the completion of the 8-week intervention using a standard spring-loaded measuring tape. Body composition was obtained using dual-energy X-ray absorptiometry (DEXA) using a total body scanner (Prodigy™, Lunar Corporation, Madison, WI, USA) (as previously described by Volek et al.,<sup>39</sup>) prior to the intervention, mid-intervention, and at the completion of the 8-week intervention. Percentage body fat was calculated as fat tissue mass divided by the total soft tissue mass plus the estimated bone mineral content. Fat-free mass was calculated as lean soft tissue plus bone mineral content. Regional body composition of the trunk, arm, and leg regions was calculated by the computer program using anatomical landmarks as boundaries. Aerobic fitness gains were validated using a "Yo-Yo Endurance Test" based on 20-meter running intervals at progressively increasing speeds. This test has been both validated to evaluate aerobic endurance and sensitive to detect changes in fitness.<sup>22,24</sup>

### Blood Collection and Biochemical Analyses

Blood samples were obtained from a forearm vein after a 12-hour overnight fast and a 24-hour abstinence from alcohol and strenuous activity prior to the intervention, mid-intervention, and at the completion of the 8-week intervention. Blood was collected into a 10-ml vacutainer tube. Whole blood was centrifuged at 1000 g for 20 min at 10°C and the resultant serum was divided into aliquots and immediately stored frozen at -80°C. Insulin was determined in duplicate using an enzyme immunoassay (APLCO Diagnostics, Salem, NH). Serum leptin was determined in duplicate using an enzyme-linked immunosorbent assay (ELISA) (Diagnostic Systems Laboratory, Webster, TX, USA). All samples were run in the same assay with an intra-assay variance of 3.2%. Serum adiponectin was determined in duplicate using an enzyme immunoassay (APLCO Diagnostics, Salem, NH). All samples for each hormone were determined in the same assay to avoid inter-assay variance and were thawed only once for each assay procedure. Assay intra-assay variance was < 5%.

### Statistical Analyses

Dependent variables were analyzed using a two-way analysis of variance or co-variance when appropriate with repeated measures with group (supplement vs. placebo) and time (0, 4, 8 wk) as main effects. All data sets were analyzed for statistical assumptions for linear statistics and if not met were appropriately transformed and then reanalyzed again prior to statistical treatment. All data sets satisfied statistical requirements for the linear approaches used. When a significant F-value was achieved, a Fisher's LSD test was used to locate the pair-wise differences between means. Pearson correla-



tion coefficients were used to determine associations of leptin, using the nQuery Advisor® software (Statistical Solutions, Saugus, MA). The statistical power for the n size used ranged from 0.80 to 0.87. The power is based on a variety of probability equations by Cohen (1988)<sup>7</sup> which represents the needed number of subjects to defend the 0.05 level of significance four fold and allow detection of a 5 to 10% treatment effect. The test retest reliability of the tests used in our laboratory showed an intra-class Rs > 0.95. All data are presented as means and standard deviations. The level of significance was set at P<0.05.

## RESULTS

Mean ± SD values for anthropometric and body composition data variables measured at baseline (week 0), midway (week 4), and after weight loss (week 8) for the supplement and placebo groups are shown in Table 1. Body weight decreased from 87.4±6.1 to 78.2±3.4 kg (p<0.05) for the supplement group and 88.3±7.2 to 82.9±4.1 kg (p<0.05) in the placebo group over the eight weeks. Additionally, over the eight weeks, the supplement group lost significantly more weight as compared to the placebo group (p<0.05 between groups) (Figure 1). Percent body fat decreased from 43.6±7.1 to 36.2±4.4 (p<0.05) for the supplement group and 44.2±8.2 to 40.9±4.3 (p<0.05) in the placebo group over the eight weeks. The supplement group lost significantly more body fat as compared to the placebo group over the eight weeks (p<0.05 between groups) (Figure 2). Body mass index (BMI) also decreased from 32.12±1.34 to 28.98±1.24 kg·m<sup>2</sup> (p<0.05) for the supplement group and 32.36±1.36 to 30.92±1.29 kg·m<sup>2</sup> (p<0.05) in the placebo group over the eight weeks. A main effect and an interaction effect for time were observed. Post Hoc testing revealed significance (p<0.05) for lower BMI values at week 8 compared to week 0 in both the supplement and placebo groups. However, no significant difference in BMI changes were observed between the supplement and the placebo groups.

Waist circumference decreased from 101.9±2.3 to 92.0±1.9 cm (p<0.05) for the supplement group and 102.2±3.5 to 96.1±2.9 cm (p<0.05) in the placebo group over the eight weeks (p<0.05 between groups) (Figure 3). Hip circumference decreased from 117.4±6.9 to 109.2±4.7 cm (p<0.05) for the supplement group and 118.2±5.1 to 110.1±4.2 cm (p<0.05) in the placebo group over the eight weeks. Thigh circumference decreased from 71.1±5.6 to 68.1±2.4 cm (p<0.05) for the supplement group and 72.2±4.6 to 69.1±3.2 cm (p<0.05) in the placebo group over the eight weeks.

Mean ± SD values for serum adiponectin, insulin, and leptin are shown in Table 2. Fasting serum adiponectin (µg/ml) increased from 12.2±3.1 to 26.2±3.4 (p<0.05) for the supplement group and 12.6±4.2 to 21.7±4.3 (p<0.05) in the placebo group over the eight weeks (p<0.05 between groups) (Figure 4). Fasting serum insulin (mU/L) decreased from 7.2±2.2 to 5.4±2.4 (p<0.05) for the supplement group and 7.7±2.2 to 5.2±3.4 (p<0.05) in the placebo group over the eight weeks. Fasting serum leptin (ng/ml) decreased from 28.2±6.1 to 16.3±3.4 (p<0.05) for the supplement group and 29.3±9.1 to 19.7±3.3 (p<0.05) in the placebo group over the eight weeks (Figure 5).

The exercise training resulted in significant increase in the number of stages performed in the Yo-Yo Test over the 8 weeks of training in both groups (supplement group; 2.8± to 3.5±1.6; placebo group 2.9± to 3.4±1.4 stages) demonstrating an increase in endurance fitness.

Symptom side effects questionnaires showed very limited side effects for both the placebo and supplement groups. No differences between frequency and type of side effects were observed between the placebo and supplement groups. There were occasional reports of symptoms such as nausea, abdominal pain, headache, ringing in ears and bloating, which may or may not be associated with supplement use. Furthermore, reported side effects were only reported on isolated days and not throughout the duration of the eight weeks.

## DISCUSSION

This study examined the effects of a dietary supplement on weight loss induced by diet and exercise on body composition, regional anthropometric changes, and serum insulin, leptin, and adiponectin. To our knowledge the present investigation is the first investigation to examine the additive role of supplementation with a proprietary blend of modified cellulose and cetylated fatty acids (Leptivin™) in a weight loss program for women. This matched double-blind, placebo controlled study demonstrated that supplementation combined with endurance exercise and a reduced-calorie diet produced significantly greater weight loss and fat loss than the diet and exercise intervention alone. A weight loss of 9.2 kg and 5.4 kg for the supplement and placebo groups, respectively, may be regarded a clinically significant weight loss in eight weeks.

Both groups lost weight over the eight weeks, with significantly more weight being lost in the group receiving the Leptivin™ compared to the placebo after eight weeks. It is possible that the differences observed in weight loss were due either to increased energy expenditure or decreased energy intake in the supplement group. Both groups followed an identical exercise program and nutritional intervention. Although both groups followed identical nutritional counseling, the supplement may have provided an appetite suppressing effect leading to a lower energy intake in the group receiving the supplement. The blend of modified cellulose component of the supplement has swellable-soluble properties that may have satiating properties to decrease caloric consumption causing a greater imbalance in the energy balance equation in individuals consuming the supplement.

Not only did the supplement group experience greater weight loss, body fat loss was also significantly greater as well. Significant improvements in body composition were observed for both groups over the eight weeks. Since both groups followed an identical exercise program, the supplement may have influenced energy substrate utilization during rest and/or exercise. BMI also significantly decreased over the 8-week program for both groups, although significant group differences were not observed. Since BMI measures do not incorporate body composition, the null finding of between group differences in BMI can likely be explained by different changes in lean and fat mass between the groups. It is also possible that the cetylated fatty acids of the supplement can explain the greater observed fat loss in the individuals taking the supplement. The cetylated fatty acid component of the supplement may have had a greater effect on lipolysis at the adipocyte level. Previous data have demonstrated that dietary fatty acids can influence overall lipid metabolism through the plasma lipid profile and body fat deposition.<sup>11</sup> In the present study it is possible that the cetylated fatty acids played a role in signaling adipocyte responses since there is evidence that fatty acids and their derivatives can function in cell signaling by acting like hormones.<sup>10</sup> Specifically, they can regulate gene expression in preadipocytes to affect adipocyte proliferation and differentiation.<sup>8</sup> As previous research has demonstrated a relationship between fat cell size and number and fatty acid composition in adipose in overweight/obese humans, the fatty acids in the supplement may play a role in the adipocyte lipolysis.<sup>11</sup>

In addition to weight loss and fat loss, experimental results indicated that leptin and insulin levels were significantly reduced, and serum adiponectin levels were significantly increased after the weight loss regimen. Furthermore, significant group differences were observed between the supplement and placebo groups for leptin and adiponectin at eight weeks. Baseline concentrations of leptin of these women who were overweight were consistent with other investigations as women generally have higher leptin concentrations than men.<sup>26,34</sup> The decreased circulating leptin concentrations during the weight loss protocol is a finding in agreement with several other investigations.<sup>12,23,25,32,34-36</sup> Since leptin is a regulator of body fat storage and mediator long-term regulation of energy of the body the decreased concentrations associated with weight loss were expected. Moreover, since leptin is produced and secreted from the adipose cells, the greater reductions in leptin concentrations in the supplement group is likely a result of the greater amounts of fat loss. Additionally, the decreased

circulating insulin concentrations during the weight loss protocol is a finding in agreement with previous research.<sup>25</sup> It is likely indicative of improved insulin sensitivity as previous research has shown both an elimination in the prevalence of insulin resistance and the metabolic syndrome in women undergoing a similar weight loss program.<sup>25</sup>

The significant increase in circulating adiponectin is a finding in agreement with Pollak et al.,<sup>30</sup> who found significant increases in high, medium, and low-molecular weight quantities of adiponectin by 5.5%, 8.5% and 18.1%, respectively, were observed ( $p < 0.05$  for all the forms) with weight loss in women. Weight loss was associated with increased total plasma adiponectin was by 36%.<sup>30</sup> Since adiponectin is involved in the regulation of glucose and fatty acid metabolism, this increase likely influences whole body insulin sensitivity. Moreover, the increased adiponectin as a result of the weight loss regimen may facilitate skeletal muscle fat oxidation.<sup>6</sup> The significantly higher adiponectin concentrations observed in the supplement group may explain the greater weight and fat loss in this group through increases in mitochondrial mass with associated increases in the fat oxidation enzymes 6 (although not measured in this investigation).

The significant reduction in circumference measurements elicited in the current study is consistent with other weight loss studies in women <sup>25,34</sup> and may be associated with improved overall health. The accumulation of fat in the intra-abdominal region is associated with a cluster of metabolic disorders including hyperinsulinemia, insulin resistance, hyperglycemia, and dyslipidemia.<sup>4</sup> In fact, abdominal obesity may be a better predictor for disease risks and all-cause mortality compared to body mass index.<sup>3,31</sup> Epidemiological data indicates that women with larger waist circumferences have significantly higher chances of having metabolic abnormalities (hypertension, diabetes, dyslipidemia, and the metabolic syndrome) compared with women with smaller waist circumferences.<sup>13,16</sup> Additionally, since waist girth decreases were significantly greater for the supplement group in the present investigation, it can be postulated that associated health improvements may also be greater. The pharmacological mechanisms underlying the anorectic effects of the proprietary blend of modified cellulose and cetylated fatty acids have not been explained. Peptides, such as leptin, insulin, and adiponectin, which act on the brain and the periphery to control appetite and feeding signals, are speculated to play a role. The greater hormonal responses in adiponectin and leptin, seen in the supplement group may be explained by the actions of the ingredients. The cetylated fatty acids and modified cellulose ingredients in the Leptivin™ supplement may influence the neural feedback loop of energy regulation and fatty acid metabolism involving these peptides, however, the precise mechanisms are currently unknown.

In summary, this study showed greater weight loss and fat loss in women receiving the Leptivin™ supplement during an 8-week weight loss program accompanied by greater effects on circulating adipocytokine concentrations. This investigation demonstrates the additive role of supplementation with a proprietary blend of modified cellulose and cetylated fatty acids in a weight loss program including diet and exercise for women. Thus, over and above diet and exercise, supplementation with Leptivin™ may facilitate the effectiveness of a weight loss program for overweight women by mechanisms that remain unclear.

## REFERENCES (maximum = 60)

1. Ballor DL, Poehlman ET: Exercise-training enhances fat-free mass preservation during diet-induced weight loss: a meta-analytical finding. *Int J Obes Relat Metab Disord* 1994;18:35-40.
2. Bell-Anderson KS, Bryson JM: Leptin as a potential treatment for obesity: progress to date. *Treat Endocrinol* 2004;3:11-18.
3. Bigaard J, Frederiksen K, Tjonneland A, Thomsen BL, Overvad K, Heitmann BL, et al.: Waist circumference and body composition in relation to all-cause mortality in middle-aged men and women. *Int J Obes (Lond)* 2005;29:778-784.
4. Bjorntorp P: Metabolic implications of body fat distribution. *Diabetes Care* 1991;14:1132-1143.
5. Bullo M, Salas-Salvado J, Garcia-Lorda P: Adiponectin expression and adipose tissue lipolytic activity in lean and obese women. *Obes Surg* 2005;15:382-386.
6. Civitarese AE, Ukropcova B, Carling S, Hulver M, DeFronzo RA, Mandarino L, et al.: Role of adiponectin in human skeletal muscle bioenergetics. *Cell Metab* 2006;4:75-87.
7. Cohen J. *Statistical Power Analysis for the Behavioral Sciences*. Hillsdale, New Jersey: Lawrence Erlbaum, 1988.
8. Duplus E, Glorian M, Forest C: Fatty acid regulation of gene transcription. *J Biol Chem* 2000;275:30749-30752.
9. Eikelis N, Esler M: The neurobiology of human obesity. *Exp Physiol* 2005;90:673-682.
10. Farnier C, Krief S, Blache M, Diot-Dupuy F, Mory G, Ferre P, et al.: Adipocyte functions are modulated by cell size change: potential involvement of an integrin/ERK signalling pathway. *Int J Obes Relat Metab Disord* 2003;27:1178-1186.
11. Garaulet M, Hernandez-Morante JJ, Lujan J, Tebar FJ, Zamora S: Relationship between fat cell size and number and fatty acid composition in adipose tissue from different fat depots in overweight/obese humans. *Int J Obes (Lond)* 2006;30:899-905.
12. Giannopoulou I, Fernhall B, Carhart R, Weinstock RS, Baynard T, Figueroa A, et al.: Effects of diet and/or exercise on the adipocytokine and inflammatory cytokine levels of postmenopausal women with type 2 diabetes. *Metabolism* 2005;54:866-875.
13. Hadaegh F, Esmailzadeh A, Azizi F: Metabolic risks in individuals with normal body mass index and normal waist circumference. *Eur J Cardiovasc Prev Rehabil* 2007;14:200-207.
14. Hara T, Fujiwara H, Nakao H, Mimura T, Yoshikawa T, Fujimoto S: Body composition is related to increase in plasma adiponectin levels rather than training in young obese men. *Eur J Appl Physiol* 2005;94:520-526.
15. Horvath TL, Diano S, Sotonyi P, Heiman M, Tschop M: Minireview: ghrelin and the regulation of energy balance--a hypothalamic perspective. *Endocrinology* 2001;142:4163-4169.
16. Janssen I, Katzmarzyk PT, Ross R: Waist circumference and not body mass index explains obesity-related health risk. *Am J Clin Nutr* 2004;79:379-384.
17. Kalra SP, Kalra PS: Neuropeptide Y: a physiological orexigen modulated by the feedback action of ghrelin and leptin. *Endocrine* 2003;22:49-56.
18. Klok MD, Jakobsdottir S, Drent ML: The role of leptin and ghrelin in the regulation of food intake and body weight in humans: a review. *Obes Rev* 2007;8:21-34.
19. Koerner A, Kratzsch J, Kiess W: Adipocytokines: leptin--the classical, resistin--the controversial, adiponectin--the promising, and more to come. *Best Pract Res Clin Endocrinol Metab* 2005;19:525-546.
20. Kraemer WJ, Volek JS, Clark KL, Gordon SE, Incledon T, Puhl SM, et al.: Physiological adaptations to a weight-loss dietary regimen and exercise programs in women. *J Appl Physiol* 1997;83:270-279.
21. Kraemer WJ, Volek JS, Clark KL, Gordon SE, Puhl SM, Koziris LP, et al.: Influence of exercise training on physiological and performance changes with weight loss in men. *Med Sci Sports Exerc* 1999;31:1320-1329.



22. Krstrup P, Mohr M, Amstrup T, Rysgaard T, Johansen J, Steensberg A, et al.: The yo-yo intermittent recovery test: physiological response, reliability, and validity. *Med Sci Sports Exerc* 2003;35:697-705.
23. Lazer S, Vermorel M, Montaurier C, Meyer M, Boirie Y: Changes in adipocyte hormones and lipid oxidation associated with weight loss and regain in severely obese adolescents. *Int J Obes (Lond)* 2005;29:1184-1191.
24. Leger LA, Lambert J: A maximal multistage 20-m shuttle run test to predict VO<sub>2</sub> max. *Eur J Appl Physiol Occup Physiol* 1982;49:1-12.
25. Lofgren IE, Herron KL, West KL, Zern TL, Brownbill RA, Ilich JZ, et al.: Weight loss favorably modifies anthropometrics and reverses the metabolic syndrome in premenopausal women. *J Am Coll Nutr* 2005;24:486-493.
26. Lonnqvist F, Nordfors L, Jansson M, Thorne A, Schalling M, Arner P: Leptin secretion from adipose tissue in women. Relationship to plasma levels and gene expression. *J Clin Invest* 1997;99:2398-2404.
27. Meier U, Gressner AM: Endocrine regulation of energy metabolism: review of pathobiochemical and clinical chemical aspects of leptin, ghrelin, adiponectin, and resistin. *Clin Chem* 2004;50:1511-1525.
28. Niswender KD, Schwartz MW: Insulin and leptin revisited: adiposity signals with overlapping physiological and intracellular signaling capabilities. *Front Neuroendocrinol* 2003;24:1-10.
29. Pellemounter MA, Cullen MJ, Baker MB, Hecht R, Winters D, Boone T, et al.: Effects of the obese gene product on body weight regulation in ob/ob mice. *Science* 1995;269:540-543.
30. Polak J, Kovacova Z, Jacek M, Klimcakova E, Kovacikova M, Vitkova M, et al.: An increase in plasma adiponectin multimeric complexes follows hypocaloric diet-induced weight loss in obese and overweight pre-menopausal women. *Clin Sci (Lond)* 2007;112:557-565.
31. Pouliot MC, Despres JP, Lemieux S, Moorjani S, Bouchard C, Tremblay A, et al.: Waist circumference and abdominal sagittal diameter: best simple anthropometric indexes of abdominal visceral adipose tissue accumulation and related cardiovascular risk in men and women. *Am J Cardiol* 1994;73:460-468.
32. Reinehr T, Kratzsch J, Kiess W, Andler W: Circulating soluble leptin receptor, leptin, and insulin resistance before and after weight loss in obese children. *Int J Obes (Lond)* 2005;29:1230-1235.
33. Shapses SA, Riedt CS: Bone, body weight, and weight reduction: what are the concerns? *J Nutr* 2006;136:1453-1456.
34. Shih LY, Liou TH, Chao JC, Kau HN, Wu YJ, Shieh MJ, et al.: Leptin, superoxide dismutase, and weight loss: initial leptin predicts weight loss. *Obesity (Silver Spring)* 2006;14:2184-2192.
35. Thompson WG, Rostad Holdman N, Janzow DJ, Slezak JM, Morris KL, Zemel MB: Effect of energy-reduced diets high in dairy products and fiber on weight loss in obese adults. *Obes Res* 2005;13:1344-1353.
36. Thong FS, Hudson R, Ross R, Janssen I, Graham TE: Plasma leptin in moderately obese men: independent effects of weight loss and aerobic exercise. *Am J Physiol Endocrinol Metab* 2000;279:E307-313.
37. Ueno N, Dube MG, Inui A, Kalra PS, Kalra SP: Leptin modulates orexigenic effects of ghrelin and attenuates adiponectin and insulin levels and selectively the dark-phase feeding as revealed by central leptin gene therapy. *Endocrinology* 2004;145:4176-4184.
38. Volek JS, Gomez AL, Love DM, Weyers AM, Hesslink R, Jr., Wise JA, et al.: Effects of an 8-week weight-loss program on cardiovascular disease risk factors and regional body composition. *Eur J Clin Nutr* 2002;56:585-592.
39. Volek JS, Sharman MJ, Love DM, Avery NG, Gomez AL, Scheett TP, et al.: Body composition and hormonal responses to a carbohydrate-restricted diet. *Metabolism* 2002;51:864-870.
40. Wadden TA, Berkowitz RI, Sarwer DB, Prus-Wisniewski R, Steinberg C: Benefits of life-style modification in the pharmacologic treatment of obesity: a randomized trial. *Arch Intern Med*

2001;161:218-227.

41. Wing RR, Koeske R, Epstein LH, Nowalk MP, Gooding W, Becker D: Long-term effects of modest weight loss in type II diabetic patients. Arch Intern Med 1987;147:1749-1753.

42. Wolfe BE, Jimerson DC, Orlova C, Mantzoros CS: Effect of dieting on plasma leptin, soluble leptin receptor, adiponectin and resistin levels in healthy volunteers. Clin Endocrinol (Oxf) 2004;61:332-338.

**Table 1.** Anthropometric responses to the weight loss intervention (Mean±SD)

		week 0		week 4		week 8	
Body Mass (kg)							
	Supplement	87.4 ± 6.1		85.3 ± 5.1 *		78.2 ± 3.4 *†‡	
	Placebo	88.3 ± 7.2		85.9 ± 5.1 *		82.9 ± 4.1 *†	
Body Fat (%)							
	Supplement	43.55 ± 7.1		40.1 ± 4.2 *		36.2 ± 4.4 *†‡	
	Placebo	44.23 ± 8.2		41.1 ± 6.1 *		40.9 ± 4.3 *	
BMI							
	Supplement	32.12 ± 1.3		30.89 ± 1.29		28.98 ± 1.2 *	
	Placebo	32.36 ± 1.4		31.88 ± 1.31		30.92 ± 1.3 *	
Circumference							
Waist	Supplement	101.9 ± 2.3		96.2 ± 3.2 *		92 ± 1.9 *†‡	
	Placebo	102.2 ± 3.5		99.2 ± 3.4 *		96.1 ± 2.9 *	
Hip	Supplement	117.4 ± 6.9		112.3 ± 5.9 *		109.2 ± 4.7 *	
	Placebo	118.2 ± 5.1		113.3 ± 3.5 *		110.1 ± 4.2 *	
Thigh	Supplement	71.1 ± 5.6		70.2 ± 4.9		68.1 ± 2.4 *	
	Placebo	72.2 ± 4.6		71.1 ± 4.1		69.1 ± 3.2 *	

\* = P < 0.05 from corresponding Week 0 value

† = P < 0.05 from corresponding Week 4 value

‡ = P < 0.05 from corresponding Placebo Group value

**Table 2.** Responses of Adipocytokines to the 8-week weight loss intervention (Mean±SD)

		week 0		week 4		week 8	
Insulin (mU/L)	Supplement	7.2 ± 2.2		4.5 ± 2.1 *		5.4 ± 2.4 *	
	Placebo	7.7 ± 2.2		4.12 ± 1.9 *		5.22 ± 2.7 *	
Adiponectin (µg/ml)	Supplement	12.2 ± 3.1		18.1 ± 3.1 *		26.2 ± 3.4 *†‡	
	Placebo	12.6 ± 4.2		19.3 ± 5.1 *		21.7 ± 4.3 *†	
Leptin (ng/ml)	Supplement	28.2 ± 6.1		22.3 ± 8.1 *		16.3 ± 3.4 *†‡	
	Placebo	29.3 ± 9.1		23.3 ± 5.1 *		19.7 ± 3.3 *†	

\* = P < 0.05 from corresponding Week 0 value

† = P < 0.05 from corresponding Week 4 value

‡ = P < 0.05 from corresponding Placebo Group value

Figure 1: Body Mass (kg) during the 8-week weight loss intervention for the supplement and placebo groups. \* =  $P < 0.05$  from corresponding Week 0 value. † =  $P < 0.05$  from corresponding Week 4 value. ‡ =  $P < 0.05$  from corresponding Placebo Group value

Figure 2: Body Composition (% Body Fat) during the 8-week weight loss intervention for the supplement and placebo groups. \* =  $P < 0.05$  from corresponding Week 0 value. † =  $P < 0.05$  from corresponding Week 4 value. ‡ =  $P < 0.05$  from corresponding Placebo Group value.

Figure 3: Waist Circumference (cm) changes during the 8-week weight loss intervention for the supplement and placebo groups. \* =  $P < 0.05$  from corresponding Week 0 value. † =  $P < 0.05$  from corresponding Week 4 value. ‡ =  $P < 0.05$  from corresponding Placebo Group value

Figure 4: Circulating Adiponectin ( $\mu\text{g/ml}$ ) during the 8-week weight loss intervention for the supplement and placebo groups. \* =  $P < 0.05$  from corresponding Week 0 value. † =  $P < 0.05$  from corresponding Week 4 value. ‡ =  $P < 0.05$  from corresponding Placebo Group value

Figure 5: Circulating Leptin (ng/ml) during the 8-week weight loss intervention for the supplement and placebo groups. \* =  $P < 0.05$  from corresponding Week 0 value. † =  $P < 0.05$  from corresponding Week 4 value. ‡ =  $P < 0.05$  from corresponding Placebo Group value

Figure 1

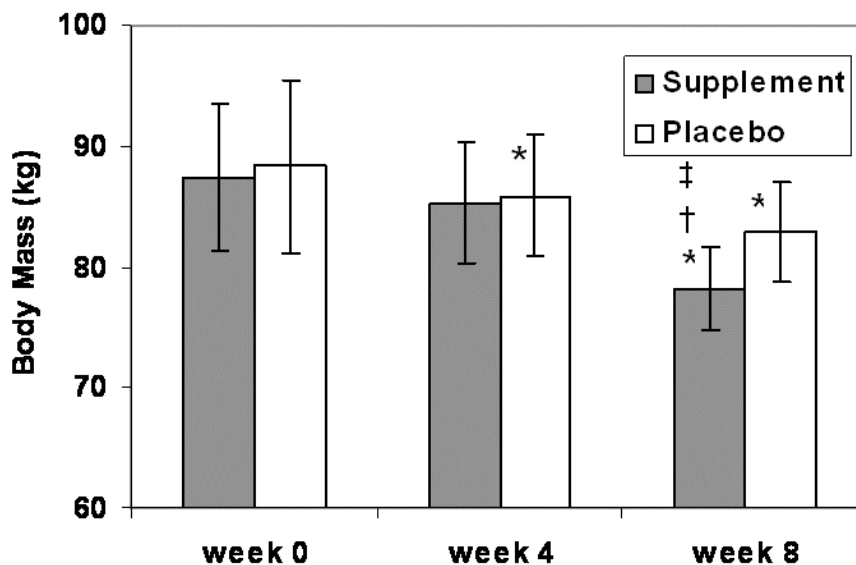


Figure 2

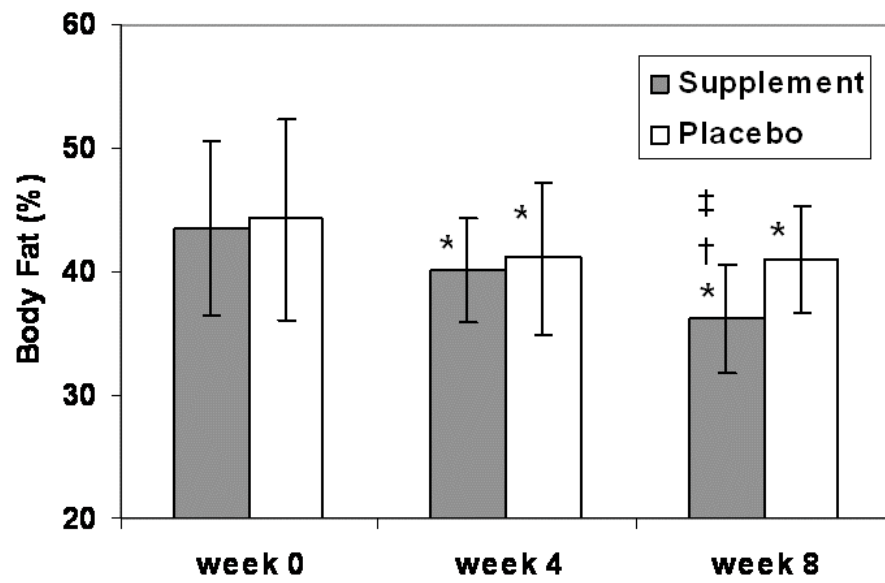


Figure 3

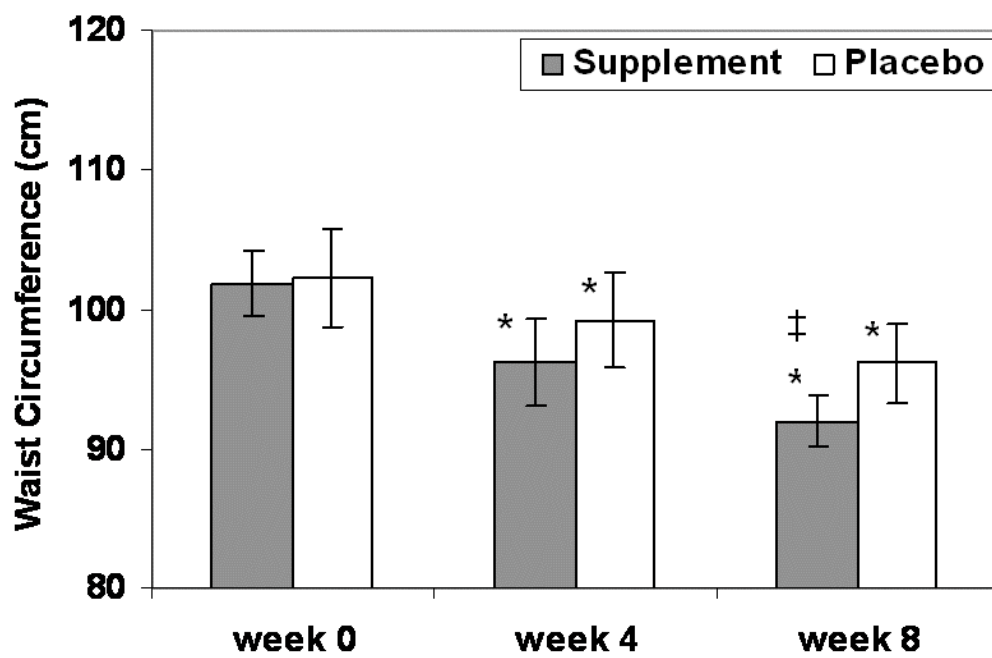




Figure 4

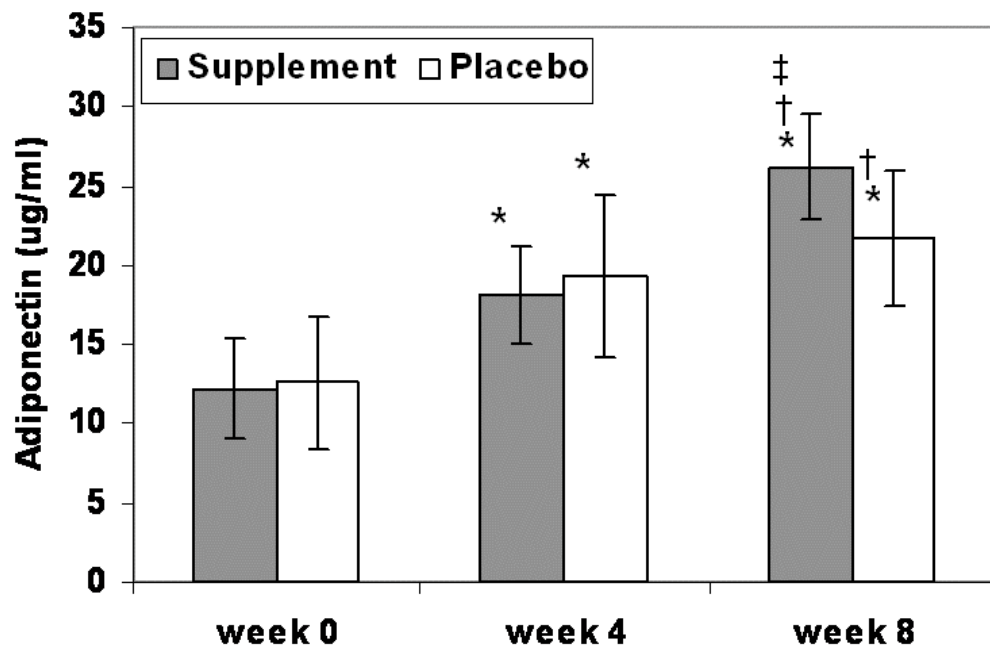


Figure 5

