

The Modifying Effects of A Calcium-rich Whey Protein Supplement (OsoLean™ Powder) on Weight Loss and Waist Circumference in Overweight Subjects: A Preliminary Study

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Abstract: This 8-week, open-label study evaluated the effects of a specialized whey protein supplement (OsoLean™ powder), rich in bioactive peptides and calcium, on weight loss in a convenience sample of healthy, overweight adults. Participants were advised to restrict their daily caloric intake and instructed to add the supplement to the beverage of their choice before breakfast and dinner. Subjects were provided a tracking log in which they were asked to record their height, daily supplement consumption, and weekly self-measurements of weight, waist circumference, and estimated daily caloric intake. BMI was calculated by the study coordinator using the self-reported height and weight measurements. Out of 112 enrolled subjects, 110 completed the first 4-week study period and 68 continued to complete the second 4 weeks. Subjects reported an average \pm standard error (SE) weight loss of 2.0 ± 0.2 kg by week 4 ($p<0.0001$) and 2.8 ± 0.3 kg by week 8 ($p<0.0001$). Mean BMI values were significantly decreased after 4 and 8 weeks, by 0.7 ± 0.1 and 1.0 ± 0.1 points, respectively. Average centimeters lost from the waist was 5.1 ± 0.3 cm at week 4 ($p<0.0001$) and 7.1 ± 0.7 cm at week 8 ($p<0.0001$). Subjects completing quality of life questionnaires reported better appetite control and improved energy levels by the end of the study. These preliminary results suggest that supplementation with this specialized whey protein product may result in significant weight loss and appetite control in overweight consumers. However, because of this study's uncontrolled, open-label methodology, cause-and-effect inferences can only be confirmed when a double-blind, randomized placebo-controlled trial is performed.

INTRODUCTION

The exploding number of overweight individuals has become a global health concern. The World Health Organization estimates that over 1 billion people worldwide are classified as overweight ($BMI\geq 25$) [1]. Currently in the U.S., about two-thirds of American adults are overweight [2], and it has been predicted that over 85% of Americans will be overweight by the year 2030 [3]. Authorities are attempting to address this problem via population-wide measures to improve dietary choices and increase physical exercise [4].

Daily dairy intake has been suggested to be important for the maintenance of healthy body weight, although results from studies have been equivocal [5]. In clinical studies of overweight adults observing calorie-restricted diets, consumption of at least three servings of dairy products a day for 6 months resulted in significantly enhanced weight and fat loss [6-8]. However, in a 12-year follow-up study of adult men in the U.S., no positive association was found between increased dairy consumption and lower long-term weight gain [9]. Consumption of different types of dairy products (e.g. high-fat vs. low-fat) has been shown to account for the inconsistencies in the reported effects of dairy on

anthropometric measures, as well as other factors associated with being overweight [10, 11]. A study showing an inverse relationship between dairy consumption and enlarged waist circumference alluded to the importance of the role of calcium and other substances found in dairy products [12]. More recent studies have thus focused on supplementation with components of dairy, such as calcium and whey protein, that show particular promise in supporting a healthy body composition.

Calcium has been shown to attenuate fat and weight gain in the absence of caloric restriction, and to accelerate fat loss during periods of reduced caloric intake [13]. A review of four clinical weight-loss studies of overweight women in their 30s or older reported a significant inverse association between calcium intake and body weight [14]. However, a recent review of randomized clinical studies looking at the effects of calcium supplementation on weight and fat loss suggests that the majority of evidence does not support these findings [5].

Clinical studies of whey protein supplementation have demonstrated significant benefits during weight loss by improving overall body composition [15]. In a 12-week, placebo-controlled, randomized clinical trial of overweight subjects adhering to a 500 calorie-reduced diet, daily consumption of a flavored, sweetened beverage including 24.4 g of intact and hydrolyzed whey protein, peptides, calcium, and other milk minerals significantly increased fat

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loss and retained lean muscle mass when compared to non-supplemented controls [16].

While dairy and calcium intake are often associated with weight and/or fat loss, the results from multiple clinical studies make it difficult to establish a clear relationship. Whey protein supplementation also appears to improve body composition, but its effects on weight loss are less studied. With an overwhelming number of weight-loss supplements on the market, it is becoming more difficult to fully assess and rely on their proposed safety and efficacies. This open-label study aimed to determine whether a version of a clinically-tested, calcium-rich whey protein supplement [16], without sweeteners or flavor additives, could positively impact weight, BMI, waist circumference, and quality of life parameters in overweight volunteers.

MATERIALS AND METHODOLOGY

Study Design

An open-label study was designed to assess the efficacy of a whey protein supplement on weight, BMI, and waist circumference over an eight-week period. Before initiation of the study, pre-screened, healthy, overweight ($BMI \geq 25$) subjects were advised to consult a healthcare professional before starting the reduced-calorie diet program and were required to sign an Informed Consent form. Subjects were advised to reduce caloric intake by 500 calories/day for the duration of the study, relying on limiting food size portions as a key to success. To improve the chances of success with caloric reduction, women were given a sample meal plan providing 1500 calories/day, and men were given a sample meal plan providing 2000 calories/day, although individual caloric intakes were expected to vary based on personal consumption. Participants were given a tracking log in which they were required to record their height and their daily whey protein supplement intake, and at the end of each week, record their estimated daily caloric intake, body weight, and waist circumference. The study was divided into two four-week periods. Subjects who completed the tracking log through week 4 were allowed to continue through week 8. A self-administered questionnaire to assess quality of life was completed on day 1 and at the end of weeks 4 and 8. Subjects were instructed to report any adverse signs or symptoms experienced while on the study product. The study protocol was reviewed and approved by an internal ethics committee and was conducted in compliance with the provisions of the Declaration of Helsinki.

Subjects

A convenience sample of healthy, overweight adults who had stable body weights over the previous two months and were not involved in another weight loss plan were eligible for study pre-screening. Pre-screening exclusion criteria included: known contraindication or hypersensitivity to any ingredients in the supplement; females who were planning to be pregnant, who were pregnant or lactating, or who were not willing to use a medically-approved birth control method; individuals for which weight loss programs were contraindicated as advised by their physician; weight loss ≥ 10 lb (4.5 kg) in the last 6 months; history or evidence of clinically significant disease such as cancer, liver disease, kidney disease, cardiovascular disease, uncontrolled hyper-

tension, thyroid disorder, and other endocrine problems; on medications known to affect weight; previous/current diagnosis of an eating disorder; current bipolar, depression, substance abuse, or dependence disorder; history of intestinal obstruction or prone to intestinal obstruction; history of any surgery on their gastrointestinal system or perforation of the stomach or intestines; history or evidence of peptic ulcer; history or evidence of Inflammatory Bowel Disease; history of brain or spinal cord injury.

Supplement

The specialized whey protein supplement (OsoLean™ powder, Mannatech, Incorporated, Coppell, TX) provided 45 calories and contained 10 g of protein and 244 mg of calcium per serving. All ingredients are classified as GRAS (Generally Recognized as Safe) by the U.S. Food and Drug Administration. Subjects were instructed to add one serving (12.2 g) of the supplement to 8 fluid oz (237 mL) of liquid and drink once in the morning, 20 minutes before breakfast, and again in the evening, 20 minutes before dinner, for a total of 24.4 g/day.

Body Measurements

Participants were given detailed instructions on how to properly measure body weight (in pounds), height (in inches), and waist circumference (in inches), and asked to record these measurements in their tracking log. Body weight was measured on bathroom scales, in bare feet and loose, non-bulky clothing (e.g. gym shorts and T shirt). If the weight fraction was less than $\frac{1}{2}$ pound, this was rounded down to the nearest whole pound; if the weight fraction was $\frac{1}{2}$ pound or greater, this was rounded up to the next whole pound. At the beginning of the study, height was measured in bare feet and loose clothing, standing on a flat surface with the head held horizontal, eyes looking directly forward with the line of vision horizontal, and chin parallel to the floor. Height was recorded to the nearest $\frac{1}{2}$ inch. Waist circumference was determined by measuring the waist at the midpoint between the lower border of the rib cage and the iliac crest, keeping arms at the side of the body, and using a tape measure against the skin, level and parallel to the floor. Waist circumference was recorded at the end of a normal, relaxed exhalation and rounded down to the nearest $\frac{1}{2}$ inch. BMI was calculated by the research staff from the weight and height measurements using the BMI calculator of the Department of Health and Human Services, National Institutes of Health [17]. For publication purposes, all measurements were converted to SI Units (International System of Units) by the research staff.

Quality of Life Assessment

Investigators developed a simple quality of life questionnaire based on other commonly used quality of life questionnaires. Subjects completed the questionnaire on day 1 and at the end of weeks 4 and 8. Ten questions were asked regarding subjects' overall health, performance of moderate activities and normal work, energy levels, ability to cope with stress, personal relationships, mood, sleep pattern, pain relief, and ability to control appetite. Answers were chosen from a scale of how often the subjects experienced positive levels of these qualities: none, a little, some, most, or all of the time.

Statistics

Body weight, BMI, and waist circumference were analyzed using a two-tailed paired t-test to compare the primary outcomes at specific time periods vs. baseline values. Data from the quality of life questionnaire was analyzed using the McNemar's chi-square test to detect if there were significant changes in subject responses. Significance was set at a $p \leq 0.05$.

RESULTS

Subject Participation

From a convenience sample of 112 participants (80 females, 32 males) who were deemed eligible and signed an Informed Consent form, 110 subjects completed the first four weeks of the study, and 68 continued through the second four weeks. Two participants dropped out on the first week. After the completion of week 4, one volunteer dropped out because of mild gastrointestinal symptoms, one complained of the taste of the product, and 40 subjects either voluntarily withdrew for personal reasons unrelated to the study or dropped out for unspecified reasons.

Estimated Caloric Intake

The subjects of this study did not appear to comply with the recommended 500 calorie/day reduction. The reported average changes in daily caloric intake were significant during the first 4 weeks of the study (range: -132 to -65 cal; $p < 0.05$), but did not remain significant through the final 4 weeks (range: -18.5 to 19.1 cal).

Effects of Whey Protein Supplementation on Body Composition

Baseline characteristics of the 112 participants who began the study are described in Table 1. During the study, body weight continuously decreased from baseline, with an average weight loss of 2.0 ± 0.2 kg at the end of week 4 and 2.8 ± 0.3 kg at the end of week 8 (Fig. 1). The average amount of weight loss became significant compared to baseline by the end of the first week and remained significant through the end of week 8 ($p < 0.0001$). The significant losses in body weight led to reductions in the mean BMIs of overweight subjects, a difference of 0.7 ± 0.1 points at the end of week 4 and a difference of 1.0 ± 0.1 points at the end of week 8 ($p < 0.0001$) (Fig. 2).

Table 1. Baseline Characteristics of Study Participants (N=112)

Age (y) (%)	18-34	8.0
	35-44	13.4
	45-54	30.4
	55-64	34.8
	65-74	12.5
	75+	0.9
Height (cm)*		167.9 ± 0.9
Weight (kg)*		87.1 ± 1.6
BMI*		30.9 ± 0.5
Waist circumference (cm)*		99.8 ± 1.3

*Reported values are means \pm SE.

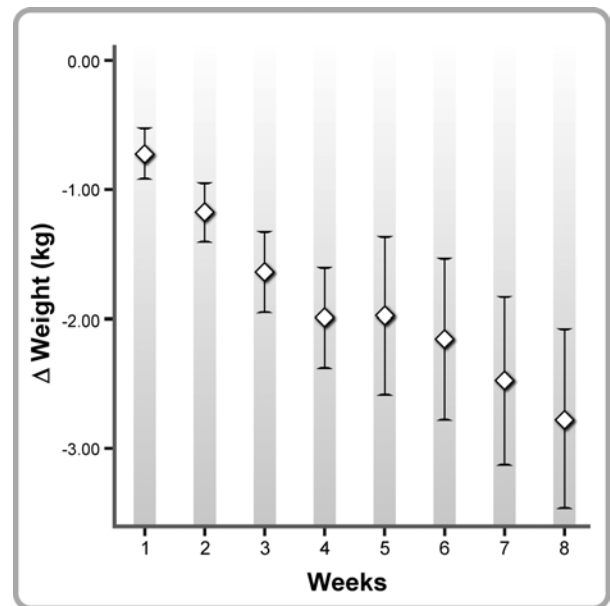


Fig. (1). Body weight was reduced in subjects consuming a specialized whey protein supplement twice a day for 8 weeks. The average body weights reported at the end of each week during the study were significantly reduced compared to baseline ($p < 0.0001$). Error bars represent \pm SE.

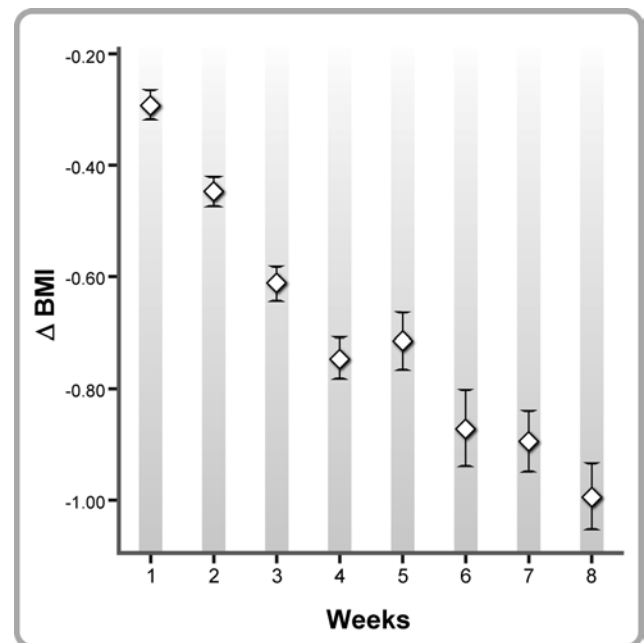


Fig. (2). Mean Body Mass Index (BMI) decreased in subjects consuming a specialized whey protein supplement twice a day for 8 weeks. Average BMI significantly decreased compared to baseline values by the end of the first week ($p < 0.0001$) and remained significant throughout the completion of the study ($p < 0.0001$). Error bars represent \pm SE.

Waist circumference is a convenient and simple measure which is unrelated to height, correlates closely with BMI, and is an approximate index of intra-abdominal fat mass and total body fat [18]. Subjects reported losing an average of 5.1 ± 0.3 cm by the end of week 4 and an average of 7.1 ± 0.7 cm by the end of week 8 ($p < 0.0001$) from their baseline

waists (Fig. 3). The changes in weight, BMI, and waist circumference were not gender specific, as all three measurements were significantly reduced in both males and females (data not shown).

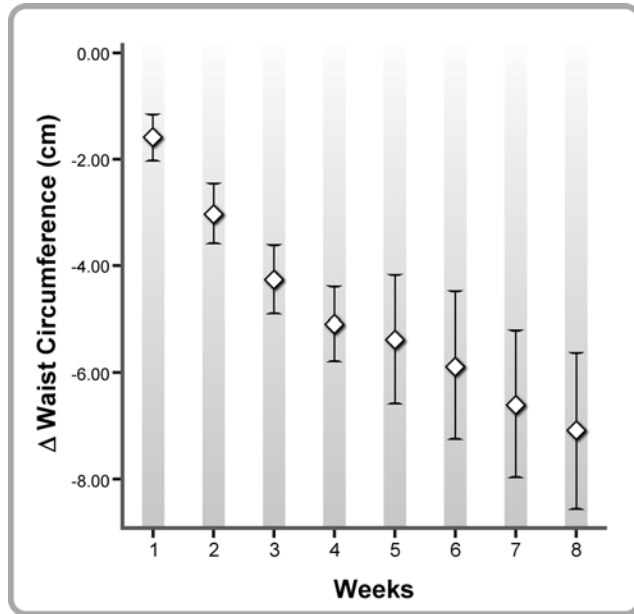


Fig. (3). Waist circumference was reduced in subjects consuming a specialized whey protein supplement twice a day for 8 weeks. The average waist circumferences reported at the end of each week during the study were significantly reduced compared to baseline ($p < 0.0001$). Error bars represent \pm SE.

Effects of Whey Protein Supplementation on Quality of Life

Analysis of the self-administered quality of life questionnaire showed interesting findings on appetite control, as well as improvements in energy levels (Fig. 4). While slight im-

provements were seen in all quality of life parameters during the duration of the study, appetite control and energy levels were enhanced the most. On day 1, 42.85% of subjects reported an ability to control appetite most or all of the time. By the end of week 8, reports of positive levels of appetite control increased by an additional 27.3% of subjects. The increase in the number of subjects reporting improvements in appetite control by the end of the study was significant ($p < 0.01$). The percentage of subjects that reported positive energy levels also largely increased by the end of week 8. The subjects' reported improvements in energy levels was statistically significant ($p < 0.01$).

Adverse Effects

The supplement was well-tolerated by the majority of the participants in this study. Adverse events were mostly mild and there were no serious adverse effects reported that required a physician's care. Nineteen subjects reported symptoms such as bloating, flatulence, and headache. Of these subjects, two dropped out before the end of week 1, six dropped out after week 4, and two after week 6. The remaining nine subjects completed all 8 weeks of supplementation, reporting that their symptoms eventually disappeared.

DISCUSSION

Supplementation with 24.4 g/day of this specialized whey protein for 8 weeks resulted in a steady loss of body weight, decrease in BMI, and decrease in waist circumference in overweight individuals. Subjects also reported better appetite control and energy levels over the study period. We suggest that the effects on weight loss, waist reduction, and appetite control were likely due to an increased daily intake of calcium and consumption of specialized bio-active peptides found in whey protein, as both changes have been associated with positive impacts on weight and body composition.

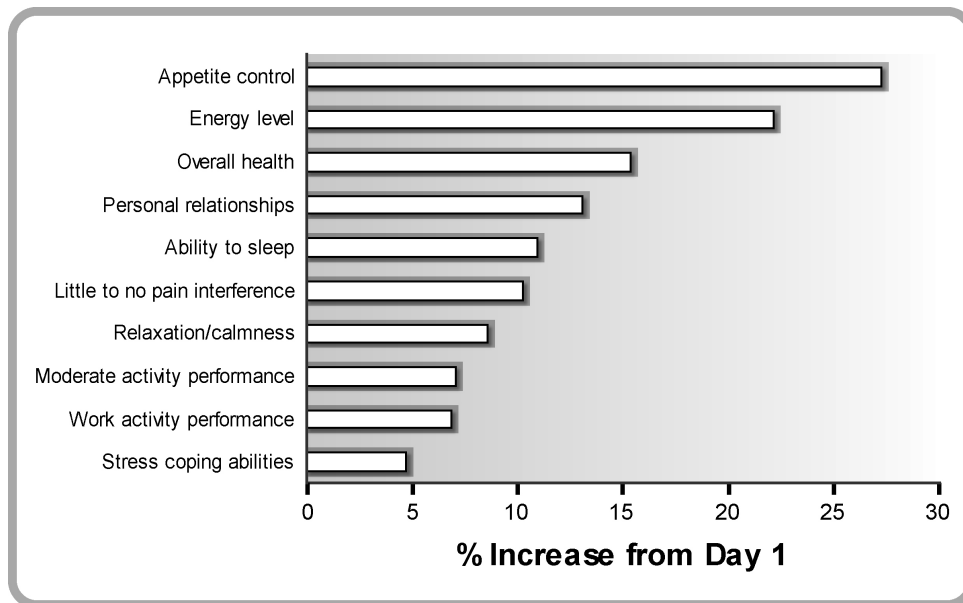


Fig. (4). Results from the Quality of Life questionnaire completed by subjects on Day 1 and at the end of the week 8. The percentages of subjects reporting positive levels of different quality of life parameters increased slightly during the duration of the study. The largest positive effects were seen with appetite control and energy levels.

Evidence suggests that consumption of an additional 300 mg calcium (~1 dairy serving) per day may result in decreased weight gain, or greater weight loss during dieting, in the range of 0.24-0.35 lbs (0.11-0.16 kg) per year [19]. Increasing calcium intake, via supplementation or dairy consumption, to an amount of 1200 mg/day for 6 months has been shown to enhance weight and fat loss during caloric reduction, with more fat lost from the abdominal region when compared to low calcium diets (500 mg/day) [6]. Many adults in the U.S. consume inadequate amounts of calcium, supporting the need for daily calcium supplementation. The Adequate Intake (AI) requirement for calcium is 1000 mg/day in adults aged 19 to 50 years, while the actual estimated median calcium intake for men in this age group is around 900 mg and for women is around 600 mg [20]. The inadequate intake of calcium becomes more significant with age: the AI for adults aged 51-70 years is 1200 mg/day and the estimated median calcium intake is only 708 mg for men and 571 mg for women [20]. Consumption of this whey protein supplement provided an additional 488 mg/day calcium, bringing intake closer to AI levels for both men and women.

The mechanisms by which calcium impacts fat and weight loss have not been clearly established. Animal and human studies have shown that increasing dietary calcium can reduce calcitropic hormone levels and increase fecal fatty acid excretion [21, 22]. Both mechanisms may be responsible for the observed fat loss seen with increased dietary calcium consumption in humans [8].

An additional component of dairy, whey protein is thought to positively impact fat and weight loss. High-protein, reduced-calorie or reduced-fat diets have been shown to be an effective means for losing weight and improving body composition in overweight adults [23], while whey protein supplementation has resulted in similar effects. Baer *et al.* [24] demonstrated that after 6 months of receiving 60 g/day of whey protein, subjects weighed less, had less body fat, and lost more inches around the waist compared to a group that consumed increased amounts of carbohydrates. In these and other weight loss studies, the amount of daily protein consumed resulted in increased intake levels above AI values. For both men and women, it is recommended that protein provides around 10-20% of total caloric intake, and most people successfully reach that mark with their normal daily diets [20]. However, consuming amounts of protein that bring the fraction of caloric intake above 20% has repeatedly been demonstrated to assist in fat loss and improvements in body composition [25]. As a result, one proposed solution for the problem of excess body weight is to increase the protein content of the food supply to provide 20-30% of energy, rather than the current 10-20%.

The mechanism(s) by which increased protein intake impacts weight and fat loss is not entirely clear. There is evidence suggesting that higher protein intake can enhance thermogenesis compared to diets lower in protein content, resulting in increased energy expenditure [26]. High protein intake may also lead to reduced energy intake at subsequent meals [25]. In addition, dairy proteins, such as whey, contain a high proportion of bioactive compounds, such as angiotensin converting enzyme (ACE) inhibitors and branched chain amino acids, which can act synergistically with calcium to enhance fat loss and maintain lean muscle mass [8].

In this study, and in two previous studies using a related specialized calcium-rich whey protein supplement, the addition of only 20 g protein per day had a positive impact on weight and/or fat loss [15, 16], an amount of protein much smaller than those used in other weight loss studies.

While the previous studies demonstrated that a specialized, calcium-rich whey protein supplement could positively impact fat loss with the addition of only a small amount of protein per day, effects on weight were not significantly altered compared to controls and effects on appetite were not assessed [15, 16]. Whey protein preloads (40-55 g) consumed 1-4 hours before a meal have been shown to significantly increase satiety when compared to preloads containing carbohydrates or other protein sources, such as casein [27-29]. To maximize the impact on appetite, this study followed the protocol established by Frestedt *et al.*, [16] instructing volunteers to consume the whey protein supplement 20 minutes before breakfast and again 20 minutes before dinner. The subjects in this study reported improvements in appetite control, indicating that consumption of this specialized whey protein supplement may also impact satiety.

The reported improvement in appetite control, and documented weight loss, would be expected to correlate with a reduction in daily energy intake. However, the subjects of this study did not appear to comply with the recommended 500 calorie/day reduction. It is important to note that dietary recall is one of the largest sources of error in weight loss research [30]. The limitations of this study were that the participants were not in-house, resulting in a high rate of attrition between the beginning and end of the study (60.7%), and that all data collection was dependent on the subjects' compliance with completion of their tracking logs. Therefore, while an attempt was duly made to assess the caloric intake of the volunteer participants in this study, the authors believe that the impact of subjects' diets on the study results cannot be properly assessed.

Successful weight and fat loss in overweight adults must include a healthy calorie-restricted diet, along with regular exercise. This study should be replicated using a randomized, double-blind, placebo-controlled design in order to exert more strict control over diet and exercise, and to further explore the novel findings of weight and centimeters lost, as well as the improvement in appetite control and energy levels reported by subjects consuming this specialized whey protein supplement.

CONCLUSION

This open-label study demonstrated that daily consumption of a specialized whey protein supplement, containing 20 g whey protein and 488 mg calcium, significantly decreased weight, BMI, and waist circumference in healthy, overweight adults over an 8 week period. Also noted by the subjects was better appetite control and improved energy levels. The whey protein supplement was demonstrated to be safe to consume over this time period. The ability to draw definitive conclusions is limited because of this study's uncontrolled, open-label methodology. In addition, failure of the subjects to report reduced caloric intake makes it difficult to fully assess the impact of diet on these effects. However, the results are very encouraging and should be explored further using a randomized, double-blind, placebo-controlled trial.

CONFLICT OF INTEREST

The study was sponsored and conducted, and the manuscript was prepared and submitted, by Mannatech, Incorporated that sells OsoLean™ powder. RAS, RLM, EDN, and JAA are employees of Mannatech, Incorporated. Data and statistical analyses were performed independently by SB and KPS, faculty at the University of North Texas Health Science Center.

ACKNOWLEDGEMENTS

RAS, RLM, and JAA participated in the design and development of the study. RLM and EDN prepared the draft of the manuscript. SB and KPS independently performed all statistical analyses. All authors read, revised, and approved the manuscript. The authors would like to thank the study participants, Jane Ramberg, Barbara Kinsey, Mary Wood, Stephen Boyd, Mo Uddin, Lisa Pennewell, Jennifer Aponte, Amy Bart, Angela Bowen, and Dana Fuller, as well as the Product Development and Quality Assurance teams at Mannatech, Incorporated for their assistance with this study.

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Received: December 18, 2008

Revised: January 16, 2009

Accepted: February 16, 2009

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