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# A high whey protein–, leucine-, and vitamin D–enriched supplement preserves muscle mass during intentional weight loss in obese older adults: a double-blind randomized controlled trial[1–3]

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**ABSTRACT**

**Background:** Intentional weight loss in obese older adults is a risk factor for muscle loss and sarcopenia.

**Objective:** The objective was to examine the effect of a high whey protein–, leucine-, and vitamin D–enriched supplement on muscle mass preservation during intentional weight loss in obese older adults.

**Design:** We included 80 obese older adults in a double-blind randomized controlled trial. During a 13-wk weight loss program, all subjects followed a hypocaloric diet (−600 kcal/d) and performed resistance training 3×/wk. Subjects were randomly allocated to a high whey protein–, leucine-, and vitamin D–enriched supplement including a mix of other macro- and micronutrients (150 kcal, 21 g protein; 10×/wk, intervention group) or an isocaloric control. The primary outcome was change in appendicular muscle mass. The secondary outcomes were body composition, handgrip strength, and physical performance. Data were analyzed by using ANCOVA and mixed linear models with sex and baseline value as covariates.

**Results:** At baseline, mean ± SD age was 63 ± 5.6 y, and body mass index (in kg/m$^2$) was 33 ± 4.4. During the trial, protein intake was 1.11 ± 0.28 g · kg body weight$^{-1}$ · d$^{-1}$ in the intervention group compared with 0.85 ± 0.24 g · kg body weight$^{-1}$ · d$^{-1}$ in the control group ($P < 0.001$). Both intervention and control groups decreased in body weight (−3.4 ± 3.6 kg and −2.8 ± 2.8 kg; both $P < 0.001$) and fat mass (−3.2 ± 3.1 kg and −2.5 ± 2.4 kg; both $P < 0.001$), with no differences between groups. The 13-wk change in appendicular muscle mass, however, was different in the intervention and control groups [+0.4 ± 1.2 kg and −0.5 ± 2.1 kg, respectively; $\beta = 0.95$ kg (95% CI: 0.09, 1.81); $P = 0.03$]. Muscle strength and function improved over time without significant differences between groups.

**Conclusion:** A high whey protein–, leucine-, and vitamin D–enriched supplement compared with isocaloric control preserves appendicular muscle mass in obese older adults during a hypocaloric diet and resistance exercise program and might therefore reduce the risk of sarcopenia. This trial was registered at the Dutch Trial Register (http://www.trialregister.nl) as NTR2751. *Am J Clin Nutr* 2015;101:279–86.

**Keywords** muscle preservation, obese older adults, randomized trial, whey protein, weight loss

## INTRODUCTION

The prevalence of obesity among older adults is increasing rapidly (1). Obesity is related to insulin resistance, high blood pressure, and dyslipidemia, which are metabolic risk factors for cardiovascular diseases and diabetes mellitus. In addition, obesity plays an important role in nonfatal physical disability in older adults (2). Weight loss leads to metabolic and functional benefits (3). However, a potential drawback of weight loss in older adults is the accompanying loss of skeletal muscle mass (4), which eventually may accelerate the development of sarcopenia (5, 6). Reduction in muscle mass and strength impairs physical function and activities of daily living and is associated with an increased risk of falling and physical disabilities (5, 6). Thus, although obese older adults may benefit from weight loss, therapy should focus on minimizing loss of muscle mass to preserve independence and quality of life (5).

Weight loss can be achieved by a reduction of calorie intake and a stimulation of physical activity. Strategies to preserve muscle mass during weight loss focus on resistance exercise and sufficient intake of high-quality protein (7–9). Resistance training is known to stimulate muscle protein synthesis in older adults, which supports muscle mass preservation and muscle function (10). High dietary protein intake, strategically timed at each meal, has also been shown to stimulate muscle protein synthesis and is another potent strategy to overcome the well-known muscle anabolic resistance in older individuals (1, 11, 12). Whey protein is a high-quality protein that has shown superiority in enhancing muscle protein synthesis compared with other protein sources in older adults (13, 14). This effect of whey is likely attributed to the faster digestion and absorption and the high content of essential amino acids, including leucine (15). Leucine is a powerful stimulator of muscle protein synthesis, and it was recently shown that leucine coingestion with a bolus of protein could further improve muscle protein synthesis (16).

The combination of a high intake of fast-digesting, high-quality protein and resistance exercise is suggested to have a synergistic effect on muscle mass preservation during weight loss (1, 17, 18), but data in obese older adults are limited (19). In addition, several studies suggest a positive effect of vitamin D on muscle protein

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metabolism (20, 21), and therefore vitamin D (800 IU) might have a potential beneficial effect on muscle mass preservation.

We therefore compared the effects of a high whey protein–, leucine-, and vitamin D–enriched nutritional supplement with an isocaloric control during a 13-wk weight-loss program consisting of a hypocaloric diet and resistance exercise training on appendicular muscle mass preservation in obese older adults.

## SUBJECTS AND METHODS

### Subjects

Obese men and women (aged ≥55 y) were recruited from the Dutch population through local flyers and advertisements. Obesity was defined as a BMI (in kg/m$^2$) >30 or as a BMI >28 with waist circumference >88 cm (women) or >102 cm (men). Potential subjects were excluded if they had any malignant diseases during the past 5 y, if they had participated in any weight loss program 3 mo before screening, if participation in the resistance training program was considered unsafe according to a physiotherapist, or when they were not able to comply with the full study protocol. A full description of the eligibility criteria is available online in the Dutch Trial Register (NTR2751; http://www.trialregister.nl). The study was approved by the Medical Ethics Committee of the VU University Medical Center Amsterdam (2010/280), and written informed consent was obtained from all subjects. The study took place from March 2011 through June 2012 at the Amsterdam University of Applied Sciences in The Netherlands.

### Design and randomization procedures

We performed a 13-wk randomized, controlled, double-blind, parallel group trial (i.e., Muscle Preservation Study). Eligible subjects were randomly allocated (1:1) to consume a high whey protein–, leucine-, and vitamin D–enriched supplement (intervention group) or an isocaloric control product (control group) by means of randomization envelopes with 4 different randomization codes stratified by sex. The randomization codes were generated by an independent statistician who was not involved in the conduct of the study. Body composition, including appendicular muscle mass, was assessed at baseline and after 13 wk of intervention. Body weight, BMI, waist circumference, muscle strength, and physical functioning were measured at baseline and after 7 and 13 wk of intervention.

### Hypocaloric diet

All subjects followed a hypocaloric diet of 600 kcal below estimated energy needs according to the Dutch guideline (22). Energy needs were estimated by multiplying measured resting energy expenditure by indirect calorimetry (Vmax Encore n29; Viasys Health Care), with physical activity level estimated by a 3-d physical activity record. This hypocaloric advice included the caloric content of 1 serving of the study products. The second serving, given only after training sessions, was provided in addition to the daily diet. At baseline, subjects received a standardized dietary plan according to the Dutch guideline (22) with a list of variation options. In the first week, subjects were called to check for compliance. Every 2 wk, subjects followed dietary counseling sessions in groups of 8–12 subjects in which experiences were shared and nutrition-related topics were discussed. Dietary intake was assessed by a 3-d food record at baseline and after 7 and 13 wk of intervention. Food records were checked for completeness during study visits, and additional information was obtained about unclear items or amounts. Total energy and macronutrient intakes were calculated by using a computerized Dutch Food Composition Table (23).

### Resistance exercise program

All subjects participated in the resistance exercise program, which was performed 3×/wk for 1 h under supervision of a qualified trainer for 13 wk. The training started with a 10-min warmup on a bicycle ergometer followed by 3 sets of 20 repetitions of the following 10 exercises: lateral pulldown, arm curl, high row, shoulder press, leg curl, horizontal row, chest press, arm extension, leg extension, and leg press. The number of repetitions was stepwise reduced to 12 repetitions, and the weights were increased to the ability of the participants. The training ended with a 5-min cool-down on a bicycle ergometer.

### Study products

Study products were provided by Nutricia Research. The composition of the study products is displayed in **Table 1**. Both products were similar in taste and appearance and provided an energetic value of 150 kcal per serving in a volume of 150 mL. Subjects were asked to consume 10 servings of the study product per week throughout the 13-wk intervention period. Subjects consumed 1 serving daily, just before breakfast, whereas 3 servings were consumed immediately after exercise training (3×/wk). Study products had to be consumed as a single bolus within 5–10 min. Subjects were asked to record product intake in a diary to check compliance.

### Measurement of body composition, muscle strength, and physical performance

Body composition, including appendicular muscle mass (primary outcome), was measured with dual-energy X-ray absorptiometry (GE Lunar Prodigy/DPX-NT; GE Healthcare). To limit within-subject variation, we performed dual-energy X-ray absorptiometry scans at the same time of the day during both visits. Appendicular muscle mass was defined as the sum of lean mass (without bone) of both arms and legs. Skeletal muscle mass index was calculated by dividing the appendicular skeletal muscle mass (kg) by height squared (m$^2$). Body weight was measured on a calibrated scale (Life Measurement). Height was measured to the nearest 0.5 cm by using a wall-mounted stadiometer (Seca 222; Seca).

For muscle strength, hand grip strength was measured with an isometric hand grip dynamometer (JAMAR 5030J1; Sammons Preston Rolyan) while the subject was in a sitting position. Three consecutive measures of hand grip strength (kg) for both hands were recorded to the nearest 0.1 kg, and all values were averaged. Physical performance was assessed with a 400-m walking test, a 4-m gait speed test, and a chair stand test (24).

### Statistical analysis

Double data entry was performed and discrepancies were solved. Treatment codes were broken after locking the database.

**TABLE 1**
Composition of the study products used in the double-blind, randomized, placebo-controlled trial of a high whey protein–, leucine-, and vitamin D–enriched supplement on preservation of muscle mass during a weight loss trial in obese older adults[1]

| Component | Intervention | Control |
|---|---|---|
| Energy | | |
| kcal | 150 | 150 |
| kJ | 635 | 635 |
| Protein, % | 55 | — |
| Carbohydrates, % | 25 | 82 |
| Fat, % | 18 | 18 |
| Fiber, % | 2 | — |
| Total protein, g | 20.7 | — |
| Total leucine,[2] g | 2.8 | — |
| Total EAA,[2] g | 10.6 | — |
| Carbohydrates | | |
| Total, g | 9.4 | 31.4 |
| Sugars, g | 4.2 | 2.6 |
| Total fat, g | 3.0 | 3.0 |
| Fiber | | |
| Total, g | 1.3 | — |
| Soluble, g | 1.3 | — |
| Minerals[3] | | |
| Sodium, mg | 150 | 142 |
| Potassium, mg | 279 | 176 |
| Chloride, mg | 70 | 344 |
| Vitamin $D_3$,[3] $\mu$g | 20 | — |

[1]Per serving of 150 mL. BCAA, branched-chain amino acids (Leu, Ile, and Val); EAA, essential amino acids (Leu, Ile, Val, Phe, Met, His, Trp, Thr, and Lys).

[2]Provided by protein and free BCAA.

[3]Intervention product also contained micronutrients: calcium (500 mg), phosphorus (250 mg), magnesium (37 mg), iron (2.4 mg), zinc (2.2 mg), copper (270 $\mu$g), manganese (0.50 mg), fluoride (0.15 mg), molybdenum (15 $\mu$g), selenium (15 $\mu$g), chromium (7.5 $\mu$g), iodine (20 $\mu$g), vitamin A (152 $\mu$g retinol equivalents), vitamin E (7.5 mg $\alpha$-tocopherol equivalents), vitamin K-1 (12 $\mu$g), vitamin B-1 (0.23 mg), vitamin B-2 (0.25 mg), niacin (8.8 mg niacin equivalents), pantothenic acid (0.81 mg), vitamin B-6 (0.76 mg), folic acid (203 $\mu$g), vitamin B-12 (3.0 $\mu$g), biotin (6.1 $\mu$g), vitamin C (32 mg), carotenoids (0.30 mg), and choline (56 mg).

Statistical analyses were performed with appendicular muscle mass as the primary outcome. In the analyses, we included all available post-baseline data for all participants independent of the level of compliance. Sample size for the present study was estimated by using data from a pilot study combining a high-protein diet with resistance exercise training in older adults with fat-free mass as a proxy for appendicular muscle mass (unpublished internal data) because no other relevant published data were available. A sample size of 40 per arm provided 80% power to detect an absolute difference of 2.0 kg fat-free mass with an SD of 1.7 kg and $P < 0.05$ (2-sided), assuming a dropout rate of 35%.

Subject characteristics and dietary intake at baseline were compared between groups by using an independent-samples *t* test or the Fisher's exact test.

Between-group differences on outcome variables that were measured at baseline and after 13 wk were analyzed with an ANCOVA by using sex and baseline value as covariates. Between-group differences on outcome variables that were measured at baseline and after 7 wk and 13 wk were analyzed by using a mixed linear model, including time (differentiating week 7 from week 13), intervention (differentiating the intervention group from the control group), and the time × intervention interaction as fixed factors; subject as a random factor; and sex and baseline value as covariates. Intervention effect $\beta$ is the estimate for the difference between the intervention and the control group at 13 wk after correction for baseline and sex.

Within-group differences were estimated by using a paired-samples *t* test (for variables that were measured at baseline and after 13 wk) or the mixed linear model (for variables that were measured at baseline and after 7 wk and 13 wk).

SAS Enterprise Guide 4.3 for Windows (SAS Institute) software was used for all statistical analyses. Data in text and tables are expressed as means ± SDs. Statistical significance was defined as a 2-tailed $P < 0.05$.

## RESULTS

### Subjects and compliance

We enrolled 80 subjects in the trial. The number of subjects screened, excluded, and randomly allocated is shown in **Figure 1**. Fifteen subjects dropped out during the study because of adverse events ($n = 6$) and personal reasons ($n = 9$), all not related to study product intake. There were no relevant differences in subjects' characteristics between the groups (**Table 2**). Compliance to study product intake was comparable between groups: consumption of at least 7 study products per week by 91% in the intervention group and 97% in the control group ($P = 0.61$). Adherence to the exercise program was comparable between groups: training on average more than 2 times per week by 72% in the intervention group and 88% in the control group ($P = 0.21$).

### Dietary intake

Baseline energy needs were calculated by using the measured resting energy expenditure and the estimated baseline level of physical activity. No differences were observed between groups (intervention: 2621 ± 437 kcal/d; control: 2473 ± 636 kcal/d; $P = 0.33$). Self-reported mean dietary intake at baseline was 2072 ± 587 kcal/d in the intervention group and slightly higher compared with the 1775 ± 574 kcal/d in the control group ($P = 0.05$). Energy intake (including supplement) at week 13 of the study was not different between groups (**Table 3**, $P = 0.76$), although both groups significantly reduced their energy intake during the trial [change: −315 ± 499 kcal/d for intervention ($P = 0.005$) and −91 ± 504 kcal/d for control ($P = 0.01$)]. Protein intake at week 13, expressed as $g \cdot kg$ body weight $(BW)^{-1} \cdot d^{-1}$, was 1.11 ± 0.28 in the intervention group compared with 0.85 ± 0.24 in the control group ($P < 0.001$), which corresponded to a higher dietary protein intake during intervention of 27.6 ± 24.9 g/d in the intervention group compared with the control group ($P < 0.001$). Contribution of carbohydrates to the total dietary intake energy percentage was higher in the control group than in the intervention group ($P < 0.001$), and there were no differences in the contribution of fat to the total dietary intake ($P = 0.92$).

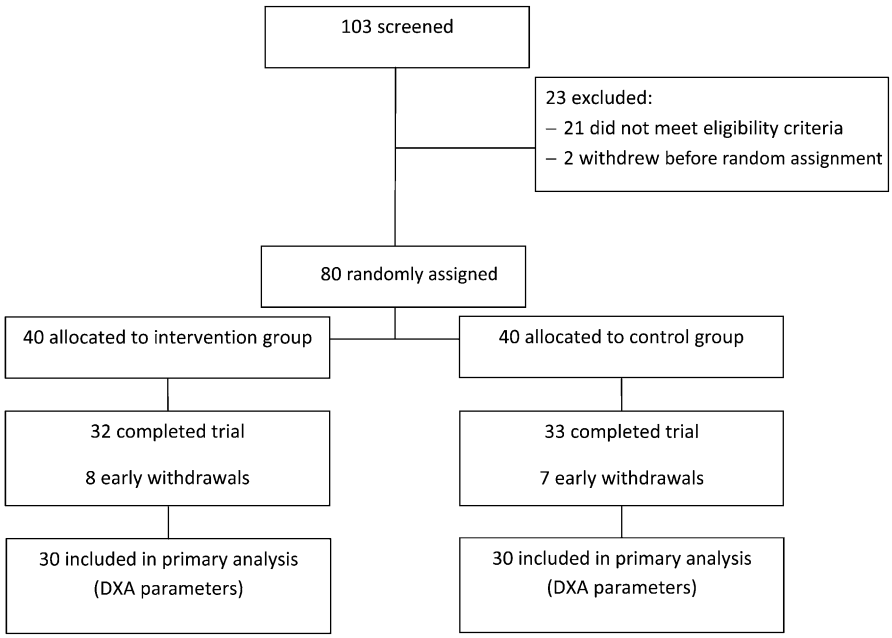


**FIGURE 1** Flowchart of a double-blind, randomized, placebo-controlled trial of a high whey protein–, leucine-, and vitamin D–enriched supplement on preservation of muscle mass during a weight loss trial in obese older adults. DXA, dual-energy X-ray absorptiometry.

### Body weight, BMI, waist circumference, and body composition

The 13-wk weight loss intervention resulted in a significantly decreased body weight and fat mass in the intervention and control groups [$-3.4 \pm 3.6$ kg and $-2.8 \pm 2.8$ kg (both $P < 0.001$) and $-3.2 \pm 3.1$ kg and $-2.5 \pm 2.4$ kg (both $P < 0.001$), respectively] without significant differences between the groups (**Table 4**). Waist circumference and BMI also decreased over time (both $P < 0.001$), with no significant differences between groups (Table 4).

### Muscle mass, muscle strength, and muscle function

After the 13-wk weight loss intervention, the change in appendicular muscle mass was different in the intervention compared with the control group [$+0.4 \pm 1.2$ kg and $-0.5 \pm 2.1$ kg, respectively; $\beta = 0.95$ kg (95% CI: 0.09, 1.81); $P = 0.03$] (**Figure 2**).

**TABLE 2**
Baseline characteristics of obese older subjects in the Muscle Preservation Study with both baseline and 13-wk measurement of the primary outcome variable, by treatment[1]

| Characteristic | Intervention group ($n = 30$) | Control group ($n = 30$) | $P$ value[2] |
|---|---|---|---|
| Male sex, $n$ (%) | 14 (47) | 14 (47) | 1.00 |
| Origin, % Caucasian | 90 | 87 | 1.00 |
| Age, y | 63.7 ± 6.0 | 63.0 ± 6.0 | 0.61 |
| Height, m | 1.71 ± 0.10 | 1.68 ± 0.07 | 0.15 |
| Body weight, kg | 95.9 ± 11.9 | 94.1 ± 14.2 | 0.60 |
| BMI, kg/m$^2$ | 32.7 ± 3.1 | 33.3 ± 4.3 | 0.54 |
| BMI <30 kg/m$^2$, $n$ (%) | 7 (23) | 6 (20) | 1.00 |
| BMI ≥30 kg/m$^2$, $n$ (%) | 23 (77) | 24 (80) | |
| Waist circumference, cm | 111 ± 10 | 110 ± 11 | 0.85 |
| Fat mass, % | 40.8 ± 7.4 | 41.4 ± 7.7 | 0.75 |
| Appendicular muscle mass, kg | 23.2 ± 4.9 | 22.6 ± 4.9 | 0.64 |
| Skeletal muscle index, kg/m$^2$ | 7.83 ± 1.18 | 7.92 ± 1.21 | 0.77 |
| Handgrip strength,[3] kg | 30.9 ± 9.8 | 29.6 ± 10.1 | 0.63 |
| 400-m walk speed,[3] m/s | 1.36 ± 0.19 | 1.33 ± 0.16 | 0.53 |
| Time to complete 5 stands, s | 15.9 ± 4.7 | 13.5 ± 3.7 | 0.04 |
| Gait speed, m/s | 1.12 ± 0.26 | 1.07 ± 0.21 | 0.37 |
| Current smoker,[3] $n$ (%) | 1 (3) | 4 (13) | 0.35 |
| Alcohol abstainers,[3] $n$ (%) | 8 (28) | 7 (23) | 0.77 |
| Alcohol consumption among users,[4] servings/d | 1.7 ± 1.1 | 1.5 ± 0.8 | 0.44 |

[1]Values are means ± SDs unless otherwise indicated.
[2]Significance level (2-sided $P$ value) for comparison between groups by using independent Student's $t$ test or Fisher's exact test (sex, origin, BMI group, current smoker, and alcohol abstainers).
[3]Intervention group, $n = 29$.
[4]Intervention group, $n = 21$; control group, $n = 23$.

**TABLE 3**
Dietary intake in intervention and control groups during intervention (including supplements)[1]

| | Intervention group ($n$ = 30) | Control group ($n$ = 32) | $P$ value[2] |
|---|---|---|---|
| Energy intake, kcal/d | 1823 ± 566 | 1662 ± 357 | 0.76 |
| Protein, g/d | 103 ± 29.0 | 75.4 ± 19.9 | <0.001 |
| Protein, g · kg BW$^{-1}$ · d$^{-1}$ | 1.11 ± 0.28 | 0.85 ± 0.24 | <0.001 |
| Protein, % of energy | 22.9 ± 3.4 | 18.3 ± 3.8 | <0.001 |
| Carbohydrate, % of energy | 42.0 ± 6.2 | 47.8 ± 5.0 | <0.001 |
| Fat, % of energy | 29.2 ± 4.0 | 29.3 ± 4.6 | 0.92 |

[1]Values are means ± SDs; intake data at week 13. BW, body weight.
[2]Significance level of differences between groups by using mixed linear models with covariates sex and baseline value.

No differences were observed in appendicular muscle mass for the intervention and control groups over time ($P$ = 0.15 and $P$ = 0.11, respectively). The 13-wk change in leg muscle mass was also different between the intervention and control groups [+0.3 ± 1.2 kg and −0.6 ± 1.8 kg, respectively; $\beta$ = 0.97 kg (95% CI: 0.24, 1.70); $P$ = 0.01]. Leg muscle mass was not different over time in the intervention group ($P$ = 0.08) and showed a trend for a decline in the control group ($P$ = 0.06) (Figure 2).

When appendicular muscle mass was adjusted for height, the skeletal muscle index still showed a significant change between the intervention and control groups [+0.1 ± 0.4 kg/m$^2$ and −0.2 ± 0.7 kg/m$^2$, respectively; $\beta$ = 0.30 kg/m$^2$ (95% CI: 0.01, 0.59); $P$ = 0.04]. Muscle strength and muscle function improved over time without differences between groups (Table 4).

## DISCUSSION

This trial is the first to show that use of a high whey protein–, leucine-, and vitamin D–enriched supplement preserves muscle mass during intentional weight loss by a hypocaloric diet combined with resistance exercise in obese older adults.

Weight loss treatment in older adults is still under discussion, due to the potential risk for permanent loss of muscle mass potentially affecting activities of daily life. Although data to support guidelines for weight loss treatment in older adults are limited, one of the main targets identified was the preservation of muscle mass by incorporating resistance exercise and increased protein consumption (1). At present, the Recommended Dietary Allowance for protein is 0.8 g/kg for all adults (25). Current expert opinion on protein requirements in the older adult or elderly population ranges from 1.0–1.2 g protein · kg BW$^{-1}$ · d$^{-1}$ (26). This implies that the intake of 0.8 g · kg BW$^{-1}$ · d$^{-1}$ during a hypocaloric diet is too low for maintenance of body protein mass (27). For overweight adults, it has been shown that preservation of fat-free mass was more effective with a high-protein diet (1.2 g · kg BW$^{-1}$ · d$^{-1}$) compared with a normal-protein diet (0.8 g · kg BW$^{-1}$ · d$^{-1}$) (28). A recent guideline for the treatment of obese elderly suggests that ingestion of 1.0 g · kg BW$^{-1}$ · d$^{-1}$ high-quality protein strategically timed at meals during a hypocaloric diet might be an approach to prevent major loss of muscle mass (1). We show preservation of skeletal muscle mass in obese older adults with an intake of 1.11 g protein · kg BW$^{-1}$ · d$^{-1}$, thus supporting the recommendation described in this guideline.

Besides the total amount of protein intake per day, the amount of protein in 1 meal, as well as the quality of the protein in the meal, seems relevant for muscle protein synthesis (8) and might explain our findings on muscle preservation in the intervention group. Several recent studies indicate that older adults are muscle anabolic resistant, which implies a blunted postprandial response to the anabolic stimuli from protein or amino acids compared with young adults (13, 29). However, providing older adults with a sufficient amount of protein or amino acid equivalent could still stimulate muscle protein synthesis (29, 30). Breen and Phillips (29) showed that the ingestion of at least 20 g protein at once leads to a significant increase of muscle protein synthesis in older adults. In addition, protein quality has major effects on the efficacy to stimulate muscle protein synthesis. It has been shown that 20 g whey protein is more effective in stimulating postprandial muscle protein accretion than casein, casein hydrolysate, or soy protein in older men (14, 15). The whey-stimulating effects on

**TABLE 4**
Outcome measures for intervention and control groups with intervention effect[1]

| | Intervention group | | | Control group | | | Intervention effect | |
|---|---|---|---|---|---|---|---|---|
| | Baseline ($n$) | Change ($n$) | $P$ value | Baseline ($n$) | Change ($n$) | $P$ value | $\beta$ (95% CI) | $P$ value |
| Body weight, kg | 96.7 ± 11.9 (32) | −3.4 ± 3.6 (32) | <0.001[2] | 93.2 ± 14.6 (33) | −2.8 ± 2.8 (33) | <0.001[2] | −0.37 (−1.68, 0.94)[3] | 0.57[4] |
| BMI, kg/m$^2$ | 32.8 ± 3.1 (32) | −1.2 ± 1.3 (32) | <0.001[2] | 33.1 ± 4.3 (33) | −1.0 ± 0.9 (33) | <0.001[2] | −0.16 (−0.61, 0.29)[3] | 0.49[4] |
| Waist circumference, cm | 111 ± 9.8 (32) | −4.4 ± 4.0 (32) | <0.001[2] | 109 ± 11 (33) | −3.7 ± 5.1 (33) | <0.001[2] | −0.69 (−2.72, 1.34)[3] | 0.50[4] |
| Fat mass, kg | 38.6 ± 7.6 (30) | −3.2 ± 3.1 (30) | <0.001[5] | 38.5 ± 9.3 (30) | −2.5 ± 2.4 (30) | <0.001[5] | −0.70 (−2.09, 0.69)[6] | 0.32[7] |
| Fat percentage | 40.8 ± 7.4 (30) | −2.3 ± 2.3 (30) | <0.001[5] | 41.4 ± 7.7 (30) | −1.6 ± 1.9 (30) | <0.001[5] | −0.62 (−1.64, 0.40)[6] | 0.23[7] |
| Handgrip strength, kg | 31.3 ± 9.9 (31) | 2.0 ± 4.6 (31) | <0.001[2] | 29.1 ± 10.1 (32) | 2.2 ± 4.1 (32) | <0.001[2] | −0.01 (−1.7, 1.68)[3] | 0.99[4] |
| 4-m gait speed, m/s | 1.12 ± 0.26 (29) | 0.11 ± 0.25 (29) | 0.003[2] | 1.04 ± 0.22 (32) | 0.11 ± 0.21 (32) | 0.007[2] | 0.02 (−0.09, 0,12)[3] | 0.77[4] |
| 400-m walk speed, m/s | 1.37 ± 0.18 (27) | 0.04 ± 0.1 (27) | 0.007[2] | 1.33 ± 0.14 (31) | 0.05 ± 0.11 (31) | 0.002[2] | −0.004 (−0.057, 0.049)[3] | 0.89[4] |
| Chair stand, s | 15.9 ± 4.7 (31) | −2.4 ± 4.0 (31) | <0.001[2] | 13.6 ± 3.8 (32) | −1.4 ± 3.1 (32) | <0.001[2] | 0.21 (−1.21, 1.64)[3] | 0.76[4] |

[1] Values are means ± SDs.
[2]Significance level of estimate of change at week 13 by using mixed linear models with covariates sex and baseline value.
[3]Estimate of intervention effect at week 13 by using mixed linear models with covariates sex and baseline value.
[4]Significance level of estimate of group difference at week 13 by using mixed linear models with covariates sex and baseline value.
[5]Significance level of estimate of change at week 13 by using a paired $t$ test.
[6]Estimate of intervention effect at week 13 by using ANCOVA with sex and baseline value as covariates.
[7]Significance level of estimate of group difference at week 13 by using ANCOVA with sex and baseline value as covariates.

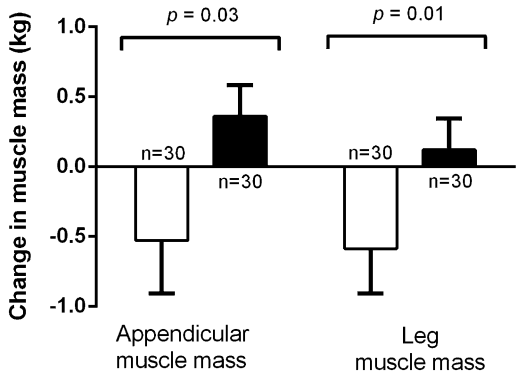


**FIGURE 2** Change in appendicular muscle mass in intervention and control groups. Data represent mean changes over 13 wk with SEM. Intervention effect and significance level are based on ANCOVA with covariates sex and baseline value. White bars represent the control group; black bars represent the intervention group.

muscle protein synthesis have been ascribed to its fast digestion, delivering amino acids in the circulation available for protein synthesis (31) and its high content of leucine, which is considered the most potent amino acid to stimulate muscle protein synthesis (32). The effect of leucine was corroborated by Wall et al. (16), showing that leucine coingestion with protein could further improve muscle protein synthesis in older adults. In this study, we therefore used a high whey protein–, leucine-enriched supplement to increase daily protein intake. The supplement was hypothesized to stimulate muscle protein synthesis in the older adult, which could tip the balance toward preservation of muscle mass compared with the usual loss of muscle mass during intentional weight loss (33).

The intervention supplement used in this study also contained 800 IU vitamin D. A low vitamin D status has been associated with impaired muscle mass and function in older adults (34), and vitamin D has also been suggested to have a positive impact on muscle protein metabolism (20, 21). Supplementing with vitamin D might therefore facilitate muscle mass preservation. However, the mechanism by which vitamin D positively affects muscle protein synthesis is not yet fully elucidated. The control supplement used in our study was matched for calories and not for specific nutrients, meaning that the observed effects should be attributed to the entire supplement, and effects of individual subcomponents cannot be determined.

Resistance exercise is a well-known facilitator that sensitizes the muscle, stimulates muscle protein synthesis, and promotes muscle hypertrophy in the older adult when performed frequently over time. Therefore, the combination of protein ingestion and resistance training enhancing muscle protein synthesis would be ideal to attenuate the loss of muscle mass (8, 35). Although in a different target group, a study with protein supplementation during a 24-wk progressive resistance exercise program in (pre) frail elderly indeed significantly increased lean mass compared with a control group (30).

Taken together, there appears to be sufficient support to emphasize additional high-quality protein supplementation in combination with resistance exercise during a weight loss program to preserve muscle mass in older adults.

A limitation of this study was the high number of subjects (25%) not available for the analysis of the primary outcome, which could bias the results compared with an intention-to-treat analysis. Baseline characteristics of the dropouts were comparable to those subjects included in the final analysis, and the dropout rate was equal in both groups. It is unknown to what extent this has influenced our findings.

Although the participants lost weight, the magnitude was below what we expected. We advised a 600-kcal/d reduction in energy intake, which was not achieved based on the analyses of the 3-d food records. In addition, the accuracy of the 3-d food record in this study seems poor, because we observed large differences between baseline estimated energy need and baseline 3-d food records, which is not unknown and has been reported earlier (36). Our findings show that it is very difficult to reach and track −600-kcal/d restriction in this target group. Dietary adherence seems strongly dependent on the counseling time with the dietitian or the research setting available. Of several previous successful weight loss trials in overweight older adults (9, 37–41), 5 had weekly group sessions with a dietitian, and in 1 trial, all meals were provided. In our study, the subjects visited the dietitian only biweekly, which may have resulted in the limited weight loss observed.

Despite a muscle-preserving effect of the supplement, we did not observe differences between groups in muscle strength and physical performance. Overall, parameters for physical performance improved in both groups. Consistent with our findings, Tieland et al. (30) showed in their randomized controlled trial that protein supplementation in (pre)frail elderly increased muscle mass during resistance-type exercise without increasing physical functioning. Generally, during the first months of a resistance training program, a steep increase in muscle strength is seen as a result of improvements in neuromuscular activation and increases in muscle quality (42, 43). Furthermore, a study by Villareal et al. (37)—a 1-y randomized controlled trial, in which the independent and combined effects of weight loss and exercise were studied in obese older adults—showed that physical performance of older obese adults significantly improved in the weight-loss group (without exercise training), losing 9.7 kg over 1 y, even though lean body mass was lost (3.2 kg). The interaction between weight loss and exercise training provided the largest improvement in physical functioning. The potential effect of preserved muscle mass attributable to the high whey–, leucine-, and vitamin D–enriched supplement on physical function might therefore be masked by the effect of training and weight loss. We speculate that the effect of preserved skeletal muscle mass will likely contribute to improve strength and functioning as time progresses.

In conclusion, a high whey protein–, leucine-, and vitamin D–enriched supplement compared with an isocaloric control supplement as part of an intentional weight loss program, including a hypocaloric diet and resistance exercise, preserves skeletal muscle mass in obese older adults. These findings support the current advice to increase protein intake of high quality and sufficient quantity during a weight loss program in obese older adults to aid in the prevention of weight loss–induced sarcopenia.

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