Artigo original

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EFFECTS OF PROTEIN SUPPLEMENTATION ON FAT-FREE MASS IN RESPONSE TO DIFFERENT WEIGHT LOSS PROGRAMS IN OBESE WOMEN

EFEITOS DA SUPLEMENTAÇÃO PROTEICA NA PRESERVAÇÃO DA MASSA MAGRA EM MULHERES OBESAS SUBMETIDAS A DIFERENTES PROGRAMAS DE EMAGRECIMENTO

RESUMO

O objetivo deste estudo foi investigar a influência da suplementação protéica na preservação da massa magra durante programas de emagrecimento. Setenta e oito mulheres adultas japonesas e obesas foram submetidas a quatro programas diferentes: dieta (D, n=24), dieta com suplementação proteica (DP, n=16), dieta com exercício físico (DE, n=17) e dieta com exercício físico e suplementação protéica (DEP, n=21). A dieta restringiu-se a 1200 kcal/dia e o programa de exercício físico compreendeu atividade aeróbica durante 90 minutos/dia, 3 dias/semana. Os participantes em suplementação proteica receberam 14 gramas/dia de proteína. A avaliação da composição corporal foi conduzida antes e depois de 14 semanas utilizando DXA. Todos os programas resultaram significante redução de massa corporal (6,9 a 9,5kg) e gordura (4,1 a 7,6%), sendo que a massa magra total diminuiu significativamente nos grupos D, DP e DE. No entanto, a redução não foi significativa para o grupo DEP. A massa magra localizada nas pernas, braços e tronco sofreu redução significativa nos grupos D e DP. Para o grupo DE, a massa magra do tronco não reduziu significativamente, assim como para o grupo DEP, a massa magra das pernas e do tronco não alterou significativamente. Entretanto, não foram observadas diferenças significantes na comparação entre os grupos com relação às mudanças obtidas na massa magra. Os resultados confirmam a eficiência da dieta e exercício físico na redução de gordura corporal. Entretanto, este estudo sugere que a suplementação proteica durante programas de emagrecimento de curta duração não se associa à preservação de massa magra.

Palavras-chave: obesidade, emagrecimento, suplementação protéica, exercício aeróbico, dieta, mulheres japonesas

ABSTRACT

The aim of this study was to investigate whether protein supplementation helps prevent the loss of fat-free mass during weight loss. The sample was composed of seventy-eight obese adult Japanese women, assigned into four different programs: diet-alone (D, n=24), diet-alone with protein supplementation (DP, n=16), diet-plus-exercise (DE, n=17), and diet-plus-exercise with protein supplementation (DEP, n=21). All participants restricted their energy intakes to 1200 kcal/day, and participants in DE and DEP had the exercise session including aerobic exercise of approximately 90 min/day, 3 day/week. Participants enrolled in protein supplementation groups received an additional 14 g/day of protein. Measures on body composition were conducted before and after the program by DXA. All programs yielded significant weight (6.9 to 9.5 kg) and fat (4.1 to 7.6%) reduction. Total fat-free mass significantly decreased in D, DP and DE groups, whereas for DEP group the decrease was not significant. Regional fat-free mass lowered for D and DP groups in leg, arms and trunk. For those in DE group, fat-free mass in trunk was not significantly decreased, and for those in DEP group, fat-free mass in leg and trunk did not differ significantly after the program. However, no significant differences of changes in fat-free mass were observed in comparisons among all groups. Our results confirmed the efficiency of weight loss intervention on fat-mass reduction through diet and exercise. However, fat-free mass does not appear to be preserved by protein supplementation, suggesting that its influence on a short-term weight reduction program is not apparent.

Key words: obesity, weight loss, protein supplementation, aerobic exercise, diet, Japanese women

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INTRODUCTION

Obesity is a condition that represents a serious worldwide threat to public health. Japanese society faces low prevalence of obesity (only 2% to 3% of the women[1]) by the World Health Organization standards[2] of body mass index (BMI) ≥ 30 kg/m^2, while for instance, ranges from 10% to 25% are reported for women in the United States[3] and 12.7% in Brazilian women from the Northeast and Southeast regions[4]. However, the prevalence of obesity-related disorders such as hypertension, dyslipidaemia, and type 2 diabetes remains high for Japanese, even with mild obesity[5]. On the basis of these studies, a distinctive definition of obesity of BMI ≥ 25 kg/m^2 has been accepted as the target for treatment among Japanese men and women[1]. In fact, obesity has become a serious concern in Japan, particularly considering the rapidly transforming lifestyles of many Japanese. Current forecasts project a higher prevalence of obesity in the future, which will be associated with serious consequence for people's health[6].

Recommendations for obesity control focus on energy balance by lifestyle therapies designed to reduce daily energy intake and increase physical activity[7,8,9]. Our previous study reported that a 1200-kcal/day diet produces significant weight reduction in obese Japanese women[10]. For successful weight reduction it is also recommended to include physical exercise because of its cumulative effects on energy expenditure over a long period of time[11]. Exercise can also contribute to the suppression of appetite prior to a meal, and it can promote health status independently of weight reduction, improve psychological functioning, minimize the loss of fat-free mass, and counteract some of the adverse metabolic consequences associated with dieting alone[12].

The foremost objective of a weight loss trial has to be the reduction in body fat. However, a concomitant decline in fat-free mass is frequently observed[13,14]. Fat-free mass is strongly influenced by change in body weight and has been closely linked with reduction in muscle strength. A longitudinal study reported that people who are able to maintain their weight naturally lose on average 1.5 kg of fat-free mass per decade, but those under weight reduction programs tend to have greater losses of fat-free mass[15]. The reduction in muscular strength is associated with increased risk of falls and hip fracture[16], and has been linked to higher rates of all-cause mortality in women in the United States[17]. Thus, losses in fat-free mass with advancing age have important health implications.

Protein is the predominant component of skeletal muscle, and dietary protein is necessary to synthesize skeletal muscle protein. Recently, attention has been directed toward the effects of protein on muscle metabolism and performance in people undergoing dietary restrictions[18,19]. By choosing appropriate foods it is possible to achieve high protein intakes, but it is also true that many high protein foods have a high fat content, which would counteract the weight loss intervention[20]. Therefore, protein supplementation is one alternative to consuming high amounts of dietary protein, and this is a strategy commonly used among people engaged in sport activities. Safety concerns regarding the appropriate application of protein supplementation and its efficacy for people undergoing a weight reduction program is still unclear[21,22]. Thus, the aim of this study was to determine whether 14 g/day of protein supplementation leads to additional improvements, preventing the loss of fat-free mass after weight loss intervention among obese Japanese women.

METHODS

Participants

Japanese women were voluntarily recruited through advertisements in Toride city, located 50 km north of Tokyo (Japan), and 83 participants, 25 to 50 years old, were selected after they met the following criteria: sedentary (engaged in exercise < 60 min/wk in the previous 6 months) and BMI ≥ 25 kg/m^2. These criteria were set according to the guidelines for obesity in the Japanese Society for the study of Obesity[1]. As determined by medical history and physical examination, none of the participants smoked or had concomitant renal, hepatic, or cardiac disease. They were all pre-menopausal and not under hormone replacement therapy. Five participants were unable to successfully complete the study. Therefore, seventy-eight women were partly randomly assigned and partly included according to their preferences into one of four weight reduction programs: diet-alone (D, n = 24), diet-alone with protein supplementation (DP, n = 16), diet-plus-exercise (DE, n = 17), and diet-plus-exercise with protein supplementation (DEP, n = 21). Exercise and diet classes were given independently among the four groups in a hospital-based fitness center. The aim and design of the study were explained to each participant and a statement of informed consent was obtained from each prior to initiation of the study. The Human Investigation Review Committee at Tsukuba Health Fitness Kenkyukai, Higashi Toride Hospital, approved the overall study protocol.

Dietary restriction

The participants were instructed to drink daily a supplemental food product (a powder dissolved in 500 ml of water; MicroDiet; SunnyHealth, Nagano, Japan) developed for very-low-energy diets (50 g per pack of 173 kcal), comprised of protein (21.0 g), carbohydrates (16.7 g), fat (2.5 g), and other 11.6 g of various amino acids, and vitamins and minerals. Two other meals were allowed per day, consisting of 240 kcal of protein, 480 kcal of carbohydrates, and 240 kcal of fat. To ensure proper and controlled daily nutrition, the subjects listened to lectures and were counseled by dieticians every week. They also kept food diaries for 1 week before the weight-loss programs and every day during the 14-week intervention period. They were asked to record the brand name and amounts (in grams) of every food and beverage ingested with each meal. To calculate energy intake (in kilocalories) and the amounts of each nutrient (fat, protein, and carbohydrate), data from 6 days (3 days: 2 weekdays and either Saturday or Sunday before the intervention period; and 3 days during the intervention period) were also collected. Skilled dieticians analyzed the data.

Exercise program

In addition to the restricted energy intake, participants from the diet-plus-exercise groups were engaged into a walking program, consisting of a 90-minute session, 3 days a week, at level that raised their heart rate corresponding to their lactate threshold. The target Borg's scale[23] (rating of perceived exertion) ranged from 13 (moderately hard) to 15 (hard). Exercise sessions were supervised by three physical educators once a week (on Saturdays). On the other two days, participants were asked to exercise for 90 minutes, at their most convenient time, measuring their heart rate by palpitation while walking and recording the intensity (heart rate or Borg's scale) of the walking during each session. The supervised program consisted of three phases: 1) 15-minute callisthenic warm-up, 2) 60-minute continuous walking, and 3) 15-minute cool-down including stretching and massage.

Protein Supplementation

Orientation of a daily consumption of one package of FeedBack WheyProtein (Meiji Dairies, Tokyo, Japan) in 200 ml of low-fat milk (118 kcal) was given to the participants under protein supplementation protocol. Each package contains 14 g of protein, and additional 2.5 g of carbohydrates, amino acids, fat, and vitamins and minerals (64 kcal). In total, participants under supplementation consumed 182 kcal more than groups without supplementation. Those enrolled in the diet-plus-exercise group were asked to drink a protein supplement immediately after exercise. This product was chosen for representing a commercially marketed product in Japan, available for the average population. Also, a similar amount of protein supplementation (between 10 and 15 g) has been previously investigated in the sports literature[22,24,25].

Anthropometric variables

Body weight was measured to the nearest 0.01 kg using a digital scale (TBF-551, Tanita, Tokyo, Japan), height was measured to the nearest 0.1 cm using a stadiometer (YG-200, Yagami, Nagoya, Japan), and BMI was calculated as weight (kilograms) divided by height squared (meters squared). Circumference of the waist was measured at the level of the umbilicus to the nearest 0.1 cm with the subjects in standing position.

Body composition by dual energy x-ray absorptiometry (DXA)

Whole body fat mass, percentage of fat, fat-free mass, and bone mass were assessed by DXA (DPX-L; Lunar, Madison, WI). Transverse scans were used for the whole-body measurement, and pixels of soft tissue were used to calculate the ratio (*R* value) of mass attenuation coefficients at 40 to 50 keV (low energy) and 80 to 100 keV (high energy), using software version 1.3Z.

Statistical analysis

Baseline difference across groups was assessed by one-way analysis of variance (ANOVA), and difference between pairs of groups by independent *t* test. Changes occurring in response to treatments (baseline *vs.* week 14) within-group were evaluated by paired Student's *t* test. Two-way ANOVA was used in order to investigate differences of changes among groups, and post-hoc comparisons (Bonferoni test) was applied by pairs of groups when significance was found. All statistical analyses were performed using SPSS 13.0 for Windows and statistical significance was set at $P < 0.05$.

RESULTS

Table 1 presents the values of baseline and week 14 of the participants grouped into the four programs. No significant difference was observed in age, weight, BMI, waist circumference and fat-free mass; whereas fat mass and percentage of fat was significantly lower among groups under protein supplementation. Average BMI values ranged among those under diet-alone groups from 28.1 kg/m^2 (DP) to 29.6 kg/m^2 (D); and among those under diet-plus-exercise from 27.9 kg/m^2 (DEP) to 29.2 kg/m^2 (DE). Attendance at the exercise programs (14 sessions) averaged 94% (range, 85% to 100%) for the diet-plus-exercise groups.

Table 2 presents absolute changes in body composition by groups. Along with all programs showed significant weight reduction despite the protein supplementation and exercise intervention. The range of weight loss was on average of 6.9 kg (the lowest, in DP) to 9.5 kg (the highest, in DEP). BMI decreased an average of 11.8% after week 14 (2.8 to 3.8 kg/m^2). Significant losses were observed in fat mass among all programs, from 4.8 kg (in DP) to 8.0 kg (in DE). A similar proportion of loss was observed in fat percentage. No significant differences were observed in bone mass among all groups.

As shown in Figure 1, after weight loss, total fat-free mass, and fat-free mass from arms, legs, trunk decreased significantly among diet-alone groups, regardless of protein supplementation. Among diet-plus-exercise, the protein supplemented group (DEP) showed a significant decrease of fat-free mass only in arms (-0.3 kg), whereas participants in DE group decreased in arms (-0.1 kg), trunk (-0.9) and total fat-free mass (-0.8). The overall analysis comparing changes among all groups (Table 2) showed higher decrease in fat mass among DE group compared to those in diet-alone (D group). However, no significant difference was observed among fat-free mass parameters in the comparison among all groups.

Table 1. Descriptive characteristics of the study participants at baseline and week 14.

			DP	D	DEP	DE	p Value†
Participant (n)			16	24	21	17	
Age (year)			39.8 ± 6.5	42.3 ± 4.1	42.1 ± 6.4	44.9 ± 5.9	NS‡
Wt (kg)		*Baseline*	68.6 ± 6.8	72.3 ± 9.1	68.1 ± 8.1	71.8 ± 6.7	NS
		Week 14	61.8 ± 5.7	63.7 ± 9.2	58.6 ± 8.3	62.4 ± 8.1	
BMI (kg/m^2)		*Baseline*	28.1 ± 2.4	29.6 ± 3.7	27.9 ± 2.2	29.2 ± 1.7	NS
		Week 14	25.3 ± 1.9	26.2 ± 3.6	24.3 ± 3.3	25.4 ± 2.6	
WC (cm)		*Baseline*	98.3 ± 3.9	97.4 ± 7.9	96.7 ± 9.5	97.0 ± 5.5	NS
		Week 14	91.4 ± 3.7	89.1 ± 9.1	86.7 ± 8.6	86.8 ± 8.4	
FM (kg)		*Baseline*	23.2 ± 5.1§¶	27.5 ± 4.7	23.5 ± 4.8§¶	28.2 ± 4.4	0.002
		Week 14	18.4 ± 3.9	20.7 ± 5.9	17.3 ± 5.5	20.2 ± 5.4	
% fat (%)		*Baseline*	33.9 ± 5.0§¶	38.5 ± 3.1	35.2 ± 4.2¶	39.9 ± 4.2	< 0.001
		Week 14	29.9 ± 5.2	32.1 ± 5.4	28.7 ± 6.3	32.3 ± 5.3	
BM (kg)		*Baseline*	2.43 ± 0.27	2.54 ± 0.25	2.47 ± 0.25	2.43 ± 0.30	NS
		Week 14	2.42 ± 0.28	2.52 ± 0.25	2.42 ± 0.25	2.41 ± 0.30	
FFM (kg)	Total	*Baseline*	42.3 ± 4.4	41.1 ± 4.8	40.3 ± 3.6	40.0 ± 5.0	NS
		Week 14	40.5 ± 4.2	40.1 ± 4.5	39.5 ± 3.7	39.2 ± 5.2	
	Arms	*Baseline*	4.1 ± 0.5	4.2 ± 0.6	3.9 ± 0.5	4.1 ± 0.6	NS
		Week 14	3.8 ± 0.5	4.1 ± 0.5	3.6 ± 0.5	4.0 ± 0.6	
	Legs	*Baseline*	14.2 ± 1.6	13.8 ± 1.7	13.8 ± 1.5	13.4 ± 2.0	NS
		Week 14	13.6 ± 1.6	13.5 ± 1.5	13.5 ± 1.3	13.5 ± 2.0	
	Trunk	*Baseline*	20.9 ± 2.3	20.0 ± 2.7	19.6 ± 1.8	19.6 ± 2.4	NS
		Week 14	20.0 ± 1.9	19.4 ± 2.5	19.3 ± 1.9	18.7 ± 2.5	

Values are presented as mean ± SD. DP, diet-alone with protein supplementation; D, diet-alone; DEP, diet-plus-exercise with protein supplementation; DE, diet-plus-exercise. †Baseline difference across groups (ANOVA one-way). ‡NS, nonsignificant difference ($p \geq 0.05$). §Differ from D, ¶differ from DE. Wt, weight; BMI, body mass index; WC, waist circumference; FM, fat mass; % fat, percentage of fat; BM, bone mass; FFM, fat-free mass.

Table 2. Changes in body composition in response to weight loss.

	DP		D		DEP		DE		p Value*
Wt (kg)	-6.9 ± 1.1	< 0.01†	-8.5 ± 0.1	< 0.01	-9.5 ± 0.2	< 0.01	-9.4 ± 1.4	< 0.01	NS‡
BMI (kg/m^2)	-2.8 ± 0.5	< 0.01	-3.4 ± 0.0	< 0.01	-3.6 ± 1.1	< 0.01	-3.8 ± 0.9	< 0.01	NS
WC (cm)	-7.0 ± 0.2	< 0.01	-8.4 ± 1.2	< 0.01	-10.0 ± 0.9	< 0.01	-10.2 ± 2.9	< 0.01	NS
FM (kg)	-4.8 ± 1.2	< 0.01	-6.8 ± 1.2	< 0.01	-6.2 ± 0.7	< 0.01	-8.0 ± 1.0	< 0.01	DE > DP
% fat (%)	-4.1 ± 0.0	< 0.01	-6.4 ± 2.3	< 0.01	-6.4 ± 2.1	< 0.01	-7.6 ± 1.2	< 0.01	NS
BM (kg)	-0.01 ± 0.02	NS	-0.02 ± 0.00	NS	-0.05 ± 0.00	NS	-0.02 ± 0.00	NS	NS
FFM (kg)									
Total	-1.8 ± 0.3	< 0.01	-1.0 ± 0.3	< 0.01	-0.8 ± 0.1	NS	-0.8 ± 0.2	< 0.01	NS
Arms	-0.3 ± 0.0	< 0.01	-0.2 ± 0.1	0.03	-0.3 ± 0.0	< 0.01	-0.1 ± 0.0	0.03	NS
Legs	-0.6 ± 0.0	< 0.01	-0.3 ± 0.2	0.01	-0.3 ± 0.2	NS	0.1 ± 0.0	NS	NS
Trunk	-0.9 ± 0.4	< 0.01	-0.6 ± 0.2	0.01	-0.2 ± 0.1	NS	-0.9 ± 0.0	< 0.01	NS

Values are presented as mean ± SD. *Differences of changes in values across all groups (ANOVA two-way). †Within-group change between baseline and week 14 (paired *t* test). ‡NS, nonsignificant difference ($p \geq 0.05$). DP, diet-alone with protein supplementation; D, diet-alone; DEP, diet-plus-exercise with protein supplementation; DE, diet-plus-exercise. Wt, weight; BMI, body mass index; WC, waist circumference; FM, fat mass; % fat, percentage of fat; BM, bone mass; FFM, fat-free mass.

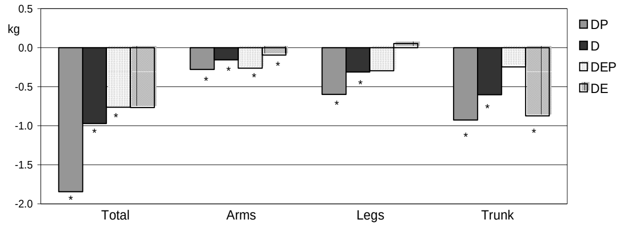

Figure 1. Fat-free mass change in response to weight loss. Significant difference between baseline and week 14 ($p < 0.05$).

DISCUSSION

Participants in this study successfully complied with the weight reduction programs, whether diet alone or combined with exercise, and with or without protein supplementation. Magnitude of obesity was significantly lower after weight loss, and 41 participants (52.6%) were no longer classified as obese (BMI < 25) at the end of the 14-week program. The decrease in weight resulted primarily from reduction of fat mass, which is consistent with many other studies of short-term weight loss programs[26,27].

After the weight loss, a significant decrease in fat-free mass was observed among participants under diet-alone program (D), whereas in combination with exercise fat-free mass did not decrease significantly in legs (DE and DEP groups) and trunk (DEP group). In fact, the slight advantage in preserving fat-free mass observed in groups under exercise program (DE, DEP) seems to be more associated with exercise than with protein supplementation. The small change in fat-free mass observed in legs among both exercise groups (DE, DEP) after 14 weeks appears to be related to the fact that the exercise program (walking) stimulates predominantly the lower body. Considering the fact that a comparison between groups with and without protein supplementation did not differ significantly in any fat-free mass item measured, our findings suggest that exercise was more efficient for preserving fat-free mass regardless of protein supplementation. Hence, our study supports the recommendation of including aerobic exercise during weight loss intervention.

A meta-analysis on exercise and fat-free mass preservation during weight loss reported that exercise reduces the percentage of weight lost as fat-free mass[28]. According to the author, low intensity exercise is appropriate for weight loss intervention also because it can generally be done daily. This, added to other convenient factors related to walking (e.g. easy access, low cost), might lead to positive behavior changes among the participants in the present study, even after the end of the intervention.

Previous investigations[29] have reported that, due to the negative energy balance, fat-free mass does not increase with inclusion of aerobic exercise intervention during a weight loss program. Indeed, most studies reported lower levels of fat-free mass after the intervention[13,14,27], which was consistent with the present study. Doi et al.[19] reported a weight reduction program by a combination of diet and light resistance exercise with and without protein supplement. The fat-free mass was significantly decreased in the control group, whereas its decrease in the supplemented group was not significant. This finding suggests that even supplementation of protein combined with anaerobic exercise program does not increase the fat-free mass during weight loss. In the study by Layman et al.[18] two different diets (high protein, low carbohydrate vs. low protein, high carbohydrate) were tested in adult women. The results demonstrated that a diet with higher protein and low carbohydrate maintained fat-free mass after a 4-month weight loss program.

The observations from this study are subject to limitations. Difficulties in defining the most appropriate amount of protein intake during a dietary restriction program was our worry, as this problem has been reported previously[30]. The 14 g protein supplement used in the present study may not be enough to stimulate muscle protein synthesis. Moreover, participants were not completely randomly assigned into the different intervention groups. In this case, direct comparison of measures between groups might partly preclude our definitive conclusions. Another limitation was that exercise mode was not directly controlled (i.e., intensity and frequency). Lastly, precise control of diet and home exercise is difficult in a study employing free-living subjects.

CONCLUSION

The present study was designed to investigate whether protein supplementation leads to additional benefits on body composition profile among pre-menopausal obese Japanese women participating in diet- and exercise-based weight reduction programs. The weight loss intervention confirmed the efficiency of diet and exercise on fat-mass reduction. However, our findings suggest that the influence of protein supplementation on fat-free mass during short-term weight reduction program is not apparent. Further investigation is needed to clarify the role of protein supplementation in weight loss interventions.

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