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Original

Effect of resistance training and hypocaloric diets with different protein content on body composition and lipid profile in hypercholesterolemic obese women

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Abstract

Lifestyle changes such as following a hypocaloric diet and regular physical exercise are recognized as effective non-pharmacological interventions to reduce body fat mass and prevent cardiovascular disease risk factors.

Purpose: **To evaluate the interactions of a higher protein (HP) *vs.* a lower protein (LP) diet with or without a concomitant progressive resistance training program (RT) on body composition and lipoprotein profile in hypercholesterolemic obese women.**

Methods: **Retrospective study derived from a 16-week randomized controlled-intervention clinical trial. Twenty-five sedentary, obese (BMI: 30-40 kg/m²) women, aged 40-60 with hypercholesterolemia were assigned to a 4-arm trial using a 2 x 2 factorial design (Diet x Exercise). Prescribed diets had the same calorie restriction (-500 kcal/day), and were categorized according to protein content as: lower protein (< 22% daily energy intake, LP) *vs.* higher protein (≥ 22% daily energy intake, HP). Exercise comparisons involved habitual activity (control) *vs.* a 16-week supervised whole-body resistance training program (RT), two sessions/wk.**

Results: **A significant decrease in weight and waist circumference was observed in all groups. A significant decrease in LDL-C and Total-Cholesterol levels was observed only when a LP diet was combined with a RT program, the RT being the most determining factor. Interestingly, an interaction between diet and exercise was found concerning LDL-C values.**

Conclusion: **In this study, resistance training plays a key role in improving LDL-C and Total-Cholesterol; however, a lower protein intake (< 22% of daily energy intake as proteins) was found to achieve a significantly greater reduction in LDL-C.**

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EFECTO DE UN ENTRENAMIENTO DE FUERZA Y DIETA HIPOCALÓRICA CON DIFERENTE APORTE PROTEICO SOBRE LA COMPOSICIÓN CORPORAL Y EL PERFIL LIPÍDICO EN MUJERES OBESAS CON HIPERCOLESTEROLEMIA

Resumen

Cambios en el estilo de vida como el seguimiento de dieta hipocalórica y práctica de ejercicio físico regular, son reconocidos como intervenciones no farmacológicas efectivas para reducir la masa grasa y prevenir enfermedades cardiovasculares.

Objetivo: **Evaluar la interacción de dietas con mayor aporte proteico (HP) *vs.* menor aporte de proteínas (LP) con o sin un programa de entrenamiento de fuerza (RT) sobre la composición corporal, y el perfil lipídico en mujeres obesas con hipercolesterolemia.**

Metodología: **Estudio retrospectivo derivado de un ensayo clínico controlado, aleatorizado de 16 semanas de intervención. 25 mujeres de entre 40-60 años, sedentarias, obesas (IMC: 30-40 kg/m²) y con hipercolesterolemia, fueron asignadas a 4 grupos, diseño factorial 2 x 2 (Dieta x Ejercicio). Las dietas, presentaban la misma restricción calórica (-500 kcal/day), y fueron categorizadas de acuerdo a su contenido proteico como: más bajas en proteínas (LP, < 22% del valor energético total) *vs.* más altas en proteínas (HP, ≥ 22% del valor energético total). La comparación del ejercicio incluyó la actividad habitual (control) *vs.* 2 sesiones/sem de entrenamiento de fuerza supervisado, durante 16 semanas.**

Resultados: **Se observaron pérdidas significativas de peso y de circunferencia de la cintura en todos los grupos. Disminución significativa de los niveles de LDL-C y colesterol total cuando la dieta LP era combinada con RT, siendo el RT el factor determinante. Se encontró una interacción entre dieta y ejercicio, en relación a los valores de LDL-C.**

Conclusión: **En este estudio, el ejercicio de fuerza juega un papel importante en la reducción de los niveles de LDL-C y Colesterol total; sin embargo, una menor ingesta de proteínas (< 22% del valor energético total) puede favorecer mayor reducciones de LDL-C.**

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Palabras clave: *Dieta. Proteínas. Obesidad. Metabolismo lipídico. Ejercicio de fuerza.*

Abbreviations

LDL-C: Low-density lipoprotein cholesterol.
RT: Resistance training.
HP: Higher protein.
LP: Lower protein.
BMI: Body mass index.
MRI: Magnetic resonance imaging.
RM: Repetition concentric maximum.
SD: Standard deviation.
SAT: Subcutaneous adipose tissue.
VAT: Visceral adipose tissue.
HDL-C: High-density lipoprotein cholesterol.
TC: Total cholesterol.
TG: Triglycerides.

Introduction

Lifestyle changes such as following a hypocaloric diet and regular physical exercise are recognized as effective non-pharmacological interventions to reduce body fat mass and prevent cardiovascular disease risk factors, e.g. hypercholesterolemia.[1,2] Moreover, the protein content in a hypocaloric diet has been associated with a decrease in triglyceride and low-density lipoprotein cholesterol (LDL-C) levels[3,4] and a fat-free mass retention during energy restriction,[5] minimizing lean tissue loss during a hypocaloric diet[6]. However, at present it is difficult to make a general standard recommendation concerning the most appropriate protein content in the daily menu to achieve an optimum response in weight loss and lipid profile.[5,7,8]

On the other hand, resistance exercise, with or without a concomitant hypocaloric diet, is gaining acceptance as a useful tool in weight reduction interventions[9], because of its proved effectiveness in decreasing body fat mass in men[10] and women.[11,12] However, in general, most intervention studies have found no improvement in lipid profiles after resistance training (RT) programs.[13] In this context, two recent reviews have concluded that resistance training does not seem to alter blood lipid and lipoprotein levels, probably because of the normal circulating lipid levels in individuals participating in those studies.[1,14] Indeed, subjects with normal lipid profiles may require greater exercise stimulus and energy expenditure, coupled with significant reductions in body weight, to further improve lipid profiles.

To date, only a few studies have examined the effects on body weight loss and lipoprotein profile of protein controlled diets with and without a concomitant resistance training in healthy obese women,[15-17] and none of these was conducted in obese women with hypercholesterolemia.

Objectives

The aims of this study were to evaluate the effects on the body fat mass loss and lipid profile of a higher protein (HP) vs. a lower protein (LP) in a restrictive diet, and their potential interactions with a resistance training program in hypercholesterolemic obese women. We hypothesized that protein content (higher protein (HP) *vs.* lower protein (LP)) within an energy-restrictive diet would interact with a resistance training program in a additive manner to bring about body fat mass loss and to improve the lipid profile in hypercholesterolemic obese women.

Methods

Subjects

Twenty-five sedentary hypercholesterolemic (> 200 mg/dl) obese (BMI ≥ 30 kg/m^2) women, aged 50 ± 6 years, participated in this study. The baseline features of the obese women appear in table I.

At the beginning of the study, all candidates were thoroughly screened by a physician using an extensive medical history, resting and maximal exercise electrocardiogram and blood pressure measurements, cardiovascular, neuromuscular, pulmonary or other debilitating diseases as determined by one or all of the screening tools were reasons for exclusion from the study. Participants were not taking any medication, have maintained the weight at least for 3 months before the intervention and were not following a particular diet prior to the enrollment in the trial. All the subjects were informed in detail about the possible risks and benefits of the project, and signed a written consent form before participating in the study. The project was approved by the ethical committee of the regional Health Department and was conform to the Code of Ethics of the World Medical Association.

Design

This is an observational study derived from a randomized controlled clinical trial lasting 16 weeks, in which participants were randomized to three groups: a control group; a diet group with a caloric restriction of 500 kcal/day without a programmed exercise; and a diet plus resistance training group with the same caloric restriction (-500 kcal/day) and a 16-week supervised whole-body resistance training program, two sessions/week. The subjects were tested on two different occasions (weeks 0 and 16) using identical protocols.[11,18] When daily caloric intake was evaluated at week 16, a deviation was noted between the initially prescribed diet (55% of calories as carbohydrates, 15% as proteins, and 30% as fat) and the real one (42% of calories as carbohydrates, 22% as proteins, and 36.5% as fat). The real diet estimation demonstrated that subjects increased the protein content and reduced the carbohydrate intake with positive results on weight, body composition and others cardiovascular risk

Table I
Baseline characteristics of participants categorized by the dietary group

	Lowe Protein Hypocaloric Diet (< 22%) (LP; LP + RT) (n = 11)	*Higher Protein Hypocaloric Diet (≥ 22%) (HP; HP + RT) (n = 14)*	*Mean Difference 95% CI*	*p value**
Age (y)	47.8 ± 6.3	51.6 ± 5,5	-3.8 (-8.7; 1.1)	0.119
Anthropometric variables				
Body Weight (kg)	88.8 ± 14.2	89.3 ± 13.8	-0.5 (-12.1; 11.1)	0.931
BMI (kg/m^2)	34.1 ± 3.4	35.3 ± 3.1	-1.2 (-3.9; 1.5)	0.364
Waist circumference (cm)	99.0 ± 6.0	101.4 ± 8.3	-2.4 (-8.5; 3.8)	0.432
WHR	0.9 ± 0.0	0.9 ± 0.0		
Abdominal MRI volume				
SAT (cm^3)	14.049 ± 3.395	15.020 ± 2.998	-970 (-3.618; 1.678)	0.456
VAT (cm^3)	3.302 ± 920	3.324 ± 1.166	-22 (-910; 867)	0.960
SAT + VAT (cm^3)	17.351 ± 3.897	18.343 ± 3.322	-992 (-3.978; 1.995)	0.499
Thigh MRI volume				
Subcutaneous fat (cm^3)	90.949 ± 23.856	97.788 ± 21.078	-6.839 (-1.570; 12.766)	0.455
Muscle (cm^3)	50.909 ± 10.228	45.311 ± 7.099	-5.598 (-25.450; 11.771)	0.120
Lipoprotein profiles				
LDL-C (mg/dl)	146.0 ± 30.1	145.1 ± 28.0	0.9 (-23.2; 25)	0.937
TG (mg/dl)	133.0 ± 42.7	119.4 ± 39.9		0.381
HDL-C (mg/dl)	70.6 ± 14.1	75.4 ± 11.6	-4.8 (-15.3; 5.9)	0.368
TC (mg/dl)	247.5 ± 38.9	250.8 ± 37.2	-3.3 (-34.9; 28.3)	0.829
HDL-C/CT (ratio)	0.3 ± 0.0	0.3 ± 0.0		

Values are expressed as means ± SD.
BMI = Body mass index; WHR = Waist/Hip ratio; MRI = Magnetic resonance imaging; SAT: Subcutaneous adipose tissue; VAT: Visceral adipose tissue; LDL-C = Low-density lipoprotein; TG = Triglycerides; HDL-C = High-density lipoprotein; TC = Total cholesterol.
**p* value from Student t or Mann-Withney U (non parametric) test for the differences between groups.

factors as described elsewhere.[11] For this reason, in this new study, only intervention groups (diet group and diet plus resistance group) were selected, and the subjects were categorized to a 4-arm trial using a 2 x 2 factorial design (Diet x RT Exercise), depending on the daily protein intake of the diets (protein intake according to the median value): Higher protein hypocaoric diet (HP); Lower protein hypocaloric diet (LP); Higher protein diet + resistance training (HP+RT); Lower protein diet + resistance training (LP+RT).

The median cutoff criteria have been previously applied[19] and is based on a valid and reliable method to assign two groups of risk in epidemiological studies.[20]

Methodology

Energy intake and energy expenditure analysis

Dietary composition was assessed by a dietitian and was based on the analysis of a validated semiquantitative food record.[11] At weeks 0 and 16 all subjects were interviewed by a trained dietitian and given instructions on how to complete food records accurately. Three-day dietary food records (including 1weekend day) were recorded being filled out on the actual day of consumption of the foods. All food records were analyzed by DIETSOURCE (DietSource program; version 1.0; Novartis, Barcelona, Spain).

Similarly, habitual physical activity was directly evaluated by accelerometry (TriTrac-R3D System, version 2.04; Madison, WI). The TriTrac-R3D was worn on a belt that was firmly attached to the anterior torso of the subject at the level of the waist. TriTrac monitoring was recorded on a minute-by-minute basis over 2 weekdays and 2 weekend days, during the days of the dietary records.

Anthropometric variables and magnetic resonance imaging

The height of the subjects was measured barefoot to the nearest 0.1 cm with a stadiometer. Body mass was measured on the same standard medical scale to an accuracy of ± 100 g. Waist and hip circumferences were measured with the subject standing erect with arms at the sides and feet together, wearing only underwear. The anthropometrist placed an inelastic tape around the subject, without compressing the skin, on a horizontal plane at the level of the last false rib and the buttocks, respectively. The measurement was recorded to the nearest 0.1 cm.

Table II
Mean nutrient intake depending on the hypocaloric dietary group

Characteristics of diet	*Lowe Protein Hypocaloric Diet (< 22%) (LP; LP + RT) (n = 11)*	*Higher Protein Hypocaloric Diet (≥ 22%) (HP; HP + RT) (n = 14)*	*Mean Difference 95% CI*	*p value**
Energy intake (kcal)	1,556 ± 329	1,473 ± 470	83	0.298
Total protein (% energy)	19.5 ± 1.7	23.5 ± 1.7	-4 (-5.5; -2.6)	0.000
Total lipid (% energy)	40.9 ± 6.2	33.2 ± 4.9	7.7 (3.1; 12.4)	0.020
Saturated fatty acid (% energy)	8.4 ± 1.2	7.1 ± 1.7	1.3 (-1.6; 2.8)	0.077
Monounsaturated fatty acid (% energy)	19.5 ± 4.2	15.2 ± 2.8	4.3 (1.4; 7.2)	0.006
Polyunsaturated fatty acid (% energy)	5.0 ± 0.7	4.6 ± 1.1	0.4	0.089
Cholesterol (mg/d)	320.6 ± 84.6	346.3 ± 44.5	-25.7	0.870
CHO (% energy intake)	39.2 ± 5.3	44.8 ± 7.8	-5.6	0.063
Fiber (g/d)	21.9 ± 5.9	22.9 ± 9.2	-1 (-7.6; 5.6)	0.752

Values are expressed as means ± SD
CHO = Carbohydrate; d = Day.
**p* value from Student t or Mann-Withney U (non parametric) test for the differences between groups.

The volumes of visceral and subcutaneous adipose tissue (abdominal and thigh) and muscle volume in the thigh were measured by magnetic resonance imaging (MRI). MRI assessment was performed with a 1T magnet equipment (Magnetom Impact Expert; Siemens, Erlangen, Germany) using body coil. Subjects were examined in a supine position with both arms positioned parallel along the sides of the body. A detailed description of the MRI procedure can be found elsewhere.[18]

Hypocaloric diets

In the original randomized clinical trial, the experimental diets were prepared by an exchange system. The energy content of the menus was individually prescribed for each subject according to a previous analysis of the individual daily energy expenditure by accelerometry, with the same caloric restriction (-500 kcal/day). These diets were designed to elicit a 0.5 kg weight loss per week. Subjects were instructed individually to follow a similar and regular pattern of meals per week according to local habits. All food groups were included in the dietary offer.

In the retrospective study, subjects were categorized by the daily protein intake of their diets: Lower protein hypocaloric diet (LP; < 22% of the daily calorie intake as protein) and Higher protein hypocaloric diet (HP; ≥ 22% daily energy from protein). The nutritional characteristics of the diets are presented in table II.

Resistance training program

Participants on the resistance group followed a progressive resistance training program. The testing and strength training program used in the prospective study has been reported previously.[11,18,21]

Briefly, lower and upper body maximal strength was assessed using 1 repetition concentric maximum (1-RM) action in a half-squat and in a bench-press position, respectively. A detailed description of the 1-RM testing procedure can be found elsewhere[21]. Women trained twice a week to perform dynamic resistance exercise for 45-60 min per session. A minimum of 2 days elapsed between two consecutive training sessions. Each training session included two exercises for the leg extensor muscles (bilateral leg press and bilateral knee extension exercises), one exercise for the arm extensor muscle (the bench-press) and four to five exercises for the main muscle groups of the body. Only resistance machines (Technogym, Gambettola, Italy) were used throughout the training period. During the first 8 weeks of the training period the subjects trained with loads of 50-70% of the individual 1-RM, and during the last 8 weeks of the training period the loads were 70-80% of the maximum. In all the individual exercise sessions performed one of the researchers was present to direct and assist each subject towards performing the appropriate work rates and loads. For all subjects average compliance with the exercise sessions was above 95%.

Statistical analysis

All data analyses were conducted using SPSS version 15.0 (SPSS Inc., Chicago, Illinois, USA). Baseline values are presented as means ± standard deviation (SD). A one-way ANOVA or Kruskal Wallis non-parametric test was used to determine any differences among the four groups' initial measurements.

Changes occurring in response to treatments (week 0-week16 value) within each group were evaluated by

Student t test when the variables followed a normal distribution or by the Wilcoxon non-parametric test for variables without a normal distribution. Week 16, delta values (Δ = week 16-baseline testing) were calculated and used for determination of delta changes variables across time in four groups (LP; HP; LP + RT; HP + RT).

To determine any differences among the four groups' delta values, one-way ANOVA or Kruskal-Wallis non-parametric tests were used. When a significant[2] was achieved, the average range was found to locate the value farthest from the averages compared. When the sample was analyzed by diet or exercise, the tests used to identify differences in delta values between groups were Student t or Mann-Withney U test (non-parametric), depending on the normality of the variables.

To evaluate the interactive effects of the diet and exercise treatments in delta values, a two-way factorial analysis of variance was applied (Diet x Exercise). The Pearson (parametric) or the Spearman (non-parametric) coefficients (r/rho respectively) were used to establish the potential relationships among variables. A multivariate regression model was applied to describe the observed change in plasma LDL-C (dependent variable), considering lipid (% energy intake) and monounsaturated fat (% energy intake) as independent variables. In all cases, the $p < 0.05$ criterion was used for establishing statistical significance.

Results

Body weight

Baseline characteristics were similar in lower protein and higher protein groups (table I). After 16 weeks of intervention, a significant decreased in weight and waist circumference was observed in LP, LP + RT, HP and HP + RT groups (table III), HP being the one with a greater Δweight (-8.9%) and Δwaist circumference (-7.9%) loss, although no differences were observed between groups (table IV). In this context, the groups with a higher protein intake (HP and HP + RT) showed a greater body weight loss than groups with a lower protein intake (LP and LP + RT) (-7.3 ± 4.5 kg, 8% *vs.* -5.2 ± 3.6 kg, 6%, respectively), although no significant difference was found. Neither the protein content of the diet nor the exercise, independently, affected anthropometric variables. The interaction between diet and exercise treatment was not significant for any anthropometric variables including body weight (table IV).

Body composition

After 16 weeks of intervention, fat mass loss was the predominant component in the decreased body weight in all the four groups (table III). The decrease in abdominal adipose tissue (ΔSAT + VAT) was similar in all groups, and it was not associated with the protein content in diet or the resistance training program (table IV).

When comparing the four groups, differences in muscle mass loss were not relevant. Nevertheless, the HP + RT group was the one with lower Δmuscle mass loss (LP:-1.3%; LP + RT:-2.0%; HP:-6.6%; HP + RT:-0.2%).

Lipoprotein profile

Baseline serum lipid profiles did not differ among experimental groups (table I). The 16 weeks of intervention were accompanied by marked changes in total cholesterol (TC) and LDL-C values only in the HP + RT group (table III). In both cases, changes were positively correlated with Δ thigh muscle (rho = 0.733, p = 0.025; and rho = 0.733, p = 0.025, respectively), and Δ visceral adipose tissue (rho = 0.717, p = 0.030; and rho = 0.650, p = 0.058, respectively). Also, a decrease in high-density lipoprotein cholesterol (HDL-C) concentration was observed only in the resistance trained groups, LP + RT (-11.7 ± 7.1, p = 0.046) and HP + RT (-7.9 ± 9.8, p = 0.043), but these changes did not correlate with any of the studied variables.

Interestingly, the greatest magnitude of change in ΔTC (-21%), ΔLDL-C (-23.7%), ΔHDL-C (-16.5%) and triglycerides (ΔTG, -13.5%) was observed in the LP + RT group, but differences among groups were found only to be significant for ΔLDL-C and ΔTC. (table IV). Changes in HDL-C and TC were affected by resistance training. In the absence of a significant interaction between exercise and diet, the resistance training effect was independent of the composition of the energy-restrictive diet (table IV). In fact, in the resistance program groups (LP + RT and HP + RT), ΔHDL-C and ΔTC decreased significant by 11.5% and 13.6%, respectively; while there was a slight increase in those who had only a dietary restriction (LP, 1%; and HP, 1.6%). However, although LDL-C concentrations were affected also by the resistance training (main effect of exercise p = 0.025), a significant interaction was identified between diet and exercise treatments (p = 0.019). In this study, though the effect of exercise was independent and additive on plasma LDL-C levels (-13.8% in groups with resistance program *vs.* +1.6% in groups with restrictive diet only) when it was combined with a restrictive diet providing a protein intake of < 22% of the daily energy intake, the decrease in LDL-C was significantly higher (-23.7%) showing a marked interaction between both interventions (fig. 1).

In addition, although a significant difference in lipid composition in diets of the two interventional groups was observed (table II), no influence on circulating LDL-C levels was found when a regression analysis was performed using either lipid (% energy intake) or monounsaturated fat intake (% energy intake) as a

Table III
Mean baseline values and changes after 16 weeks in obese women by the hypocalorics diets and the resistance training

	Baseline (week 0)				Within-groups changes after 16 weeks			
	Protein intake lower < 22% of energy (LP) (n = 11)		Protein intake higher ≥ 22% of energy (HP) (n = 14)		Portein intake lower < 22% of energy		Portein intake higher ≥ 22% of energy	
Variable	*No RT*	*RT*	*no RT*	*RT*	*No RT*	*RT*	*No RT*	*RT*
	mean ± standard deviation				mean difference (95% CI)			
Anthropometric								
Body Weight (kg)	90.7 ± 17.1	85.6 ± 8.2	84.1 ± 12.8	92.2 ± 14.1	-4.3 (0.66, 7.86)*	-6.8 (2.32, 11.38)*	-7.6 (0.86, 14.41)*	-7.1 (3.85, 10.46)*
BMI (kg/m²)	34.5 ± 4.19	33.5 ± 1.6	34.7 ± 2.4	35.7 ± 3.5	-1.6 (0.31, 2.80)*	-2.6 (1.15, 4.14)*	-3.0 (0.66, 5.50)*	-2.7 (1.63, 3.81)*
Waist (cm)	100.4 ± 6.9	96.8 ± 3.8	102.2 ± 6.4	101 ± 9.5	-5.6 (0.75, 10.39)*	-5.9 (3.17, 8.58)*	-8.0 (1.33, 14.67)*	-6.5 (3.89, 9.11)*
Abdominal MRI volume								
SAT (cc)	14,115 ± 4,144	13,934 ± 2,010	13,404 ± 1,851	15,917 ± 3,218	-1,982 (684, 3,279)*	-3,556 (120, 6,993)*	-3,075 (1,447, 4,703)*	-3,262 (1,710, 4,813)*
VAT (cc)	3,204 ± 667	3,474 ± 1,366	3,530 ± 1,371	3,209 ± 1,109	-582 (83, 1,080)*	-873 (147, 1,599)*	-663 (291, 1,034)*	-561 (175, 947)*
SAT + VAT (cc)	17,319 ± 4,593	17,408 ± 2,900	16,935 ± 3,118	1,9126 ± 3,338	-2,564 (889, 4,238)*	-4,429 (740, 8,118)*	-3,738 (1,912, 5,564)*	-3,823 (2,205, 5,441)*
Thigh MRI volume								
SAT (cc)	90,473 ± 26,914	91,782 ± 21,138	77,063 ± 18,675	109,303 ± 1,140	-11,304 (1,179, 21,430)*	-14,512 (5,170, 23,855)*	-12,495 (1,574, 23,415)*	-17,227 (12,065, 22,388)*
Muscle (cc)	49,886 ± 11,152	52,698 ± 9,655	44,700 ± 4,856	45,651 ± 835	-678 (-1,002, 2,358)	-986 (-2,354, 4,326)	-2,605 (156, 5,053)*	-88 (-1,130, 1,307)
Lipoprotein profiles								
LDL (mg/dl)	136.3 ± 27.3	163.0 ± 30.3	164.0 ± 28.5	134.6 ± 22.8	6.4 (-31.86, 19.0)	-40.5 (-1.69, 82.69)	-12.6 (-17.68, 42.88)	-11.4 (0.10, 22.79)*
TG (mg/dl)	138.3 ± 49.8	123.8 ± 30.6	144.4 ± 50.9	105.6 ± 26.3	-23.4 (p = 0.735)[a]	-22.0 (p = 0.273)[a]	-2.2 (p = 0.893)[a]	-2.4 (p = 0.483)[a]
HDL-c (mg/dl)	69.6 ± 17.5	72.5 ± 6.4	74.2 ± 10.1	76.0 ± 12.9	1.8 (-13.07, 9.36)	-11.7 (0.40, 23.10)*	-3.0 (-8.17, 14.17)	-7.9 (0.33, 15.44)*
TC (mg/dl)	235.9 ± 39.4	267.8 ± 32.9	270 ± 41.9	240.1 ± 31.8	9.4 (-47.38, 28.52)	-58.5 (-2.53, 119.53)	-10.6 (-34.13, 55.33)	-23.8 (6.81, 40.74)*

Within-group (95% confidence interval [CI] changes are presented for the four groups.
RT = Resistance Training; BMI = Body mass index; SAT: Subcutaneous adipose tissue; VAT: Visceral adipose tissue; LDL-C= Low-density lipoprotein; TG = Triglycerides; HDL-C= High-density lipoprotein; TC = Total cholesterol.
[a]Determinaded by Wilcoxon test (non parametric).
Statistical significance: * $p < 0.05$.

Table IV

Between-group changes after 16 weeks of intervention and interaction of a higher protein (HP) vs. a lower protein (lP) diet with or without a concomitant resistance training program (RT) on body composition and lipoprotein profile in hypercholesterolemic obese women

	Between-group changes after 16 weeks					*Diet and exercise interaction*		
	Protein intake lower < 22% of energy (LP)		*Protein intake higher ≥ 22% of energy (HP)*			*D*	*RT*	*D x RT*
	No RT	*RT*	*No RT*	*RT*	*p value*[a]		*p value*	
			Δ values (week 16-week 0)					
Anthropometric								
Body Weight (kg)	-4.3 ± 3.9	-6.8 ± 2.8	-7.6 ± 5.4	-7.1 ± 4.3	0.488	0.315	0.562	0.400
BMI (kg/m²)	-1.6 ± 1.3	-2.6 ± 0.9	-3.0 ± 1.9	-2.7 ± 1.1	0.296	0.256	0.554	0.249
Waist (cm)	-5.6 ± 5.2	-5.9 ± 1.7	-8.0 ± 5.4	-6.5 ± 3.4	0.792	0.402	0.741	0.618
Abdominal MRI volume								
SAT (cm³)	-1,982 ± 1,404	-3,556 ± 2,160	-3,075 ± 1,311	-3,262 ± 2,018	0.430	0.595	0.247	0.359
VAT (cm³)	-582 ± 539	-873 ± 456	-663 ± 299	-561 ± 502	0.726	0.568	0.640	0.336
SAT + VAT (cm³)	-2,564 ± 1,811	-4,429 ± 2,318	-3,738 ± 1,470	-3,823 ± 2,101	0.438	0.732	0.247	0.289
Thigh MRI volume								
SAT (cm³)	-11,304 ± 10,948	-14,512 ± 5,871	-12,495 ± 795	-17,227 ± 6,715	0.545	0.587	0.274	0.832
Muscle (cm³)	-678 ± 1,817	-986 ± 2,099	-2,605 ± 1,972	-88 ± 1,585	0.173	0.505	0.160	0.077
Lipoprotein profiles								
LDL-C (mg/dl)	6.4 ± 27.5	-40.5 ± 26.55	-12.6 ± 24.4	-11.4 ± 14.7	0.028	0.603	0.025	0.019
TG (mg/dl)	-23.4 ± 71.3	-22.0 ± 42.6	-2.2 ± 32.1	-2.4 ± 10.9	0.737	0.284	0.975	0.964
HDL-C (mg/dl)	1.8 ± 12.1	-11.7 ± 7.1	-3.0 ± 9.0	-7.9 ± 9.8	0.147	0.907	0.040	0.314
TC (mg/dl)	9.4 ± 41.0	-58.5 ± 38.5[b]	-10.6 ± 36.0	-23.8 ± 22.0	0.032	0.607	0.009	0.065

Between-group changes data are presented as mean difference ± SD.
The boldface values depict statistical significance *($p < 0.05$)*.
[a]Determinaded by one-way ANOVA test (parametric) or Kruskal Wallis (non parametric) test.
[b]Different from LP, HP, HP+RT, *$p < 0.05$*.

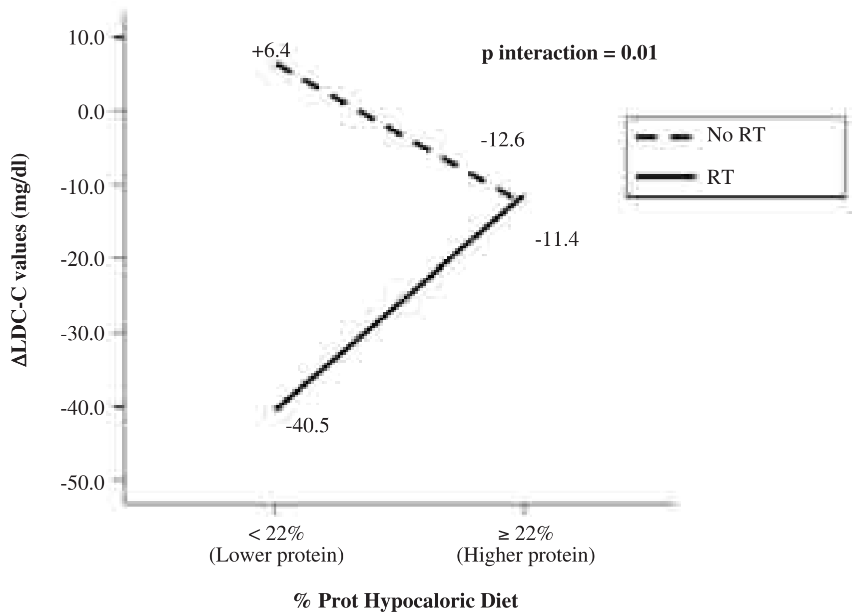

Fig. 1.—Interaction between Exercise an Hypocaloric diet on the changing (Δ LDL-C) values.

covariate. The protein content in diet was a predictive factor of ΔLDL-C in RT groups, independently of lipid and monounsaturated fat content in diet. Furthermore, no differences were found when an analysis using as a cutoff the median concerning fat intake was performed.

Discussion

Positive interaction between the protein content of the restrictive diet and the resistance training on plasma LDL-C levels during weight loss

The most relevant outcome of this study was the finding of an interaction between the protein content of the restrictive diet and the resistance training on the circulating levels of LDL-C.

Indeed, resistance training significantly improved plasma LDL-C in all patients (main effect of exercise $p = 0.025$). In addition, when RT was combined with a lower protein- hypocaloric diet (LP + RT group) a greater effect was observed (p for interaction = 0.019). However, a higher daily protein intake (HP + RT group) did not show any effect on lipid profile (fig. 1).

To our knowledge, this is the first study evaluating the influence of the diet composition with a concomitant resistance training on the lipid profile of obese women with hypercholesterolemia. So far, no interactions between a hypocaloric diet and a RT program on normal plasma levels of LDL-C in obese women has been reported.[15-17] In the present study, a hypocaloric diet was ineffective at modifying lipid and lipoprotein profiles in women who were obese and hypercholesterolemic. The main effect on TC and LDL-C was due to resistance exercise. This result is consistent with other studies that have reported improvements in LDL-C, TC and TG after a resistance training program,[22] while it differed from other reports[1,14] showing that resistance training does not seem to alter blood lipid and lipoprotein levels in normolipidemic subjects. A possible explanation for the lack of significant lipoprotein-lipid changes with resistance training may be the fact that TC values for most study groups have been < 200 mg/dl at study entry. Individuals with normal lipid profiles may require greater exercise stimulus and energy expenditure, coupled with significant reductions in body weight, to further improve lipid profiles. As to the role played by the protein content in the diet, Layman et al.[16] observed, in agreement with our results, that the higher the diet protein content, the lower is the decrease in LDL-C and TC levels. Of note, in our study no significant correlation was observed between the amount and type of fat in diet, or the percentage of carbohydrate, with changes in LDL-C in the LP+RT group.

On the other hand, whereas no clear dose–response relation between weight loss and circulating lipids modulations could be determined, it would appear that trials that experience a weight reduction > 5% of initial body weight seem to produce the most significant changes in TC and LDL-C concentrations.[23] However, this situation was not the case with our hypocaloric diet groups (LP and HP), where a decrease of ~6% of body weight was not translated into a significant improvement in lipid profile. In view of these findings, it may be assumed that in our study a chronic resistance exercise was the main factor responsible for the lipid profile improvement in both resistance groups (LP + RT and

HP + RT). This benefit in circulating lipids could be explained partly by further reductions in weight and body fat mass, usually associated with ameliorations in lipid profile in woman.[24,25] Interestingly, significant correlations were observed between LDL-C and VAT (rho = 0.56, p = 0.04) only in exercise groups (LP + RT and HP + RT groups), when segmented by exercise. These correlations are in agreement with the results of Fahlman et al.[22] who reported that 10 weeks of resistance training, three sessions per week, in overweight older women significantly improved the lipid profile without concurrent changes in weight or diet.

Absence of a main effect of protein content in diet on weight loss and body composition

After 16 weeks of intervention, a significant loss in weight and a decrease in waist circumference and body fat was observed in all groups. However, our results showed no significant effect of both protein content in diet and exercise on weight loss, anthropometric variables and subcutaneous adipose tissue (SAT) and visceral adipose tissue (VAT) measures (table III). Indeed, although greater changes did occur in groups with a higher content of protein in diet (HP and HP + RT) compared with women on less daily protein intake (LP and LP + RT), changes were not statistically significant. These results differ from those reported by others[16,17,26] and a possible explanation could be related to a different nutritional composition of diets and the low number of subjects. Indeed, owing to the eating habits of our population, high protein diet means no more than 25% of daily calorie intake, whereas in the mentioned studies the protein content of a high protein diet was always over 30% of daily energy intake. Moreover, our findings are consistent with the results of a meta-analysis[5] based on 84 dietary trials showing that after controlling for energy intake, neither a lower protein diet (< 1.06 g/kg) nor a higher protein diet (> 1.06 g/kg) were significant predictors of changes in body weight and body fat mass, although a high protein intake was associated with a lower lean mass loss. Like Clifton,[6] we observed no differences in lean body mass in our groups of study when we considered only the composition of the diet. On the other hand, the absence of a main effect of resistance exercise on body weight agrees with the results obtained by others[16,27,28] and could be explained because during weight loss RT can maintain or increase lean tissue, reducing changes in total body weight. Likewise, in this study, in accordance with others[15,29,30] resistance training had no significant effect on BMI, waist circumference or body fat mass. These findings suggest that exercise alone can not adequately promote greater changes in these anthropometric variables, although it prevents the decline in fat-free mass and resting metabolic rate. Along these lines, we observed further decreases in muscle mass in LP and HP groups of women (3.5%) than in resistance trained groups (LP + RT and HP + RT) (0.8%), although the differences were not significant.

Limitations

Limitations of this study include the small sample size, and the short duration of the intervention. Comparison between our study and others is difficult because of differences in study designs (age, gender, study duration, training frequency and intensity, duration of exercise, composition of the restrictive diet) and because literature is scarce regarding RT studies.

The results should be understood as translational pilot data that warrant further in-depth studies to determine the robustness of these interventions and the extent to which their findings can be generalized.

Conclusion

This study provides support for the effectiveness of combining resistance training and a lower content protein in an energy-restricted diet (< 22% of daily energy) to promote a significantly greater reduction in LDL-C nivels.

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