# Hospitalaria



# Original Effects of dietary restriction combined with different exercise programs or physical activity recommendations on blood lipids in overweight adults

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#### Abstract

*Background and aim:* Many exercise studies, although generally showing the beneficial effects of supervised aerobic, resistance or combined exercise on blood lipids, have sometimes reached equivocal conclusions. The aim of this study is to evaluate the impact of different programs that combined exercise and dietary restriction on blood lipids versus a clinical practice intervention for weight loss, in overweight adults.

*Methods:* For this study 66 subjects participated in a supervised 22 weeks training program, composed of three sessions per week and they were randomized in three groups: strength training (S; n = 19), endurance training (E; n = 25), a combination of E and S (SE; n = 22). Eighteen subjects served as physical activity group (PA) that followed a clinical intervention consisted of physical activity recommendations. All groups followed the same dietary treatment, and blood samples were obtained for lipids measurements, at the beginning and end of the study.

*Results:* Lipid profile improved in all groups. No significant differences for baseline and post-training values were observed between groups. In general, SE and PA decreased low-density lipoprotein cholesterol (LDL-C) values (p < 0.01). S decreased triglyceride levels (p < 0.01) and E, SE, and PA decreased total cholesterol levels (p < 0.05, p < 0.01 and p < 0.01, respectively).

*Conclusions:* These results suggest that an intervention program of supervised exercise combined with diet restriction did not achieved further improvements in blood lipid profile than diet restriction and physical activity recommendations, in overweight adults.

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Key words: Lipoprotein. Overweight. Strength training. Aerobic training. Combined training. Supervised exercise. Physical activities recommendations.

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#### EFECTOS DE LA RESTRICCIÓN DIETÉTICA COMBINADA CON DIFERENTES PROGRAMAS DE EJERCICIO O ACTIVIDAD FÍSICA SOBRE LOS LÍPIDOS SANGUÍNEOS EN ADULTOS CON SOBREPESO

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#### Resumen

Antecedentes y objetivo: Muchos estudios sobre ejercicio han proporcionado en ocasiones conclusiones equívocas, si bien, por lo general, han demostrado los efectos beneficiosos del ejercicio supervisado aeróbico, de resistencia o combinado. El propósito de este estudio fue evaluar el impacto de diferentes programas que combinan ejercicio y restricción dietética sobre los lípidos sanguíneos frente a una intervención de la práctica clínica de pérdida de ejercicio en los adultos con sobrepeso.

*Métodos:* En este estudio participaron 66 individuos en un programa de entrenamiento supervisado de 22 semanas, compuesto por tres sesiones semanales y, posteriormente, se les distribuyó al azar en tres grupos: entrenamiento de fuerza (F; n = 19), entrenamiento de resistencia (R; n = 25), o una combinación de R y F (FR; n = 22). Dieciocho sujetos sirvieron como grupo de actividad física (AF) que siguió una intervención clínica que consistía en recomendaciones de actividad física. Todos los grupos siguieron el mismo tratamiento dietético y se obtuvieron muestras de sangre para medición de lípidos al inicio y al final del estudio.

*Resultados:* El perfil lipídico mejoró en todos los grupos. No se observaron diferencias significativas basales ni tras el entrenamiento entre los grupos. Por lo general, el FR y la AF disminuyeron los valores de lipoproteínas de densidad baja-colesterol (LDL-C) (p < 0,01). El F disminuyó los valores de triglicéridos (p < 0,01) y el R, FR y AF disminuyeron las concentraciones de colesterol total (p < 0,05, p < 0,01 y p < 0,01, respectivamente).

*Conclusiones:* Estos resultados sugieren que la intervención con un programa de ejercicio supervisado combinado con la restricción dietética no proporcionó mejorías en el perfil lipídico con respecto a la restricción dietética y las recomendaciones de actividad física, en adultos con sobrepeso.

(Núm. de registro en Clinical Trials Gov: NCT01116856).

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Palabras clave: Lipoproteína. Sobrepeso. Entrenamiento de fuerza. Entrenamiento aeróbico. Entrenamiento combinado. Ejercicio supervisado. Recomendaciones de actividades físicas.

### Abbreviations

S: Strength training group.

E: Endurance training group.

SE: Strength + Endurance combined training group.

PA: Diet and physical activity recommendations group.

VO, peak: Peak oxygen uptake.

PRONAF: Programas de Nutrición y Actividad Física para el tratamiento del sobrepeso y la obesidad). HULP: Hospital Universitario La Paz. HRR: Heart rate reserve.

HRmax: Maximal heart rate.

RM: Repetition máximum.

DEE: Daily energy expenditure.

HPA: Physical activity

# Introduction

Many exercise training studies, although generally showing the beneficial effects of aerobic, resistance or combined exercise on plasma lipids and lipoproteins, have sometimes reached equivocal conclusions. The results of initial studies of strength training in coronary risk factors showed evidence that high intensity strength training improved blood lipid profile, with any diet counseling.<sup>1</sup> Strength training protocol suggested that this exercise without diet could improve HDL-C levels in overweight and obesity men, while diet alone did not.<sup>2</sup> Other study without diet intervention have reported increases in HDL-C ranging from 9% to 15% with endurance training, while strength and combination did not results in any significant changes in any of the blood lipid variables <sup>3</sup>. Some investigators found no differences in the blood lipid profile improvements obtained among endurance, strength, combined training and diet and diet alone intervention.4.5 Previous studies have suggested a relationship between endurance and combined training, without diet intervention, and favorable blood lipid profile in men and women.6 On the other hand, dietary and caloric intake interventions alone have shown clinical evidence about its influence on plasma lipid and lipoprotein levels.4,5,7,8

At present there are many studies including behavior strategies based in diet and exercise advice, but there is limited evidence that such "standard lifestyle interventions" including specific recommendations for informal physical activity, improve metabolic abnormalities.<sup>9,10</sup> Therefore, it is not so far clear if supervised exercise may contribute to reach health goals better than following standard recommendations.

The PRONAF Study (*Programas de Nutrición y Actividad Física para el tratamiento del sobrepeso y la obesidad*) is a research project in nutrition and physical activity programs for overweight and obesity, developed in Spain in several years of intervention. The design and protocol includes three exercise modes (endurance, strength and combined training) and diet

restriction, in a randomized control trial concerning diverse health status variables. A particular characteristic in the methodology is to include a group that follows the principles of hospital clinical practice for lifestyle changes (diet and physical activity recommendations) when treating patients for weight loss management. This way the PRONAF study will provide harmonized and comparable data on exercise effects on blood lipid profile, which is the purpose of this particular study here presented.

Therefore our study attempts to evaluate the impact of different supervised exercise modes with diet restriction on lipid profile versus the habitual weight loss intervention clinical practice with physical activity recommendations included, in overweight men and women.

# Materials and methods

# **Participants**

A group of 119 middle-aged (range 18-50 years) men (n = 46) and women (n = 73) volunteered to participate in this study (table I). Figure 1 shows the flow diagram of the PRONAF study. Of the initial 1568 subjects who reported to the advertisement, 84 only completed the study and were included in the final analysis. The subjects were recruited using a wide variety of techniques, including newspapers, radio, mailers, and posters. Inclusion criteria required to be healthy, overweight [body mass index (BMI) 25-29.9 kg·m<sup>-2</sup>], nonsmokers, sedentary (one or less exercise bout per week), not on a diet program, normoglycemic and women had regular menstrual cycles. The voluntary participants who fulfilled the inclusion criteria and passed the baseline physical examination were stratified by age and sex and randomly divided into four groups: diet and supervised strength training [strength training group (S)], diet and supervised endurance training [endurance training group (E)], diet and supervised combined strength and endurance training [combined strength and endurance training group (SE)] or diet and physical activity recommendations (PA).

After receiving written and oral information about the project and possible risks and benefits of the experimental protocol, the participants read and signed an institutionally approved informed consent document, in agreement with the guidelines of the Declaration of Helsinki regarding research on human subjects. The study was approved by the Human Research Review Committee of the *Hospital Universitario La Paz* (HULP) (PI-643).

# Study design

This study was an intervention trial of 24 week duration. The measurements took place in the first week

		<b>Tabl</b> Subjects' character	<b>e I</b> ristics at base	eline		
		Men		Women		Total
		<i>n</i> = 37		<i>n</i> = 47		<i>n</i> = 84
	n	Mean ± SD	n	$Mean \pm SD$	n	Mean ± SD
Age (years)						
S	7	$35.5 \pm 9.2$	12	$36.4 \pm 8.6$	19	$36.1 \pm 8.7$
E	10	$34.6 \pm 8.3$	15	$36.4 \pm 7.9$	25	$35.8 \pm 8$
SE	12	$37.1 \pm 7.7$	10	$34.7 \pm 6.9$	22	$36 \pm 7.3$
PA	8	$36.2 \pm 8.6$	10	$37.2 \pm 9.3$	18	$36.8 \pm 8.9$
Weight (kg)						
S		$92.7 \pm 10.7$		$76.2 \pm 8.6$		$81.4 \pm 12,0$
Е		$89.3 \pm 10$		$74.9 \pm 6.6$		$80.1 \pm 10.5$
SE		$86.1 \pm 8.8$		$72.5 \pm 7.6$		$79.6 \pm 10.6$
PA		$88.1 \pm 7.2$		$74.8 \pm 5.8$		$80.4 \pm 9.2$
$BMI(kg/m^2)$						
S		$29.9 \pm 2.7$		$29.8 \pm 1.7$		$29.8 \pm 2$
E		$29.2 \pm 1.1$		$28.8 \pm 1.9$		$29 \pm 1.7$
SE		$28.2 \pm 1.8$		$28.2 \pm 1.5$		$28.2 \pm 1.7$
PA		$29 \pm 1.4$		$27.7 \pm 1$		$28.3 \pm 1.3$
Body fat (%)						
S		$39.5 \pm 6.8$		$40.5 \pm 6.8$		$40.1 \pm 6.7$
E		$39.5 \pm 5.7$		$39.9 \pm 6.4$		$39.7 \pm 6.1$
SE		$39.2 \pm 6.8$		$36.5 \pm 5.6$		$37.8 \pm 6.2$
PA		$40.8 \pm 3.2$		$40 \pm 6.8$		$40.4\pm5.2$
Body fat free (kg)						
S		$48.9 \pm 9$		$46.1 \pm 11.4$		$47.2 \pm 10.4$
E		$44.9 \pm 8.9$		$46 \pm 8.8$		$45.6 \pm 8.6$
SE		$45.4 \pm 8.1$		$50.1 \pm 10$		$47.8 \pm 9.3$
PA		$45.6 \pm 6.8$		$46.4 \pm 9.2$		$46 \pm 7.9$
VO <sub>2neak</sub> (mL/kg/min)						
S		$36,0 \pm 8.7$		$27.5 \pm 3.6$		$30.5 \pm 7.1$
E		$38.4 \pm 4.7$		$27.1 \pm 3.2$		$30.9 \pm 6.5$
SE		$38.3 \pm 5.3$		$29.5 \pm 5.7$		$34.1 \pm 7$
PA		$30.7 \pm 6.1$		$30.7 \pm 6.1$		$30.7 \pm 6.1$

Data are shown as mean  $\pm$  SD.

S: Strength group; E: Endurance group; SE: Combined strength and endurance group; PA: Physical activity recommendations group. BMI: Body Mass Index; VO2peak: Peak oxygen uptake.

(baseline values) for all subjects before starting training, and 22 weeks of training later, in 24<sup>th</sup> week (post-training values). Once the first group started the pre-evaluation week, each group started sequentially (fig. 2) maintaining the same periodization. Adherence criteria for diet and exercise were took into account to determine final analyzed subjects (table I).

# Diet

Diet prescription was performed for all patients by expert dieticians in the Nutrition Department of HULP. All groups underwent an individualized and hipocaloric diet (between 1,200 and 3,000 kcal). Diet was lowered a 25% from Daily Energy Expenditure (DEE)<sup>11</sup> measured using SenseWear Pro Armband<sup>TM</sup> data. Macronutrient distribution consisted of 29-34% of energy from fat, 12-18% from protein, and 50-55% from carbohydrates, according to recommendations.<sup>12</sup> A dietitian interviewed each participant at baseline, 3 months, and 6 months and reviewed a 3-day food record diary. An adherence to diet of 90% was elicited and was calculated with 72-hour recall.<sup>13</sup>

# Exercise training

The different exercise groups followed the corresponding, supervised training program of 22 weeks, which consisted in all cases of three sessions per week. All training sessions were carefully supervised by certified personal trainers. An adherence to training of 90% was demanded. The exercise programs were designed taking into account each subject's muscle strength (MS) and the heart rate reserve (HRR). MS



Fig. 1.—Participant flow diagram in the PRONAF study. A total of 1,568 were screened, of whom 119 were randomized into the PRO-NAF study. The dropout rates in the groups were: strength group (S) was 36.6%, endurance group (E) was 16.6%, combined group (SE) was 26.6% and physical activity recommendations group (PA) was 37.9%.

was measured using the 15-repetition maximum (15 RM) testing method,<sup>14</sup> in the S and SE groups (both of which involved strength training). The 15 RM for each exercise in each program was recorded twice on different

days during the pre-intervention subject strength evaluation period. The intraclass correlation coefficient of reliability for all exercises was ICCr = 0.995 and ICCr = 0.994for the men and women respectively (groups S and SE



Fig. 2.—Study design. Pre and post-evaluation (baseline and post-training evaluation) week consisted of the same tests: peak oxygen uptake test ( $O_{2peak}$ ) test, habitual physical activity, blood analysis, body composition, diet prescription and 15RM. Top bar shows intensity (HRR and RM) and volume (minutes) progression during 22 week.

subjects together). All the assessments and trainings were carried out with the same machines and free weights (Johnson Health Tech. Iberica, Matrix, Spain). Heart rate reserve (HRR) was also calculated to prescribe exercise intensity plus resting heart rate for E and SE interventions programs.<sup>15,16</sup>

The intensity of exercise was increased over the study period. In weeks 2-5 exercise was at an intensity of 50% of the 15RM and HRR, and lasted an overall 51 min and 15 s (twice around the circuit, lasting 7 min 45 s each lap). In weeks 6-14 exercise was performed at an intensity of 60% of 15RM and HRR, again with a duration of 51 min and 15 s (again, twice around the circuit). Finally, in weeks 15-23, exercise was performed at an intensity of 60% of 15RM and HRR, with a duration of 64 minutes (three times around the circuit). The recovery period between circuits was set at 5 min. Participants performed 15 repetitions (45 s) of each exercise with a rest period of 15 seconds between them.

Each training session for the strength, endurance + combined strength and endurance training commenced with a 5 min aerobic warm-up, followed by the main session exercises, and concluded with 5 min of cooling down and stretching exercises. In addition, each session was monitored for HR and Rate of Perceived Exertion (RPE) scale. In all sessions the exercise rhythm was controlled by instructions recorded on a compact disk. The cadence for the resistance exercises was fixed at 1:2 (concentric-eccentric phase).

Feedbacks for training loads were done once a month with the RPE to subjectively evaluate each session and determine where the participant considered the intensity to be at, following a similar methodology as used elsewhere.<sup>17</sup>

Endurance training group (E). The E group training involved the use of a treadmill, exercise bike or cross trainer.

Strength training group (S). The S group followed a circuit involving the following eight exercises: shoulder

press, squat, barbell row, lateral split, bench press, front split, biceps curl, and French press for triceps.

Strength and endurance training group (SE). The SE group performed a combination of cycle ergometry, treadmill or cross trainer work, plus weight training with the following exercises intercalated: squat, row machine, bench press and front split.

Physical activity recommendations group (PA). These participants followed the habitual hospital clinical practice. This means the same dietary intervention as the training groups plus general recommendations in physical activity (ACSM),<sup>18</sup> without being supervised and regulated. The control of lifestyle changes was registered with accelerometer, just as real clinical health practitioners at hospital units.

# Data collection

*Body composition.* Body composition was assessed by dual-energy x-ray absorptiometry DXA (GE Lunar Prodigy; GE Healthcare, Madison, WI, GE Encore 2002, version 6.10.029 software) and was used to measure total body fat (%) and body fat free (kg) mass.

Anthropometric measures included height (stadiometer SECA; range 80-200 cm), body mass (BC-420MA. Bio Lógica. Tecnología Médica SL) and body mass index (BMI) calculated as [body weight (kg)/(height (m))<sup>2</sup>].

Peak oxygen uptake test. Peak oxygen uptake  $(O_{2_{peak}})$  was measured using the modified Bruce protocol. The test was conducted on a computerized treadmill (H/P/COSMOS 3P<sup>®</sup> 4.0, H/P/Cosmos Sports & Medical, Nussdorf-Traunstein, Germany). Volume and composition of expired gas measure was carried out with a gas analyzer Jaeger Oxycon Pro (Erich Jaeger, Viasys Healthcare, Germany) and continuous 12-lead electrocardiographic monitoring. The exercise test was maintained until exhaustion. The mean of the

last three highest rates of oxygen consumption was used as  $\mathrm{O}_{_{\mathrm{2neak.}}}$ 

*Habitual physical activity.* Habitual physical activity (HPA) was assessed with a SenseWear Pro3 Armband<sup>TM</sup> (Body Media, Pittsburgh, PA). DEE was calculated using a generalized proprietary algorithm (Innerview Research Software Version 6.0). Subjects were instructed to wear the monitors continuously for 5 days including weekend days and weekdays following general recommendations<sup>19</sup> at baseline and posttraining intervention. Data was recorded by 15min intervals. All subjects were instructed to continue their habitual daily activities as before and were provided with an exercise and HPA diary to log the type, duration, and intensity of any physical activity or exercise undertaken during intervention.

*Blood analysis.* All blood samples were taken after 12 h fast between 7:00 and 9:00 a.m. at baseline and post-training intervention (week 1 and week 24). All post-training samples were obtained 72 hours after the last training day to avoid acute effects of training on blood lipids. All blood samples were drawn from the antecubital vein and handled according to standardized laboratory practice at HULP.

Blood lipids and lipoprotein. Serum biochemicals (total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), and triglycerides (TG) were determined using enzymatic methods with Olympus reagents by automated spectrophotometry performed on Olympus AU 5400 (Olympus Diagnostica, Hamburg, Germany). Menstrual cycle was controlled by diary to define the follicular and luteal phases when blood samples were taken.<sup>20</sup>

# Statistical analysis

SPSS version 15.0 for Windows was used for statistical analyses (SPSS Inc., Chicago, Illinois, USA). Standard statistical methods were used for the calculation of the means and standard deviation. Two way analysis of variance (ANOVA) (group x measurement [baselinepost]) for repeated measures was used to determine any differences between four groups and differences in baseline and post-training values in each group assessed. Bonferroni's post-hoc test was employed to locate specific differences. The delta percentage was calculated through the standard formula: change (%) = [(post-test)]score-pre-test score]  $\times$  100. The effect of menstrual cycle on lipid profile was assessed by impaired T-test. The effect of ApoE on lipid profile was assessed by univariate analysis of variance (ANOVA). The significance level was set at  $\alpha = 0.05$ .

# Results

Data is presented separated in men and women, and total. Even though the purpose of this study was not to

observe gender differences, we decided to show both ways in order to discuss results clearly, taking into account that most of the previous studies are performed with only men or women.

#### Anthropometric and Fitness Measurements

Baseline characteristics of the participants revealed no significant differences for weight, percentage body fat, body fat free mass and  $O_{2peak}$ . Baseline BMI was significantly different between S and PA (table I). Weight change results showed a significant decrease in men and women of all groups (p < 0.01).

#### Blood lipids and lipoproteins

Table II shows changes in plasma lipid and lipoprotein concentrations in four groups before and after the intervention period. There were no statistically significant differences between groups at baseline and posttraining values.

Significant changes can be observed between baseline and post-training values in each group. No significant changes were found in the HDL-C, LDL-C and CT levels in SE group, while a significant decrease was observed for TG levels (22.8%; p < 0.01). No significant changes were found in the HDL-C, LDL-C and TG levels in E group, but TC decreased significantly (4.4%; p < 0.05). For SE group, significant changes were found for LDL-C (10.6%; p < 0.01) and TC levels (8.8%; p < 0.01). LDL-C (10.4%; p < 0.01) and TC (7.6%; p < 0.01) levels significantly decreased in the PA group, with no significant changes in HDL-C and TG values.

There were no differences in lipid profile values between luteal and non-luteal (follicular) phase at baseline and post-training measurements (data not shown). Regarding ApoE groups, there were no differences in serum lipids and lipoprotein concentrations at baseline (data not shown).

## Dietary analyses

Measurements on macronutrient and energy intakes at baseline and post-training are shown in table III. There were no statistically significant differences between groups at baseline and post-training values. Carbohydrate percentage intake increased significantly in S and E group (p < 0.01). The lipid percentage intake showed a significant decrease in all four groups p < 0.01). E and SE groups obtained significant increased levels of protein percentage intake (p < 0.05).

## Daily Energy Expenditure

There were no differences between groups neither at baseline nor at post-training for daily energy expendi-

		Men					Women					Total		
		n = 37					n = 47					n = 84		
2	Baseline	Post-training	Change (00)	D value	2	Baseline	Post-training	Change (06)	D victure	5	Baseline	Post-training	Change (00)	D victure
2	$Mean \pm SD$	$Mean \pm SD$	Cumige ( 70)	I - Value	u	$Mean \pm SD$	$Mean \pm SD$	(m) agumu	r -vaue	2	$Mean \pm SD$	$Mean \pm SD$	Cumige ( 10)	r -vaude
g/dL)														
7	$41.9 \pm 7.8$	$45 \pm 9.3$	7.4	0.18	12	$59 \pm 9.6$	$59.5 \pm 12.2$	0.8	0.77	19	$53.5 \pm 12.1$	$54.9 \pm 13.1$	2.6	0.35
10	$48,0 \pm 6.8$	$51.8 \pm 5.4$	7.9	0.07	15	$54.3 \pm 10.9$	$52.9 \pm 9.8$	-2.6	0.37	25	$52 \pm 10$	$52.5 \pm 8.4$	1	0.72
12	47.1±7.6	$51.1 \pm 6.7$	8.5	0.03	10	$62.3 \pm 8.5$	$57.4 \pm 8.9$	-7.9	0.01	22	$54 \pm 11$	$54 \pm 8.2$	0	0.97
8	$46.1 \pm 6.4$	$48.7 \pm 8.4$	5.6	0.27	10	$57.4 \pm 11.4$	57.5 ± 12.5	0.2	0.92	18	$53 \pm 11.1$	$54.1 \pm 11.7$	2.1	0.47
g/dL)														
	$138.2 \pm 25.2$	$138.8 \pm 24.9$	0.4	0.91		$121 \pm 18.8$	$120.5 \pm 23$	-0.4	0.0		$125.9 \pm 21.7$	$125.8 \pm 24.4$	-0.1	0.97
	$128.9 \pm 29$	$122.8 \pm 26.8$	-4.7	0.21		$117.3 \pm 26.3$	$127.9 \pm 21.8$	6	0.01		$121.4 \pm 27.3$	$126.1 \pm 23.3$	3.9	0.13
	$142 \pm 31.8$	$127 \pm 29.9$	-10.6	0.01		$126.2 \pm 19.7$	$112.8 \pm 25.3$	-10.6	0.01		$134.8 \pm 27.6$	$120.5 \pm 28.2$	-10.6	0.01
	$144.9 \pm 35.5$	$138.3 \pm 36.1$	-4.6	0.24		$125.6 \pm 25.9$	$107 \pm 28.4$	-14.8	0.01		$133.1 \pm 30.5$	$119.2 \pm 34.4$	-10.4	0.01
(														
	$158.3 \pm 91.1$	$118.1 \pm 70.8$	-25.4	0.02		$111.3 \pm 45.7$	$87.8 \pm 33.3$	-21.1	0.05		$126.3 \pm 65.3$	$97.5 \pm 48.8$	-22.8	0.01
	85.2±12	$85.6 \pm 19.2$	0.5	0.98		$96.3 \pm 44.3$	$94.4 \pm 69.1$	-2	0.87		$92.3 \pm 36.1$	$91.2 \pm 55.9$	-1.2	0.9
	$114.3 \pm 80$	$100.7 \pm 38.6$	-11.9	0.31		$87 \pm 20.7$	$84.4 \pm 21.3$	-3	0.86		$101.9 \pm 61.1$	$93.3 \pm 32.3$	-8.4	0.38
	$120 \pm 54.6$	$135.9 \pm 63.4$	13.3	0.36		$114.2 \pm 75.9$	$96.8\pm48.9$	-15.2	0.21		$116.4 \pm 66.7$	$112 \pm 56.6$	-3.8	0.68
	$204.6 \pm 26.4$	$192 \pm 42.2$	-6.2	0.15		$197.7 \pm 25.8$	$197.6 \pm 32$	-0.1	0.99		$199.9 \pm 25.6$	$195.8 \pm 34.6$	-2.1	0.42
	$209.8 \pm 36.4$	$191.6 \pm 26.1$	-8.7	0.02		$199.5 \pm 38$	$197.4 \pm 26.5$	-1.1	0.72		$203.2 \pm 37$	194.3 ± 26	4.4	0.02
	$213.5 \pm 41.1$	$198.2 \pm 29.6$	-7.2	0.02		$209.6 \pm 25.8$	$187.1 \pm 26.1$	-10.7	0.01		$211.7 \pm 34.2$	$193.1 \pm 28$	-8.8	0.01
	$218.3 \pm 48.9$	$213.3 \pm 49.7$	-2.3	0.56		$207.9 \pm 32.9$	$184.6 \pm 31.5$	-11.2	0.01		$211.9 \pm 38.8$	$195.8 \pm 40.8$	-7.6	0.01

		Chang	es in weigh	t. Daily En	ergy Exp	Table IIIenditure an	d diet macr	onutrient a	fter interve	ntion				
		Men					Women					Total		
		n = 37					n = 47					n = 84		
u	Baseline	Post-training	Change (%)	P-value	u	Baseline	Post-training	Change (%)	P-value	и	Baseline	Post-training	Change (%)	P-value
	$Mean \pm SD$	Mean±SD				$Mean \pm SD$	Mean ± SD				$Mean \pm SD$	$Mean \pm SD$		
Weight (kg)														
S 7	$92.7 \pm 10.7$	$84.4 \pm 9.5$	6-	0.01	12	$76.2 \pm 8.6$	$70.6 \pm 9.1$	-7.3	0.01	19	81.4±12	$75 \pm 11.1$	-7.9	0.01
E 10	$89.3 \pm 10$	$80.9 \pm 10.4$	-9.4	0.01	15	$74.9 \pm 6.6$	$68.6 \pm 7.8$	-8.4	0.01	25	$80.1\pm10.5$	$73 \pm 10.5$	-8.9	0.01
SE 12	$86.1\pm8.8$	$77.1 \pm 8.4$	-10.5	0.01	10	72.5±7.6	$65.5 \pm 9.2$	-9.7	0.01	22	$79.6 \pm 10.6$	$71.6 \pm 10.4$	-10.1	0.01
PA 8	88.1±7.2	$81.4 \pm 9$	-7.6	0.01	10	$74.8 \pm 5.8$	$69.6 \pm 7.2$	-7.0	0.01	18	$80.4 \pm 9.2$	74.5 ± 9.8	-7.3	0.01
Daily Energy Expenditure (kcal/d)														
S	$3,024 \pm 335.6$	$2,799 \pm 335.1$	-7.4	0.1		$2,432 \pm 331,3$	$2,354 \pm 266.4$	-3.2	0.4		$2,621 \pm 430$	$2,496 \pm 352.8$	-4.8	0.11
н	$2,660^{\circ} \pm 317$	$2,935 \pm 247.9$	10.4	0.02		$2,246\pm 285,1$	$2,209 \pm 355.1$	-1.6	0.69		$2,401 \pm 355.5$	$2,482 \pm 476.7$	3.3	0.28
SE	$2,893 \pm 387.8$	$2,872 \pm 506.1$	-0.7	0.84		$2,273 \pm 197.9$	$2,198 \pm 276.9$	-3.3	0.53		$2,628 \pm 443.9$	$2,583 \pm 537$	-1.7	0.57
PA	$2,924 \pm 434.9$	$2,672 \pm 146.4$	-8.6	0.05		$2,403 \pm 245$	$2,264 \pm 343.3$	-5.8	0.18		2,612±415.7	$2,427 \pm 343.9$	-7.1	0.02
Carbolydrate (%)														
S	$41.4 \pm 5.3$	51±7.2	23.2	0.01		$39.1 \pm 7.4$	$43.5 \pm 5.5$	11.3	0.04		$39.8 \pm 6.8$	$45.8\pm6.9$	15.1	0.01
н	$38 \pm 5.5$	$42.7 \pm 5.4$	12.4	0.1		$36.5 \pm 4.9$	$46.8 \pm 7.5$	28.2	0.01		$37 \pm 5.1$	45.4 ± 7	22.7	0.01
SE	$40.3 \pm 7$	44.2±4.6	9.7	0.12		$39.7 \pm 4$	$41.9 \pm 4.6$	5.5	0.38		$40 \pm 5.5$	$43.1 \pm 4.6$	7.8	0.09
PA	$33 \pm 8.7$	$41 \pm 5.8$	24.2	0.01		$37.8 \pm 5.2$	$39.9 \pm 4.7$	5.6	0.44		$35.4 \pm 7.3$	$40.4 \pm 5.1$	14.1	0.01
Protein (%)														
S	$17 \pm 2.5$	$17.2 \pm 0.7$	1.2	0.93		$17.3 \pm 2.6$	$18.9 \pm 2.7$	9.2	0.22		$17.2 \pm 2.5$	$18.4 \pm 2.4$	7.0	0.28
Ш	$17.7 \pm 3.5$	$19.7 \pm 2.8$	11.3	0.23		$16.3 \pm 2.2$	$18.4 \pm 3$	12.9	0.09		$16.8 \pm 2.7$	$18.9 \pm 2.9$	12.5	0.04
SE	$15.7 \pm 2.4$	$17.8 \pm 2.1$	13.4	0.18		$16.3 \pm 2.6$	$19.5 \pm 2.1$	19.6	0.04		$16 \pm 2.4$	$18.6 \pm 2.2$	16.3	0.02
PA	$21.9 \pm 13.5$	$20.7 \pm 2.8$	-5.5	0.49		$16.2 \pm 3.1$	$19.4 \pm 3$	19.8	0.06		$19 \pm 9.9$	$20.1 \pm 2.9$	5.8	0.01
Fat(%)														
S	$36.1 \pm 7.1$	$27.9 \pm 7.9$	-22.7	0.01		$39.1 \pm 5.9$	$34.4 \pm 4.6$	-12,0	0.03		$38.2 \pm 6.3$	$32.4 \pm 6.3$	-15.2	0.01
Е	$38.7 \pm 3.3$	$32.3 \pm 7.7$	-16.5	0.02		$43.9 \pm 5.1$	$30.5 \pm 7.7$	-30.5	0.01		$42.1 \pm 5.2$	$31.1 \pm 7.5$	-26.1	0.01
SE	$39.5 \pm 7.2$	$35 \pm 3.5$	-11.4	0.07		$40.3 \pm 5.5$	$35.2 \pm 3.8$	-12.7	0.04		$39.9 \pm 6.2$	$35.1 \pm 3.6$	-12,0	0.01
PA	$41.1 \pm 7.4$	$34.3 \pm 5.4$	-16.5	0.02		$43.3 \pm 6.3$	$37.5 \pm 5.3$	-13.4	0.04		$42.2 \pm 6.7$	$35.9 \pm 5.4$	-14.9	0.01
Significant difference with baseline $(p \le 0.5)$ S: Strength group; E: Endurance group; S	0.05). *Significant of SE: Strength and end	lifference with di durance group; P.	et and physical a A: Physical activ	ctivity recomm	endations group	oup (C). p ≤ 0.05								

ture (DEE). Table III shows data from daily energy expenditure without the supervised training sessions. No significant changes were found between baseline and post-training values for each group, except to PA group that decreased significantly (p < 0.05). When comparing these data adding the energy expenditure from the training sessions there were not significant differences: S (p = 0.40), E (p = 0.33), SE (p = 0.26).

# Discussion

The main finding of the present study was that adding supervised training to diet restriction did not obtain further improvements on lipid profile versus diet restriction and physical activity recommendations usual in clinical practice.

# Training effects on lipids

# Effects on HDL-C concentration

Measurements on HDL-C in men, after 22 weeks intervention in this study, indicated a mild improvement in S, E and PA groups, and a significant increase in SE (8.5%). Previously, other studies had confirmed improvements in HDL-C with exercise programs, without following a hipocaloric diet.<sup>2</sup> These results suggest that exercise alone could improve HDL-C levels in overweight and obese men, but not diet alone. In contrast, women in our study after the intervention did not improve HDL-C levels as reported by other authors.2 A relevant clinical review exposes that, on average, women have HDL-C levels approximately 10 mg/dL higher than men.<sup>21</sup> This higher initial value may be responsible for women not enhancing HDL-C levels in our study. Possibly, resistance training or endurance training are not an effective stimulus to increase HDL-C in women or subjects with high baseline values.<sup>22</sup> Nevertheless, the importance of increased HDL-C should be emphasized, even though these changes could not appear to be significant. The Framingham Heart Study concluded that for every 1 mg/dL rise in HDL-C levels, there was a 3% reduction in cardiovascular risk.23

# Effects on LDL-C concentration

Measurements on LDL-C levels showed significant greater reductions in SE compared to the rest. A similar study reported significant improvements in LDL cholesterol in both diet-only and diet-plus exercise conditions in men and women.<sup>4</sup> The decrease in body fat percentage and increase in body fat free mass may explain the decrease observed in LDL.<sup>24</sup> A quantitative analysis in the study of Durstine et al. (2001), about adaptation of blood lipids and lipoproteins with contribution of exercise, suggest that LDL-C reductions could occur with intensities of exercise > 60% of maximal heart rate (HRmax).25 Our results are in agreement with this observation except for E women. There are some studies which observed that LDL-C levels increased despite the contribution of exercise and the main reasons that seem to explain this result are related to LDL-C particle size and increased resistance to oxidation of LDL-C. These studies concluded that other factors than concentration must be considered when determining the efficacy of a given intervention. such as exercise training, on LDL.26 On the other hand, our results showed greater improvements in LDL-C in the SE group, in agreement with others, where a combined exercise program demonstrated higher efficacy on LDL-C levels in subjects independently if they were engaged in a diet program.27

# Effects on TG concentrations

The results of the present study exhibit a favorable response of TG levels both in men and women for exercise groups, but only S group obtained significant decreased levels. A recent review and meta-analyses<sup>28</sup> indicated that strength training leads to lower TG concentrations in men and women, independent of changes in body weight or body composition. It seems that strength training improves TG levels better than other exercise modalities but we should take into account also that in our study S group started with worst levels, fact that could have determined the significant changes.29 A positive response of TG levels to exercise compared to diet alone has been observed previously.<sup>30</sup> In addition, it is necessary to refer to studies that show the efficacy of LDL-TG removal from circulation.<sup>31</sup> These suggest that there could be a increase of the triglyceride content of LDL secreted by the liver, which in turn may affect intravascular lipolysis of LDL-triglyceride and LDL-triglyceride plasma clearance, because larger, triglyceride-rich particles are more susceptible to lipolysis by lipoprotein lipase than smaller particles.<sup>31</sup>

# Effects on TC concentration

In our study, TC levels after intervention were maintained or reduced in all groups compared with the data of pretraining time. Significant reductions were occurred in E, SE and PA groups. Several studies about blood lipid and exercise concluded that TC levels infrequently change with exercise training and, as happened with LDL levels, it appears to be an intensity threshold to obtain reductions.<sup>25</sup> Studies with similar protocols to our study<sup>4,5</sup> found no differences between groups, but also achieved significant changes in all groups between baseline and post-training values. On the other hand, diet intervention must be taken into account, following previous study, where plasma TC levels were directly related to total fat intake.<sup>32</sup>. Therefore, reduction in blood TC seems to be attributable in a manner to dietary advice resulting to improvements in fat intake.<sup>33</sup>

# Lipid profile and diet treatment

In our study, macronutrient components in the diet of all participants tended to improve the distribution towards the current recommendations. Even though changes in macronutrient distributions were only significantly different in women of E compared with PA (p < 0.05), general distribution improved in a great manner for all macronutrients in all groups getting closer to the ideal recommendations.<sup>34</sup>

There is considerable clinical evidence about the influence of changes in dietary and caloric intake on plasma lipid and lipoprotein levels.<sup>8</sup> In our study, all groups achieved to lose weight and to improve plasma lipid and lipoprotein concentrations. Our data are in agreement with similar studies<sup>4,5</sup> that presented 10-15% weight losses which obtained with diet alone and/or different exercise protocols and associated with significant improvements in serum lipid profiles.

Findings from accelerometer-measured daily physical activity indicated that there were not significant changes in any group (non-training activity) after 6 months of intervention, except for men in the E and PA (p < 0.05). No differences between groups were found, including PA. Even though participants in the PA group may have tried to engage in different activities following the ACSM recommendations received, it was not enough to increase their lifestyle activity significantly. On the other hand, training groups did not result in a more active lifestyle outside training intervention.

As the flow diagram shows, the PA group showed up with the highest dropouts percentage (37.9 %). Recent studies try to investigate predictive variables for weight loss programs abandons, meaning that is a big matter of concern.<sup>35</sup> Our results showed that supervised exercise did not obtain any additive effects to diet restriction and physical activity recommendations, but it seems that was helpful in sustaining adherence in order to finish the intervention program. Hospital units tend to supervise with often feedback the dietary modifications, but poor counseling in the exercise recommendation is done. When analyzing previous interventions developed to promote changes in diet and/or physical activity in adults it seems to be clear that the key elements to successful behavioral change are frequent contact and support.36,37

Menstruation and ApoE were measured as confounding variables. Previous studies suggest the existence of a variation in the blood lipid levels during the menstrual cycle, with increases in TC, LDL-C and HDL-C during the follicular phase when compared to the luteal phase.<sup>20</sup> On the other hand, ApoE is a glycoprotein that

plays a fundamental role in the lipid metabolism. Many studies assessing the role of ApoE polymorphism on plasma lipids have shown that the presence of the 4 allele is associated with elevations in LDL-C, while the presence of 2 is associated with decreased levels of LDL-C.<sup>38</sup> Therefore, both menstruation and ApoE polymorphism were included in the statistical analysis allowing excluding these variables interference or influencing on the results obtained in this study. Given that there was no statistically significant difference when including menstrual cycle and ApoE, the alterations found after the intervention period would appear to have been caused by the diet and/or exercise program.

Unfortunately, our sample size could have turned to be too small to detect significant changes in all variables due to a higher experimental death than estimated. The sample size of the PRONAF study was calculated to detect a main effect of training and diet on body fat (%) with 80% statistical power at 5% significance, assuming a 0.80 correlation between repeated measures and an estimated percentage of experimental death. The strengths of the present study include the randomized-controlled design, the long supervised training period and the lifestyle and genetic factors controlled.

We also assumed that, as other studies suggested previously,<sup>39</sup> supervised training protocols may have not achieved enough intensity in order to obtain significant improvements versus diet and unsupervised regular physical activity recommendations. These could be due also because the baseline lipid profile concentrations in this study were considered as "low risk" according to the guidelines published by the expert panel report,40 and it is known that better baseline values determine post-intervention changes.<sup>29</sup> Moreover, baseline macronutrient distribution of the subjects' diet was far away from recommended guidelines.<sup>34</sup> This means that a considerable lifestyle change was made in this way, dietary intake changed in all groups in great manner and diet adherence was also high. Although dieting alone generally achieves beneficial results, regular exercise add salutary effects and therefore both strategies support counseling by health practitioners.41

In conclusion, an intervention program of endurance, strength or combined supervised training protocol with diet restriction did not achieved further improvements in lipid profile than diet restriction and usual physical activity recommendations developed in clinical practice in overweight men and women. Future research is required in order to investigate if higher intensity of any supervised training protocol mode can add improvements to dietary modification.

# Statement of authorship

The contributions of the authors to the manuscript are as follows. EMM: study design, data collection,

data analysis and writing of the manuscript; BRM, PJB and ABP.: study design and data collection; MGG, CGC and CFF reviewing the manuscript; PJB: study design and reviewing the manuscript. All authors read and approved the manuscript.

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#### **Conflict of interest statement**

The authors have no conflicts of interest.

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