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Beef and soy-based food supplements differentially affect serum lipoprotein-lipid profiles because of changes in carbohydrate intake and novel nutrient intake ratios in older men who resistive-train☆

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Abstract

Objective: We examined if the predominant source of dietary protein influenced the lipoprotein-lipid profile in older men who performed resistive exercise training (RT).
Design: This is a 14-week, randomized, repeated-measures study with a 12-week period of RT with supplementation of different sources of dietary protein (beef and soy).
Setting: Nutrition, Metabolism, and Exercise Laboratory, Central Arkansas Veteran's Healthcare System, North Little Rock, Ark.
Subjects: Twenty-six healthy men were recruited, and 21 men (age 65 ± 5 years, body mass index 28.2 ± 2.6 kg/m^2) completed the study.
Interventions: For 14 weeks, all men were counseled to self-select a lacto-ovo-vegetarian diet. For 2 weeks (baseline), all men also consumed 0.6 g-protein/kg per day from portioned quantities of soy-based texturized vegetable protein foods. For the next 12 weeks, 11 men were randomized to continue with texturized vegetable protein foods (VEG group), whereas 10 men were randomized to receive 0.6 g-protein/kg per day from portioned quantities of beef (BEEF group) and continue their otherwise lacto-ovo-vegetarian diet. All men participated in RT 3 d/wk during this 12-week period. Assessments of upper and lower body muscle strength and power, serum lipoprotein-lipid profile, and dietary nutrient intakes were made at baseline and week 12 of RT (POST).
Results: The BEEF and VEG groups increased ($P < .05$) overall muscle strength and muscle power with RT, with no differences between groups. From baseline to POST, the BEEF group had increased concentrations of high-density lipoprotein cholesterol ($P = .025$; HDL-C), low-density lipoprotein cholesterol ($P = .027$; LDL-C), and total cholesterol ($P = .015$: CHOL), with no changes ($P > .05$) in triacylglycerol (TG), the CHOL/HDL-C ratio, or the TG/HDL-C ratio. The VEG group did not experience within-group changes ($P > .05$) in any lipoprotein-lipid parameter. At POST, the concentrations of HDL-C, LDL-C, and CHOL were greater in the BEEF group compared with the VEG group. There were significant interaction effects for HDL-C ($P = .004$) and the TG/HDL-C ratio ($P = .022$). Multiple regression analysis determined that, regardless of intervention, change in the saturated fat/fiber ratio (SF/fiber) predicted CHOL (adjusted $R^2 = 0.34$); the SF/fiber ratio predicted LDL-C (adjusted $R^2 = 0.36$); the cholesterol/fiber intake ratio predicted HDL-C (adjusted $R^2 = 0.26$), and the change in carbohydrate intake predicted the CHOL/HDL-C ratio (adjusted $R^2 = 0.37$) and TG (adjusted $R^2 = 0.44$).
Conclusions: These results suggest that the lipoprotein-lipid profile in these older men was differentially affected by supplementation with beef versus soy-based foods during RT. Regardless of group, the lipoprotein-lipid changes were predicted by differences in the SF/fiber ratio and cholesterol/fiber ratio and increases in carbohydrate intake over time.

1. Introduction

It is well established that muscle mass, strength, and power decrease with advancing age in adult human beings [1]. Age-associated sarcopenia and decreased muscular function are associated with physical disability and a decrease in functional capacity [2]. Older adults must

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consume adequate dietary protein to minimize diet as a contributing factor to sarcopenia [3-5]. Moreover, it has been suggested that protein from meat sources enhances net protein synthesis relative to soy protein [6,7].

Protein intake from different sources might confer differential health attributes because of the nonprotein constituents in the respective foods. Other nutrients that tend to be ingested in greater quantities in diets containing red meat are saturated fat and cholesterol [8]. High intakes of saturated fat are associated with an increased risk for cardiovascular [8] and diabetic complications [9,10]. From an epidemiological perspective, meat intake was positively associated with serum cholesterol concentrations [8], and the risk of acquiring cardiovascular disease was associated with lipoprotein levels [11,12]. Conversely, recent data demonstrate that increasing saturated fat intake and decreasing fiber intake, via a low-carbohydrate diet, decrease the risk of cardiovascular disease and insulin resistance via decreased triacylglycerol (TG) levels and a decrease in the TG/high-density lipoprotein cholesterol (HDL-C) ratio [13], which is an indicator of insulin resistance [14].

To attenuate sarcopenia and improve functional capacity, it is recommended that older adults engage in strength or resistive exercise training (RT). Relative to lipoproteins, RT has yielded equivocal results [15,16] but has been shown to increase levels of HDL-C and improve the lipoprotein-lipid profile including the total cholesterol (CHOL) to HDL-C ratio (CHOL/HDL-C) [17-19]. However, it has yet to be determined whether RT with a controlled diet containing differing sources of protein (and other intrinsic nutrients) differentially affects lipoprotein-lipid levels. We previously found that RT in conjunction with controlled beef intake did not differentially affect body composition or improvements in muscle hypertrophy [20] but did differentially affect iron-dependent hematological indices [21]. Therefore, the objective of this portion of the study was to determine whether the controlled beef intake, compared with texturized vegetable protein (TVP), affected the lipoprotein-lipid profile in older men after an otherwise lacto-ovo-vegetarian (LOV) diet and 12-week RT program. To determine whether dietary factors besides protein (eg, saturated fat and fiber) predicted the lipoprotein-lipid results, nutrient intake was also assessed.

2. Subjects and methods

2.1. Subjects

Twenty-six men were recruited, provided written informed consent in accord with the institutional review board at the University of Arkansas for Medical Sciences before participating, and were enrolled into the study. Results of a prestudy physical examination, written medical history, resting electrocardiogram, and clinical blood and urine chemistries were used to exclude men with medical conditions that might place them at risk for participating in or completing the study. A total of 21 men completed the protocol (age 65 ± 5 years, height 178 ± 6 cm, weight 89.2 ± 7.4 kg, body mass index 28.2 ± 2.6 kg/m^2).

2.2. Experimental design and dietary control

The 14-week protocol started with a 2-week period of dietary control (baseline), followed immediately by a 12-week period of RT with the dietary control continuing throughout RT. Testing was completed at week 2 of baseline and week 12 of RT (POST).

During baseline, all subjects were asked to consume portioned quantities of soy-based TVP products that provided 0.6 g-protein/kg · per day and were counseled by a registered dietitian to self-select the remainder of their weight-maintenance diet from LOV food and beverage choices. The TVP products, individually weighed to the nearest 0.1 g, included breakfast patties, grillers, chik patties, and veggie dogs from Morningstar Farms (Worthington Foods, Inc, Worthington, Ohio). Throughout the 12-week period of RT, all men continued the counseling-based selection of the LOV diet. Based on randomization, 11 of the men were assigned to continue eating the 0.6 g-protein/kg per · day of TVP products (VEG group), whereas the other 10 men were assigned to consume 0.6 g-protein/kg · per day from beef-containing foods (cube steak, ground beef, and beef tips; BEEF group) instead of the TVP products, with the remainder of their diet coming from LOV foods. The daily portions of the TVP or beef foods were distributed to the subjects on a weekly basis in frozen form. Each subject cooked and prepared these foods based on personal preference. Throughout the dietary intervention, the subjects were routinely asked about their adherence to the diet and counseled to adjust, if necessary, their self-selected energy intake based on changes in their body weight (measured before each exercise session, 3 d/wk) to remain weight-stable.

During the baseline and POST testing periods, all of the men recorded what they consumed, including the supplemented food products over 24 hours for 3 consecutive days (1 weekend day and 2 weekdays) to assess total energy, macronutrient, and micronutrient intakes. These diet records were analyzed using Nutritionist V (N-Squared Computing, First Data Bank, Inc, San Bruno, Calif) software. One subject forgot to record his diet on 1 weekend day during the last week of the study.

2.3. Resistive training

After baseline testing, all of the men began a 12-week RT program as previously described [20]. One-repetition maximum (1-RM) testing was done at the first exercise session to establish the resistance to be used in subsequent exercise sessions. Training sessions were performed 3 d/wk on nonconsecutive days. The exercises consisted of unilateral seated leg extension and flexion, bilateral leg press, arm pull, and seated chest press on pneumatically adjusted resistance exercise equipment (K400, Keiser, Inc, Fresno, Calif). The performances on the leg exercises were combined to determine lower body strength, and the arm exercises were combined to determine upper body strength. The subjects performed a warm-up set, then performed 2 sets of 8 repetitions at 80% of their 1-RM and then completed a third set to voluntary fatigue. The resistance for subsequent sessions was increased by 5% to 10% when 12 or more repetitions were performed during the third set. The 1-RM procedure was repeated on the final day of the RT program. All subjects completed at least 31 of the 33 possible exercise sessions. These results have been previously published [20].

Each subject was tested for upper body (arm pull) and lower body (knee extension) power output (Newton per second for upper body and Newton meter · per second for lower body) using a previously described protocol [22]. Briefly, each subject performed 3 maximal repetitions at 4 intensities (20%, 40%, 60%, and 80% of 1-RM) relative to their 1-RM at the time of testing. The average of the 3 movements at each intensity was used as the value for that intensity (upper body coefficient of variation [CV] = 4.5%, lower body CV = 5.7%).

2.4. Blood collection and analyses

At baseline and POST, each man arrived at the laboratory between 6:30 and 7:00 AM after a 10-hour overnight fast. Blood was collected between 8:00 and 8:30 AM into evacuated tubes with serum separator tube (SST) gel to be analyzed for serum lipoprotein and lipid concentrations. Samples were allowed to clot at room temperature before centrifugation. Samples were analyzed at the John C. McClellan Veterans Administration Hospital (Little Rock, Ark) for total cholesterol (CHOL), HDL-C, and triacylglycerol (TG). The analyzer (Dimension RXL, Dade-Behring, Inc, Newark, Del) subsequently calculated the low-density lipoprotein cholesterol (LDL-C) using the following equation [23]: LDL-C = CHOL − HDL-C − (TG/5).

2.5. Statistical analyses

Unless otherwise noted, values are expressed as mean ± SD. Time (baseline and POST), group (VEG and BEEF), and interaction (group × time) effects were assessed using a repeated-measures analysis of variance. Independent *t* tests were used to determine between-group differences, and paired *t* tests were used to assess within-group differences for each variable at baseline and POST. Linear multiple regression analyses were performed using lipoprotein-lipid values as the dependent variables with descriptive data, body composition data, and dietary data entered in a stepwise fashion. The analysis of variance and mean comparisons were completed using Statistica computer software (Statsoft, Tulsa, Okla), whereas the multiple regression analyses were completed using SPSS (v11.5, Chicago, Ill). Statistical significance was set at $P \leq .05$.

3. Results

3.1. Diet

There were no differences in reported intakes of energy, carbohydrate, fat, protein, cholesterol, fiber, or zinc between groups at baseline (Table 1). Compared with baseline, the BEEF group reported increased intakes of saturated fat, cholesterol, and zinc and a decreased consumption of fiber at POST. The BEEF group ingested a greater (P = .001) percent of their fat from saturated fat (37% ± 2%) relative to the VEG group (28% ± 7%) at POST. The reported intakes of zinc and fiber at POST for the BEEF group were different than the VEG group. The reported nutrient intakes for the VEG group did not change from baseline to POST.

3.2. Lipoprotein-lipid profile

At baseline, there were no differences between groups for any of the lipid or lipoprotein variables (Table 2). The BEEF group experienced increases in HDL-C, LDL-C, and CHOL after the 12-week diet and RT interventions. There were no changes from baseline to POST in VEG for any of

Table 1
Reported nutrient intakes after the initial 2-week lacto-ovo-vegetarian diet (baseline) and after the 12-week RT program (POST)

	Baseline	POST	*P* value[a]
Total energy intake (MJ/d)			.789
VEG	9.37 ± 1.8	9.33 ± 1.4	
BEEF	9.09 ± 2.1	9.07 ± 2.3	
Protein (g/kg per day)			.225
VEG	1.17 ± 0.1	1.15 ± 0.1	
BEEF	1.10 ± 0.2	1.03 ± 0.3	
Carbohydrate (g/d)			.338
VEG	274 ± 67	266 ± 52	
BEEF	282 ± 79	238 ± 71[b]	
Fat (g/d)[c]			.215
VEG	85 ± 26	79 ± 29	
BEEF	73 ± 21	95 ± 27[b]	
Cholesterol (mg/d)			.017
VEG	217 ± 146	250 ± 199	
BEEF	254 ± 164	326 ± 153[d]	
Saturated fat (g/d)[c]			.026
VEG	26 ± 11	23 ± 11	
BEEF	22 ± 11	34 ± 11[b,d]	
Fiber (g/d)			.000
VEG	32 ± 8	27 ± 6	
BEEF	26 ± 4	14 ± 6[b,d]	
Saturated fat (g) to fiber (g) ratio			.001
VEG	0.9 ± 0.4	1.0 ± 0.6	
BEEF	0.9 ± 0.6	3.0 ± 1.4[b,d]	
Cholesterol (mg) to fiber (g) ratio			.006
VEG	9.0 ± 6.0	8.0 ± 5.1	
BEEF	9.0 ± 6.5	32.8 ± 23.1[b,d]	
Zinc (mg/d)			.000
VEG	6 ± 2	7 ± 4	
BEEF	5 ± 2	13 ± 3[b,d]	

Mean ± SD.
[a] *P* value represents between-group differences at POST.
[b] Significantly different from baseline value.
[c] Significant interaction (group × time) effect.
[d] Significantly different from VEG at same time point.

the lipid or lipoprotein variables, although there was a trend ($P < .10$) for decreased HDL-C and increased CHOL/HDL-C ratio. As for a differential effect of the diet intervention, there were group × time interaction effects for HDL-C ($P = .004$) and the TG/HDL-C ratio ($P = .022$). Aside from interaction effects, there were no differences between or within groups for TG, TG/HDL-C ratio, or the CHOL/HDL-C ratio.

The multiple regression analyses revealed that several dietary intake parameters predicted the lipoprotein-lipid outcomes (Table 3). Total serum cholesterol levels were only predicted by the change in the saturated fat/fiber intake (SF/fiber) ratio from baseline to POST. The SF/fiber ratio was also determined to be the best predictor of LDL-C levels. HDL-C was only predicted by the change in the cholesterol/fiber intake ratio from baseline to POST. TG concentrations and the CHOL/HDL-C ratio were both predicted by the change in carbohydrate intake from baseline to POST.

3.3. Strength

There were no differences at baseline between groups for lower body (VEG = 1633 ± 230 Nm; BEEF = 1508 ± 361 Nm) or upper body (VEG = 1074 ± 100 N; BEEF = 1060 ± 204 N) strength. Both groups increased lower body (VEG = 1958 ± 170 Nm; BEEF = 1900 ± 381 Nm) and upper body (VEG = 1248 ± 137 N; BEEF = 1283 ± 150 N) strength after the 12-week RT program. There were no differences in strength gains between groups over time.

Table 2
Changes in serum lipoprotein-lipid profile during 12 weeks in older men consuming self-selected vegetarian diets supplemented with texturized vegetable protein (VEG) or beef (BEEF)

	Baseline	POST	P value[a]	Δ[b]	P value[c]
CHOL (mmol/L)					
VEG	4.51 ± 0.8	4.45 ± 0.9	0.797	−0.06 ± 0.7	.160
BEEF	5.11 ± 0.6	5.42 ± 0.6[d,e]	0.015	0.30 ± 0.3	
HDL-C (mmol/L)[f]					
VEG	1.16 ± 0.3	1.06 ± 0.2	0.053	−0.10 ± 0.1	.004
BEEF	1.17 ± 0.2	1.24 ± 0.3[d,e]	0.025	0.08 ± 0.1[e]	
LDL-C (mmol/L)					
VEG	2.88 ± 0.8	2.80 ± 0.8	0.685	−0.08 ± 0.6	.102
BEEF	3.35 ± 0.5	3.65 ± 0.5[d,e]	0.027	0.30 ± 0.4	
CHOL/HDL-C ratio					
VEG	4.08 ± 1.0	4.41 ± 1.4	0.069	0.33 ± 0.6	.098
BEEF	4.57 ± 1.2	4.51 ± 1.0	0.613	−0.07 ± 0.4	
TG (mmol/L)					
VEG	1.03 ± 0.5	1.30 ± 0.8	0.164	0.27 ± 0.6	.078
BEEF	1.31 ± 0.6	1.15 ± 0.4	0.301	−0.16 ± 0.5	
TG/HDL-C ratio[f]					
VEG	0.99 ± 0.6	1.35 ± 1.0	0.095	0.36 ± 0.6	.022
BEEF	1.19 ± 0.6	0.99 ± 0.5	0.108	−0.20 ± 0.4[e]	

Mean ± SD; $P < .05$.
[a] P values for within-group comparisons.
[b] Change, POST − baseline.
[c] P values for comparisons of change between groups.
[d] Significant difference from baseline.
[e] Significantly different from VEG.
[f] Significant interaction effect.

Table 3
Multiple regression analysis to determine the independent variable that best predicts the posttraining serum lipid-lipoprotein levels for all subjects

	Adjusted R^2	P value
Total cholesterol		
SF/fiber ratio change	0.34	.004
LDL-C		
SF/fiber ratio	0.36	.003
HDL-C		
Cholesterol/fiber change	0.26	.022
TG		
Carbohydrate change	0.37	.006
Cholesterol/HDL-C ratio		
Carbohydrate change	0.44	.002
TG/HDL-C ratio		
Carbohydrate change	0.42	.002

n = 20; List of variables in analysis: age, body fat (kg), energy (MJ), carbohydrate (g), protein (g), fiber (g), total fat (g), saturated fat (g), cholesterol (mg), zinc (mg), SF/fiber ratio, cholesterol/fiber ratio, and change from baseline (POST − baseline) for each.

3.4. Power output

There was no difference between groups for upper body or lower body power output at baseline or POST. The BEEF group experienced increases in upper body power output at 20%, 60%, and 80% of 1-RM, whereas the VEG group experienced increased power at each workload. For lower body power output, the BEEF group increased power output at 20%, whereas the VEG group increased power output at 20% and 40% of 1-RM.

4. Discussion

The results from this randomized clinical trial support the results from studies that have investigated the effects of meat intake on nutrient intake [24], the association of meat intake on lipoprotein-lipid concentrations [8,25], and a weight loss study that observed decreased TG levels and the TG/HDL-C ratio with higher intakes of saturated fat [13]. Contrary to recommendations pertaining to high–saturated fat intake and low-fiber intake, beef intake compared with TVP intake may decrease the risk for developing or progressing of the metabolic syndrome given the increase in HDL-C levels and the interaction effects for HDL-C and TG/HDL-C ratio [13,14]. Furthermore, although CHOL and HDL-C levels increased over time in the BEEF group, this did not result in a change in the CHOL/HDL-C ratio. This is important as the CHOL/HDL-C ratio has been suggested as a better predictor of cardiovascular disease risk than individual lipoprotein values [26]. Thus, the rise in HDL-C levels and decrease in the TG/HDL-C ratio in the BEEF group may offset the health risks associated with individual increases in CHOL and LDL-C levels. With regard to the metabolic syndrome, the BEEF group decreased their risk relative to HDL-C, as it increased in this group. As for the VEG group, consuming a LOV diet did not change lipoprotein-lipid levels in these men, but significant interaction effects were present indicating potential health risk differences.

With regard to the differences in fiber and saturated fat intake and their influence on the lipoprotein-lipid profiles, it appears that the ratio of each may be more critical than the absolute amounts of each as the individual variables did not predict any of the lipoprotein-lipid values. Consequently, we derived 2 novel ratios (SF/fiber ratio and the cholesterol/fiber ratio) that predicted CHOL, LDL-C, and HDL-C levels. However, studies employing these ratios in relation to morbidity and mortality are needed to validate whether this approach is justifiable from a public health perspective. This is a very significant issue given the current deliberations pertaining to low-carbohydrate diets and the likely increased intake of saturated fat and decreased intake of fiber that tends to occur with these popular diets.

The supplemented beef products contained less fiber (0 g of fiber/100 g of beef) than the vegetable protein products (4-6 g of fiber/100 g of product)—an 89-kg subject would have consumed 248 g/d of beef (0 g of fiber) on average and 292 g/d of TVP (14 g of fiber). Hence, the observed changes in CHOL and LDL-C levels for the BEEF group were expected [8], and the increased TG/HDL-C ratio is supported by a weight loss study using a higher fat and protein diet [13]. However, based on the regression results, it seems that fiber and saturated fat intake were not important individual dietary components, as only the ratio of saturated fat intake to fiber intake predicted CHOL and LDL-C levels. Likewise, an observation relative to current low-carbohydrate diet trends was that the change in carbohydrate intake predicted both TG levels and the CHOL/HDL-C ratio at the end of the study. This demonstrates that those who consumed less carbohydrate over time, regardless of intervention group, were inclined to have decreased TG levels, TG/HDL-C ratio, and CHOL/HDL-C ratio, suggesting a decreased risk for cardiovascular and metabolic diseases [14,26]. Of clinical relevance, the increase in HDL-C levels and the decreased TG/HDL-C ratio in the BEEF group, relative to the VEG group responses, occurred without weight loss, indicating that changes in lifestyle alone decrease specific diagnostic criteria for the metabolic syndrome.

The alteration in the intakes of zinc between groups was expected, as beef contains larger quantities of this nutrient than the TVP products. Although zinc intake has been shown to be positively associated with LDL-C [27], zinc intake was not selected as a strong predictor of any of the lipoprotein-lipid values in the present study.

The changes in strength and power output observed are supported by others who reported similar improvements in these variables in older men [22,28,29]. Based on the present results, improvements in strength and power were similar during the 12 weeks of RT regardless of the source of dietary protein. This is contrary to the hypothesis, which was based on previous results demonstrating that protein metabolism was attenuated soy protein [6,7]. Thus, a lacto-ovo-vegetarian diet is capable of achieving equal gains in muscle performance; however, further research with vegetarian diets is needed to verify the contribution of whey, casein, and egg-derived proteins on net muscle balance and muscle hypertrophy. This is critical because proteins from dairy foods or eggs may have compensated for the potential decreased net protein balance caused by the soy isoflavones genistein and/or genistin [7].

In conclusion, the incorporation of beef, at a level of 0.6 g-protein/kg · per day, into the diets of resistive-training older men consuming an otherwise LOV diet, leads to changes in lipoprotein-lipid concentrations and intakes of selected nutrients with no change in the CHOL/HDL-C ratio. The interaction effects for HDL-C and TG/HDL-C indicate that beef intake decreased risk factors for the metabolic syndrome compared with similar intakes of TVP supplements. The lower lipoprotein-lipid levels were predicted by decreased carbohydrate intake over time and increased animal-based nutrients (saturated fat and cholesterol) to fiber intake ratios. The improvements in upper and lower body strength and power were not different between groups, indicating that both diets are equally effective at increasing muscle strength and power output.

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