

A Reduced Ratio of Dietary Carbohydrate to Protein Improves Body Composition and Blood Lipid Profiles during Weight Loss in Adult Women^{1,2}

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ABSTRACT Claims about the merits or risks of carbohydrate (CHO) vs. protein for weight loss diets are extensive, yet the ideal ratio of dietary carbohydrate to protein for adult health and weight management remains unknown. This study examined the efficacy of two weight loss diets with modified CHO/protein ratios to change body composition and blood lipids in adult women. Women ($n = 24$; 45 to 56 y old) with body mass indices $>26 \text{ kg/m}^2$ were assigned to either a CHO Group consuming a diet with a CHO/protein ratio of 3.5 (68 g protein/d) or a Protein Group with a ratio of 1.4 (125 g protein/d). Diets were isoenergetic, providing 7100 kJ/d, and similar amounts of fat ($\sim 50 \text{ g/d}$). After consuming the diets for 10 wk, the CHO Group lost $6.96 \pm 1.36 \text{ kg}$ body weight and the Protein Group lost $7.53 \pm 1.44 \text{ kg}$. Weight loss in the Protein Group was partitioned to a significantly higher loss of fat/lean ($6.3 \pm 1.2 \text{ g/g}$) compared with the CHO Group (3.8 ± 0.9). Both groups had significant reductions in serum cholesterol ($\sim 10\%$), whereas the Protein Group also had significant reductions in triacylglycerols (TAG) (21%) and the ratio of TAG/HDL cholesterol (23%). Women in the CHO Group had higher insulin responses to meals and postprandial hypoglycemia, whereas women in the Protein Group reported greater satiety. This study demonstrates that increasing the proportion of protein to carbohydrate in the diet of adult women has positive effects on body composition, blood lipids, glucose homeostasis and satiety during weight loss. *J. Nutr.* 133: 411–417, 2003.

KEY WORDS: • obesity • body fat • blood lipids • insulin

Obesity is an important public health concern in the United States (1). Accumulating body fat is associated with the onset of diverse health risks including type 2 diabetes, cardiovascular disease, cancer and osteoarthritis (2). Although obesity is recognized as a disorder of energy balance, causes and solutions remain elusive. At the center of the debate about controlling obesity is the optimal balance of macronutrients for adult health.

To maintain or reduce body weight, diets must control energy intake. Diets high in fat are assumed to contribute to obesity because they are usually highly palatable and energy dense. Current dietary guidelines advocate a daily intake of macronutrients with carbohydrates accounting for $\geq 55\%$ of dietary energy, fats limited to $\leq 30\%$ of dietary energy, and protein at $\leq 15\%$ of energy (3–5). However, the putative balance of dietary fat, carbohydrates and protein leading to

obesity has been challenged by evidence from epidemiologic (6,7), clinical (8–10) and experimental studies (11–14). These researchers report that high carbohydrate diets reduce oxidation of body fat (11,12), increase blood triglycerides (8,10,13) and reduce satiety (14). These reports raise new questions about the ideal ratios of macronutrients to balance energy needs for adults.

Generally, the debate about an optimal ratio of macronutrients for adults focuses on carbohydrates vs. fat; however, there are increasing questions about the role of protein in the adult diet (7,8,15). Three independent groups reported beneficial effects on body composition and blood lipids derived from direct substitution of protein for carbohydrates in adult diets (8–10). Although these researchers found positive effects of increasing dietary protein and reducing carbohydrates, the reports lack a fundamental hypothesis to explain the metabolic need for protein above current recommended dietary allowances (RDA)⁴ levels. In a companion paper (16), we proposed that an optimal ratio of carbohydrate to protein could be evaluated on the basis of a theory of glucose homeostasis.

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⁴ BMI, body mass index; BHB, β -hydroxybutyrate; CHO, carbohydrate; DXA, dual X-ray absorptiometry; GNG, gluconeogenesis; HDL-C, HDL cholesterol; LDL-C, LDL cholesterol; TAG, triacylglycerol; T4, thyroxine; T3, triiodothyronine.

Under conditions of high CHO intake (55% of energy), the body must adapt to the disposal of large quantities of dietary glucose and relies on insulin to manage changes in blood glucose. Under conditions of lower carbohydrate (CHO) intake (<200 g/d), the body relies on hepatic production to maintain blood glucose. Glucose released from the liver can be derived from glycogenolysis and gluconeogenesis (GNG); however the larger component is GNG, with dietary amino acids providing the primary source of carbon substrates (17,18). Applying this hypothesis to a weight loss study, we found that obese women consuming a diet with a CHO/protein ratio <1.5 for 10 wk minimized fasting and postprandial changes in blood glucose and enhanced insulin sensitivity (16). These findings provide a conceptual basis on which to test the efficacy of diets with reduced ratios of CHO/protein.

The objective of this study was to compare the effects of two reduced energy diets with modified ratios of protein and carbohydrates on changes in body weight, body composition, blood lipids and satiety. This study used a 10-wk, highly controlled nutrition protocol with adult women to evaluate the effects of weight loss diets with moderate differences in protein and carbohydrates.

SUBJECTS AND METHODS

Women ($n = 24$; 45 to 56 y old) with body weights >15% above ideal body weight were recruited to participate in a weight loss study as previously described (16). All subjects participated in a baseline evaluation period that included a 3-d weighed dietary record and measurement of body composition, blood lipids, glucose and insulin. This period served as an initial control period for each subject. After the baseline evaluation, subjects were divided into two groups ($n = 12$) based on age (50.1 ± 1.1 y), body weight (85.2 ± 3.6 kg) and body mass index (BMI; 30.3 ± 1.0 kg/m²).

One group of 12 women was assigned to a moderate protein diet (Protein Group) designed to provide dietary protein at 1.6 g/(kg · d) with a CHO/protein ratio of ~1.4 and dietary lipids at <30% energy intake. The second group was assigned to a high carbohydrate diet (CHO Group) designed to provide dietary protein at 0.8 g/(kg · d) with a CHO/protein ratio >3.5 and dietary lipids at <30% energy intake. The two diets were designed to be equal in energy (~7100 kJ/d; 1700 kcal/d), total fat intake (~50 g/d) and fiber (~20 g/d). With these general criteria, we developed a 2-wk menu plan for each group with meals for each day meeting established nutritional requirements and lipid guidelines for the STEP I diet. The dietary differences between groups were designed to reflect a direct substitution of foods in the protein groups (meats, milk, cheese, eggs and nuts) for foods in the refined grain/starch groups (breads, rice, cereals, pasta and potatoes). For the CHO Group, the diet followed the guidelines of the USDA Food Guide Pyramid diet (4), which emphasizes the use of breads, rice, cereals and pasta. For the Protein Group, the diets substituted foods from the protein groups that emphasized animal proteins including combinations of red meats, milk, cheese and eggs with a requirement for a minimum of seven beef-containing meals each week. Both diets included extensive use of vegetables (5 to 6 servings/d). Physical activity was monitored with written questionnaires and was kept constant for each subject throughout the study.

The overall experimental design included a 1-wk Initial Control period providing baseline data for all subjects about their usual dietary patterns. The baseline period was followed by a 10-wk diet study that consisted of an initial 4 wk of a highly controlled diet with subjects receiving all food in our food research laboratory followed by 6 wk with subjects continuing to follow the 2-wk diet rotation at home. During the first 4 wk of the study, all food was prepared in the food research laboratory and all meals were weighed by the research staff and also by the subjects to evaluate reliability and reproducibility of the subject weighed food records. During the laboratory-based diet period, subjects also received daily instruction by a research dietitian about the menus, food substitutions, portion sizes and procedures for

maintaining weighed diet records. During the final 6 wk of the study, subjects continued to use the 2-wk menu rotation at home. Each week, subjects were required to report to the research laboratory for measurement of body weight and to review their 3-d food records with the dietitian.

At times 0, 2, 4 and 10 wk, body composition, blood and urinary chemistries, and food records were evaluated. Measurements included body weights by electronic scale, body composition by dual X-ray absorptiometry (DXA), plasma glucose, insulin, thyroid hormones (T4, thyroxine and T3, triiodothyronine), urea, β -hydroxybutyrate (BHB) and lipid profiles and urinary acetoacetate and urea. Plasma glucose was analyzed by a glucose oxidase-peroxidase automated method (YSI model 2300 analyzer, Yellow Springs Instruments, Yellow Springs, OH). Insulin and thyroid hormones were determined by commercial RIA kits (07-26102 and 06B254221, respectively, ICN Pharmaceuticals, Costa Mesa, CA). Serum total cholesterol, HDL cholesterol (HDL-C) and triacylglycerols (TAG) were determined by standardized methods (19) by the Washington University School of Medicine Core Laboratory for Clinical Studies (St. Louis, MO). LDL cholesterol (LDL-C) was calculated using the Friedewald equation (20). Plasma urea nitrogen and urinary urea were determined with a blood urea nitrogen (BUN) kit (Sigma, St. Louis, MO), BHB was determined by enzymatic conversion to acetoacetate with measurement of NADH (Ketone Kit, Sigma) and urinary acetoacetate was tested with a clinical urine stick (Sigma). Nutrient intakes were evaluated as mean daily intakes from the 3-d weighed records using Nutritionist V software (First DataBank, San Bruno, CA). Individual feelings and satisfaction were evaluated with a questionnaire completed on d 3 of the 3-d food records throughout the study. The questionnaire contained 11 quantitative questions with a 7-point response range addressing feelings about satiety, energy level and hunger.

On measurement days, subjects reported to the research facility at 0700 h after a 12-h overnight fast. Body weights were determined and blood samples were drawn. Then subjects were given a standard breakfast meal providing 1670 kJ (400 kcal) with the macronutrient composition designed to reflect the respective diet treatments (e.g., Protein Group received 33 g protein, 39 g carbohydrate and 13 g fat; CHO Group received 10 g protein, 57 g of carbohydrate and 12 g fat). Subjects had a postprandial blood sample drawn 2 h after completion of the meal.

Data were evaluated using a one-way ANOVA with repeated measures with diet treatment and time as independent variables (SAS Institute, Cary, N.C.). When significant treatment or treatment \times time effects were observed ($P < 0.05$), differences were further evaluated using Fisher's Least Significant Difference test to determine differences between diet treatments or differences within each diet treatment over time. A paired t test was used for fasting vs. postprandial comparisons. Values are means \pm SEM.

RESULTS

The experimental diets provided similar intakes of energy, fat and fiber (Table 1) and produced weight loss in both groups (Table 2). The protein intake for the high protein diet averaged 125 g/d [~ 1.5 g/(kg · d)] with carbohydrate intake of 171 g/d. The CHO Group received an average of 68 g of protein per day [0.8 g/(kg · d)] and 239 g of carbohydrate. The relative proportions of energy in the Protein Group were 30% protein, 41% carbohydrate and 29% fat with a ratio of carbohydrate to protein (CHO/protein) of 1.4, whereas the proportion in the CHO Group was 16% protein, 58% carbohydrate and 26% fat with a ratio of 3.5. The Protein Group consumed 239 ± 8 mg/d of cholesterol, whereas the CHO Group consumed 115 ± 5 mg/d. Saturated fatty acid intake was higher in the Protein Group, but both groups reduced their intakes of saturated fatty acids compared with the initial baseline period.

Body weight changes did not differ between the groups (Table 2). After consuming the diets for 10 wk, the Protein Group had a total weight loss of 7.53 ± 1.44 kg and the CHO Group lost 6.96 ± 1.36 kg.

Changes in body composition indicated that the weight loss was predominately body fat (Table 2). After 10 wk, women in the Protein Group lost 5.60 ± 0.52 kg of body fat or 14.4% of initial body fat, whereas women in the CHO Group lost 4.74 ± 0.65 kg of body fat or 12.2% of initial body fat. Loss of lean body mass tended to be greater for the women in the CHO Group (1.21 ± 0.58 kg) compared with those in the Protein Group (0.88 ± 0.33 kg) ($P = 0.07$). When changes in body composition were expressed as a ratio of fat/lean loss (Fig. 1), weight loss was targeted progressively to body fat in both groups. The ratio of fat/lean loss demonstrated that the higher protein diet partitioned a significantly greater percentage of the weight loss to body fat while sparing lean tissue. Specifically, the Protein Group achieved a fat/lean loss of 6.36 ± 0.85 at 10 wk compared with the CHO Group with a fat/lean loss of 3.92 ± 0.79 ($P < 0.05$).

Thyroid hormones in the two groups responded differently. Throughout the study, plasma T4 concentrations (Fig. 2A) increased in both groups from initial baseline values, whereas the energy restriction decreased plasma T3 levels in both groups. T3 levels differed between the two groups at wk 2 and 4 with the Protein Group having a greater concentration during weight loss. Although T3 levels declined, the precursor T4 levels increased with the increase at wk 10 greater in the Protein Group.

Both weight loss diets produced changes in serum lipids (Table 3). The initial values indicated that women assigned to the Protein Group had baseline values for total cholesterol and LDL-C that were significantly greater than those of the CHO Group. HDL-C and TAG did not differ between groups. Changes in blood cholesterol associated with the dietary treatments reflected the energy restriction and weight loss with significant decreases in both total cholesterol and LDL-C. After the first 4 wk of the treatments with food intake monitored in the research facility, total cholesterol was decreased in the Protein Group by 10.0% (0.58 mmol/L) and LDL-C by 10.5% (0.40 mmol/L). Similarly, total cholesterol was decreased by 11.2% (0.55 mmol/L) in the CHO Group and LDL-C by 14.3% (0.45 mmol/L). HDL-C values decreased in both groups at wk 2 and 4, but were not different from baseline at wk 10. Fasting TAG were reduced significantly in the Protein Group, with the values ranging from 16 to 23% below initial baseline values.

TABLE 1

Dietary intakes for adult women at baseline and during weight loss while consuming reduced energy diets with a ratio of carbohydrate (CHO)/protein of 3.5 (CHO Group) or 1.4 (Protein group)¹

	Baseline ²	Protein group	CHO group
Energy, kJ/d (kcal/d)	8196 ± 267 (1959)	6987 ± 197 (1670)	6941 ± 167 (1659)
Protein, g/d	75 ± 3	125 ± 3	68 ± 2
Carbohydrate, g/d	246 ± 10	171 ± 7	239 ± 5
Total fat, g/d	75 ± 5	54 ± 2	48 ± 2
Cholesterol, mg/d	207 ± 17	239 ± 8	115 ± 5
SFA, g/d	25 ± 2	18 ± 1	14 ± 1
Dietary fiber, g/d	14 ± 1	19 ± 1	25 ± 1

¹ Values are means \pm SEM, $n = 12$.

² Initial values were not different between groups, and data for all 24 subjects were combined for the baseline period (wk 0). SFA = saturated fatty acids.

Blood and urinary ketone levels were not altered by the dietary treatments. The initial fasting concentration of plasma BHB had a mean of 98.9 ± 11.6 μ mol/L for both groups and concentrations of 112.8 ± 15.2 and 166.2 ± 10.1 μ mol/L after 10 wk of consuming the Protein and CHO diets, respectively. Acetoacetate was not detectable in the urine at any time point.

Blood urea concentrations were affected by diet treatment, by fed vs. fasting state and by the duration of the study (Table 4). The baseline values for the two groups differed, but the magnitude of this difference declined over the course of the study. At wk 4 and 10, fasting blood urea did not differ between groups. Postprandial blood urea concentrations reflected the nitrogen intakes. For the protein Group, the postprandial responses at wk 2, 4 and 10 had mean increases of 43% ($P < 0.05$) and the CHO Group had a mean increase of 27% ($P < 0.05$). Urinary urea also reflected differences in nitrogen intake between the two diet groups with a mean for wk 2, 4 and 10 for the Protein Group of 511 ± 39 mmol/d and the CHO Group of 279 ± 37 mmol/d ($P < 0.05$).

Plasma glucose and insulin were measured under both fasting and postprandial conditions. After 10 wk, fasting insulin levels did not differ between groups (Table 5). However, fasting glucose was lower in the CHO Group. Insulin was elevated 2 h after ingestion of the test meal, whereas plasma glucose was lower than the corresponding fasting concentration in both groups. In the CHO Group, the 2-h postprandial insulin concentration remained >2 times the fasting value and was associated with a lower level of blood glucose.

Both Groups expressed general satisfaction with the diet plans, but the Protein Group consistently reported a higher level of satisfaction (Table 6). Subjects in the Protein Group expressed a feeling of more energy, and they reported a higher level of satiety. There were no differences in hunger between the two groups.

DISCUSSION

The ideal ratios of dietary protein, carbohydrate and fat for adult health and weight management remain unknown. Claims about the merits or risks of diets with high protein are extensive (21), but there are few experimental studies. This study evaluated the effect of substitution of protein for carbohydrate as components of weight loss diets.

Changes in body weight require changes in energy balance (energy intake – energy expenditure). In this study, physical activity was constant; therefore, changes in body weight should be in proportion to food intake. Subjects in this study consumed diets with equal energy, fat and fiber but with different CHO/protein ratios of either 3.5 or 1.4. After 10 wk, subjects in both groups lost significant body weight.

Although changes in body weight did not differ between diet groups, the higher protein diet was more effective in improving body composition. Changes in the ratio of fat loss/lean loss (Fig. 1) indicated that the higher protein diet improved utilization of body fat while maintaining lean body mass. The mechanism for these differential effects on body composition is unknown. However, we found that substituting dietary protein for carbohydrate in an energy-restricted diet maintained levels of thyroid hormones T3 and T4 (Fig. 2) and reduced the insulin response to a test meal (Table 5). These endocrine differences are consistent with higher rates of lipolysis (22). Further, an increased amount of dietary protein has been shown to reduce nitrogen losses associated with very low energy diets (23), and we reported that increased use of proteins or specifically branched-chain amino acid serves to main-

TABLE 2

Body weight and composition of adult women consuming either moderate protein or high carbohydrate (CHO) weight loss diets¹

	Time, wk				Total loss
	0	2	4	10	
	kg				
Body weight					
Protein group	84.83 ^a ± 3.64	82.13 ^{a,b} ± 2.70	80.97 ^b ± 3.85	77.30 ^b ± 3.50	7.53 ± 1.44
CHO group	85.68 ^a ± 2.77	83.47 ^{a,b} ± 2.21	82.46 ^b ± 3.22	78.72 ^b ± 2.46	6.96 ± 1.36
Body fat					
Protein group	38.97 ^a ± 2.86	37.15 ^{a,b} ± 2.80	35.92 ^b ± 2.77	33.37 ^b ± 2.81	5.60 ± 0.52
CHO group	38.92 ^a ± 2.18	37.41 ^{a,b} ± 2.18	36.32 ^b ± 2.07	34.18 ^b ± 1.99	4.74 ± 0.65
Lean body mass					
Protein group	42.69 ± 1.04	42.14 ± 1.05	42.14 ± 0.99	41.81 ± 0.99	0.88 ± 0.33
CHO group	43.53 ± 1.24	42.66 ± 1.17	42.51 ± 1.23	42.32 ± 1.16	1.21 ± 0.58

¹ Values are means ± SEM, *n* = 12. Means for a variable without a common letter differ, *P* < 0.05.

tain muscle protein synthesis during catabolic conditions (24–26). Hence, the changes in body composition associated with the higher protein diet may be associated with either targeting of body fat or sparing of muscle protein, or both.

Similar findings for changes in body composition have been reported in other studies in which dietary fat was constant (9,10). Parker et al. (10) examined weight loss in 66 subjects with type 2 diabetes and BMI of 34 kg/m². These investigators utilized diets with equal energy (6690 kJ/d) and equal fat (~27% of energy) with CHO/protein ratios of 3.4 or 1.5. After 8 wk, subjects had similar weight losses (4.5 kg) but those consuming the high protein diet lost more body fat (5.3 kg vs 2.8 kg; *P* < 0.05).

In the study from Denmark (9), investigators selected 65 individuals with a mean age of 39 y and BMI of 30 kg/m². Individuals were assigned to either a high carbohydrate diet (CHO/protein = 4.9, fat = 29% of energy) or high protein diet (CHO/protein = 1.9, fat = 29%) with all food provided by the researchers but with self-selection by the subjects and home preparation. Subjects were allowed to consume their food ad libitum. After 6 mo, the subjects receiving the higher protein diet consumed 17% less energy per day, lost more body

weight and lost more body fat than the high carbohydrate group. In the Denmark study, subjects self-selected energy intake based on appetite, whereas subjects in the present study were restricted to equal energy intakes. However, both studies

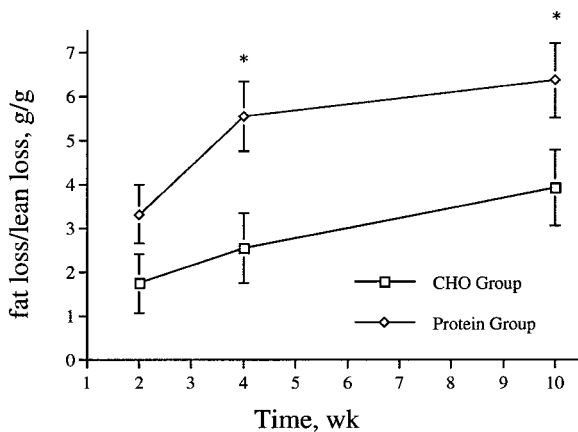


FIGURE 1 Time course changes for the ratio of loss of body fat compared with loss of lean body mass (fat/lean) during weight loss for adult women consuming diets with a carbohydrate (CHO)/protein ratio of 3.5 (CHO Group) or 1.4 (Protein Group). Values are means ± SEM, *n* = 12. Means without a common letter differ, *P* < 0.05.

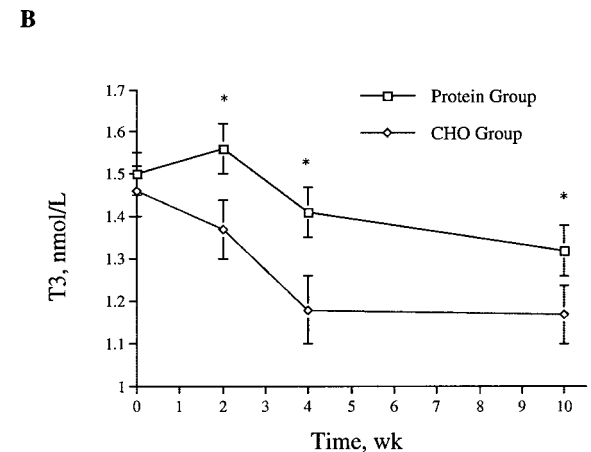
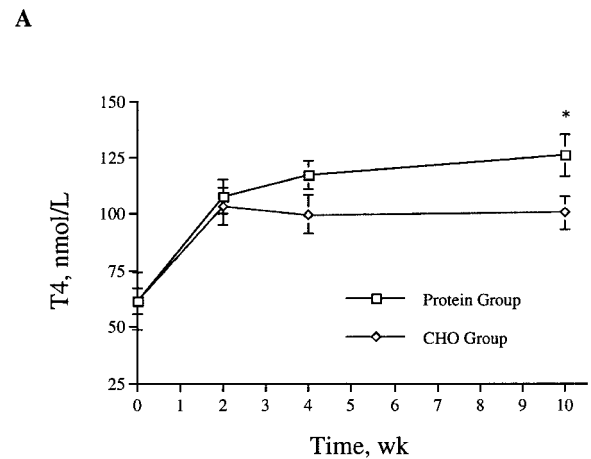


FIGURE 2 Response of thyroid hormones thyroxine (T4) and triiodothyronine (T3) during weight loss in adult women consuming diets with a carbohydrate (CHO)/protein ratio of 3.5 (CHO Group) or 1.4 (Protein Group). Values are means ± SEM, *n* = 12. *Different from the CHO Group, *P* < 0.05.

TABLE 3

Serum lipid concentrations in adult women at baseline (wk 0) and during weight loss while consuming reduced energy diets with a ratio of carbohydrate (CHO)/protein of 3.5 (CHO group) or 1.4 (protein group)¹

	Time, wk			
	0	2	4	10
	mmol/L			
Total cholesterol				
Protein group	5.80 ^a ± 0.16	5.24 ^b ± 0.26	5.22 ^b ± 0.19	5.38 ^b ± 0.21
CHO group	4.92 ^c ± 0.29	4.46 ^d ± 0.27	4.37 ^d ± 0.27	4.40 ^d ± 0.29
LDL-C				
Protein group	3.81 ^a ± 0.17	3.46 ^b ± 0.23	3.41 ^b ± 0.19	3.45 ^b ± 0.19
CHO group	3.15 ^c ± 0.24	2.77 ^d ± 0.21	2.70 ^d ± 0.22	2.64 ^d ± 0.22
HDL-C				
Protein group	1.43 ^a ± 0.07	1.35 ^b ± 0.08	1.33 ^b ± 0.09	1.46 ^a ± 0.10
CHO group	1.33 ^a ± 0.09	1.23 ^b ± 0.07	1.20 ^b ± 0.07	1.28 ^a ± 0.07
Triacylglycerol				
Protein group	1.21 ^a ± 0.15	0.92 ^b ± 0.13	1.02 ^{a,b} ± 0.17	0.95 ^b ± 0.13
CHO group	1.05 ^a ± 0.09	1.01 ^{a,b} ± 0.11	1.01 ^{a,b} ± 0.09	1.05 ^a ± 0.11

¹ Values are means ± SEM, *n* = 12. Means for a variable without a common letter differ, *P* < 0.05.

found that diets with CHO/protein ratios <2.0 partitioned weight loss toward body fat. In a subsequent study (27), these researchers reported that individuals consuming a high protein diet had higher 24-h energy expenditure than when consuming a high carbohydrate diet, and that energy expenditure was higher when the protein was derived from animal proteins compared with plant proteins.

A major concern about using higher protein diets, particularly those rich in animal products, has been the association of cholesterol and saturated fatty acids with cardiovascular disease. There is a clear relationship of abnormal blood lipoprotein patterns with onset of heart disease. Further, there is increasing evidence concerning the causal relationship of total dietary lipids and saturated fatty acids with the onset of atherosclerosis (28). However, the independent roles of protein and carbohydrates have received less attention. It is often assumed that animal proteins are atherogenic, whereas diets high in complex carbohydrates reduce the risk of heart disease. However, both of these associations are confounded by other components of the diet including total fat, total energy and dietary fiber. A recent report from the Nurses' Health Study by

Hu and colleagues (7) provides epidemiologic evidence that replacing carbohydrates in the diet with protein enhanced the overall quality of the diet and lowered the risk of ischemic heart disease in adult women.

The current study directly evaluated the relationship between protein and carbohydrate by substituting foods in the protein groups (meats, dairy, eggs and nuts) for foods in the high carbohydrate group (breads, rice, pasta, and cereals) while maintaining total energy, total fat and fiber constant. Using this protocol, both diet groups exhibited ~10% reductions in total cholesterol and LDL-C even though the Protein Group had more than twice the cholesterol intake. These data suggest that other factors such as energy balance (weight loss), total energy intake, total fat or saturated fat were more important than dietary cholesterol. These findings are consistent with previous clinical reports (8,29) and with the current understanding of regulation of endogenous cholesterol synthesis (30).

Although changes in total cholesterol and LDL-C were similar in the two groups, fasting serum TAG concentrations differed. After 2 wk, the Protein Group exhibited a 20%

TABLE 4

Fasting and post-prandial plasma and urinary urea in adult women at baseline (wk 0) and during weight loss while consuming reduced energy diets with a ratio of carbohydrate (CHO)/protein of 3.5 (CHO group) or 1.4 (Protein group)¹

	Time, wk			
	0	2	4	10
Fasting plasma urea, mmol/L				
Protein group	4.32 ^a ± 0.21	3.75 ^b ± 0.21	3.39 ^c ± 0.14	3.14 ^c ± 0.18
CHO group	3.46 ^{b,c} ± 0.28	2.39 ^d ± 0.21	3.32 ^{b,c} ± 0.25	3.07 ^c ± 0.28
Post-prandial plasma urea, mmol/L				
Protein group	4.49 ^b ± 0.21	5.53 ^{a*} ± 0.18	4.57 ^{b*} ± 0.14	4.60 ^{b*} ± 0.18
CHO group	3.50 ^c ± 0.28	3.25 ^{c*} ± 0.21	4.60 ^{b*} ± 0.25	3.32 ^c ± 0.28
Urinary urea, mmol/d				
Protein group	339 ^b ± 28	468 ^a ± 32	482 ^a ± 25	585 ^a ± 64
CHO group	307 ^b ± 25	218 ^c ± 28	253 ^{b,c} ± 53	368 ^b ± 32

¹ Values are means ± SEM, *n* = 12. Means for a variable without a common letter differ, *P* < 0.05. * Indicates the value for post-prandial is different from fasting, *P* < 0.05.

TABLE 5

Fasting and post-prandial plasma glucose and insulin concentrations in adult women after 10 wk consuming reduced energy weight loss diets with a ratio of carbohydrate (CHO)/protein of 3.5 (CHO group) or 1.4 (protein group)¹

	Glucose		Insulin	
	Fasting	Post-prandial ²	Fasting	Post-prandial ²
	mmol/L		pmol/L	
Protein group	4.89 ^a ± 0.11	4.34 ^b ± 0.15	176 ^c ± 18	251 ^b ± 21
CHO group	4.33 ^b ± 0.10	3.77 ^c ± 0.14	178 ^c ± 18	384 ^a ± 27

¹ Values represent means ± SEM, *n* = 12. Means for a variable without a common letter differ, *P* < 0.05.

² Post-prandial values were determined 2 h after consumption of a 1670 kJ breakfast. The macronutrient content of the breakfast reflects the respective dietary treatments. Data adapted from Layman et al. (16).

reduction in TAG, whereas the CHO Group did not change. These changes in TAG are in agreement with other studies (7–9,13,31) that found that high carbohydrate diets are linked to increased hepatic production of TAG (11,31) and increased levels of circulating TAG (7,8,13). Similarly, dietary substitution of protein for carbohydrates appears to have positive effects on the ratio of TAG/HDL-C. The Bezafibrate Coronary Atherosclerosis Intervention study (32) reported that decreases in the ratio of TAG/HDL-C correlate directly with reductions in coronary heart disease without changes in LDL-C levels. In the present study, the ratio of TAG/HDL-C decreased in the Protein Group (0.846 ± 0.021 to 0.651 ± 0.016 mmol/L; *P* = 0.031), whereas the ratio did not change in the CHO Group (0.789 ± 0.004 to 0.820 ± 0.004 mmol/L).

The effect of dietary protein intake on renal function has been debated for 50 y (33). In cases with compromised renal function, reduced levels of dietary protein can retard the

progression to renal failure. Conditions such as diabetes, hypertension, infection or renal surgery often lead to changes in renal physiology, including increases in glomerular capillary pressure and blood flow rates (34). Restriction of dietary protein appears to reduce the “renal workload” and minimize glomerular perfusion. By extrapolation, it is often suggested that adults avoid high protein intakes to minimize glomerular filtration rates. However, there is no known association of protein intake with progressive renal insufficiency during aging (34,35). Further, a recent study by Poortmans and Dellalieux (36) reported no negative effects on renal function of long-term daily protein intakes ranging from 1.2 to 2.0 g protein/kg body weight. Evaluation of urea data in the current study suggests that subjects adapted rapidly to the different nitrogen intakes. There were no differences between the groups in fasting blood urea, with the values becoming more consistent throughout the course of the study. Both groups exhibited a postprandial increase in blood urea that was in proportion to the protein intake. Similarly, urinary excretion of urea was in proportion to the dietary intake. These data suggest that with dietary intakes of protein ranging from 0.8 to 1.5 g/kg, renal clearance of nitrogen is rapid and efficient.

The roles of specific macronutrients in satiety and control of energy intake remain poorly understood. There is increasing evidence that diets high in some carbohydrates, particularly refined grains, produce high postprandial levels of blood glucose. This meal response of blood sugar is termed the glycemic index (37). More important, the rapid rise in blood glucose evokes an equally intensive response of insulin that can lead to a period of hypoglycemia and increased hunger (37–39). This hypoglycemic response typically occurs ~2 h after a meal, depending on meal size and rates of gastric emptying (38). Lipids appear to increase satiety both as dietary lipids that slow gastric emptying (40) and as fatty acids free in the blood that reduce hunger (41). Proteins made up of 20 individual amino acids are more complex, but clearly have the potential to effect neurotransmitters and satiety (42). Still other factors affecting satiety include dietary fiber and fluid intakes (43). This study was designed to maintain levels of fiber, fat, fluids and total energy constant and compare effects of protein vs. carbohydrates. Overall, subjects in the Protein Group reported higher satiety and greater energy when consuming the diet with a lower CHO/protein ratio. Subjects in the CHO Group exhibited increased postprandial insulin and reduced glucose concentrations. These findings are consistent with the hypothesis about the role of carbohydrates in postprandial hypoglycemia and hunger (39) and the effect of the glycemic index on blood glucose and insulin (38).

TABLE 6

Satisfaction surveys for adult women consuming reduced energy weight loss diets with a ratio of carbohydrate (CHO)/protein of 3.5 (CHO group: CHO) or 1.4 (protein group: protein)¹

My satisfaction level with this dietary plan is: (Very unsatisfied 1–7 Very satisfied)	
Protein	6.08 ± 0.13*
CHO	5.48 ± 0.13
My energy level is: (Very low 1–7 Very high)	
Protein	5.72 ± 0.08*
CHO	4.90 ± 0.11
Between meals on this dietary plan my hunger is: (Very low 1–7 Very high)	
Morning:	
Protein	2.84 ± 0.16
CHO	2.96 ± 0.15
Afternoon:	
Protein	3.42 ± 0.14
CHO	3.30 ± 0.14
Evening:	
Protein	3.07 ± 0.14
CHO	2.87 ± 0.15
The satiety value of this dietary plan is: (Very low 1–7 Very high)	
Protein	6.05 ± 0.12*
CHO	5.32 ± 0.14

¹ Values are means ± SEM for averages of weekly surveys, *n* = 12.

* Different from CHO group, *P* < 0.05.

In summary, both diets were effective weight loss plans and improved the blood lipid profile. However, the protein diet produced greater improvements in body composition with an increased ratio of fat/muscle loss. Further, the protein diet, with a reduced CHO/protein ratio, produced positive changes in blood lipids with reduction of TAG levels and the ratio of TAG/HDL-C. Subjects consuming the higher protein diet reported greater satiety. Although it is unlikely that any one diet will be ideal for all individuals, these results indicate that changes in the ratio of protein to carbohydrate toward a higher protein diet can be effective in the control of body weight with parallel improvements in blood lipids.

LITERATURE CITED

- Kuczmarski, R. J., Flegal, K. M., Campbell, S. M. & Johnson, C. L. (1994) Increasing prevalence of overweight among US adults. *J. Am. Med. Assoc.* 272: 205–211.
- Must, A., Spadano, J., Coakley, E. H., Field, A. E., Colditz, G. & Dietz, W. H. (1999) The disease burden associated with overweight and obesity. *J. Am. Med. Assoc.* 282: 1523–1529.
- Surgeon General's Report on Nutrition and Health (1988) USDHHS Publication no 88–50210. Public Health Service, Washington, DC.
- U. S. Department of Agriculture/Department of Health and Human Services (1995) Dietary Guidelines for Americans, 4th ed., Home and Garden bulletin 232. DHHS, Washington, DC.
- American Heart Association Scientific Statement (2000) AHA Dietary Guidelines Revision 2000. *Circulation* 102: 2284–2299.
- Seidell, J. C. (1998) Dietary fat and obesity: an epidemiologic perspective. *Am. J. Clin. Nutr.* 67 (suppl.): 546S–550S.
- Hu, F. B., Stampfer, M. J., Manson, J. E., Rimm, E., Colditz, G. A., Speizer, F. E., Hennekens, C. H. & Willett, W. C. (1999) Dietary protein and risk of ischemic heart disease in women. *Am. J. Clin. Nutr.* 70: 221–227.
- Wolfe, B. M. & Giovannetti, P. M. (1991) Short-term effects of substituting protein for carbohydrate in diets of moderately hypercholesterolemic human subjects. *Metabolism* 40: 338–343.
- Skov, A. R., Toubro, S., Ronn, B., Holm, L. & Astrup, A. (1999) Randomized trial on protein vs carbohydrate in ad libitum fat reduced diet for the treatment of obesity. *Int. J. Obes.* 23: 528–536.
- Parker, B., Noakes, M., Luscombe, N. & Clifton, P. (2002) Effect of a high-protein, monounsaturated fat weight loss diet on glycemic control and lipid levels in type 2 diabetes. *Diabetes Care* 25: 425–430.
- McGarry, J. D. (1998) Glucose-fatty acid interactions in health and disease. *Am. J. Clin. Nutr.* 67 (suppl.): 500S–504S.
- Wolfe, R. R. (1998) Metabolic interactions between glucose and fatty acids in humans. *Am. J. Clin. Nutr.* 67 (suppl.): 519S–526S.
- Sidossis, L. S., Mittendorfer, B., Walsler, E., Chinkes, D. & Wolfe, R. R. (1998) Hyperglycemia-induced inhibition of splanchnic fatty acid oxidation increases hepatic triacylglycerol secretion. *Am. J. Physiol.* 275: E798–E805.
- Ludwig, D. S., Majzoub, J. A., Al-Zahrani, A., Dallal, G. E., Blanco, I. & Roberts, S. B. (1999) High glycemic index foods, overeating, and obesity. *Pediatrics* 103: E261–E266.
- El-Khoury, A. E., Fukagawa, N. K., Sanchez, M., Tsay, R. H., Gleason, R. E., Chapman, T. E. & Young, V. R. (1994) The 24-h pattern and rate of leucine oxidation, with particular reference to tracer estimates of leucine requirements in healthy adults. *Am. J. Clin. Nutr.* 59: 1012–1020.
- Layman, D. K., Shiue, H., Sather, C., Erickson, D. J. & Baum, J. (2003) Increased dietary protein modifies glucose and insulin homeostasis in adult women during weight loss. *J. Nutr.* 133: 405–410.
- Jungas, R. L., Halperin, M. L. & Broxnan, J. T. (1992) Quantitative analysis of amino acid oxidation and related gluconeogenesis in humans. *Physiol. Rev.* 72: 419–448.
- Balalabramanyam, A., McKay, S., Nadkarni, P., Rajan, A. S., Farza, A., Pavlik, V., Herd, J. A., Jahorr, F. & Reeds, P. J. (1999) Ethnicity affects the postprandial regulation of glycogenolysis. *Am. J. Physiol.* 40: E905–E914.
- Myers, G., Cooper, G., Winn, C. & Smith, S. (1989) The Centers for Disease Control-National Heart, Lung and Blood Institute Lipid Standardization Program. An approach to accurate and precise lipid measurements. *Clin. Lab. Med.* 9: 105–135.
- Friedewald, W., Levy, R. & Frederickson, D. (1972) Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin. Chem.* 18: 499–502.
- American Heart Association Science Advisory (2001) Dietary protein and weight reduction. *Circulation* 104: 1869–1873.
- Tepperman, J. (1980) The thyroid. In: *Metabolic and Endocrine Physiology*, 4th ed. Year Book Medical Publishers, Chicago, IL.
- Bistrrian, B. R., Winterer, J., Blackburn, G. L., Young, V. & Sherman, M. (1977) Effect of a protein-sparing diet and brief fast on nitrogen metabolism in mildly obese subjects. *J. Lab. Clin. Med.* 89: 1030–1035.
- Hong, S. C. & Layman, D. K. (1984) Effects of leucine on in vitro protein synthesis and degradation in rat skeletal muscle. *J. Nutr.* 114: 1204–1212.
- Gautsch, T. A., Anthony, J. C., Kimball, S. R., Paul, G. L., Layman, D. K. & Jefferson, L. S. (1998) Availability of eIF4E regulates skeletal muscle protein synthesis during recovery from exercise. *Am. J. Physiol.* 274: C406–C414.
- Anthony, J. C., Gautsch-Anthony, T. & Layman, D. K. (1999) Leucine supplementation enhances skeletal muscle recovery in rats following exercise. *J. Nutr.* 129: 1102–1106.
- Mikkelsen, P. B., Toubro, S. & Astrup, A. (2000) Effect of fat-reduced diets on 24-h energy expenditure: comparisons between animal protein, vegetable protein, and carbohydrate. *Am. J. Clin. Nutr.* 72: 1135–1141.
- National Cholesterol Education Program (2001) Third Report of the Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) NIH Publication no. 01–3670, Washington, DC.
- Wolf, R. N. & Grundy, S. M. (1983) Influence of exchanging carbohydrate for saturated fatty acids on plasma lipids and lipoproteins in men. *J. Nutr.* 113: 1521–1528.
- Spady, D. K., Woollett, L. A. & Dietschy, J. M. (1993) Regulation of plasma LDL-cholesterol levels by dietary cholesterol and fatty acids. *Annu. Rev. Nutr.* 13: 355–381.
- Katan, M. B. (1998) Effect of low-fat diets on plasma high-density lipoprotein concentrations. *Am. J. Clin. Nutr.* 67: 573S–576S.
- Ericsson, C. G., Mamsten, A., Nilsson, J., Grip, L., Svane, B. & De Faire, U. (1996) Angiographic assessment of effects of bezafibrate on progression of coronary artery disease in young male postinfarction patients. *Lancet* 347: 849–853.
- Addis, T. (1948) *Glomerular Nephritis: Diagnosis and Treatment*. Macmillan, New York, NY.
- Brenner, B. M., Meyer, T. W. & Hostetter, T. H. (1982) Dietary protein intake and the progressive nature of kidney disease. *N. Engl. J. Med.* 307: 652–659.
- Brandle, E., Sieberth, H. G. & Hautmann, R. E. (1996) Effect of chronic dietary protein intake on the renal function in healthy subjects. *Eur. J. Clin. Nutr.* 50: 734–740.
- Poortmans, J. R. & Dellalieux, O. (2000) Do regular high protein diets have potential health risks on kidney function in athletes? *Int. J. Sport Nutr. Exerc. Metab.* 10: 28–38.
- Ludwig, D. S. (2000) Dietary glycemic index and obesity. *J. Nutr.* 130: 280S–283S.
- Roberts, S. B. (2000) High-glycemic index foods, hunger, and obesity: is there a connection? *Nutr. Rev.* 58: 163–169.
- Cryer, P. E. (1999) Symptoms of hypoglycemia thresholds for their occurrence, and hypoglycemia unawareness. *Endocrinol. Metab. Clin. N. Am.* 28: 495–500.
- Sidery, M. B., Macdonald, I. A. & Blackshaw, P. E. (1994) Superior mesenteric artery blood flow and gastric emptying in humans and the differential effects of high fat and high carbohydrate meals. *Gut* 35: 186–190.
- Ludwig, D. S., Majzoub, J. A., Al-Zahrani, A., Dallal, G. E., Blanco, I. & Roberts, S. B. (1999) High glycemic index foods, overeating, and obesity. *Pediatrics* 103: E261–E266.
- Fernstrom, J. D. (1994) Dietary amino acids and brain function. *J. Am. Diet. Assoc.* 94: 71–77.
- Burton-Freeman, B. (2000) Dietary fiber and energy regulation. *J. Nutr.* 130: 272S–275S.