See corresponding editorial on page 989.

See corresponding CME exam on page 1205.

Separate effects of reduced carbohydrate intake and weight loss on atherogenic dyslipidemia[1–3]

Ronald M Krauss, Patricia J Blanche, Robin S Rawlings, Harriett S Fernstrom, and Paul T Williams

ABSTRACT

Background: Low-carbohydrate diets have been used to manage obesity and its metabolic consequences.

Objective: The objective was to study the effects of moderate carbohydrate restriction on atherogenic dyslipidemia before and after weight loss and in conjunction with a low or high dietary saturated fat intake.

Design: After 1 wk of consuming a basal diet, 178 men with a mean body mass index (in kg/m^2) of 29.2 ± 2.0 were randomly assigned to consume diets with carbohydrate contents of 54% (basal diet), 39%, or 26% of energy and with a low saturated fat content (7–9% of energy); a fourth group consumed a diet with 26% of energy as carbohydrate and 15% as saturated fat. After 3 wk, the mean weight loss (5.12 ± 1.83 kg) was induced in all diet groups by a reduction of ≈1000 kcal/d for 5 wk followed by 4 wk of weight stabilization.

Results: The 26%-carbohydrate, low-saturated-fat diet reduced triacylglycerol, apolipoprotein B, small LDL mass, and total:HDL cholesterol and increased LDL peak diameter. These changes were significantly different from those with the 54%-carbohydrate diet. After subsequent weight loss, the changes in all these variables were significantly greater and the reduction in LDL cholesterol was significantly greater with the 54%-carbohydrate diet than with the 26%-carbohydrate diet. With the 26%-carbohydrate diet, lipoprotein changes with the higher saturated fat intakes were not significantly different from those with the lower saturated fat intakes, except for LDL cholesterol, which decreased less with the higher saturated fat intake because of an increase in mass of large LDL.

Conclusions: Moderate carbohydrate restriction and weight loss provide equivalent but nonadditive approaches to improving atherogenic dyslipidemia. Moreover, beneficial lipid changes resulting from a reduced carbohydrate intake were not significant after weight loss. *Am J Clin Nutr* 2006;83:1025–31.

KEY WORDS Saturated fat, lipoproteins, carbohydrates, weight loss

INTRODUCTION

Excess body weight can result in changes in plasma lipids and lipoproteins that increase the risk of atherosclerotic cardiovascular disease (CVD), ie, increases in triacylglycerols and small, dense LDL particles, with variable increases in total LDL cholesterol, and decreases in HDL cholesterol (1). Atherogenic dyslipidemia associated with excess adiposity is highly correlated with reduced insulin sensitivity and is a major feature of the metabolic syndrome (2, 3).

Dietary carbohydrates, especially simple sugars, can also promote atherogenic dyslipidemia, in large part because of effects on the metabolism of plasma triacylglycerol-rich lipoproteins (4). High-carbohydrate, low-fat diets have been shown to induce increased concentrations of small, dense LDL and expression of the small, dense LDL particle phenotype (LDL subclass pattern B) in a high proportion of healthy men (5, 6).

Recently, attention has focused on the use of very-low-carbohydrate diets to achieve weight loss (7–12). Reductions in dietary carbohydrate intake in such diets are achieved both by reductions in total calorie intake and by substitution of protein, fat, or both. Resulting metabolic changes include reductions in plasma triacylglycerol and reductions or a lack of increase in LDL cholesterol, despite relatively high dietary contents of saturated fat and cholesterol (7–11). Other studies have examined the effect on lipoprotein metabolism of less extreme reductions in carbohydrate intake in conjunction with weight loss (13–16). These studies, however, have not distinguished the effects of changes in dietary macronutrients from those of weight loss. The main objective of the present study was to test the effects of moderate reductions in carbohydrate intake with and without weight loss on atherogenic dyslipidemia in overweight and mildly obese men. In addition, we tested the extent to which changes in lipoprotein with reductions in carbohydrate intake are influenced by variations in saturated fat content.

SUBJECTS AND METHODS

Study design and diets

The study design is presented in **Figure 1**. All subjects consumed the basal diet (54% of energy as carbohydrate) for 1 wk, after which they were randomly assigned to the basal diet or 1 of

[1] From the Children's Hospital Oakland Research Institute, Oakland, CA (RMK, PJB, RSR, and HSF), and the Life Science Division, Lawrence Berkeley National Laboratory, Berkeley, CA (RMK, PJB, RSR, HSF, and PTW).

[2] Supported by a grant from the National Dairy Council.

[3] Reprints not available. Address correspondence to RM Krauss, Children's Hospital Oakland Research Institute, 5700 Martin Luther King Jr Way, Oakland, CA 94609. E-mail: rkrauss@chori.org.

Received August 8, 2005.

Accepted for publication December 1, 2005.

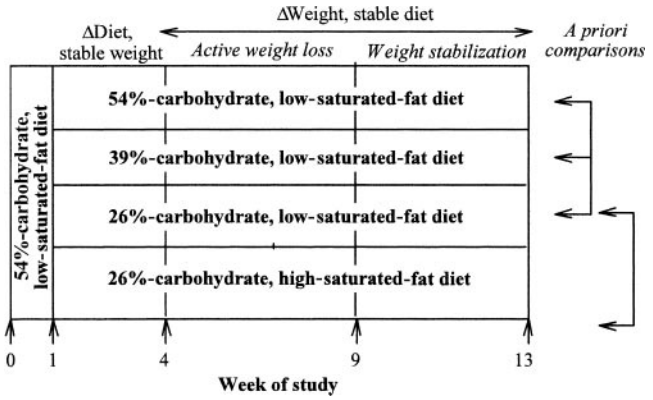

FIGURE 1. Study design. SF, saturated fat.

the 3 low-carbohydrate diets. After 3 wk (first stable-weight period), weight loss was induced by a reduction of ≈1000 kcal/d in each diet for 5 wk, after which energy intake was adjusted to stabilize weight for 4 wk (final stable-weight period).

The participants were free-living and were instructed not to alter their physical activity during the study. All participants received menus designed to provide nutrient intake over 6-d cycles as specified in **Table 1**. Nutrient calculations were based on the nutritional database and algorithms of THE FOOD PROCESSOR software (version 7.3; ESHA Research, Salem, OR). Frozen, prepared entrees fortified with vitamins and minerals to meet the Recommended Dietary Allowances (LifeSpring Home Nutrition, Irvine, CA) were provided for lunch and dinner. The participants prepared their own breakfasts and snacks according to menus, and the participants were weighed weekly by the staff, who adjusted energy intakes if necessary. Adherence was promoted through frequent telephone contacts and weekly meetings with the dietitians. Compliance was assessed via analysis of a daily checklist of foods eaten. None of the participants were eliminated for noncompliance (defined as daily diet deviations averaging >5% of total energy).

Subjects

Subjects were recruited through mailed solicitations to households within a reasonable driving distance to our outpatient clinic in Berkeley, CA, through commercially available mailing lists. Financial and logistical constraints on the number of subjects that could be managed during the 3-y study required the inclusion of only one sex to satisfy statistical power calculations. Men were chosen because of their higher prevalence of LDL pattern B, which was the major outcome variable on which the power calculations were based.

The subjects had no history of CVD or other chronic disease and none were taking drugs known to affect lipid metabolism. Other eligibility criteria included a body mass index (BMI; kg/m^2) ≥26–35, total and LDL-cholesterol concentrations below the 95th percentile for age and sex, a triacylglycerol concentration ≤500 mg/dL (5.65 mmol/L), a fasting glucose concentration ≤125 mg/dL (6.94 mmol/L), a systolic blood pressure <150 mm Hg, and a diastolic blood pressure <90 mm Hg. None of the men smoked, and no alcohol was consumed during the study. All subjects gave informed consent under a protocol approved by the Institutional Review Boards of Children's Hospital Oakland and the University of California, Berkeley.

The subjects were assigned to groups according to directions in sealed sequentially numbered envelopes that were the result of random permutations of subjects into treatment conditions for randomly determined blocks of 4, 8, 12, 16, 20, or 24 subjects. Of 256 subjects found eligible after screening, 35 declined to enroll and 43 discontinued after beginning the dietary protocol (**Table 2**). In most cases, discontinuation resulted from difficulties meeting the time requirements of the study.

Blood samples were obtained after an overnight fast after the 1-wk basal diet, after the first stable-weight period of the experimental diets (week 4), and after weight stabilization after weight loss (week 13; Figure 1). Plasma was kept at 4 °C for ≤3 d before processing. At each visit, body weight was measured, and percentage body fat was determined by bioimpedance (model TBF-551; Tanita, Skokie, IL).

Laboratory measurements

Plasma total cholesterol and triacylglycerol concentrations were determined by enzymatic procedures on an Express 550 Plus analyzer (Ciba Corning, Oberlin, OH). These measurements were consistently in control as monitored by the Centers for Disease Control and Prevention–National Heart, Lung, and Blood Institute standardization program. HDL cholesterol was measured after dextran sulfate precipitation of plasma (17), and LDL cholesterol was calculated from the formula of Friedewald et al (18). Apolipoproteins A-I (apo A-I) and B (apo B) were measured by immunoturbidometric assay (Express 550 Plus analyzer) (19).

Measurements of mass concentrations of LDL subfractions were performed by analytic ultracentrifugation as described elsewhere (20). LDL I-IV subfractions, ranging from the largest and

TABLE 1
Macronutrient composition of the diets[1]

	54% Carbohydrate (basal)	39% Carbohydrate	26% Carbohydrate	26% Carbohydrate, high saturated fat
			% of energy	
Carbohydrate	54	39	26	26
Protein	16	29	29	29
Total fat	30	31	46	45
Saturated	7	8	9	15
Monounsaturated	13	13	27	20
Polyunsaturated	8	8	5	6

[1] Each diet was controlled for other nutrients as follows: cholesterol, 150 mg/1000 kcal; *trans* fatty acids, 2% of energy; fiber, 25 g/2000 kcal plus 2.5 g/500 kcal above 2000; carbohydrate, 50% simple and 50% complex; protein, 50% animal and 50% vegetable; dairy intake, 3 portions of milk, yogurt, or cheese per day.

TABLE 2
Body weight and plasma lipid and lipoprotein measurements at baseline and changes during the low- and high-saturated-fat (SF) diets[1]

	Low-SF diet (7–9%)			High-SF diet (15%)	*P*	
	54% CHO	39% CHO	26% CHO	26% CHO	Low-SF diet (ANOVA)[2]	High- vs low-SF diet (*t* test)[3]
Subjects (*n*)						
Recruited	57	56	59	52	—	—
Dropped out	8	14	12	12	—	—
Final sample size	49	42	47	40	—	—
Weight (kg)						
Baseline	91.6 ± 9.0	92.7 ± 9.1	93.6 ± 9.3	95.6 ± 10.1	NS	NS
ΔDiet, stable weight	−0.0 ± 0.4	−0.2 ± 0.2	−0.9 ± 0.2[4]	−0.5 ± 0.2	0.005	NS
ΔWeight, stable diet	−5.3 ± 0.3	−5.0 ± 0.4	−5.4 ± 0.3	−4.8 ± 0.4	NS	NS
Body fat (%)						
Baseline	27.3 ± 4.0	28.4 ± 4.6	27.5 ± 4.3	28.0 ± 4.2	NS	NS
ΔDiet, stable weight	−0.2 ± 0.1	−0.3 ± 0.1	−0.4 ± 0.1	−0.5 ± 0.2	0.02	NS
ΔWeight, stable diet	−3.5 ± 0.3	−3.5 ± 0.3	−3.6 ± 0.2	−3.3 ± 0.3	NS	NS
Total cholesterol (mg/dL)						
Baseline	203.2 ± 34.6	202.1 ± 23.2	201.1 ± 31.7	203.0 ± 34.8	NS	NS
ΔDiet, stable weight	−7.4 ± 3.0	−9.4 ± 2.8	−21.4 ± 3.2[4,5]	−10.7 ± 3.6	0.002	0.03
ΔWeight, stable diet	−10.6 ± 2.5	−2.1 ± 2.9	7.0 ± 3.1[6]	2.2 ± 3.3	0.0001	NS
LDL cholesterol (mg/dL)						
Baseline	130.1 ± 30.2	125.5 ± 23.1	129.1 ± 25.7	127.8 ± 32.0	NS	NS
ΔDiet, stable weight	−2.6 ± 3.1	−0.6 ± 3.3	−11.2 ± 2.7	−0.7 ± 3.9	0.04	0.03
ΔWeight, stable diet	−8.9 ± 2.5	−1.2 ± 2.5	4.3 ± 2.7[4]	1.1 ± 2.7	0.002	NS
Triacylglycerol (log mg/dL)						
Baseline	2.16 ± 0.20	2.19 ± 0.23	2.10 ± 0.24	2.18 ± 0.25	NS	NS
ΔDiet, stable weight	−0.05 ± 0.02	−0.12 ± 0.03	−0.19 ± 0.03[7]	−0.20 ± 0.03	0.0004	NS
ΔWeight, stable diet	−0.07 ± 0.02	−0.06 ± 0.02	0.01 ± 0.02	−0.03 ± 0.02	0.04	NS
HDL cholesterol (mg/dL)						
Baseline	41.7 ± 8.7	41.6 ± 9.0	43.1 ± 12.4	41.0 ± 11.1	NS	NS
ΔDiet, stable weight	−1.3 ± 0.7	0.6 ± 0.6	0.4 ± 0.9	3.0 ± 1.0	NS	0.06
ΔWeight, stable diet	1.9 ± 0.7	2.0 ± 0.7	2.4 ± 0.8	2.5 ± 0.9	NS	NS
Apolipoprotein B (mg/dL)						
Baseline	102.3 ± 21.7	102.6 ± 18.4	100.0 ± 21.2	104.2 ± 24.7	NS	NS
ΔDiet, stable weight	−4.9 ± 2.0	−9.5 ± 1.8	−15.8 ± 1.9[7]	−12.5 ± 2.1	0.0004	NS
ΔWeight, stable diet	−6.4 ± 1.8	−0.9 ± 2.4	2.3 ± 1.5[4]	−1.4 ± 2.0	0.004	NS
Apolipoprotein A-I (mg/dL)						
Baseline	113.8 ± 15.8	114.0 ± 15.5	111.0 ± 16.4	111.2 ± 14.3	NS	NS
ΔDiet, stable weight	−3.1 ± 1.3	0.8 ± 1.6	0.2 ± 1.7	2.3 ± 1.9	NS	NS
ΔWeight, stable diet	−0.9 ± 1.5	−0.8 ± 1.5	2.9 ± 1.9	0.8 ± 1.7	NS	NS
Total:HDL cholesterol						
Baseline	5.03 ± 1.17	5.09 ± 1.25	4.93 ± 1.30	5.30 ± 1.80	NS	NS
ΔDiet, stable weight	−0.05 ± 0.10	−0.31 ± 0.10	−0.62 ± 0.12[7]	−0.7 ± 0.14	0.001	NS
ΔWeight, stable diet	−0.45 ± 0.08	−0.29 ± 0.11	−0.03 ± 0.09[4]	−0.16 ± 0.08	0.005	NS
LDL peak particle diameter (Å)						
Baseline	258.8 ± 8.7	258.5 ± 8.0	260.3 ± 8.5	258.9 ± 9.2	NS	NS
ΔDiet, stable weight	0.2 ± 0.8	3.5 ± 0.9[8]	3.6 ± 0.9[8]	5.1 ± 1.1	0.007	NS
ΔWeight, stable diet	3.9 ± 1.0	1.3 ± 0.8	0.9 ± 0.7[8]	1.9 ± 0.8	0.02	NS

[1] All baseline values are $\bar{x} \pm$ SD, and all changes (Δ) are $\bar{x} \pm$ SE. To convert cholesterol and triacylglycerol concentrations to SI units (mmol/L), multiply by 0.0259 and 0.0113, respectively. CHO, carbohydrate. There were no significant differences between groups at baseline.
[2] 54%-CHO vs 39%-CHO vs 26%-CHO diet.
[3] 26%-CHO diet only.
[4,6–8] Significantly different from the 54%-CHO control diet (Scheffe test): [4]$P < 0.01$, [6]$P < 0.0001$, [7]$P < 0.001$, [8]$P < 0.05$.
[5] Significantly different from the 39%-CHO diet, $P \leq 0.05$ (Scheffe test).

most buoyant to the smallest and most dense, were estimated as lipoproteins of Svedberg flotation rate (S_f) 7–12, 5–7, 3–5, and 0–3, respectively (20). Nondenaturing polyacrylamide gradient gel electrophoresis with lipid staining of plasma was performed as described previously for determination of peak LDL particle diameter and LDL subclass patterns A and B (19).

Statistical procedures

Comparisons were made by analysis of variance or covariance, and post hoc analyses of significance by Scheffe's tests. All statistical procedures were performed by using STATVIEW software (version 5.0.1, SAS Institute, Cary, NC). Group averages at baseline are reported as means ± SDs, and group changes are reported as means ± SEs. Mean changes during the Δdiet, stable-weight phase were calculated as the differences between study week 4 and week 1, and the mean changes during the Δweight, stable-diet phase were calculated as the differences between study week 13 and week 4 (Figure 1). Log transformations of plasma triacylglycerol concentrations were used to attain normal distributions. The study was designed to be able to detect

a 50% reduction in the prevalence of LDL-subclass pattern B between groups.

RESULTS

No significant differences in lipids, lipoproteins, and apolipoproteins were observed between the experimental groups at baseline (Table 2). The mean (±SD) ages of the men assigned to low-saturated-fat diets with 54% (50.3 ± 9.8 y), 39% (51.5 ± 11.6 y), or 26% carbohydrate (49.2 ± 9.3 y) or high-saturated-fat diets with 26% carbohydrate (50.1 ± 11.3 y) did not differ significantly ($P = 0.94$). Repeated-measures analysis of variance showed significant time-by-diet interactions for all lipoprotein variables except LDL-III: total cholesterol ($P = 0.0008$), LDL cholesterol ($P = 0.005$), triacylglycerol ($P = 0.0006$), HDL cholesterol ($P = 0.005$), apo B ($P = 0.008$), apo A-I ($P = 0.04$), total:HDL cholesterol ($P = 0.001$), LDL peak particle diameter ($P = 0.007$), LDL-I ($P = 0.003$), LDL-II ($P = 0.004$), LDL-III ($P = 0.59$), and LDL-IV ($P = 0.04$). Consistent with the design, body weight, and percentage body fat changed with time ($P < 0.0001$ for both), but there was no significant time-by-diet interaction.

In the initial Δdiet, stable-weight phase of the study, the 26%-carbohydrate, low-saturated-fat diet resulted in reductions from baseline in total cholesterol, triacylglycerol, apo B, and total:HDL cholesterol that were greater than the changes observed in the group remaining on the 54%-carbohydrate diet. However, the difference in the change in LDL cholesterol between the 26%-carbohydrate diet and the control diet was not significant by post hoc analysis ($P = 0.13$). Despite our effort to maintain constant weight, the 26%-carbohydrate, low-saturated-fat diet group lost more weight than did the 54%-carbohydrate group during the stable-weight period. There was also a trend for a greater reduction in percentage body fat with the lower-carbohydrate diets ($P < 0.02$, analysis of variance). The significance of the lipoprotein differences between the 26%- and 54%-carbohydrate groups persisted after adjustment for the change in body weight for total cholesterol ($P = 0.01$), triacylglycerols ($P = 0.02$), apo B ($P = 0.001$), and total:HDL cholesterol ($P = 0.002$), whereas differences in LDL cholesterol and LDL-IV between groups became marginal ($P = 0.09$ and $P = 0.11$, respectively). In contrast with the findings during the initial Δdiet, stable-weight phase, weight loss and stabilization led to reductions in each of these variables that were significantly greater with the 54%-carbohydrate diet than with the 26%-carbohydrate, low-saturated-fat diet.

Repeated-measures analysis of variance showed significant changes in HDL cholesterol over time that did not differ significantly by diet. For all diets combined, no significant change in HDL cholesterol was observed during the stable-weight phase ($P = 0.16$). In contrast, significant increases in HDL cholesterol after weight loss were observed for all diets combined ($P < 0.0001$). No significant changes in apo A-I were observed with either of the low-carbohydrate diets before or after weight loss.

In the initial stable-weight phase, LDL peak particle diameter increased to a significantly greater extent with both the 39%- and 26%-carbohydrate, low-saturated-fat diets than with the 54%-carbohydrate diet (Table 2). These differences remained significant when adjusted for the change in weight. After weight loss and stabilization, there were minimal further changes with the lower-carbohydrate diets and a significantly greater increase with the 54%-carbohydrate diet. These changes could be related to corresponding differences in plasma mass concentrations of LDL subfractions as measured by analytic ultracentrifugation (**Table 3**).

During the initial stable-weight phase, the 26%-carbohydrate, low-saturated fat diet resulted in significantly greater decreases in small LDL-III and LDL-IV than did the 54%-carbohydrate diet. Adjustment for the modest changes in weight reduced the significance of the LDL-IV reduction ($P = 0.07$) but not the LDL-III reduction ($P = 0.01$). After weight loss and stabilization, there were minimal additional changes with the 26%-carbohydrate, low-saturated-fat diet but significantly greater decreases in LDL-III and LDL-IV with the 54%-carbohydrate diet. Changes with the 39%-carbohydrate diet were of intermediate magnitude between those of the 2 other diets and were significantly different from those with the 26%-carbohydrate, low-saturated-fat diet for LDL-III in the initial stable-weight period. For both intervention periods, changes from baseline in LDL-III and LDL-IV were correlated with changes in triacylglycerol: $r = 0.37$ and 0.36, respectively, before weight loss, and $r = 0.52$ and $r = 0.48$ after weight loss ($P < 0.0001$ for all).

Changes in peak LDL diameter (Table 2) and mass concentrations of LDL subfractions (Table 3) induced by each of the diets were reflected by changes in the proportions of subjects exhibiting LDL subclass pattern B (**Figure 2**). There were linear reductions in the prevalence of pattern B as a function of reduced carbohydrate intake after both the stable-weight and weight-loss periods. However, the slopes of these relations differed ($P = 0.04$) such that the magnitude of the reduction in expression of pattern B induced by weight loss increased in association with the percentage of carbohydrate intake.

Lipid, lipoprotein, and apolipoprotein values did not differ significantly between the men who consumed the 26%-carbohydrate diet that contained 15% saturated fat and the men who consumed the 26%-carbohydrate that contained 9% saturated fat before or after weight loss (Table 2), except for changes in total and LDL cholesterol during the initial stable-weight period. The higher-saturated-fat diet led to a smaller decrease in total and LDL cholesterol than did the lower-saturated-fat diet.

DISCUSSION

The atherogenic dyslipidemia associated with adiposity is characterized by relative increases in plasma triacylglycerol, reductions in HDL, and increased concentrations of small, dense LDL particles (1, 21). Weight loss has been shown to improve these lipid variables as well as the insulin resistance that often occurs concomitantly (1). The benefits of a reduction in carbohydrate intake below currently recommended intakes have also been reported, although many studies have examined only the effects of relatively extreme carbohydrate restriction and have not distinguished the effects of changes in macronutrient composition from those of concurrent weight loss (7–11).

In the present study, we investigated separately the effects of moderate reductions in carbohydrate intake and body weight on the components of atherogenic dyslipidemia in moderately overweight and mildly obese but otherwise healthy middle-aged men. As has been shown for diets with more extreme reductions in carbohydrate, placing men on a diet with 26% carbohydrate resulted in reductions in plasma triacylglycerol concentrations,

TABLE 3
Plasma LDL subfraction total mass concentrations at baseline and changes during the low- and high-saturated-fat (SF) diets[1]

	Low-SF diet (7–9%)			High-SF diet (15%)	*P*	
	54% CHO (n = 49)	39% CHO (n = 42)	26% CHO (n = 47)	26% CHO (n = 40)	Low-SF diet (ANOVA)[2]	High- vs low-SF diet (t test)[3]
LDL-I						
Baseline	90.2 ± 35.1	93.0 ± 32.6	95.4 ± 29.9	94.2 ± 37.1	NS	NS
ΔDiet, stable weight	−3.8 ± 3.6	2.7 ± 3.8	6.3 ± 4.1	16.1 ± 5.4	NS	NS
ΔWeight, stable diet	−0.3 ± 3.9	6.2 ± 3.8	1.1 ± 4.7	−1.6 ± 4.8	NS	NS
LDL-II						
Baseline	100.0 ± 35.1	99.2 ± 30.4	101.5 ± 26.0	98.1 ± 33.0	NS	NS
ΔDiet, stable weight	1.3 ± 3.3	1.8 ± 4.2	−5.5 ± 3.9	3.6 ± 5.5	NS	NS
ΔWeight, stable diet	−7.3 ± 4.1	−0.6 ± 3.6	2.3 ± 4.0	−5.3 ± 4.5	NS	NS
LDL-III						
Baseline	75.3 ± 39.4	71.7 ± 34.9	75.7 ± 38.3	77.9 ± 42.8	NS	NS
ΔDiet, stable weight	−5.3 ± 4.1	−7.3 ± 4.2	−24.5 ± 4.1[4,5]	−24.3 ± 5.1	0.002	NS
ΔWeight, stable diet	−16.1 ± 3.5	−11.6 ± 4.7	−1.2 ± 3.6[6]	−9.6 ± 3.8	0.02	NS
LDL-IV						
Baseline	18.3 ± 15.5	14.1 ± 14.7	16.6 ± 13.5	22.0 ± 21.4	NS	NS
ΔDiet, stable weight	−1.1 ± 1.3	−1.8 ± 2.2	−7.2 ± 1.8[6]	−9.6 ± 2.6	0.03	NS
ΔWeight, stable diet	−6.2 ± 2.0	−2.0 ± 1.8	1.4 ± 1.1[4]	−2.4 ± 1.3	0.007	0.03

[1] All baseline values are $\bar{x}$ ± SD, and all changes (Δ) are $\bar{x}$ ± SEM. CHO, carbohydrate. There were no significant differences between groups at baseline.
[2] 54%-CHO vs 39%-CHO vs 26%-CHO diet.
[3] 26%-CHO diet only.
[4,6] Significantly different from the 54%-CHO control diet (Scheffe test): [4]$P < 0.01$, [6]$P < 0.05$.
[5] Significantly different from the 39%-CHO diet, $P \leq 0.05$.

consistent with the known effects of carbohydrate intake on the metabolism of triacylglycerol-rich lipoproteins (4). Lower carbohydrate intakes have also been shown to be associated with reduced plasma apo B concentrations and a reduced ratio of total cholesterol to HDL. The reduction in these variables as a function of carbohydrate intake was significant only for the 26%-carbohydrate diet group. In comparing the 54%- and 26%-carbohydrate, low-saturated-fat diets, it is not possible to discriminate the effects of the addition of protein and monounsaturated fat from those due to the reduction in carbohydrate. However, when the monounsaturated fat content of the 26%-carbohydrate, low-saturated fat diet was reduced from 27% to 20%, primarily by replacement with saturated fat, the changes in triacylglycerol, apo B, and total:HDL cholesterol were not significantly different. This finding suggests that these changes were not due primarily to the higher monounsaturated fat content of the diet.

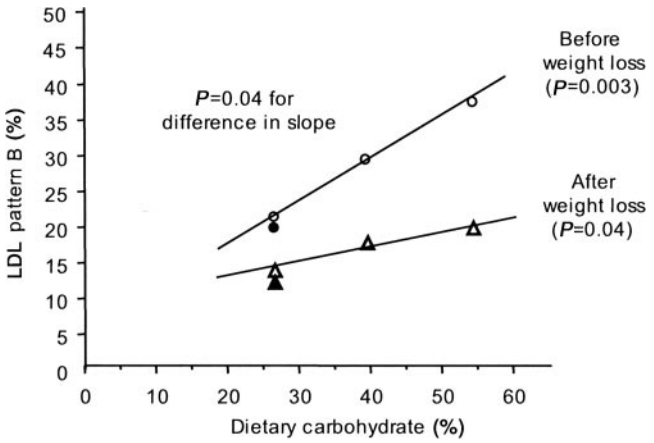

FIGURE 2. Prevalence of LDL subclass pattern B as a function of dietary carbohydrate content for each experimental diet before and after weight loss and stabilization with the diets. Closed symbols represent the low-saturated-fat diet group (n = 49, 42, and 47 for the 54%-, 39%-, and 26%-carbohydrate diets, respectively), and open symbols represent the high-saturated-fat diet group (n = 40).

We showed a linear relation of carbohydrate intake to the prevalence of LDL pattern B between the diets (Figure 2), including 2 with similar fat contents (54% and 39%) and 2 with similar protein contents (39% and 26%). Hence, it is most likely that a reduction in carbohydrate intake was the primary dietary determinant of changes in pattern B expression. Although the ratio of simple to complex carbohydrate as well as total fiber content were kept constant with all diets, the possibility of differential effects of certain classes of carbohydrates, particularly sugars and more hydrolysable starches, remains (4).

There was a reduction in LDL cholesterol with the 26%-carbohydrate, low-saturated-fat diet, although the differences compared with the higher-carbohydrate diets were of borderline statistical significance. Examination of changes in LDL subfraction distribution, however, showed that the low-carbohydrate diet resulted in significantly lower concentrations of small, dense LDL. Moreover, the reductions were significantly correlated with reduced plasma triacylglycerol concentrations, consistent with the pathways connecting triacylglycerol-rich lipoproteins to the production of small, dense LDL (21). Hence, the overall effects of low-carbohydrate diets on the standard LDL-cholesterol measurement are dependent on the extent to which such diets differentially modify the metabolism of larger compared with smaller LDL subclasses.

Analysis of LDL subfractions also helps to clarify the differing effects on LDL cholesterol of low-carbohydrate diets with higher compared with lower contents of saturated fatty acids. Although the reductions in small, dense LDL with the 2 diets were similar,

the higher saturated fat diet produced a somewhat larger offsetting increase in large buoyant LDL (Table 3), which resulted in a smaller net change in total LDL cholesterol. Although the difference in the response of large LDL between the 2 diets was not significant, the results are consistent with previous evidence that saturated fat intake results in an increase of larger LDL rather than smaller LDL particles (22) that may result from selective changes in both production and clearance of larger LDL (23).

Concentrations of apo B, a measure of total atherogenic particle concentrations, as well as total:HDL cholesterol, an integrated measure of CVD risk, decreased similarly with both the higher- and lower-saturated-fat diets. Moreover, the changes in LDL cholesterol observed here for both the lower- and higher-saturated-fat diets (−11 and 1 mg/dL, respectively) were considerably more beneficial than were those predicted on the basis of studies that used diets with a more conventional macronutrient composition (−1 and 9 mg/dL, respectively) (24). Overall, these findings suggest that for most overweight or obese men, the favorable effect of a reduced carbohydrate intake on pathways leading to the production of small, dense LDL can substantially modify the effect of changes in dietary fat composition on LDL cholesterol.

We found that, with weight loss, there were reductions in major lipid and lipoprotein indicators of CVD risk (LDL cholesterol, triacylglycerol, apo B, total:HDL cholesterol, and small, dense LDL), and that these reductions were much greater with the higher- than with the lower-carbohydrate diet. Because the estimated energy intake for weight maintenance after weight loss is estimated to be only several hundred kilocalories less per day than that before weight loss, the absolute reduction in carbohydrate intake after weight loss in the subjects who consumed the 54%-carbohydrate diet was much less than that achieved in the subjects who consumed the 26%-carbohydrate diet during the Δdiet, stable-weight phase. Hence, although this difference in carbohydrate intake could have contributed to some extent to the differences in lipoprotein changes after weight loss with the higher versus lower carbohydrate diets, the results indicate that there is an interaction between the effects of carbohydrate intake and weight loss on these lipoprotein measurements. It is possible that this finding reflects a convergence of the effects of adiposity and carbohydrate intake on common pathways that affect these lipoprotein measures, such that reductions of either can achieve similar results. However, at least under the conditions studied here, the effects are not additive—a result that is consistent with earlier observations (25).

Reductions in small, dense LDL with weight loss have been reported to be similar on diets with either a high-carbohydrate or high–monounsaturated fat content (14). This finding is similar to that of the present study, although, as described above, the effect of weight loss per se is much greater with diets with higher than with lower carbohydrate contents. Our data are also consistent with observations by Noakes and Clifton (26) that weight reduction with a low-saturated-fat diet produces significantly greater reductions in LDL cholesterol than does weight reduction with a high-saturated-fat diet, although we found that the difference in fatty acid composition, rather than caloric restriction and weight loss, was the major determinant of this effect.

The interaction of carbohydrate intake and weight loss is also reflected in their relations to the expression of LDL subclass pattern B, a phenotype that has been associated with increased CVD risk (21, 27–29). As shown in Figure 2, the expression of this atherogenic metabolic phenotype can be suppressed by either intervention, but the magnitude of the effect of reduced carbohydrate intake is diminished by sufficient weight loss and vice versa. These observations have implications for the management of atherogenic dyslipidemia in that low-carbohydrate diets may be particularly efficacious in those persons who are not able to achieve or maintain an adequate reduction in adiposity.

Limitations of the present study include the fact that it was restricted to men and that it was of relatively short duration. It has been shown, however, that a 4-wk period of weight maintenance after weight loss with a moderate-fat diet is sufficient to stabilize plasma lipids and lipoproteins (13). Moreover, evidence from recent studies (7, 10) indicates that plasma lipid profiles associated with very-low-carbohydrate diets can persist for as long as 1 y, even after weight loss with these diets has been attenuated by virtue of reduced dietary compliance.

Concerns with carbohydrate restriction include the potential for inadequate intake of certain beneficial dietary components, such as fiber and some vitamins, and the possibility of diminished effectiveness over time. However, with moderate carbohydrate limitation, such as the 26%-carbohydrate, low-saturated-fat diet studied in the present study, it is possible to meet the Recommended Dietary Allowances for all essential nutrients and to maintain adequate fiber intake with appropriate supplementation. Note that increased physical activity without dietary carbohydrate reduction can also result in improvements in the components of atherogenic dyslipidemia (30) and can have the additional benefit of helping to maintain reductions in body weight.

We thank Laura Holl, Joseph Orr, Bahareh Sahami, and Arnelio Acosta for technical assistance; Barbara Middleton for clinical help; Kathy Lundberg Bourdet for creating the diet menus; Barbara Sutherland for help with administering the dietary protocols; Linda Abe for data management; and Carol Procter and Megan Burnett for assistance with subject recruitment.

RMK was the principal investigator. PJB was the laboratory manager. RSR was the clinical coordinator. HSF was the dietitian. PTW provided statistical consultation. The National Dairy Council had no role in the design or conduct of the study or in the analysis or interpretation of the study results.

REFERENCES

1. Howard BV, Ruotolo G, Robbins DC. Obesity and dyslipidemia. Endocrinol Metab Clin North Am 2003;32:855–67.
2. Grundy SM, Small LDL. atherogenic dyslipidemia, and the metabolic syndrome. Circulation 1997;95:1–4.
3. Reaven GM. Syndrome X: 6 years later. J Intern Med Suppl 1994;736:13–22.
4. Parks EJ, Hellerstein MK. Carbohydrate-induced hypertriacylglycerolemia: historical perspective and review of biological mechanisms. Am J Clin Nutr 2000;71:412–33.
5. Krauss RM, Dreon DM. Low-density-lipoprotein subclasses and response to a low-fat diet in healthy men. Am J Clin Nutr 1995;62(suppl):478S–87S.
6. Dreon DM, Fernstrom HA, Williams PT, Krauss RM. A very low-fat diet is not associated with improved lipoprotein profiles in men with a predominance of large, low-density lipoproteins. Am J Clin Nutr 1999;69:411–8.
7. Foster GD, Wyatt HR, Hill JO, et al. A randomized trial of a low-carbohydrate diet for obesity. N Engl J Med 2003;348:2082–90.
8. Brehm BJ, Seeley RJ, Daniels SR, D'Alessio DA. A randomized trial comparing a very low carbohydrate diet and a calorie-restricted low fat diet on body weight and cardiovascular risk factors in healthy women. J Clin Endocrinol Metab 2003;88:1617–23.
9. Samaha FF, Iqbal N, Seshadri P, et al. A low-carbohydrate as compared with a low-fat diet in severe obesity. N Engl J Med 2003;348:2074–81.
10. Stern L, Iqbal N, Seshadri P, et al. The effects of low-carbohydrate versus

conventional weight loss diets in severely obese adults: one-year follow-up of a randomized trial. Ann Intern Med 2004;140:778–85.
11. Yancy WS Jr, Olsen MK, Guyton JR, Bakst RP, Westman EC. A low-carbohydrate, ketogenic diet versus a low-fat diet to treat obesity and hyperlipidemia: a randomized, controlled trial. Ann Intern Med 2004; 140:769–77.
12. Willett WC. Reduced-carbohydrate diets: no roll in weight management? Ann Intern Med 2004;140:836–7.
13. Pelkman CL, Fishell VK, Maddox DH, Pearson TA, Mauger DT, Kris-Etherton PM. Effects of moderate-fat (from monounsaturated fat) and low-fat weight-loss diets on the serum lipid profile in overweight and obese men and women. Am J Clin Nutr 2004;79:204–12.
14. Archer WR, Lamarche B, St-Pierre AC, et al. High carbohydrate and high monounsaturated fatty acid diets similarly affect LDL electrophoretic characteristics in men who are losing weight. J Nutr 2003;133: 3124–9.
15. Colette C, Percheron C, Pares-Herbute N, et al. Exchanging carbohydrates for monounsaturated fats in energy-restricted diets: effects on metabolic profile and other cardiovascular risk factors. Int J Obes Relat Metab Disord 2003;27:648–56.
16. Luscombe-Marsh ND, Noakes M, Wittert GA, Keogh JB, Foster P, Clifton PM. Carbohydrate-restricted diets high in either monounsaturated fat or protein are equally effective at promoting fat loss and improving blood lipids. Am J Clin Nutr 2005;81:762–72.
17. Warnick GR, Nguyen T, Albers AA. Comparison of improved precipitation methods for quantification of high-density lipoprotein cholesterol. Clin Chem 1985;31:217–22.
18. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem 1972;18:499–502.
19. Dreon DM, Fernstrom HA, Williams PT, Krauss RM. Reduced LDL particle size in children consuming a very-low-fat diet is related to parental LDL-subclass patterns. Am J Clin Nutr 2000;71:1611–6.
20. Krauss RM, Blanche PJ. Detection and quantitation of LDL subfractions. Curr Opin Lipidol 1992;3:377–83.
21. Berneis KK, Krauss RM. Metabolic origins and clinical significance of LDL heterogeneity. J Lipid Res 2002;43:1363–79.
22. Dreon DM, Fernstrom HA, Campos H, Blanche P, Williams PT, Krauss RM. Change in dietary saturated fat intake is correlated with change in mass of large low-density-lipoprotein particles in men. Am J Clin Nutr 1998;67:828–36.
23. Krauss RM. Hold the antioxidants and improve plasma lipids? J Clin Invest 2004;113:1253–5.
24. Mensink RP, Zock PL, Kester AD, Katan MB. Effects of dietary fatty acids and carbohydrates on the ratio of serum total to HDL cholesterol and on serum lipids and apolipoproteins: a meta-analysis of 60 controlled trials. Am J Clin Nutr 2003;77:1146–55.
25. Hulley SB, Wilson WS, Burrows MI, Nichaman MZ. Lipid and lipoprotein responses of hypertriglyceridaemic outpatients to a low-carbohydrate modification of the A.H.A. fat-controlled diet. Lancet 1972;2:551–5.
26. Noakes M, Clifton PM. Changes in plasma lipids and other cardiovascular risk factors during 3 energy-restricted diets differing in total fat and fatty acid composition. Am J Clin Nutr 2000;71:706–12.
27. Austin MA, King MC, Vranizan KM, Krauss RM. Atherogenic lipoprotein phenotype. A proposed genetic marker for coronary heart disease risk. Circulation 1990;82:495–506.
28. Lamarche B, Tchernof A, Moorjani S, et al. Small, dense low-density lipoprotein particles as a predictor of the risk of ischemic heart disease in men. Prospective results from the Quebec Cardiovascular Study. Circulation 1997;95:69–75.
29. Gardner CD, Fortmann SP, Krauss RM. Association of small low-density lipoprotein particles with the incidence of coronary artery disease in men and women. JAMA 1996;276:875–81.
30. Williams PT, Krauss RM, Vranizan KM, Wood PD. Changes in lipoprotein subfractions during diet-induced and exercise-induced weight loss in moderately overweight men. Circulation 1990;81:1293–304.