Effect of Weight Loss on LDL and HDL Kinetics in the Metabolic Syndrome

Associations with changes in plasma retinol-binding protein-4 and adiponectin levels

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OBJECTIVE — The purpose of this study was to examine the effect of weight loss on LDL and HDL kinetics and plasma retinol-binding protein-4 (RBP-4) and adiponectin levels in men with the metabolic syndrome.

RESEARCH DESIGN AND METHODS — LDL apolipoprotein (apo)B-100 and HDL apoA-I kinetics were studied in 35 obese men with the metabolic syndrome at the start and end of a 16-week intervention trial of a hypocaloric, low-fat diet ($n = 20$) versus a weight maintenance diet ($n = 15$) using a stable isotope technique and multicompartmental modeling.

RESULTS — Consumption of the low-fat diet produced significant reductions ($P < 0.01$) in BMI, abdominal fat compartments, and homeostasis model assessment score compared with weight maintenance. These were associated with a significant increase in adiponectin and a fall in plasma RBP-4, triglycerides, LDL cholesterol, and LDL apoB-100 concentration ($P < 0.05$). Weight loss significantly increased the catabolism of LDL apoB-100 (+27%, $P < 0.05$) but did not affect production; it also decreased both the catabolic (−13%) and production (−13%) rates of HDL apoA-I ($P < 0.05$), thereby not altering plasma HDL apoA-I or HDL cholesterol concentrations. VLDL apoB-100 production fell significantly with weight loss ($P < 0.05$). The increase in LDL catabolism was inversely correlated with the fall in RBP-4 ($r = -0.54$, $P < 0.05$) and the decrease in HDL catabolism with the rise in adiponectin ($r = -0.56$, $P < 0.01$).

CONCLUSIONS — In obese men with metabolic syndrome, weight loss with a low-fat diet decreases the plasma LDL apoB-100 concentration by increasing the catabolism of LDL apoB-100; weight loss also delays the catabolism of HDL apoA-I with a concomitant reduction in the secretion of HDL apoA-I. These effects of weight loss could partly involve changes in RBP-4 and adiponectin levels.

Diabetes Care 30:2945–2950, 2007

Dyslipoproteinemia is a cardinal feature of central obesity and the metabolic syndrome (1). It is characterized by elevated plasma concentrations of apolipoprotein (apo)B-100, reflecting the accumulation of LDLs, and decreased plasma concentrations of apoA-I, reflecting low concentrations of HDLs. Both elevated LDL cholesterol and low HDL cholesterol are major predictors of cardiovascular events in subjects with the metabolic syndrome. The plasma ratio of apoB-100 to apoA-I is also positively associated with cardiovascular events across populations (2). Dyslipoproteinemia results from hepatic oversecretion of VLDL apoB-100, decreased catabolism of LDL apoB-100, and accelerated catabolism of HDL apoA-I (3). Weight regulation remains the cornerstone of treatment.

In general, moderately low–fat diets lower plasma triglyceride and LDL cholesterol concentrations while maintaining or lowering HDL cholesterol concentrations (4). In contrast with low-fat diets, low-carbohydrate, high-protein weight loss diets consistently increase HDL cholesterol but also elevate plasma LDL cholesterol (5). Previous studies have shown that weight loss with a low-fat diet decreases insulin resistance and cholesterol synthesis (6). Because the expression of hepatic LDL receptors is inversely related to insulin resistance (7) and the availability of cholesterol (8), weight loss could have a major effect in increasing the catabolism of LDL apoB-100. By decreasing plasma triglyceride levels, weight loss may also alter the metabolic fate of HDL particles. In a preliminary report of seven subjects with the use of isotopic ratio mass spectrometry to measure tracer enrichment (6), we suggested that weight loss increases catabolism of LDL apoB-100. However, the kinetic effects of a low-fat diet on LDL apoB-100 and HDL apoA-I in subjects with metabolic syndrome have not yet been formally investigated in a controlled study.

Retinol-binding protein-4 (RBP-4) and adiponectin are two important adipocytokines that may relate to insulin resistance and dyslipidemia in metabolic syndrome (9,10). Weight loss has been shown to lower plasma RBP-4 and elevate adiponectin levels (11,12). These effects may account for improvement in dyslipidemia with weight loss by regulating hepatic output and catabolism of VLDL, with associated remodeling of both LDL and HDL particles. The extent to which both RBP-4 and adiponectin are associ-

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Received for publication 20 April 2007 and accepted in revised form 25 July 2007.

Published ahead of print at http://care.diabetesjournals.org on 8 August 2007. DOI: 10.2337/dc07-0768.

Abbreviations: apo, apolipoprotein; ATM, adipose tissue mass; CETP, cholesteryl ester transfer protein; FCR, fractional catabolic rate; FFM, fat-free mass; HOMA, homeostasis model assessment; IDL, intermediate density lipoprotein; NEFA, nonesterified fatty acid; PLTP, phospholipid transfer protein; RBP-4, retinol-binding protein-4.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

ated with lipoprotein kinetics after weight loss in obesity remains to be clarified. Moreover, the remodeling of these lipoprotein particles is also regulated by cholesteryl ester transfer protein (CETP) and phospholipid transfer protein (PLTP) (13). However, the effect of weight loss on CETP and PLTP activities (14) and the corresponding impact on LDL and HDL metabolism are also unclear.

We hypothesized that short-term weight loss in obese men with metabolic syndrome improves the kinetics of LDL and HDL metabolism by increasing the catabolism of LDL apoB-100 and delaying the catabolism of HDL apoA-I and that these effects relate to changes in plasma RBP-4 and adiponectin levels. We also explored the corresponding relationship with alterations in plasma CETP and PLTP activities. Although our focus was LDL and HDL kinetics, for completeness we also confirmed the effect of weight loss on VLDL apoB-100 kinetics.

RESEARCH DESIGN AND METHODS

We studied 35 nonsmoking, centrally obese Caucasian men with metabolic syndrome (15). None had diabetes, the apoE2/E2 or E4/E4 genotype, macroproteinuria, creatinemia (>120 μmol/l), hypothyroidism, or abnormal liver enzymes or consumed >30 g alcohol/day. None reported cardiovascular disease or taking agents affecting lipid metabolism. The study was approved by the Royal Perth Hospital Ethics Committee. Seven subjects had participated previously in a pilot study of the effect of weight loss on LDL apoB-100 kinetics (6).

Study design and clinical protocols

Subjects entered a randomized, controlled dietary intervention study. After weight stabilization for 4 weeks, they were randomly assigned to either a hypocaloric diet for 14 weeks immediately followed by a 2-week weight stabilization period or to weight maintenance with consumption of an isocaloric diet for 16 weeks. All tests were performed, at baseline and after 16 weeks, when subjects were at a stable body weight. Body weight, height, waist circumference, and blood pressure were recorded. Body composition was estimated using a Holtain Body Composition Analyser (Holtain, Dyfed, U.K.) from which total fat mass and fat-free mass (FFM) were derived (6). Subcutaneous abdominal adipose tissue and visceral adipose tissue volumes and masses were estimated after magnetic resonance imaging, as described previously (16). All subjects were studied after a 14-h fast. Venous blood was collected for biochemical measurements before stable isotope infusion. LDL apoB-100 and HDL apoA-I kinetics were measured using primed (1 mg/kg), constant (1 $mg \cdot kg^{-1} \cdot h^{-1}$) intravenous infusion of [1-^{13}C]leucine (99.5% enrichment; Tracer Technologies, Somerville, MA) for 10 h (6). Blood samples for lipoprotein kinetic estimates were collected before and after isotope injection at 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, 6, 7, 8, 9, and 10 h. Subjects were studied in a semirecumbent position and allowed water only.

Nutritional protocols

For the first 14 weeks of weight loss, the diet prescribed provided 6,143 kJ/day (as a percentage of energy intake: 25% of energy from fat, 55% from carbohydrate, and 20% from protein). This diet was immediately followed by a 2-week isocaloric diet. In comparison, the dietary composition of the weight maintenance group was maintained constant during the study (35% of energy from fat, 40% from carbohydrate, and 20% from protein). We attempted to maintain the composition of the diet during the weight maintenance period in the intervention group to be similar to that during the weight stabilization run-in phase. Subjects were requested to maintain their usual level of physical activity and alcohol intake. These were assessed by 7-day recall questionnaires and alcohol diaries. Three-day dietary diaries were completed every 3 weeks by both groups and analyzed using DIET 4 Nutrient Calculation Software (Xyris Software, Brisbane, Australia); glycemic load and index were estimated from published tables (17).

Isolation and measurement of isotopic enrichment of LDL apoB-100 and HDL apoA-I

VLDL apoB-100, LDL apoB-100, and HDL apoA-I were isolated from plasma by sequential ultracentrifugation. The procedures for isopropanol precipitation, delipidation, hydrolysis, and derivatization of apoB-100 were described previously (3). ApoA-I was isolated from the HDL fraction by SDS-PAGE and blotted onto a polyvinylidene difluoride membrane; apoA-I bands were excised from the polyvinylidene difluoride membrane, hydrolyzed overnight (6 M hydrochloric acid, 110°C), and dried for derivatization. Isotopic enrichment of apoB-100 and apoA-I was determined using negative chemical ionization by gas chromatography–mass spectrometry.

Biochemical measurements

After precipitation of apoB-100 with isopropanol, LDL apoB-100 concentrations were determined by a modified Lowry method as described previously (6) (coefficient of variation [CV] <4.0%). Total plasma apoB-100 and apoA-I concentrations were determined by immunonephelometry (Dade Behring BN_2 nephelometer) (interassay CVs <4.3%). ApoB-100 was quantified from three pooled plasma samples during the isotope infusion; other biochemical assays were performed at baseline before the infusion. Plasma adiponectin and RBP-4 were determined using enzyme immunoassay kits according to the manufacturer's instructions (interassay CV <7%, Quantikine; R&D Systems, Minneapolis, MN; and interassay CV <10%; Immunodiagnostik, Bensheim, Germany). Plasma CETP activity was analyzed by an exogenous assay (Roar Biomedical, New York, NY). PLTP activity was determined by measuring the transfer of radiolabeled phosphatidylcholine ([^{14}C]dipalmitoylphosphatidyl choline) from unilamellar vesicles to isolated HDL, precipitating the vesicles with a $MnCl_2$/heparin solution and counting the [^{14}C]dipalmitoylphosphatidyl choline remaining in the supernatant (interassay CV <10%). Cholesterol, triglyceride, and HDL cholesterol were determined by standard enzymatic methods. LDL cholesterol was calculated using the Friedewald equation or by direct measurement with triglycerides >4.5 mmol/l. Plasma nonesterified fatty acids (NEFAs) were measured by an enzymatic method (CV <3%; Boehringer Mannheim, Mannheim, Germany). Glucose was measured by a hexokinase method (CV <3%; Bayer Diagnostics, Sydney, Australia) and insulin by an enzyme-linked immunosorbent assay (CVs <8%; Boehringer Mannheim). Insulin resistance was estimated by homeostasis model assessment (HOMA) score (18). Plasma lathosterol and campesterol concentrations were measured by gas chromatography–mass spectrometry (CV <6.0%; Hewlett Packard 5890) (19).

Models for apoB-100 and apoA-I metabolism

Tracer-to-tracee ratios were modeled using SAAM-II (University of Washington, Seattle, WA) from which fractional catabolic rates (FCRs) of LDL apoB-100 and

Table 1—*Clinical and biochemical anthropometric characteristics before and after intervention in the weight reduction and weight maintenance groups*

	Weight reduction (n = 20)		Weight maintenance (n = 15)	
	0	16 weeks	0	16 weeks
Weight (kg)	109.3 ± 2.3	96.0 ± 2.7*	105.1 ± 2.9	109.0 ± 2.2
BMI (kg/m^2)	35.2 ± 1.0	30.5 ± 0.7*	33.4 ± 0.7	35.2 ± 0.9
Waist (cm)	112.1 ± 1.7	102.6 ± 1.9*	112.9 ± 1.7	112.5 ± 1.7
Total fat mass (kg)	42.6 ± 2.7	30.0 ± 1.9*	38.8 ± 1.8	44.1 ± 2.8
FFM (kg)	65.4 ± 1.9	62.5 ± 2.0	63.9 ± 1.7	64.0 ± 1.8
Visceral ATM (kg)	7.1 ± 0.5	5.4 ± 0.4*	6.9 ± 0.4	6.7 ± 0.4
Total subcutaneous ATM (kg)	8.4 ± 0.7	6.5 ± 0.4*	9.6 ± 0.7	9.9 ± 0.7
Mean arterial pressure (mmHg)	95.4 ± 2.8	86.4 ± 2.8†	96.6 ± 3.0	94.7 ± 3.1
Cholesterol (mmol/l)	5.95 ± 0.31	5.24 ± 0.24†	5.98 ± 0.18	6.00 ± 0.21
Triglyceride (mmol/l)	3.46 ± 0.58	1.97 ± 0.18*	2.92 ± 0.58	2.71 ± 0.40
HDL cholesterol (mmol/l)	1.03 ± 0.04	1.07 ± 0.05	0.96 ± 0.04	0.95 ± 0.04
LDL cholesterol (mmol/l)	3.30 ± 0.18	3.03 ± 0.20‡	3.85 ± 0.20	3.89 ± 0.19
Total apoB-100 (g/l)	1.21 ± 0.06	1.02 ± 0.06†	1.24 ± 0.06	1.20 ± 0.05
Total apoA-I (g/l)	1.29 ± 0.05	1.26 ± 0.04	1.20 ± 0.04	1.23 ± 0.02
ApoB–to–apoA-I ratio	0.95 ± 0.04	0.82 ± 0.05‡	1.04 ± 0.06	0.99 ± 0.05
Glucose (mmol/l)	5.66 ± 0.16	5.32 ± 0.12	5.44 ± 0.18	5.45 ± 0.31
Insulin (mU/l)	14.0 ± 1.8	8.2 ± 0.7*	18.4 ± 2.7	15.5 ± 1.5
HOMA score	3.65 ± 0.54	1.99 ± 0.18†	4.57 ± 0.78	3.99 ± 0.64
NEFAs (mmol/l)	0.96 ± 0.07	0.86 ± 0.08	0.81 ± 0.05	0.82 ± 0.05
Lathosterol (μmol/l)	17.4 ± 3.4	11.9 ± 2.4‡	14.5 ± 2.1	14.4 ± 2.0
Campesterol (μmol/l)	7.23 ± 0.95	6.14 ± 0.80	5.68 ± 0.54	6.45 ± 0.61
Adiponectin (μg/ml)	3.92 ± 0.42	4.60 ± 0.44‡	3.40 ± 0.34	3.50 ± 0.37
RBP-4 (μg/ml)	38.1 ± 1.9	30.4 ± 2.1‡	35.9 ± 2.9	35.0 ± 2.9
CETP activity (nmol · ml^{-1} · h^{-1})	31.2 ± 3.4	32.4 ± 3.3	37.4 ± 2.8	39.0 ± 2.5
PLTP activity (nmol · ml^{-1} · h^{-1})	1,839 ± 270	1,841 ± 198	1,834 ± 224	1,729 ± 204

Data are means ± SEM. Significant differences in the weight loss group relative to weight maintenance group were analyzed using general linear modeling: *P < 0.001; †P < 0.01; ‡P < 0.05.

HDL apoA-I were estimated from the best fit of the model to the data. The apoB-100 model consisted of seven compartments (20). Compartment 1 represents the input of the tracer, which is connected to an intrahepatic compartment (compartment 2) that accounts for synthesis and secretion of apoB-100 into the VLDL pool (compartment 3). Compartments 3 and 4 account for the kinetics of apoB-100 in the VLDL fraction. Compartments 5 and 6 account for the kinetics of apoB-100 in the intermediate-density lipoprotein (IDL) and LDL fractions, respectively. The apoA-I multicompartmental model consisted of three compartments (21). Compartment 1 represents the tracer input, which is incorporated into an intrahepatic compartment (compartment 2) that accounts for the synthesis and secretion of apoA-I into the HDL fraction (compartment 3). LDL apoB-100 and HDL apoA-I transport rates were calculated by multiplying the FCR by pool size (milligram per kilogram of FFM per day).

Statistical analyses

All analyses were performed using SPSS version 15 (SPSS, Chicago, IL). Skewed data were log-transformed where appropriate. Treatment effects of the weight loss group relative to the weight maintenance group were analyzed using general linear modeling with adjustment for the dependent variable at baseline (i.e., end of study variable = baseline variable + treatment group + constant). Statistical significance was defined as $P < 0.05$.

RESULTS — Table 1 shows the clinical and biochemical characteristics of the subjects studied. On average, they were middle-aged, obese, dyslipidemic, and insulin resistant. There were no significant group differences in these characteristics at baseline. With the weight loss diet, there was a significant reduction in body weight (−12.2%, $P < 0.001$), waist circumference (−8.5%, $P < 0.001$), total fat mass (−29.6%, $P < 0.001$), visceral (−23.5%, $P < 0.001$) and subcutaneous (−22.5%, $P < 0.001$) abdominal adipose tissue masses (ATMs), and mean arterial pressure (−9.43%, $P < 0.01$), but no significant changes in FFM. Compared with weight maintenance, the weight loss diet significantly ($P < 0.05$) lowered plasma concentrations of total cholesterol (−12%), triglycerides (−43%), LDL cholesterol (−8%), and total apoB-100 (−17%); ratios of LDL cholesterol to HDL cholesterol (−9%) and of apoB-100 to apoA-I (−14%); and lathosterol (−23%), as well as insulin (−34%) and HOMA score (−40%). With weight loss there was also a significant ($P < 0.05$) increase and decrease in plasma levels of adiponectin (+17%) and RBP-4 (−20%), respectively. However, there were no significant effects of weight loss on plasma concentrations of NEFAs, glucose, and HDL cholesterol or on plasma CETP and PLTP activities.

Table 2 summarizes the dietary composition and nutrient intake of subjects during the study. There was no significant difference in dietary intake between groups at baseline. Subjects in the weight

Table 2—*Dietary composition and nutrient intake in the weight reduction and weight maintenance groups*

	Weight reduction (n = 20)			Weight maintenance (n = 15)		
	0	6 weeks	14–16 weeks	0	6 weeks	14–16 weeks
Total energy (kJ)	9,782 ± 438	6,143 ± 363*	6,979 ± 312*	10,246 ± 634	9,914 ± 705	9,738 ± 397
Fat (g)	102 ± 5	55 ± 7*	67 ± 6*	99 ± 8	103 ± 9	103 ± 6
Fat (% energy)	37 ± 1	26 ± 2*	35 ± 3	35 ± 2	37 ± 1	39 ± 2
Carbohydrate (g)	218 ± 11	177 ± 9†	159 ± 13†	245 ± 21	234 ± 21	220 ± 13
Carbohydrate (% energy)	37 ± 2	48 ± 2*	39 ± 4	40 ± 2	39 ± 2	38 ± 2
Protein (g)	112 ± 5	74 ± 5†	83 ± 5†	121 ± 6	112 ± 9	114 ± 6
Protein (% energy)	19 ± 1	21 ± 1	20 ± 1	20 ± 1	19 ± 1	20 ± 1
Alcohol (g)	23 ± 7	12 ± 4	14 ± 4	17 ± 4	15 ± 5	12 ± 5
Alcohol (% energy)	10 ± 5	5 ± 1	6 ± 1	5 ± 1	4 ± 2	4 ± 1
Glycemic index	56 ± 2	57 ± 2	54 ± 2	52 ± 5	57 ± 1	58 ± 1
Glycemic load	124 ± 10	89 ± 7†	94 ± 7†	134 ± 14	129 ± 10	132 ± 9

Data are means ± SEM. Significant differences in the weight loss group at 6 week and during weight stabilization period (14–16 weeks) relative to weight maintenance group were analyzed using general linear modeling: *$P < 0.001$; †$P < 0.01$.

loss group significantly reduced their total energy and fat and significantly increased carbohydrate consumption during the active weight loss period. Energy and nutrient intake did not change in the subjects in the weight maintenance group. That the subjects on the weight loss diet consumed an isocaloric diet from weeks 14 to 16 was supported by the fact that body weight did not vary by >1% during this period. Glycemic load decreased significantly in the weight loss group compared with that in the weight maintenance group, but the glycemic index did not. There was also no change in reported physical activity levels during the study in either the weight loss or weight maintenance groups (data not shown).

Table 3 shows the kinetic indexes for VLDL, LDL, and HDL metabolism in the two groups. There were no significant group differences in lipoprotein kinetics at baseline. As before (13), weight loss significantly decreased the pool size (−41%, $P = 0.007$), concentration (−47%, $P = 0.003$), and production rate (−47%, $P < 0.05$) of VLDL apoB-100 but did not change VLDL apoB-100 FCR. There was a significant decrease ($P < 0.05$) in the weight loss group in the plasma LDL apoB-100 concentration (−24%) and pool size (−23%), as well as a significant increase in the LDL apoB-100 FCR (+27%), but no change in the LDL apoB-100 production rate. Weight loss was also associated with an increase in the percent conversion of VLDL apoB-100 to LDL apoB-100 (+23%, $P < 0.01$), and this

Table 3—*Lipoprotein kinetics in the subjects before and after weight reduction and weight maintenance*

	Weight reduction (n = 20)		Weight maintenance (n = 15)	
	0	16 weeks	0	16 weeks
VLDL apoB				
Pool size (mg)	358 ± 57	218 ± 28*	287 ± 66	318 ± 48
Concentration (mg/l)	106 ± 19	56 ± 6*	86 ± 18	96 ± 14
Fractional catabolic rate (pools/day)	4.26 ± 0.79	4.61 ± 0.73	8.05 ± 2.57	5.11 ± 0.92
Production rate (mg · kg FFM^{-1} · day^{-1})	23.8 ± 3.01	15.6 ± 1.63*	30.0 ± 4.65	28.1 ± 3.92
LDL apoB				
Pool size (mg)	1,347 ± 127	1,038 ± 89†	1,217 ± 116	1,270 ± 110
Concentration (mg/l)	390 ± 33	296 ± 24†	348 ± 38	385 ± 29
Fractional catabolic rate (pools/day)	0.51 ± 0.05	0.65 ± 0.06†	0.63 ± 0.07	0.51 ± 0.05
Production rate (mg · kg FFM^{-1} · day^{-1})	14.1 ± 1.90	12.1 ± 1.17	16.5 ± 1.49	15.6 ± 2.17
HDL apoA-I				
Pool size (mg)	4,394 ± 210	4,427 ± 204	4,038 ± 147	3,929 ± 165
Concentration (mg/l)	1,290 ± 46	1,256 ± 44	1,199 ± 35	1,228 ± 25
Fractional catabolic rate (pools/day)	0.23 ± 0.01	0.20 ± 0.01†	0.25 ± 0.01	0.24 ± 0.01
Production rate (mg · kg FFM^{-1} · day^{-1})	15.1 ± 1.12	13.1 ± 0.97†	15.5 ± 1.01	15.1 ± 1.13
Lipoprotein channeling				
% of total	67.7 ± 6.8	83.5 ± 5.5*	70.5 ± 8.3	64.4 ± 7.8
% of direct	12.2 ± 3.5	19.4 ± 5.2	10.1 ± 2.1	14.6 ± 2.6
% via IDL	55.5 ± 6.9	64.2 ± 5.6‡	60.4 ± 8.2	49.8 ± 7.6

Data are means ± SEM. Significant differences in the weight loss group relative to weight maintenance group were analyzed using general linear modeling: *$P < 0.01$; †$P < 0.05$; ‡$P = 0.06$.

increase was chiefly attributed to channelling via IDL (+16%, $P = 0.06$). The increase in LDL apoB-100 FCR was significantly correlated with the decrease in the pool size of LDL apoB-100 ($r = -0.60$, $P < 0.01$). Compared with weight maintenance, weight loss decreased HDL apoA-I production (−13%, $P < 0.05$) and FCR (−13%, $P = 0.02$), with no significant changes in the plasma concentration or pool size of HDL apoA-I. The changes in HDL apoA-I FCR and production rate were highly correlated ($r = 0.72$, $P < 0.001$). However, the changes in LDL and HDL FCR with weight loss were not statistically correlated.

The increase in LDL apoB-100 FCR was significantly correlated with the fall in RBP-4 ($r = -0.546$, $P < 0.05$) but not with changes in adiponectin or insulin; in a regression model including all three variables, the regression coefficient for RBP-4 as a predictor of LDL apoB-100 FCR was significant (β coefficient = −0.583, $P = 0.01$). The association between LDL apoB-100 FCR and RBP-4 also remained significant in regression models including RBP-4 and two extra predictors selected from changes in visceral ATM, subcutaneous ATM, total ATM, triglycerides, NEFAs, and lathosterol. The decrease in HDL apoA-I FCR was significantly correlated with changes in adiponectin ($r = -0.561$, $P < 0.05$), but not with changes in RBP-4 or insulin; in a regression model including all three variables, the regression coefficient for adiponectin as a predictor of HDL apoA-I FCR was significant (β coefficient = −0.555, $P = 0.014$). This association also remained significant in regression models including adiponectin and two extra predictors selected from changes in visceral ATM, subcutaneous ATM, total ATM, triglycerides, NEFAs, and lathosterol.

CONCLUSIONS — We extend previous reports by examining a larger number of obese subjects with the metabolic syndrome in a placebo-controlled study design investigating the effect of weight loss with a moderately low-fat diet on LDL apoB-100 and HDL apoA-I metabolism. We focused on LDL and HDL kinetic changes and confirmed our previous demonstration that weight loss decreases hepatic secretion of VLDL apoB-100 (6). Our new findings were that weight loss had favorable and opposing effects on the fractional catabolism of LDL apoB-100 and HDL apoA-I that were related to changes in plasma RBP-4 and adiponectin levels, respectively. The increase in the fractional catabolism of LDL without change in LDL production accounted for the decrease in LDL apoB-100 and LDL cholesterol. The fall in fractional catabolism of HDL apoA-I was tightly correlated with the fall in its secretion, so that HDL apoA-I and HDL cholesterol concentrations remain unaltered. We did not confirm that the foregoing kinetic changes were related to changes in plasma lipid transfer protein activities.

Data interpretation: mechanisms

The increase in fractional catabolism of LDL apoB-100 with weight loss could involve multiple mechanisms, including a decrease in hepatic de novo cholesterol synthesis, in hyperinsulinemia, and in liver fat content. LDL receptor synthesis is regulated by a feedback mechanism linked to cellular cholesterol content (8). An improvement in insulin resistance decreases cholesterol synthesis, thereby increasing LDL receptor activity (7,8). RBP-4 levels are directly related to liver fat content (22), consistent with experimental data suggesting that impaired retinoic acid signaling can lead to hepatic steatosis (23), and this may involve inhibition of hepatic peroxisome proliferator–activated receptor-α. Hence, the inverse association we report between LDL apoB-100 FCR and RBP-4 may reflect changes in hepatic fat content, including decreased availability of cholesterol substrate, as well as fatty acids that per se can have a direct impact on cholesterol synthesis (24). Although plasma free fatty acid levels did not alter in our study, these may not reflect the corresponding portal or hepatic concentrations that regulate apoB-100 metabolism. Whether an LDL-lowering effect of RBP-4 with weight loss also involves a reduction in proprotein convertase subtilisin/kexin type 9 expression merits investigation (25). By decreasing VLDL triglycerides, weight loss leads to the formation of larger size LDL particles that are catabolized more rapidly (26). Increase in LDL size could also partially explain our finding of accelerated LDL apoB-100 FCR. However, changes in plasma lipid transfer protein activities with weight loss do not appear to contribute to the lipoprotein kinetic changes, consistent with reports indicating that plasma lipid transfer protein activities do not alter with weight loss (14). Despite a reduction in the hepatic secretion of VLDL apoB-100, we did not observe decreased production of LDL apoB-100. This result may be explained by our finding of increased conversion of VLDL to LDL apoB-100 and may be a consequence of increased lipoprotein lipase activity.

The catabolic changes in HDL with weight loss could relate to an increase in HDL particle size, which in turn may be a consequence of a reduction in the plasma VLDL triglyceride pool available for exchange with HDL (27). Increased adiponectin can inhibit hepatic lipase activity (28), which could account for the partial correlation in our study between changes in plasma adiponectin and HDL apoA-I FCR. A "balancing feedback" mechanism probably accounts for the tight correlation between changes in catabolism and production of HDL apoA-I after weight loss. Furthermore, the fact that HDL underproduction offset the HDL-elevating effect of depressed HDL catabolism could in part reflect the impact of lowered dietary fat intake on the hepatic expression and secretion of apoA-I (29). However, we found no significant correlation between the changes in HDL apoA-I production rate and dietary saturated fat intake in our weight loss group. That there was no significant correlation between the changes in LDL and HDL FCR suggests that different mechanisms underlie these alterations in lipoprotein metabolism after weight loss.

Limitations

We did not estimate the kinetics of LDL subspecies. Because in the present context the reduction in hepatic VLDL apoB-100 secretion with weight loss is likely to reflect chiefly $VLDL_1$ apoB-100 secretion (26), we suggest that the production of LDL_2 would have also decreased with weight loss. We only examined the short-term effect of weight loss followed by a 2-week isocaloric weight-stabilizing period, but we and others have shown favorable effects on lipoprotein metabolism with this regimen (6). More prolonged periods of weight maintenance can lead to rebound changes in plasma lipids that could mask the full benefit of weight loss. Our use of a primed-constant infusion of isotope (10 h) may lack precision for measurement of LDL or HDL kinetics, but this methodology has been well correlated with a bolus injection technique (20).

Implications

The reduction in the ratio of apoB-100 to apoA-I could translate into a significant decrease in risk of cardiovascular disease in metabolic syndrome (2). Although we

may have seen an increase in HDL concentration with a longer period of weight maintenance, our data suggest that achieving a similar effect in the short-term would require other treatments, such as peroxisome proliferator–activated receptor-α agonists (3) that increase apoA-I secretion. Rimonabant also incrementally increases HDL relative to weight loss and may partly achieve this by increasing plasma adiponectin.

In summary, in men with the metabolic syndrome, short-term weight loss with a low-fat diet increases the catabolism of LDL apoB-100 and decreases the catabolism of HDL apoA-I. The full benefit on HDL metabolism is offset, however, by reduced secretion of HDL apoA-I. Further studies should be conducted to explore the mechanism and effect of weight loss with different diets and lifestyle modifications on apoB-100 and apoA-I kinetics in a wider group of subjects and the incremental benefits of selected pharmacotherapies, as well as the effect of more extended periods of weight loss.

Acknowledgments— This work was supported by the National Heart Foundation of Australia and National Health and Medical Research Foundation (NHMRC). D.C.C. is an NHMRC Career Development Fellow. P.H.R.B. is an NHMRC senior research fellow and is supported in part by National Institutes of Health Grant NIBIB P41-EB-001975.

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