Nutritional regulation of cholesterol synthesis and apolipoprotein B kinetics: studies in patients with familial hypercholesterolemia and normal subjects treated with a high carbohydrate, low fat diet

Peter W. Stacpoole,[1,*,†] Klaus von Bergmann,[††] Laura L. Kilgore,[*] Loren A. Zech,[***] and Waldo R. Fisher[*,]**

Departments of Medicine (Endocrinology and Metabolism),[*] Pharmacology,[†] and Biochemistry,[**] University of Florida, College of Medicine, Gainesville, FL 32610; Department of Medicine,[††] (Clinical Pharmacology), University of Bonn, 5300 Bonn 1, Germany; and Laboratory of Mathematical Biology,[***] National Institutes of Health, Bethesda, MD 20892

Abstract High carbohydrate, low fat diets decrease plasma low-density lipoprotein cholesterol (LDL-C) and apolipoprotein B (apoB) mass in normal subjects and in patients with familial hypercholesterolemia (FH). To investigate the mechanisms for these effects, four normal, four FH heterozygous, and one FH homozygous subjects were studied on a basal (45% carbohydrate, 40% fat) diet and during continuous nasogastric infusion of Vivonex (90% carbohydrate, 1% fat). For the entire group, the mean changes in total cholesterol, LDL-C, high-density lipoprotein cholesterol (HDL-C) and triglycerides were −90, −95, −14 (all $P < 0.01$) and +114 ($P < 0.02$) mg/dl, respectively. Fecal sterol balance measurements demonstrated a 24% decrease in whole body cholesterol synthesis in normals, from 8.4 ± 4.4 (mean ± SD) to 6.4 ± 1.3 mg/kg per day and in FH subjects, a 58% decrease, from 11.4 ± 5.6 to 4.8 ± 1.7 mg/kg per day (both $P < 0.05$). ApoB kinetic studies were performed using a [^{3}H]leucine tracer in two normals and three FH heterozygotes on both basal and Vivonex regimens, and the results were analyzed by compartmental modeling using the SAAM program. Total apoB production was not altered in a consistent manner by carbohydrate feeding. ApoB secretion, however, was shifted from the production of small VLDL/IDL-like particles to large VLDL by Vivonex, with an accompanying increase in intrahepatic assemblage time before secretion. In the two normal subjects, Vivonex induced an increase in apoB loss as VLDL/IDL; however, in the FH patients no such loss occurred. A decrease ($P < 0.05$) in the residence time of LDL-apoB occurred for all subjects and was the primary determinant of the fall in plasma LDL concentration, since LDL-apoB transport did not change consistently. Thus, in FH patients, a high carbohydrate, low fat diet results in suppression of cholesterol synthesis and a fall in plasma LDL concentration due to an increased plasma clearance rate for LDL.—**Stacpoole, P. W., K. von Bergmann, L. L. Kilgore, L. A. Zech, and W. R. Fisher.** Nutritional regulation of cholesterol synthesis and apolipoprotein B kinetics: studies in patients with familial hypercholesterolemia and normal subjects treated with a high carbohydrate, low fat diet. *J. Lipid Res.* 1991. **32:** 1837–1848.

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High carbohydrate, low fat diets have long been known to lower serum total and low density lipoprotein (LDL) cholesterol in humans and to induce at least a transient elevation of circulating triglycerides (1–3). Changes have also been recorded in the lipid and apolipoprotein composition of lipoprotein particles, the magnitude of which appear to relate in part to the amount and type of carbohydrate in the diet and the underlying genotype of the individual (4–8).

We previously reported that administration of a very high carbohydrate, low fat liquid diet (Vivonex) in two patients with receptor-negative homozygous familial hypercholesterolemia (FH) reduced circulating LDL cholesterol levels and led in one patient to marked suppression of total body cholesterol synthesis (5, 9). Animal studies showed that high carbohydrate solid or liquid diets inhibited hepatic 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase activity and cholesterolgenesis (10–12).

In this investigation we examined the cholesterol-lowering response to high carbohydrate, low fat feedings in normal subjects and patients with homozygous or heterozygous FH, using the combined techniques of whole body cholesterol balance and tracer kinetics, in which

Abbreviations: FH, familial hypercholesterolemia; VLDL, very low density lipoproteins; IDL, intermediate density lipoproteins; LDL, low density lipoproteins; HDL, high density lipoproteins; SAAM, simulation, analysis and modeling; apoB, apolipoprotein B; apoE, apolipoprotein E; HMG-CoA, 3-hydroxy-3-methylglutaryl coenzyme A; FCR, fractional catabolic rate.

[1]To whom correspondence should be addressed at: Box J-226, University of Florida, College of Medicine, Gainesville, FL 32610.

tritiated leucine was used as an endogenous tracer for apolipoprotein B (apoB) metabolism.

Serum total and LDL cholesterol levels fell in both healthy controls and FH patients on a high carbohydrate, low fat regimen. The decrease in circulating cholesterol was associated with an inhibition of endogenous cholesterol synthesis and a stimulation of LDL apolipoprotein B-100 (apoB-100) clearance. The data demonstrate a shift in the secretion of apoB from cholesteryl ester-enriched to triglyceride-enriched particles. In FH heterozygotes, however, the primary mechanism accounting for the reduction of LDL-cholesterol levels was an increased clearance of LDL, presumably by the liver, which may not depend entirely on the integrity of the classical LDL (apoB,E) receptor pathway.

METHODS

Subjects

This investigation was approved by the Institutional Review Board of the Health Center of the University of Florida. Participants in this study included four healthy male volunteers, aged 33–58 years, and five patients with FH, aged 20–65 years. Four FH subjects were heterozygotes and one was a compound heterozygote whose combined defects in LDL receptor number and activity rendered him functionally similar to FH homozygotes with less than 2% LDL receptor activity (J. L. Goldstein, personal communication). Drugs known to alter lipid metabolism were discontinued at least 1 month prior to study.

Diet

Studies were conducted on the Clinical Research Center ward of the Shands Hospital under two isocaloric, metabolically steady-state conditions. Each diet was administered for 1 month, with at least a 3 month wash-out period occurring between each diet. The initial (basal) regimen was weight-maintaining and had a caloric distribution of 45% carbohydrate, 40% fat, and 15% protein. The polyunsaturated:saturated fat ratio was approximately 1.0 and the daily cholesterol intake was approximately 100 mg/day. The diet was divided into three meals and a bedtime snack. The subsequent (test) diet was Vivonex (Norwich-Eaton Pharmaceuticals), administered as a constant, 24-h infusion through a small-bore (No. 8 French) nasogastric feeding tube. Vivonex contains 90.5% of calories as glucose or glucose oligosaccharides, 8.2% as crystalline amino acids, and 1.3% as safflower oil (containing 80% linoleic acid). The diet was supplemented with vitamins and minerals and contained no cholesterol. Ferrous sulfate (325 mg) was administered twice daily throughout each diet period.

Lipids and lipoproteins

Blood was obtained during each kinetic study for serum lipid and lipoprotein determinations after 11- to 12-h overnight fasts (basal diet) or during constant feedings (Vivonex). Serum total cholesterol and triglycerides were measured by standard, automated procedures. High density lipoprotein (HDL) cholesterol was determined by heparin-manganese precipitation (13), and LDL cholesterol was estimated by the Friedewald equation (14).

Sterol balance

Stools were collected daily and aliquots were taken over the final 10–12 days of each diet for analysis of neutral (15) and acidic (16) steroids. Neutral steroid data included determination of cholesterol, coprostanone, coprostanol, and epicholesterol. Beta-sitosterol (225 mg twice daily) and chromic oxide (60 mg 3 times daily) were administered throughout each diet period and were used to calculate cholesterol balance, as described previously (9).

ApoB kinetics

The study protocol and methods for apoB isolation and specific radioactivity measurement have been described (17). Briefly, after 7–8 days of inpatient dietary equilibration, 5 μCi/kg [^{3}H]leucine was injected into a peripheral vein, and timed blood samples were obtained over the next 13 days for isolation of VLDL and LDL. The apoB was recovered from these lipoproteins, and its specific radioactivity was determined. At each sampling time, plasma was also processed for the recovery of free leucine, and plasma leucine concentration and its specific radioactivity were measured.

The kinetic data were analyzed by mathematical, compartmental modeling using the SAAM/CONSAM computer programs (18). The apoB model (**Fig. 1**) used in analyzing these data has been described in detail (17).

Statistical analyses

Since subjects served as their own controls, all data were analyzed as the means of paired differences, and the analysis tested the differences against the null hypothesis using a two-tailed t-test. Where indicated, logarithmic transformation of the data was performed.

RESULTS

Serum lipids and lipoproteins

Table 1 summarizes the changes in serum lipids and lipoproteins induced by the high carbohydrate, low fat diet. Every normal and FH subject sustained a decrease in total and LDL cholesterol. Even the homozygous FH patient showed a 43% fall in LDL cholesterol, from 596

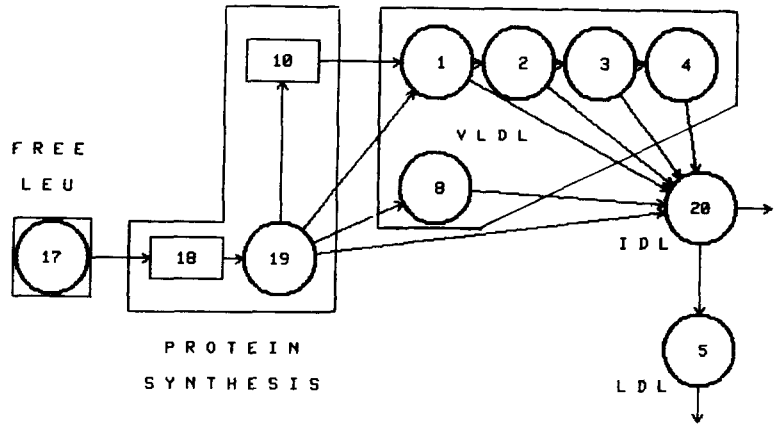


Fig. 1. Compartmental model of apolipoprotein B metabolism. [^{3}H] leucine tracer is injected into plasma (C17) and is incorporated in cellular biosynthetic pathways with production of apoB (C18→C19→C10). Nascent apoB enters VLDL through three pathways and also enters IDL (C20). See text and ref. 17 for further description of model.

mg/dl on the basal diet to 331 mg/dl on Vivonex. HDL cholesterol concentrations also decreased in all subjects with carbohydrate feeding, while the LDL cholesterol/HDL cholesterol ratio was variably affected. As anticipated, short-term administration of a high carbohydrate regimen increased serum triglycerides.

Sterol balance

Table 2 illustrates the total body cholesterol synthesis measured in four normal and three FH patients, one of whom was the homozygous subject. Daily cholesterol intake averaged 100 mg of the basal diet. On this regimen, excretion of neutral (cholesterol) and acidic (bile acid)

TABLE 1. Changes in serum lipids and lipoproteins induced by a high carbohydrate, low-fat diet in normal subjects and patients with familial hypercholesterolemia

Diagnosis Subjects (Sex)	Weight	Age	Diet	Total Chol	Triglycerides	LDL-Chol	HDL-Chol	LDL-Chol/ HDL-Chol
	kg	*yr*				*mg/dl*		
Normal								
N1 (M)	61	33	Basal	152	98	91	42	2.2
			Vivonex	114	159	44	38	1.2
N2 (M)	72	46	Basal	209	112	152	34	4.5
			Vivonex	145	126	94	26	3.6
N3 (M)	90	58	Basal	250	203	ND[a]	ND[a]	ND[a]
			Vivonex	192	546			
N4 (M)	72	33	Basal	136	56	60	66	0.9
			Vivonex	103	117	49	32	1.5
Mean difference				43	120	39	15	1.6
SD				7.5	75	14	9.4	0.54
Probability				<0.01	NS	NS	NS	NS
FH								
FH1 (F)	62	52	Basal	426	106	358	47	7.6
			Vivonex	349	175	274	39	7.0
FH2 (F)	78	40	Basal	549	134	430	92	4.7
			Vivonex	374	222	267	63	4.2
FH3 (M)	69	52	Basal	232	171	161	44	3.7
			Vivonex	162	137	98	35	2.8
FH4 (M)	68	20	Basal	645	181	596	20	29.8
			Vivonex	436	331	342	13	26.3
FH5 (F)	66	61	Basal	455	131	455	51	7.4
			Vivonex	373	409	373	31	8.4
Mean difference				123	110	129	15	0.9
SD				29	51	36	4.3	0.7
Probability				<0.025	NS	<0.025	<0.05	NS
Data for total population								
Mean difference				90	114	95	15	0.7
SD				20	41	27	4.0	0.5
Probability				<0.01	<0.025	<0.01	<0.01	NS

The means of at least three individual determinations of serum lipid and lipoprotein levels from each subject were used to calculate the value for each entered datum.

[a]ND, not determined because of lipemia occuring during Vivonex administration.

TABLE 2. Changes in total body cholesterol and bile acid synthesis induced by a high carbohydrate, low-fat diet in normal subjects and patients with familial hypercholesterolemia

Diagnosis Diet	Weight	Dietary Cholesterol Intake (A)	Fecal Sterol Excretion						Cholesterol Balance (B-A)	
			Neutral Steroids		Acidic Steroids		Total Steroids (B)			
	kg	*mg/d*	*mg/d*	*mg/kg/d*	*mg/d*	*mg/kg/d*	*mg/d*	*mg/kg/d*	*mg/d*	*mg/kg/d*
Normal, n = 4										
Control	74.1 ± 12.4	115 ± 19	499 ± 215	6.6 ± 2.0	223 ± 142	3.0 ± 1.9	772 ± 328	9.6 ± 3.6	632 ± 369	8.4 ± 4.4
Vivonex	72.0 ± 19.2	0	296 ± 116	4.0 ± 0.9	165 ± 131	2.4 ± 1.4	468 ± 164	6.4 ± 1.3	468 ± 164	6.1 ± 1.3
FH, n = 3										
Control	70.8 ± 5.9	93 ± 10	699 ± 422	9.6 ± 5.0	227 ± 56	3.6 ± 1.7	926 ± 466	12.8 ± 5.4	832 ± 470	11.4 ± 5.6
Vivonex	71.0 ± 62.0	0	264 ± 139	3.6 ± 1.7	80 ± 7	1.1 ± 0.1	347 ± 140	4.8 ± 1.7	347 ± 140	4.8 ± 1.7
Mean ± SD of the difference				4.4 ± 1.1		1.6 ± 0.4		5.3 ± 1.4		3.9 ± 1.6
P value				<0.01		<0.005		<0.025		<0.05

sterols was slightly greater in FH individuals than in healthy subjects. Changing to a high carbohydrate, low fat diet decreased fecal cholesterol and bile acid production and inhibited whole body cholesterol formation in the healthy subjects and, even more strikingly, in every FH patient. During Vivonex administration, cholesterol synthesis fell 24%, from 8.4 ± 4.4 to 6.4 ± 1.3 mg/kg per day in the controls and 58%, from 11.4 ± 5.6 to 4.8 ± 1.7 mg/kg per day, in the FH subjects.

ApoB kinetics on a high carbohydrate, low fat diet

ApoB production varied among the subjects during the control diet (**Table 3**). During Vivonex administration, apoB production decreased in the two normal individuals and in one FH patient but increased in the other two FH subjects. Thus carbohydrate feeding is not a primary determinant of apoB production.

As previously reported (17), apoB may be secreted by way of a VLDL delipidation pathway [C(1)→C(2)→C(3)→C(4)], by which large, triglyceride-enriched VLDL enter plasma, or as smaller VLDL or IDL, C(8) and C(20), by the IDL pathway (Fig. 1, and ref 1). **Fig. 2** illustrates the resolution of these two pathways in fitting the VLDL kinetic data from one subject while receiving Vivonex. **Fig. 3** presents experimental data and model-derived computer fits for VLDL-apoB for the five subjects studied on a control diet and after receiving Vivonex.

As shown in **Fig. 4A,** the high carbohydrate, low fat diet shifted apoB secretion from the IDL to the delipidation pathway ($P = 0.02$). Total apoB secretion, however, did not change (**Table 4**). Under control conditions, apoB secreted as large VLDL generally resides within the hepatocyte for the same time as it does when secreted as small VLDL or IDL, i.e., these three secretory pathways originate directly in C(19) of Fig. 1. Alternately, VLDL-apoB may be secreted after an additional delay within the hepatocyte, represented by compartment 10 in the model, by the pathway [C(19)→C(10)→C(1)→C(2)→C(3)→C(4)→]. In normal and FH subjects this pathway was most prominent during carbohydrate feeding, when VLDL-triglyceride secretion is known to increase (19, 20). The data in Table 4 and Fig. 4B demonstrate that Vivonex induced a significant ($P < 0.05$) shift in apoB secretion from the C(19)→C(1) to the C(19)→C(10)→C(1) pathway by which large VLDL are believed to be secreted. However, the increase in intrahepatic residence time for apoB traversing this pathway was highly variable (Table 3). Thus the induction of hypertriglyceridemia altered the metabolism of apoB by shifting the plasma input of apoB from the IDL

TABLE 3. Residence time of apoB on control and Vivonex diets

Subjects	ApoB Synthesis	Secretory Delay	VLDL	LDL
	hours			
Normal				
N1				
Control	1.2	0.0	3.4	52.1
Vivonex	1.2	0.0	4.6	34.7
Diff	0.0	0.0	−1.2	17.4
N2				
Control	2.1	0.0	2.5	38.5
Vivonex	1.0	3.3	1.8	27.5
Diff	1.1	−3.3	0.7	11.0
FH subjects				
FH1				
Control	0.6	0.6	19.0	227.0
Vivonex	0.8	0.8	6.6	69.7
Diff	−0.2	−0.2	12.4	157.3
FH2				
Control	2.0	0.0	1.1	68.3
Vivonex	1.2	0.8	4.9	58.2
Diff	0.8	−0.8	−3.8	10.1
FH3				
Control	1.1	0.0	4.5	55.2
Vivonex	1.0	0.3	5.6	31.2
Diff	0.1	−0.3	−1.1	24.0
Mean differences	0.36	−0.32	1.4	44.0
SD	0.25	0.13	2.8	28.4
Probability	NS	NS	NS	<0.05[a]

[a]Performed after logarithmic transformation.

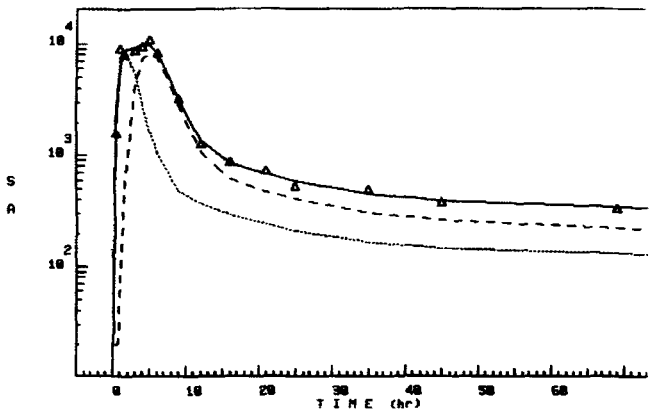


Fig. 2. Computer fit of VLDL-apoB kinetic data for subject N2, a normal subject receiving Vivonex. The model-generated curve (—) is resolved into two components, a fast input component (· · · ·) fitting the early portion of the data (C19→C8), and a component (----) with a delayed input that enters a plasma delipidation chain (C19→C10→C1→C4). Data (△) are specific activity of apoB (dpm/mg); time in hours.

to the delipidation pathway and by lengthening the time within the hepatocyte required to secrete a VLDL particle before its entry into the delipidation chain.

In the three FH patients, Vivonex did not induce any catabolic loss of apoB as VLDL- or IDL-sized particles, although such was again demonstrable in the control subjects (Table 4). Nor was a change in VLDL residence time observed (Table 3). In both healthy subjects and in two FH patients, VLDL-apoB mass increased while Vivonex was administered, although the magnitude of the change was variable and not significant (**Table 5**).

In all subjects the residence time of LDL-apoB decreased during the Vivonex feeding, reflecting an increased rate for the removal of LDL-apoB from plasma (Table 3). The LDL-apoB mass also decreased in all subjects (Table 5). The magnitude of this change, however, was not well correlated with the change in the residence time of LDL, due to variable production rates of LDL-apoB. In each individual, therefore, the decrease in LDL residence time induced by Vivonex was sufficient to determine that a decrease in the mass of LDL-apoB occurred, but the magnitude of the decrease in mass among the subjects was a function of the differences in the LDL transport rates for these individuals.

DISCUSSION

The focus of this study was to investigate the mechanism of the cholesterol-lowering action of high carbohydrate, low fat diets. In so doing, we also tested the hypothesis that the fall in plasma cholesterol induced by carbohydrate feeding reflected changes in the metabolism of VLDL and LDL.

Diets enriched in carbohydrate exert complex effects on the concentrations and metabolism of lipids and lipoproteins. Many, if not most, normolipemic or hypercholesterolemic individuals who consume over 50% of their daily calories as carbohydrate demonstrate a significant reduction in serum total cholesterol, compared to subjects in whom carbohydrates make less of a caloric contribution (1–3). The magnitude of this change, however, relates to many factors, including the amount and type of carbohydrate, duration of feeding, route of administration, lipoprotein phenotype (5–8), and degree of glucose tolerance (4, 21–23). The fall in total cholesterol concentration is usually the net result of decrements in both LDL cholesterol and HDL cholesterol and an increase in VLDL cholesterol. It is also frequently associated with at least a transient elevation of serum triglycerides (6, 7, 21, 23–26). Despite, or because of, these changes in circulating lipids and lipoproteins, epidemiologic studies indicate that populations whose diets are higher in carbohydrate and lower in fat than those currently consumed by most Western cultures may be at reduced risk for atherosclerosis (26–30). What has not been determined is whether the change in the amount of either of the specific nutrients (carbohydrate or fat) exerts the greater influence on cholesterol metabolism.

The decrease in circulating cholesterol observed in this study was attended by marked suppression of cholesterol synthesis. This was particularly striking in the FH individuals, in whom sterol balance data revealed a mean 58% decrease in whole body cholesterol production with Vivonex. These findings extend our previous observations (5, 9) and those of others (24, 31) on the cholesterol-lowering effect of orally or parenterally administered glucose in normal or FH subjects. In addition, they firmly establish that one mechanism accounting for the reduction in serum cholesterol is inhibition of total body cholesterol synthesis. Animal and human studies have shown that glucose and other monosaccharides are relatively poor substrates for hepatic cholesterolgenesis, compared to fatty acids or lactate (32–36). Indeed, when rats are fed increasing amounts of carbohydrate in place of fat, the activity of hepatic HMG-CoA reductase, the rate-limiting enzyme of cholesterolgenesis, falls and the rate of hepatic cholesterol synthesis is reduced (10–12). One result of this dietary perturbation should, therefore, be a reduction in the cholesterol content of the hepatocyte.

In contrast, increased dietary carbohydrate stimulates hepatic triglyceride formation and the activities of several rate-determining enzymes of lipogenesis, such as fatty acid synthetase and malic enzyme (37, 38). These changes in the relative rates of hepatic triglyceride and cholesterol production are consistent with observations in humans that carbohydrate feeding leads to relative enrichment in triglyceride of VLDL, LDL, and HDL particles (1, 7, 39, 40).

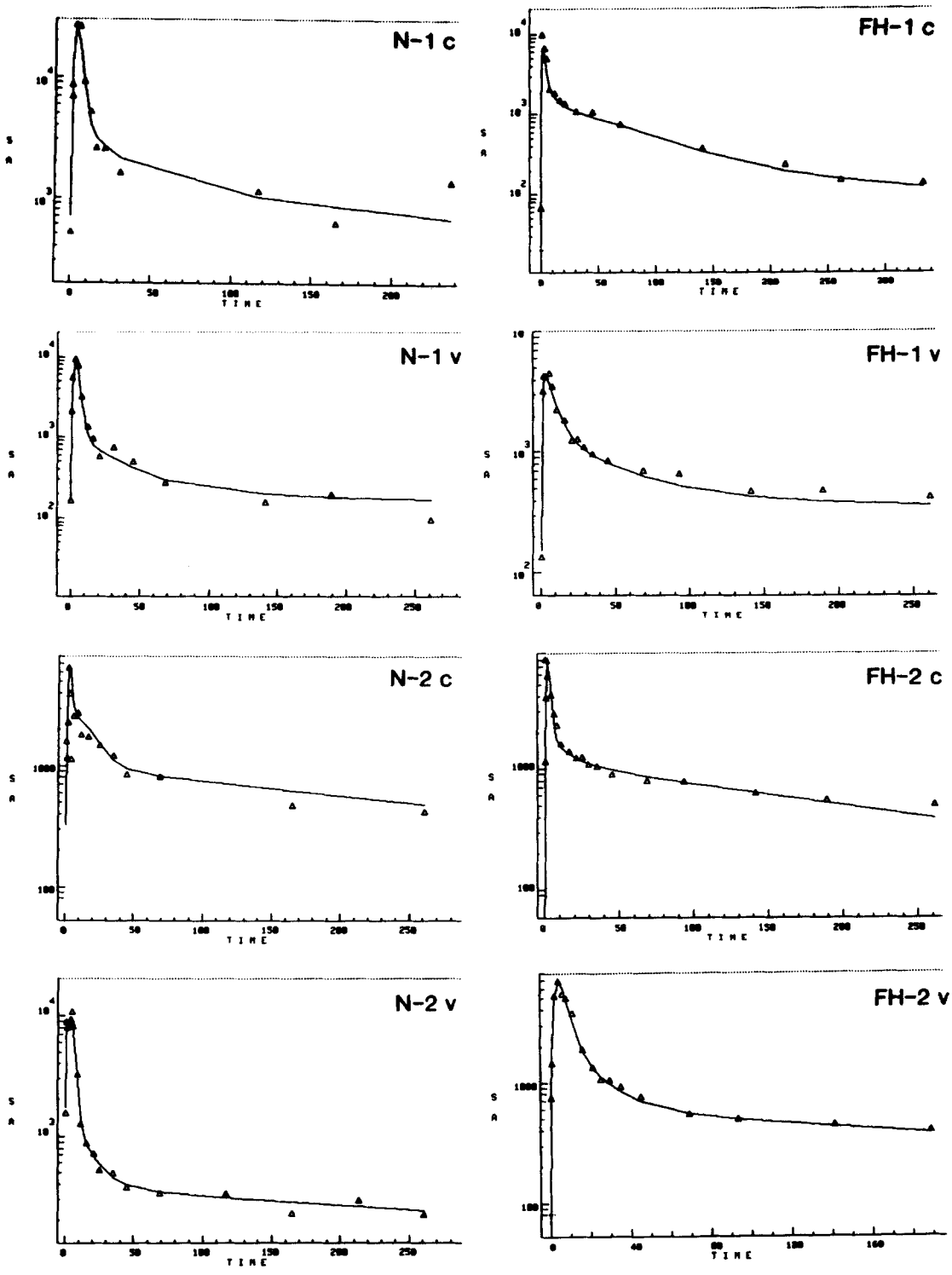
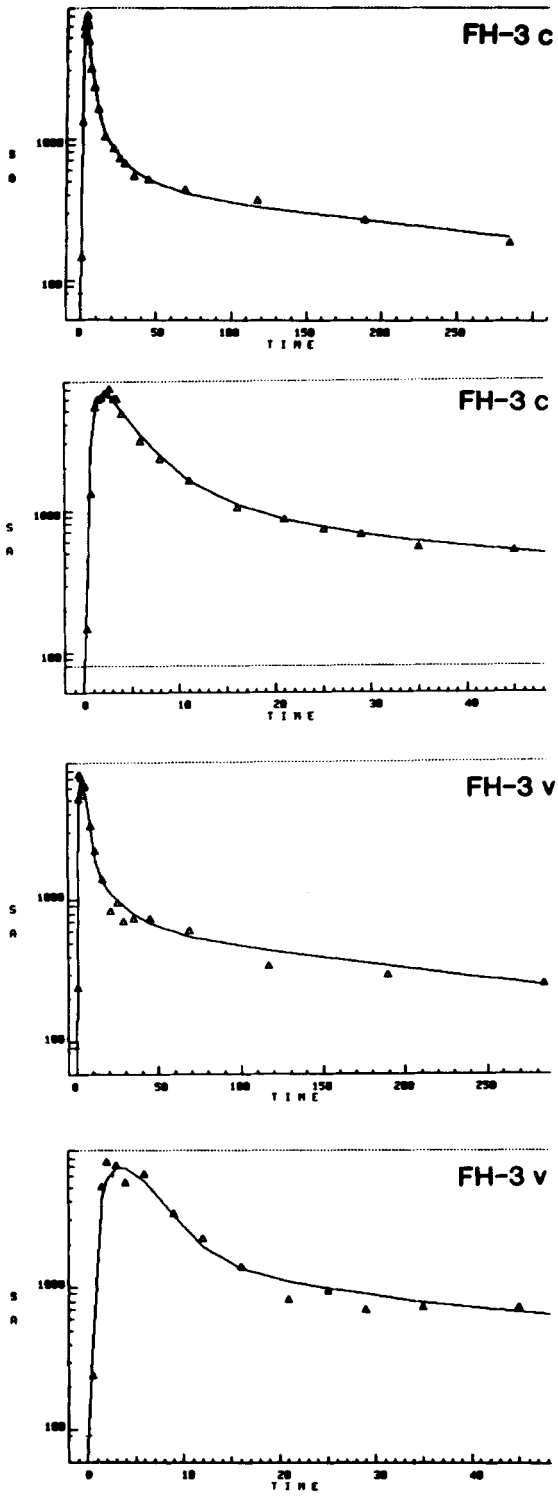


Fig. 3. VLDL apoB kinetic data plotted versus time (h) for the two normal (N) and three familial hypercholesterolemic (FH) subjects on the basal (C) diet and on Vivonex (V). In each case the data (△) and model-derived computer solution are shown. For two studies, FH-3C and FH-3V, the early data are presented on an expanded time scale to provide a better visualization of the computer-generated fits, while Fig. 2 is a similarly expanded plot for study N-2V. All data are specific activities (dpm/mg apoB). Time axis is in hours.



The kinetic investigations reported here provide new insight into the mechanism of the carbohydrate-induced changes in the metabolism of the major cholesterol transport molecules, VLDL and LDL. Plasma apoB metabolism for normal and FH subjects has been resolved into two pathways using the leucine tracer (17). ApoB may be secreted as large, triglyceride-rich VLDL that are metabolized along a delipidation pathway to LDL, or as smaller, IDL/LDL-like particles that are rapidly converted to LDL via the IDL pathway. We speculate that the VLDL delipidation pathway is the primary route for triglyceride secretion by the hepatocyte, and the transport of apoB by this pathway is determined by hepatic triglyceride load. It is well documented that the neutral lipid content of hepatocytes determines the core lipid content of secreted lipoproteins (41–44), and in vitro conditions that promote hepatic triglyceride synthesis are associated with increased rates of VLDL triglyceride secretion, despite no change in total apoB production (41). Similar findings were obtained in our subjects fed a high carbohydrate, low fat diet. Thus apoB secretion adapted to increased hepatic triglyceride synthesis by shifting the flux of apoB from the IDL pathway to the VLDL delipidation pathway with no net change in apoB production (Fig. 3A and ref. 5). It has also been reported that net apoB production in diabetic, nondiabetic, and hypertriglyceridemic humans is not altered when triglyceride secretion is increased on a high carbohydrate diet (19, 20). By using a leucine tracer, however, a shift in apoB secretion from the IDL to the delipidation pathway can be shown to be the mechanism by which the increase in triglyceride transport occurs, at least in FH subjects (Fig. 4A).

Concomitant with this shift in pathways is a further secretory delay for those triglyceride-enriched VLDL entering the delipidation pathway (Fig. 4B). Such a delay is consistent with reports that apoB is retained within hepatic organelles, giving rise to a prolonged cellular residence time compared with other secretory proteins. This delay presumably reflects the time needed to assemble an intact lipoprotein particle (reviewed in ref. 41).

The hypothesis that there is separate metabolic control for the production and catabolism of apoB secreted as large, triglyceride-rich VLDL and as LDL was proposed by us for hypertriglyceridemic subjects (45), and has been expanded upon and tested in hypertriglyceridemic and familial combined hyperlipoproteinemic subjects by Kissebah (46). This hypothesis is also consistent with the findings from hepatic cell culture studies that apoB is synthesized constitutively and that only a portion of the protein produced is secreted, the rest being catabolized (41, 47–49). By implication, that portion of apoB required for lipid transport is utilized, while the remainder is degraded. The observations by Ginsberg et al. (50) in mildly hypertriglyceridemic subjects fed a high carbohy-

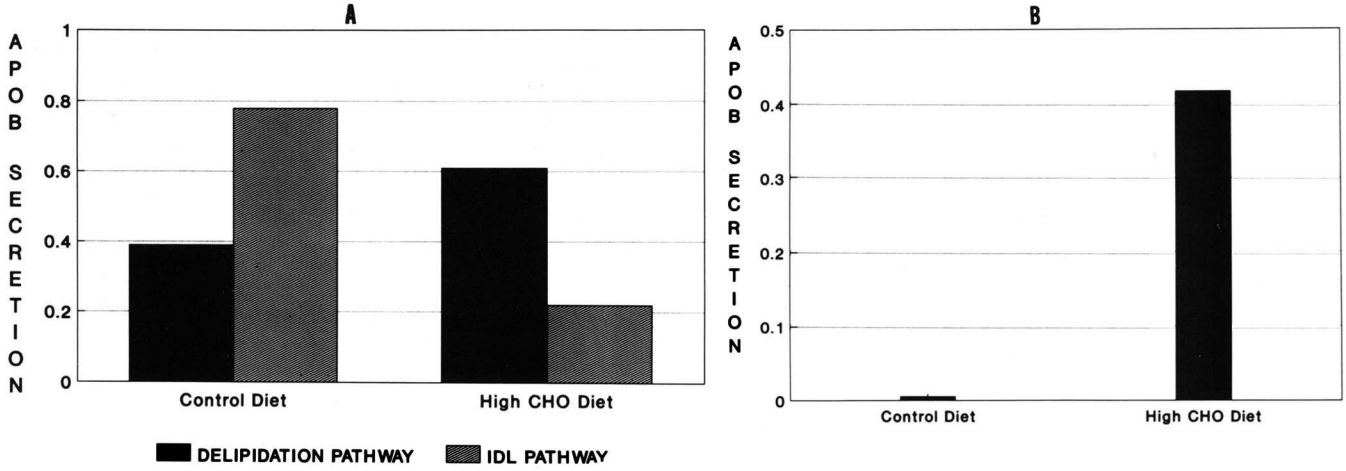


Fig. 4. A: Shift in the fractional transport of apoB from the IDL to the VLDL delipidation pathway, after institution of a high carbohydrate, low fat diet ($P = 0.02$). B: Fraction of total apoB secreted via the delayed, secretory pathway (C19→C10→C1) before and after institution of a high carbohydrate, low fat diet ($P < 0.05$).

drate diet are directly relevant. On a control diet, 38–50% of VLDL-apoB was catabolized by these subjects, while on a high carbohydrate diet, where VLDL-triglyceride secretion increased, 58–66% of VLDL-apoB was catabolized and not converted to LDL. Similarly Abbott et al. (19) reported that, in diabetic and nondiabetic Pima Indians, the observed reduction in LDL-cholesterol on a high carbohydrate diet was primarily a consequence of increased VLDL catabolism, with a concomitant reduction in the conversion of VLDL-apoB to LDL. An increased catabolic rate for LDL was also noted following carbohydrate feedings but this was felt to be of lesser significance.

Although the catabolic loss of the greater portion of apoB as VLDL or IDL is a consistent observation in nor-

TABLE 4. Transport of apoB in subjects on control and Vivonex diets

Subjects	Total ApoB Secretion	VLDL Secretory Delay Pathway	VLDL Secretion	VLDL/IDL Loss	IDL/LDL Secretion	LDL Transport
			mg/kg/day			
Normals						
N1						
Control	26.3	0.0	26.3	15.1	0.0	11.2
Vivonex	34.4	0.0	32.6	27.0	1.8	7.5
Diff	−8.1	0.0	−6.3	−11.9	−1.8	3.7
N2						
Control	36.7	0.0	34.2	15.9	2.4	20.7
Vivonex	52.7	27.8	51.5	26.7	1.2	26.0
Diff	−16.0	−27.8	−17.3	−10.8	1.2	−5.3
FH subjects						
FH1						
Control	9.6	0.2	2.2	0.0	7.4	9.6
Vivonex	25.1	12.5	12.5	0.0	12.6	25.1
Diff	−15.5	−12.3	−10.3	0.0	−5.2	−15.5
FH2						
Control	45.7	0.0	12.6	0.0	33.1	45.7
Vivonex	31.9	11.4	22.0	0.0	9.9	31.9
Diff	13.8	−11.4	−9.4	0.0	23.2	13.8
FH3						
Control	17.9	0.0	17.5	0.0	0.4	17.9
Vivonex	11.2	7.8	9.5	0.0	1.7	11.2
Diff	6.7	−7.8	8.0	0.0	−1.7	6.7
Mean differences	−3.8	−11.9	7.1	−4.5	3.1	0.7
SD	6.0	4.5	4.2	2.8	5.3	5.1
Probability (%)	NS	0.06	NS	NS	NS	NS

TABLE 5. Plasma mass of LDL-apoB in subjects on control and Vivonex diets

Subjects	VLDL-ApoB	LDL-ApoB
	mg	
Normal		
N1		
Control	147	1,984
Vivonex	326	660
Diff	−179	1,324
N2		
Control	218	2,780
Vivonex	275	2,125
Diff	−57	655
FH subjects		
FH1		
Control	122	6,125
Vivonex	242	4,903
Diff	−120	1,222
FH2		
Control	45	10,028
Vivonex	352	6,050
Diff	−307	3,978
FH3		
Control	223	2,800
Vivonex	152	1,008
Diff	71	1,792
Mean differences	−105	1,640
SD	70	633
Probability	NS	<0.025[a]

[a]Performed after logarithmic transformation.

mal and hypertriglyceridemic subjects, in the FH heterozygote there appears to be no catabolic loss of apoB proximate to LDL (17, 45). Thus, in our three FH patients, Vivonex did not induce loss of VLDL or IDL, whereas in the two normal subjects an increased catabolism of these lipoproteins occurred (Table 4). Our observations are consistent with those of Abbott et al. (19) and of Melish et al. (20) and emphasize a fundamental difference in apoB metabolism in the FH patient.

In all five subjects, Vivonex increased the catabolism of LDL, presumably by enhancing hepatic receptor uptake. This is consistent with both the results of our sterol balance measurements and with in vitro experiments that demonstrate that dietary carbohydrate reduced cholesterol synthesis (11). Thus, one interpretation of our data is that the Vivonex-induced inhibition of cholesterol synthesis leads to relative depletion of intracellular cholesterol in the liver, which responds by augmenting the receptor-mediated uptake of LDL. This mechanism is similar to that invoked to explain the cholesterol-lowering effects of bile acid-binding resins and inhibitors of HMG-CoA reductase (51, 52) and is consistent with the hypothesis that the cholesterol-raising effect of diets high in saturated fat is due, at least in part, to suppression of hepatic apoB,E receptor activity (53).

Additional insight into the mechanisms by which a high carbohydrate diet may increase LDL-apoB clearance is provided by the studies of Keidar et al. (54). Their findings demonstrated that such a diet modulates epitope expression on LDL-apoB, as shown by an altered reactivity of a monoclonal antibody that specifically blocks binding of apoB to the apoB,E receptor. Similarly, these LDL also have increased affinity for the fibroblast receptor and increased rates of degradation.

Modulation of apoB,E receptor activity by dietary carbohydrate, however, is unlikely to account entirely for its effects on LDL metabolism. The ability of orally or parenternally administered liquid formulas, high in glucose oligosaccharides and low in fat, to cause striking reductions in serum total and LDL cholesterol levels has now been demonstrated in both FH heterozygotes and homozygotes (5, 9). Since this effect can occur in FH homozygotes with few or no functional B,E receptors, it implies that receptor-mediated clearance of LDL cannot be the sole explanation for the fall in circulating cholesterol. Under such circumstances, the suppression of endogenous cholesterol synthesis by dietary carbohydrate must be sufficient to alter plasma cholesterol metabolism so as to reduce LDL-cholesterol levels through mechanisms other than enhanced B,E receptor clearance. Such processes may assume greater importance in mediating apoB clearance under conditions of carbohydrate feeding, either because of diet-induced compositional changes in circulating VLDL or LDL particles or by somehow directly regulating the function of non-apoB,E receptor pathways.

In this regard, it is noteworthy that Woollett and coworkers (53) have shown that diet-induced suppression of apoB,E receptor activity in hamsters results in a relative increase in LDL uptake by apoB,E receptor-independent processes. It is conceivable, therefore, that such pathways may assume greater importance in mediating apoB clearance under conditions of carbohydrate feeding, either because of diet-induced compositional changes in circulating VLDL or LDL particles or by somehow directly regulating the function of these non-apoB,E receptor processes. Indeed, high carbohydrate diets are reported to increase the apoE content of VLDL in humans (55) and in vitro enrichment of VLDL in apoE promotes their subsequent plasma clearance when injected into hypercholesterolemic (Watanabe) rabbits with defective apoB,E receptor function (56). Presumably, this facilitated removal of VLDL occurs by means of enhanced uptake of these particles by hepatic apoE receptors (57), but whether such a process contributes to the Vivonex-induced decrease in LDL concentration cannot be determined from present data.

Regardless of the precise mechanisms by which high carbohydrate, low fat diets exert their cholesterol-lowering action, their effectiveness in even homozygous forms of

FH raise therapeutic implications for this disease, in which standard diet or drug therapy offers little or no benefit. Still uncertain is whether frequent or continuous feeding of liquid formulas containing mostly glucose are substantially more effective than solid diets of varied carbohydrate composition that are consumed during a more traditional meal schedule. In addition, concern about the long-term human safety of high carbohydrate, low fat liquid formulas (58) warrants careful scrutiny of this dietary intervention, particularly in children.

In summary, the major finding of this investigation is that plasma LDL-cholesterol concentration and LDL-apoB mass in FH heterozygotes both decrease as a result of carbohydrate feeding. In contrast to normal and hypertriglyceridemic individuals, in whom substantial catabolism of VLDL-apoB occurs, these decreases in FH are the result of an increased plasma clearance of LDL. An increased hepatic clearance of LDL is consistent with our findings in these same subjects of a decrease in whole body cholesterol synthesis. The magnitude of the decrease in the plasma LDL-cholesterol concentration, however, is not proportional to the decreases in LDL residence time, due to a variable response in apoB production to Vivonex.

Our observations also demonstrate that a diet capable of stimulating triglyceride secretion, while depleting the liver of cholesterol, results in a shift in hepatic lipoprotein secretion from the production of cholesterol-rich IDL/LDL lipoproteins to the production of large VLDL. These findings are consistent with the hypothesis that the neutral lipid content of the hepatocyte determines the core lipid content of secreted lipoproteins.

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