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In Vivo Evidence for the Role of Lipoprotein Lipase Activity in the Regulation of Apolipoprotein AI Metabolism: A Kinetic Study in Control Subjects and Patients with Type II Diabetes Mellitus*

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ABSTRACT

The aim of this study was to delineate the role of lipoprotein lipase (LPL) activity in the kinetic alterations of high density lipoprotein (HDL) metabolism in patients with type II diabetes mellitus compared with controls. The kinetics of HDL were studied by endogenous labeling of HDL apolipoprotein AI (HDL-apo AI) using a primed infusion of D_3-leucine. The HDL-apo AI fractional catabolic rate (FCR) was significantly increased (0.32 ± 0.07 *vs.* 0.23 ± 0.05 pool/day; $P < 0.01$), and HDL composition was changed [HDL cholesterol, 0.77 ± 0.16 *vs.* 1.19 ± 0.37 mmol/L ($P < 0.05$); HDL triglycerides, 0.19 ± 0.12 *vs.* 0.10 ± 0.03 mmol/L ($P < 0.05$)] in diabetic patients compared with healthy subjects. HDL-apo AI FCR was correlated to plasma and HDL triglyceride concentrations ($r = 0.82$; $P < 0.05$ and $r = 0.80$; $P < 0.05$, respectively) and to homeostasis model assessment ($r = 0.78$; $P < 0.05$). Postheparin plasma LPL activity was decreased in type II diabetes (6.8 ± 2.8 *vs.* 18.1 ± 5.2 μmol/mL postheparin plasma·h; $P < 0.005$) compared with that in healthy subjects and was correlated to the FCR of HDL-apo AI ($r = -0.63$; $P < 0.05$). LPL activity was also correlated with HDL cholesterol ($r = 0.78$; $P < 0.05$), plasma and HDL triglycerides ($r = -0.87$; $P < 0.005$ and $r = -0.83$; $P < 0.05$, respectively), and homeostasis model assessment ($r = -0.79$; $P < 0.05$). In addition, the LPL to hepatic lipase ratio was correlated with the catabolic rate of HDL ($r = -0.76$; $P < 0.06$). These results suggest that a decrease in the LPL to hepatic lipase ratio in type II diabetes mellitus, mainly related to lowered LPL activity, could induce an increase in HDL catabolism. These alterations in HDL kinetics in type II diabetes proceed to some extent from changes in their composition, probably linked to an increase in triglyceride transfer from very low density lipoprotein particles, in close relationship with LPL activity and resistance to insulin. (*J Clin Endocrinol Metab* **86:** 1962–1967, 2001)

THE INVERSE RELATIONSHIP between high density lipoprotein (HDL) cholesterol level and the incidence of coronary heart disease (1) underlines the need to understand the processes regulating synthetic and catabolic rates of HDL. Low levels of HDL cholesterol as well as increased HDL-apo AI fractional catabolic rate (FCR) are often associated with hypertriglyceridemia (2), and changes in lipoprotein lipase (LPL) and hepatic lipase (HL) activities may partly explain this atherogenic potential (3). HDL2 particles are actually formed from the LPL-catalyzed hydrolysis of very low density lipoprotein triglycerides (VLDL-TG) as apolipoproteins (apo) and surface phospholipids released from VLDL merge with preexisting HDL3 particles (4, 5). However, although a direct linkage between LPL activity and HDL concentration has been observed in healthy subjects (6–9), in dyslipidemia (10, 11), and in diabetes mellitus (12–14), intervention of the lipase in the kinetic perturbations of HDL metabolism remains poorly understood. Zech *et al.* (15) as well as Magill *et al.* (10) reported higher FCRs of apo AI in three subjects with familial LPL deficiency compared with those in control subjects. Similar results were reported in monkeys when anti-LPL antibodies were injected to reduce LPL activity (16). Indirect evidence also supports the role of LPL activity in HDL catabolism. For example, in type II diabetes, in which a decrease in LPL activity related to insulin resistance is usually reported (13, 14, 17–19), we observed an increased catabolic rate of HDL-apo AI (20). Such a correlation was also reported in patients with impaired glucose tolerance (21). In addition, in patients with low HDL cholesterol levels and hypertriglyceridemia, the LPL to HL ratio was correlated with the clearance rate of HDL (3). This study therefore aims to test the hypothesis that decreased LPL activity is related to increased HDL catabolism.

Subjects and Methods

Subjects

Kinetic studies of apolipoprotein AI metabolism were performed in seven healthy subjects (normal LPL activity group) and seven type II diabetic patients (low LPL activity group). Some relevant clinical and physiological characteristics are shown in Tables 1 and 2. None of the subjects had taken any medication that could affect lipid for at least 2 months before the study. All women were postmenopausal. Diabetic patients had no proteinuria or hypothyroidism, and were not regular cigarette smokers or alcohol consumers. They had never been treated with probucol and were not receiving insulin. The subjects were in-

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TABLE 1. Clinical and physiological characteristics of study subjects

Subject no.	Age (yr)	Gender	BMI (kg/m^2)	FBG (μmol/mL)	Insulinemia	HbA_{1c}	HOMA
1	45	Male	29.0	10.0	7.0	6.7	3.1
2	58	Male	32.0	6.1	4.1	7.6	1.1
3	50	Male	27.4	10.9	6.0	8.1	2.9
4	64	Male	33.1	9.0	16.1	8.8	6.4
5	65	Female	30.1	7.6	14.5	7.8	4.9
6	57	Female	28.4	12.4	6.8	10.2	3.7
7	50	Female	35.7	9.4	16.8	7.2	7.0
8	47	Male	28.9	12.4	13.5	6.7	7.4
Diabetic patients (mean)	55	3/5	30.6	9.7	10.6	7.9	4.6
SD	8	F/M	2.8	2.2	5.1	1.2	2.2
9	24	Male	21.8	5.0	4.9		1.1
10	24	Male	21.8	4.6	7.7		1.6
11	48	Male	31.6	4.4	6.3	4.9	1.2
12	47	Female	29.3	5.0	7.5		1.7
13	47	Male	29.6	5.9	15.5	4.3	3.9
14	40	Male	29.6	5.9	6.7	5.3	1.8
15	51	Male	33.5	6.2	7.6	5.0	2.1
Control subjects (mean)	40	1/6	28.2	5.3	8.0	4.9	1.9
SD	12	F/M	4.6	0.7	3.4	0.4	0.9

FBG, Fasting blood glucose; Hba_{1c}. glycosylated hemoglobin.

TABLE 2. Lipase activities, plasma and HDL levels of lipids (millimoles per L), and apo AI (milligrams per dL) in study subjects

Subjects no.	LPL activity	HL activity	LPL/HL	Plasma apo AI	Plasma CH	Plasma TG	HDL-CH	HDL-TG
1	3.6	20.0	0.18	128	6.72	3.25	0.82	0.16
2	5.2	31.9	0.16	93	7.03	2.83	0.70	0.15
3	10.7	55.6	0.19	125	5.66	3.17	0.82	0.12
4	11.4	49.0	0.23	129	8.22	4.19	0.80	O.10
5	4.6	10.1	0.45	134	6.72	3.58	0.83	0.22
6	6.1	28.4	0.21	149	7.47	4.66	0.79	0.26
7	6.3	23.8	0.27	151	8.29	1.69	1.01	0.09
8	6.1	20.5	0.30	111	5.43	3.81	0.43	O.46
Diabetic patients (mean)	6.8	29.9	0.25	127.5	6.94	3.40	0.77	0.19
SD	2.8	15.3	O.09	19.O	1.05	0.91	0.16	012
9	25.5	42.8	0.60	106	2.92	0.73	1.29	0.08
10	23.3	41.5	0.56	100	4.44	0.82	1.52	0.08
11	20.1	22.3	0.90	110	3.59	1.20	O.73	0.10
12	10.9	19.2	0.57	129	6.02	2.23	1.17	O.16
13	17.6	49.7	0.35	111	5.12	0.89	0.94	0.05
14	15.2	26.3	0.58	211	5.74	0.73	1.79	0.10
15	13.9	25.1	0.55	116	6.05	1.34	0.93	0.10
Control subjects (mean)	18.1	32.4	0.59	126.1	4.84	1.13	1.19	0.10
SD	5.2	11.9	0.16	38.5	1.23	0.54	0.37	0.03
P^a	<0.005	NS	<0.005	NS	<0.05	<0.005	<0.05	<0.05

LPL and HL activities, in minsonoles per mL post heparin plasma/h; CH, cholesterol.
[a] Diabetic patients (no. 1–8) *vs.* control subjects (no. 9–15).

structed by a dietician to eat a weight maintenance diet composed of 50% of the usual daily caloric intake as carbohydrate, 35% as fat, and 15% as protein for at least 1 week before the study. The experimental protocol was approved by the ethical committee of Nantes University Hospital, and informed consent was obtained before the study was started.

Infusion of stable isotope tracer

The kinetic protocol was described in a previous study (20). Briefly, the endogenous labeling of apo AI was performed by the administration of L-[5,5,5-$^2H_3$]leucine (99.8 atom % $^2H_3$; Cambridge Isotope Laboratories, Andover, MA), which was dissolved in a 0.9% saline solution and tested for sterility and the absence of pyrogens before the study. All subjects fasted overnight for 12 h before the study and remained fasting during the entire protocol. Each subject received an iv priming dose of 10 μmol/kg tracer, immediately followed by a constant tracer infusion (10 μmol/kg·h) for 14 h. Venous blood samples were withdrawn in ethylenediamine tetraacetate tubes (Venoject, Paris, France) at baseline, every 15 min during the first hour, every 30 min during the next 2 h, and then hourly until the end of the study. Plasma was immediately separated by centrifugation for 30 min at 4 C; sodium azide, an inhibitor of bacterial growth, and Pefabloc SC (Interchim, Montluçon, France), a protease inhibitor, were added to blood samples at final concentrations of 1.5 mmol/L and 0.5 mmol/L, respectively.

Analytical procedures

Measurement, isolation, and preparation of apo. VLDL (density, <1.006 g/mL) were isolated from 3 mL plasma by sequential ultracentrifugation using an angle rotor at 40,000 rpm for 24 h at 10 C (Himac CP70, Hitachi, Hialeah, FL). HDL2 (1.063 < density < 1.125 g/mL) and HDL3 (1.125 < density < 1.210 g/mL) were then isolated by a modified method of density gradient ultracentrifugation (22), using a swinging bucket rotor at 40,000 rpm for 24 h at 10 C (Centrikon T 2060, Kontron Instruments Ltd., Zurich, Switzerland). Cholesterol and TG levels in plasma and the

HDL fraction were measured using commercially available enzymatic kits (Roche Molecular Biochemicals, Mannheim, Germany). The apo AI concentration was measured in plasma by immunonephelometry (Behring, Rueil Malmaison, France). The apo AI pool size (milligrams per kg) was calculated by multiplying the mean plasma apo AI concentration by 0.032–0.048 L/kg, assuming a plasma volume of 3.2–4.8% of body weight according to the age, gender, and body weight of each subject (23). The plasma apo AI concentration was taken to be the HDL apo AI concentration, with the assumption that more than 90% of plasma apo AI resides in the HDL fraction (24).

HDL-apo AI and VLDL-apo B100 were concentrated (25) and isolated from other apolipoproteins by SDS-PAGE using a 4–5-10% discontinuous gradient. Apolipoproteins were identified by comparing migration distances with those of known molecular weight standards (cross-linked phosphorylase b markers, Sigma, St. Louis, MO; electrophoresis calibration kit, Pharmacia LKB, Biotechnology, Inc., Piscataway, NJ). Apo bands were excised from polyacrylamide gels and dried in a vacuum (RC 10–10 Jouan, Saint Herblain, France). The desiccated gel slices were hydrolyzed with 1 mL 4 N HCl (Sigma, St. Quentin Fallavier, France) at 110 C for 24 h. Hydrolysates were then evaporated to dryness, and the amino acids were purified by cation exchange chromatography using Temex 50W-X8 resin (Bio-Rad Laboratories, Inc., Richmond, CA). Plasma amino acids were esterified with propanol/acetyl chloride and further derivatized using heptafluorobutyric anhydride (Fluka Chemie AG, Buchs, Switzerland) before analysis.

Determination of tracer to tracee ratios. Chromatographic separations were carried out on a 30-m × 2.52-mm (id) DB-5 capillary column (J&W Scientific, Rancho Cordova, CA). The column temperature program was as follows: initial temperature was held at 80 C, then increased at 10 C/min to a final temperature of 180 C. Electron impact gas chromatography-mass spectrometry was performed on a 5891 A gas chromatograph connected to a 5971 A quadrupole mass spectrometer (Hewlett-Packard Co., Palo Alto, CA). The isotopic ratio was determined by selected ion monitoring at m/z 282 and 285. Calculations of apo AI kinetic parameters were based on the tracer to tracee mass ratio (26).

Determination of lipase activities. On the day of the kinetic study, pre- and postheparin blood samples were drawn into ice-cold ethylenediamine tetraacetate tubes before and 10 min after iv injection of 100 IU heparin/kg BW. This bolus of heparin was injected at the end of the tracer infusion to avoid any effect on VLDL metabolism. Plasma was separated at 4 C and was stored frozen until assayed. Lipase activities were measured following the method described by Iverius and Brunzell (27). The assay was performed using glycerol tri-[1-^{14}C]oleate (NEN Life Science Products, Boston, MA) emulsified with Triton X-100 as substrate. LPL HL activities were, respectively, inhibited by high salt concentration and SDS, as previously described (28). Lipases activities were expressed as micromoles of free fatty acids hydrolyzed by 1 mL postheparin plasma during 1 h of incubation at 37 C.

Insulin sensitivity estimate. The insulin resistance level was estimated with the homeostasis model assessment (HOMA) (29) using the following formula: HOMA = [insulin]/(22.5 $e^{-\ln [glucose]}$).

The plasma insulin concentration (microinternational units per mL) was measured by radioimmunometric assay (Sanofi Pharmaceuticals, Inc., Marnes-La-Coquette, France). The fasting blood glucose concentration (micromoles per mL) was evaluated using a glucose oxidase enzymatic assay (BioMérieux, Marcy-l'Etoile, France).

Modeling

For HDL modeling, we used a one-compartment model, as previously described (20). Kinetic analysis of the tracer to tracee ratio was achieved by computer software for simulation, analysis, and modeling (SAAM II version 1.0.1, Resource Facility for Kinetic Analysis, SAAM Institute, Seattle, WA). VLDL-apo B100 and HDL-apo AI data were kinetically analyzed using a monoexponential function (26): $A(t) = Ap[1 - \exp(-k(t - d))]$, where A(t) is the tracer to tracee ratio at time t, Ap is the tracer to tracee ratio at the plateau of the VLDL apo B100 curve, d is the delay between the beginning of the experiment and the appearance of tracer in the apolipoprotein, and k is the fractional production rate (FPR) of the apolipoprotein. For the estimation of apo AI synthesis, we used the plateau of VLDL-apo B100 tracer to tracee ratio as the precursor pool enrichment. It was assumed that this plateau value, obtained using a monoexponential function, corresponded to the tracer to tracee ratio of the leucine precursor pool. This estimation is based upon the assumption that apo B100 and the majority of apo AI are synthesized by the liver (30). We estimated the FPR, *i.e.* the proportion of apo AI entering the pool per unit time (days), and the absolute production rate (APR), *i.e.* the amount of apolipoprotein AI entering the pool per unit time (milligrams per kg/day). APR was the product of FPR multiplied by the apo AI mass in the HDL fraction. The apo AI pool was considered to be constant, as no significant variation was observed between measurements made at three different infusion times (data not shown). Under these steady state conditions, FPR equals the FCR.

Statistical analysis

Data are reported as the mean ± SD unless otherwise specified. Statistical analysis was performed using Instat Software package (GraphPad Software, Inc., San Diego, CA). The Mann-Whitney U test was used to compare clinical and kinetic data between type II diabetes and controls. Linear regression and correlation analyses were performed with a linear correlation analysis, using the StatView 4.5 software package (Abacus Concepts, Berkeley, CA). A two-tailed *P* level of 0.05 was accepted as statistically significant.

Results

Kinetic data

Enrichment in plasma free leucine reached a plateau after 30 min of infusion and remained stable to the end of the study (data not shown). The mean tracer to tracee ratio curves in HDL are shown in Fig. 1. VLDL-apo B100 isotopic enrichments reached steady state conditions within the infusion period regardless of the subject investigated (data not shown). Kinetic parameters of apo AI are shown in Table 3. Patients with type II diabetes mellitus showed increased HDL-apo AI FCR (0.32 ± 0.07 *vs.* 0.23 ± 0.05 pool/day; $P < 0.01$), whereas HDL-apo AI APR was not altered (15.8 ± 3.3 *vs.* 12.3 ± 5.5 mg/kg·day; P = NS). The FCR of HDL-apo AI was correlated to HOMA ($r = 0.78$; $P < 0.05$; Table 4).

Apo and lipid concentrations

Individual data for plasma and HDL composition are presented in Table 2. Patients with type II diabetes mellitus

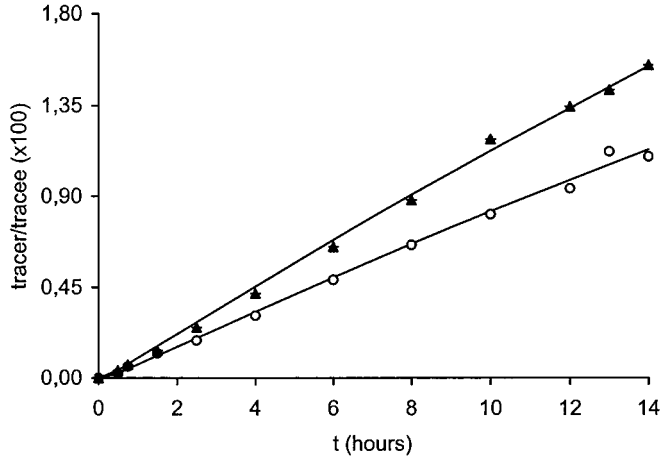

FIG. 1. Mean experimental values (*symbols*) of the tracer to tracee ratio for apo AI-HDL in controls (○) and patients with type II diabetes mellitus (▲). Fits (*lines*) were calculated using monocompartmental analysis during a primed constant infusion of [$^2H_3$]leucine. Data are shown as the mean ± SEM.

TABLE 3. Apo Al pool size and HDL kinetic parameters in study subjects

Subjects no.	FCR HDL-apo Al	APR HDL-apo	Apo Al pool size
1	0.31	16.4	52.5
2	0.31	11.5	37.7
3	0.29	15.9	55.0
4	0.44	21.3	48.4
5	0.27	13.5	50.3
6	0.30	17.0	56.6
7	0.25	12.2	48.3
8	0.40	18.2	45.5
Diabetic patients (mean)	0.32	15.8	49.3
SD	0.07	3.3	6.0
9	0.16	7.8	50.4
10	0.17	8.4	48.5
11	0.23	10.0	44.6
12	0.28	14.5	51.0
13	0.28	12.9	46.1
14	0.26	23.4	90.7
15	0.21	9.2	44.1
Control subjects (mean)	0.23	12.3	53.6
SD	0.05	5.5	16.6
P^a	<0.01	NS	NS

FCR, fractional catabolic rate (pool/day); APR, absolute production rate (milligrams per (no. kg/day); apo AI pool size in (milligrams per kg).
[a] Diabetic patients (no. 1–8) *vs.* control subjects (no. 9–15).

TABLE 4. Correlation analysis in study subjects

	FCR HDL-apo Al	LPL activity
Plasma TG	0.82^a	-0.87^b
HDL-TG	0.80^a	-0.83^a
HOMA	0.78^a	-0.79^b
LPL/HL	-0.76^c	
HDL-CH		0.78^a
Fasting blood glucose		-0.87^b

FCR, Fractional catabolic rate (pool/day).
[a] $P < 0.05$.
[b] $P < 0.005$.
[c] $P < 0.06$.

showed characteristically higher plasma lipids levels compared with controls [total cholesterol, 6.94 ± 1.05 *vs.* 4.84 ± 1.23 mmol/L ($P < 0.05$); TG, 3.40 ± 0.91 *vs.* 1.13 ± 0.54 mmol/L ($P < 0.005$)]. HDL composition was also changed [HDL cholesterol, 0.77 ± 0.16 *vs.* 1.19 ± 0.37 mmol/L ($P < 0.05$); HDL-TG, 0.19 ± 0.12 *vs.* 0.10 ± 0.03 mmol/L ($P < 0.05$)]. Plasma apo AI pool size was not significantly lower in diabetic patients (49.3 ± 6.0 *vs.* 53.6 ± 16.6 mg/kg; P = NS). The plasma and HDL-TG levels were correlated with the catabolic rate of HDL-apo AI (r = 0.82; $P < 0.05$ and r = 0.80; $P < 0.05$; Table 4).

Post-HL activities

LPL activity (Table 2) was decreased in diabetic patients (6.8 ± 2.8 *vs.* 18.1 ± 5.2 μmol/mL postheparin plasma·dL; $P < 0.005$) and was correlated with the FCR of apo AI (r = −0.63; $P < 0.05$; Fig. 2). LPL activity was also correlated with HDL-cholesterol (r = 0.78; $P < 0.05$) and plasma and HDL-TG levels (r = −0.87; $P < 0.005$ and r = −0.83; $P < 0.05$, respectively; Table 4). Correlations were observed between

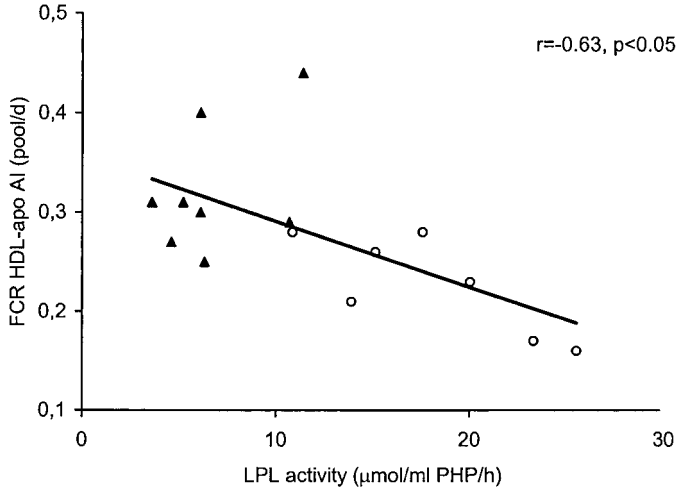

FIG. 2. Relationship between HDL-apo AI FCR (pool per day) and LPL activity (micromoles per mL postheparin plasma/h) in type II diabetic patients and controls.

LPL activity and fasting blood glucose (r = −0.87; $P < 0.005$) or HOMA (r = −0.79; $P < 0.05$; Table 4).

HL activity was similar in diabetic patients and controls (29.9 ± 15.3 *vs.* 32.4 ± 11.9 μmol/mL postheparin plasma·dL; P = NS).

The LPL to HL ratio was decreased in type II diabetes mellitus (0.25 ± 0.09 *vs.* 0.59 ± 0.16; $P < 0.005$), and correlated with the FCR of HDL-apo AI (r = −0.76; $P < 0.06$) and the plasma TG level (r = −0.92; $P < 0.001$).

Discussion

This study was designed to further delineate *in vivo* the relationship between LPL activity and the kinetic aspects of HDL-apo AI metabolism in control subjects and type II diabetic patients. We found a negative correlation between LPL activity and the clearance rate of HDL. As expected, postheparin LPL activities were lower in type II diabetic patients compared with controls and were close to those obtained in diabetic patients with moderate hypertriglyceridemia (14, 17). In this group the FCR of HDL-apo AI was significantly increased. Both plasma levels and HDL composition were altered in type II diabetes mellitus, with an increased HDL-TG level and a decrease in the HDL cholesterol concentration. The HDL-TG level was correlated with the FCR of HDL-apo AI. In addition, LPL activity was inversely correlated with plasma and HDL-TG levels and with HOMA, but was positively related to the HDL cholesterol concentration, as previously reported (6–13).

Study subjects were recruited according to their potential level of LPL activity; type II diabetes mellitus was theoretically considered a model of low LPL-mediated hydrolysis of VLDL-TG, compared with that in control subjects (13, 14, 17, 18). None of them had been included in our previous study (20). The heparin assay we performed showed, as expected, that diabetic patients actually presented with low LPL activity, whereas controls had normal LPL levels. In addition, there was an overlap in the activities observed in the two groups. Therefore, the correlations we found were not related to two different sets of data, but, rather, corresponded

to a homogenous plot of points. Although gender and age do not appear to be key parameters in the control of HDL catabolism (24, 31, 32), the lack of absolute matching of the two study groups according to these parameters could constitute a limitation of the study. We performed an endogenous labeling of apo AI by infusion of leucine labeled with a stable isotope because this procedure avoids any change in lipoprotein kinetics related to potential alterations of the protein's characteristics due to the exogenous labeling (33). Our experimental enrichment data could not be adjusted on a two-pool model, as was sometimes previously done, because our study was designed with a constant infusion of tracer and our period of sampling did not allow characterization of tracer exchanges with a second pool. Therefore, as in other apo AI kinetic studies (21, 34), we applied a single HDL compartment to our modeling design. As an estimate of apo AI leucine precursor pool enrichment, we considered VLDL-apo B100 enrichment at the plateau, which was reached at the end of the infusion period. This assumed that apo AI was mainly synthesized by the liver (30), which is likely to occur in the fasting state.

Our data, although partly speculative, contribute to a global overview of the metabolic processes that link HDL to TG-rich lipoproteins. In type II diabetes, the reduced LPL activity previously reported (13, 14, 17–19) induces a defect in the clearance of TG-rich lipoparticles from the circulation (10, 35–37). This combined with the typical overproduction of VLDL consequently lead to an increase in VLDL-TG. This may enhance cholesterol ester transfer protein (CETP)-mediated TG-cholesteryl ester exchanges, leading to alterations in HDL composition. The negative correlation between LPL activity and HDL concentration corroborates this hypothesis (6–13). This is also in keeping with an *in vivo* study in an animal model (38). In transgenic mice expressing the CETP transgene, LPL activity was correlated with the HDL cholesterol level, but not in the absence of CETP. However, whereas LPL activity appears to play a strong role in HDL composition, its effect on HDL-apo AI metabolism in humans has been poorly studied. A 28% increase in the HDL clearance rate was also observed by Magill *et al.* in one subject with LPL deficiency after exogenous labeling of [^{125}I]HDL (10). Furthermore, Goldberg and co-workers, by infusing specific monoclonal antibodies into female cynomolgus monkeys to inhibit LPL, observed that the HDL-apo AI catabolic rate in LPL-inhibited animals was more than double that in control rabbits (16). Thus, they suggested that the variations in apo AI level and clearance rate might be a consequence of differences in LPL-mediated lipolysis of TG-rich lipoproteins. We now report that LPL activity is correlated to HDL composition and catabolism in humans, and therefore we suggest that impaired lipase activity on VLDL could induce an increased CETP-mediated efflux of TG on HDL, leading to alterations in both their composition and their clearance rate. This hypothesis is in agreement with the positive correlation between plasma TG levels and HDL-apo AI FCR that we previously observed in type II diabetes (20). In addition, as previously reported (17, 36), HL activities were similar in the two study groups. As HL activity is increased and HDL2 cholesterol seems to be specifically reduced in obesity (39), we would have probably observed lowered HL activities among a control group composed of lean subjects. Furthermore, the LPL to HL ratio was decreased in diabetic patients and correlated to the FCR of HDL-apo AI, as previously reported in patients with low HDL cholesterol levels (3). HL and LPL have opposing effects on HDL composition; LPL activity catalyzes the degradation of TG-rich lipoproteins and induces transfer of lipid surface components to HDL, whereas HL catabolizes HDL phospholipids. Thus, a low LPL to HL ratio should promote a depletion of HDL surface components and an enrichment of these particles in TG, which is in agreement with their enhanced clearance (3).

As previously reported (40–43), we found similar plasma apo AI levels in diabetic patients and controls, contrasting with other studies (20, 44). The clinical characteristics of healthy subjects, matched for mean age and body mass index with diabetic patients in the current study, could explain this discrepancy. Furthermore, in our previous study the plasma apo AI level was decreased in diabetes mellitus as the result of an increased clearance rate and unchanged production rate of HDL (20). In the current study the slight increase in HDL APR was sufficient to restore a normal plasma apo AI level. The heterogeneity of apo AI production rates related to genetic or environmental factors could therefore be a key factor in the control of the plasma HDL concentration in the case of enhanced clearance, and this aspect needs to be clarified in further studies.

The insulin resistance of patients with type II diabetes may additionally contribute to the down-regulation of LPL activity (45, 46) and the increase in HDL-apo AI FCR (32). The correlations between HOMA and LPL activity or HDL clearance rate support this hypothesis. Therefore, LPL activity may be a target of hypolipidemic treatment to restore a normal HDL cholesterol level in type II diabetic patients with low HDL. Studies have actually shown that fibrates enhanced the expression of LPL by activating transcription factors of the peroxisome proliferator-activated receptors (47). Other treatments, such as weight loss or biguanides, may directly act upon insulin resistance to recover suitable LPL activities.

In conclusion, these results support the hypothesis that reduced LPL activity, related to resistance to insulin, may play a major role in disorders of HDL metabolism in humans. In fact, impaired lipase activity on VLDL could induce an increased efflux of TG on HDL, leading to alterations in both their composition and their clearance rate. This study, therefore, provides further information about the coordinate regulation of HDL and TG metabolism.

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