Decreased Activities of Lipoprotein Lipase and Hepatic Triglyceride Lipase in Patients With Gout

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Postheparin plasma lipoprotein lipase (LPL) and hepatic triglyceride lipase (HTGL) activities were measured in 30 male primary gout patients as well as in control subjects. The activities of these lipolytic enzymes were significantly decreased in the patients as compared with the controls (gout *v* control; LPL, 5.4 ± 0.4 *v* 7.9 ± 0.9 U; HTGL, 14.6 ± 2.0 *v* 17.9 ± 3.4 U) when matched with serum triglyceride concentration. Further, LPL activity was negatively correlated with serum- and very–low-density lipoprotein (VLDL)-triglyceride in gout patients, while that of HTGL was negatively correlated with low-density lipoprotein (LDL)-triglyceride in both gout patients and control subjects. These results suggest that decreased activities of LPL and HTGL may contribute, in part, to the increased concentrations of serum-, VLDL-, and LDL-triglyceride seen in gout patients, leading to a higher risk for coronary atherosclerotic diseases in gout.

HYPERTRIGLYCERIDEMIA, ONE OF the risk factors of coronary artery disease, is frequently seen in patients with gout,[1] however, its underlying mechanism still remains undetermined despite many studies. Some investigators have suggested the effect of obesity, as well as excessive alcohol or sucrose intake, on the development of hypertriglyceridemia in gout,[2-4] while others have concluded that some undetermined intrinsic factor(s) are contributors.[5-7] In a previous study, we demonstrated that hypertriglyceridemia exists in gout independent of body weight or alcohol consumption.[7] In our search for the intrinsic factor(s) of hypertriglyceridemia in gout, the activities of those lipolytic enzymes, which catalyze the hydrolysis of lipoprotein-triglycerides, lipoprotein lipase (LPL; EC 3.1.1.34), and hepatic triglyceride lipase (HTGL; EC 3.1.1.3), were measured in conjunction with serum lipids and lipoproteins. Moreover, their relationships to serum-, very–low-density lipoprotein (VLDL)-, and low-density lipoprotein (LDL)-triglyceride were also investigated.

MATERIALS AND METHODS

Subjects

Thirty male patients with primary gout, aged 41.8 ± 1.6 years (mean ± SE), and 30 male adults as controls aged 46.0 ± 1.9 years (mean ± SE), were studied on an outpatient basis. Gout patients were selected to match the control subjects in terms of alcohol consumption and serum triglyceride level. The diagnosis of primary gout was made on the basis of criteria advocated by the American Rheumatism Association (ARA) (Table 1).[8] All patients fulfilled at least 6 of the 12 clinical, laboratory, and x-ray findings listed in Table 1. Information concerning the consumption of alcoholic beverages over the previous month was taken to be representative of the drinking habits of each individual by means of a questionnaire on frequency, quantity, and type, and then converted to daily alcohol consumption.[9] Control subjects were randomly selected from applicants for an annual health check-up. They were judged as normal from results of a physical examination and laboratory findings, except for mild hypertriglyceridemia observed in some subjects. Prior to the study, all medications that were considered to affect serum lipid profiles were withheld for at least 1 month. All patients and subjects underwent a 75-g oral glucose tolerance test, and those who showed a diabetic pattern were excluded. Apolipoprotein C-II, a cofactor of LPL, was not abnormally low in any who participated in the study.

Serum Lipids Analysis and Lipoprotein Isolation

Venous blood samples were obtained after an overnight fast. Lipoproteins were then isolated from fresh serum by sequential ultracentrifugation[10]: VLDL at density (d) less than 1.006, IDL at $1.006 < d < 1.019$, LDL at $1.019 < d < 1.063$, and high-density lipoprotein (HDL) at $1.063 < d < 1.210$. Concentrations of triglyceride (TG), total cholesterol (T-chol), and HDL-cholesterol (HDL-chol) in both serum and lipoprotein fractions were measured using the respective commercial kits: TG, Triglyceride T-test Wako; T-chol, Nescot TC kit; and HDL-chol, HDL-C kit (Wako Pure Chemicals Industries, Osaka, Japan).

Determination of LPL and HTGL Activities in Postheparin Plasma

Postheparin blood was collected in a tube containing EDTA-2Na 10 minutes after an intravenous injection of 30 U/body weight of heparin (Novo, Nordisk A/S, Denmark), after which postheparin plasma (PHP) was immediately separated by low speed centrifugation (1,700×*g*) at 4°C for 10 minutes and stored at −70°C until analysis. Activities of LPL and HTGL in PHP were measured with an emulsion of triolein (9, 10-^{3}H) in gum arabic as a substrate.[11] LPL activity was determined after inhibition of HTGL activity by 50 mmol/L of sodium dodecyl sulfate (SDS), and HTGL activity was determined following inhibition of LPL activity by 1 mol/L of sodium chloride. Both lipase activities were expressed as free fatty acid (FFA) formed per hour, and 1 U of enzyme activity was defined as μg FFA/h/mL PHP.

Determination of LPL Mass in PHP

LPL mass in PHP was determined by enzyme-linked immunosorbent assay (ELISA)[12] using a Markit F LPL kit (Dainippon Pharmaceutical, Tokyo, Japan).

Statistical Analyses

Data are expressed as mean ± SE. Observed differences between groups were analyzed by Student's *t* test and after logarithmic transformation of the variable with nonparametric distribution. Correlations between variables were assessed by Spearman's rank sum test. A *P* value less than .05 was considered to be statistically significant.

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Table 1. Proposed Criteria for Acute Arthritis of Primary Gout

(A) The presence of characteristic urate crystals in the joint fluid
(B) A tophus proved to contain urate crystals by chemical or polarized light microscopy
(C) 1. More than 1 attack of acute arthritis
2. Maximum inflammation developed within 1 day
3. Monoarthritis attack
4. Redness observed over joints
5. First metatarsophalangeal joint painful or swollen
6. Unilateral first metatarsophalangeal joint attack
7. Unilateral tarsal joint attack
8. Tophus (proven or suspected)
9. Hyperuricemia
10. Asymmetric swelling within a joint on x-ray
11. Subcortical cysts without erosions on x-ray
12. Joint fluid culture negative for organisms during attack

NOTE. Diagnostic criteria for primary gout were (A) the presence of characteristic urate crystals in the joint fluid, and/or (B) a tophus proved to contain urate crystals by chemical or polarized light microscopy, and/or (C) the presence of 6 of the 12 clinical, laboratory, and x-ray findings.

Data from Wallace et al.[8]

RESULTS

As shown in Table 2, there were no significant differences between groups with regard to age (41.8 ± 1.6 years *v* 46.0 ± 1.9 years, not significant [NS]), body mass index (BMI) (24.4 ± 0.6 *v* 23.7 ± 0.4, NS), systolic blood pressure (128.7 ± 2.9 mm Hg *v* 126.7 ± 2.7 mm Hg, NS), diastolic blood pressure (75.6 ± 1.7 mm Hg *v* 73.8 ± 1.5 mm Hg, NS), alcohol consumption (29.6 ± 4.8 g/d *v* 23.9 ± 3.6 g/d, NS), creatinine (0.085 ± 0.003 mmol/L *v* 0.078 ± 0.002 mmol/L, NS), T-chol (5.14 ± 0.16 mmol/L *v* 5.03 ± 0.18 mmol/L, NS), or HDL-chol (1.51 ± 0.10 mmol/L *v* 1.44 ± 0.06 mmol/L, NS) or regarding serum concentrations of triglyceride (1.79 ± 0.12 mmol/L *v* 1.54 ± 0.15 mmol/L, NS), VLDL-triglyceride (1.12 ± 0.11 mmol/L *v* 0.89 ± 0.13 mg/dL, NS), or LDL-triglyceride (0.26 ± 0.02 mmol/L *v* 0.22 ± 0.02 mmol/L, NS). The activities of both LPL and HTGL were significantly decreased in the gout patients as compared with the control subjects (5.4 ± 0.9 U *v* 7.9 ± 0.9 U, $P < .01$ and 14.6 ± 2.0 U *v* 17.9 ± 3.4 U, $P < .05$) (Fig 1A and B). In addition, LPL mass was significantly decreased in gout (200.0 ± 10.0 ng/mL *v* 228.0 ± 10.0 ng/mL, $P < .05$) (Fig 1C) and was significantly correlated with LPL activity ($r = .57$, $P < .0001$, n = 60). As indicated in Table 3, HTGL activity was negatively correlated with LDL-triglyceride in both the gout patients ($r = -.46$, $P < .05$) and control subjects ($r = -.42$, $P < .05$), while LPL activity was negatively correlated with serum- and VLDL-triglyceride concentrations in the gout patients ($r = -.39$ and $-.37$, $P < .05$, respectively). However, no correlation was observed between LPL activity or serum- and VLDL-triglyceride concentrations in the control subjects. HTGL mass was not measured.

Table 3. Relationship Between Lipase Activities and Serum-, VLDL-, and LDL-Triglyceride Concentrations

	Gout (n = 30)	Control (n = 30)
LPL activity (log LPL)		
v serum-TG	$r = -.39$, $P < .05$	NS
VLDL-TG	$r = -.37$, $P < .05$	NS
HTGL activity (log HTGL)		
v LDL-TG	$r = -.46$, $P < .05$	$r = -.42$, $P < .05$

NOTE. Abbreviations are the same as in Table 2.

DISCUSSION

Many studies have investigated hypertriglyceridemia in gout, however, no definite conclusion has been obtained as to the underlying cause(s). Hypertriglyceridemia develops from an imbalance between production and lipolysis, both intrinsically and extrinsically. In addition to the extrinsic factors leading to hypertriglyceridemia, such as the excessive alcohol and sucrose consumption and/or obesity frequently observed in gout patients, a link between uric acid and triglyceride produc-

Table 2. Clinical and Laboratory Data

	Gout (n = 30)	Control (n = 30)	*P* Value
Age (yr)	41.8 ± 1.6	46.0 ± 1.9	NS
BMI (kg/m^2)	24.4 ± 0.6	23.7 ± 0.4	NS
SBP (mm Hg)	128.7 ± 2.9	126.7 ± 2.7	NS
DBP (mm Hg)	75.6 ± 1.7	73.8 ± 1.5	NS
Alcohol intake (g/day)	29.6 ± 4.8	23.9 ± 3.6	NS
Uric acid (mmol/L)	0.55 ± 0.02	0.32 ± 0.02	<.001
S-Cr (mmol/L)	0.085 ± 0.003	0.078 ± 0.002	NS
TG (mmol/L)	1.79 ± 0.12 (0.58 − 3.23)	1.54 ± 0.15 (0.38 − 3.38)	NS
T-chol (mmol/L)	5.14 ± 0.16	5.03 ± 0.18	NS
HDL-chol (mmol/L)	1.51 ± 0.10	1.44 ± 0.06	NS
HDL-TG (mmol/L)	0.20 ± 0.01	0.18 ± 0.01	NS
LDL-TG (mmol/L)	0.26 ± 0.02	0.22 ± 0.02	NS
VLDL-TG (mmol/L)	1.12 ± 0.11	0.89 ± 0.13	NS

NOTE. Data are expressed as mean ± SE. Parenthesis show the range of serum triglyceride concentration.

Abbreviations: BMI, body mass index: S-Cr, serum creatinine; DBP, diastolic blood pressure; TG, triglyceride; T-chol, total cholesterol; HDL-chol, high-density lipoprotein cholesterol; HDL-TG, high-density lipoprotein triglyceride; LDL-TG, low-density lipoprotein triglyceride; NS, not significant; SBP, systolic blood pressure; VLDL-TG, very low-density lipoprotein.

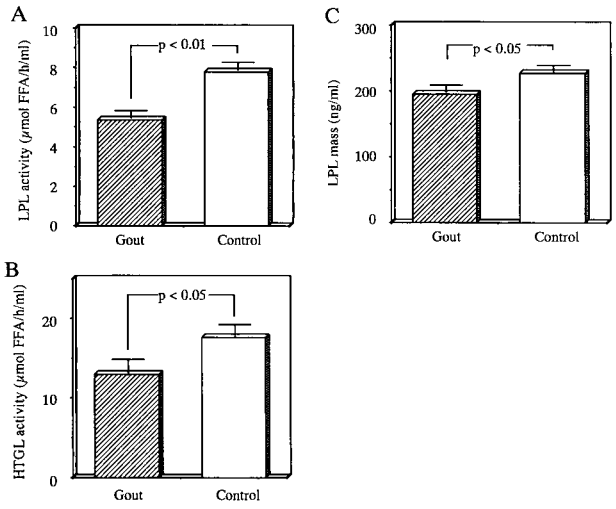

Fig 1. LPL and HTGL activities (A and B) and LPL mass (C) in patients and control subjects. Both LPL and HTGL activities were significantly decreased in gout patients as compared with the controls (5.4 ± 0.9 U *v* 7.9 ± 0.9 U, *P* < .01, and 14.6 ± 2.0 U *v* 17.9 ± 3.4 U, *P* < .05). In addition, LPL mass was significantly decreased in gout (200.0 ± 10.0 ng/mL *v* 228.0 ± 10.0 ng/mL, *P* < .05).

tion has been demonstrated in primary gout.[13] On the other hand, several studies have attempted to clarify the cause of hypertriglyceridemia from the viewpoint of defective lipolysis.[14,15]

LPL acts during the first step of hydrolyzing triglyceride in chylomicrons and VLDL on vascular endothelial cells, while HTGL acts to hydrolyze triglyceride in lipoproteins, such as IDL, LDL, and HDL in hepatic sinusoidal endothelial cells.[16] Thus, it is probable that decreased activities by these lipases may contribute to the development of hypertriglyceridemia in gout.

In the present study, we examined LPL and HTGL in gout to determine whether their decreased activities may serve as intrinsic factors causing hypertriglyceridemia. In a previous study, decreased activity of HTGL in gout was suggested,[14] however, because the number of subjects was too few (9 cases), we investigated the activities of LPL and HTGL in gout together with serum lipids and lipoproteins in a relatively large number of gout patients. Our results demonstrated that LPL and HTGL activities were significantly decreased even in normal or mildly hypertriglyceridemic gout patients, and that the activities of these lipolylic enzymes were negatively correlated with the concentrations of serum-, VLDL-, and LDL-trigylceride, respectively. We concluded that elevated serum- and LDL-triglyceride concentrations in gout may be partly ascribable to the decreased activities of these enzymes, with obesity and excessive alcohol consumption being adjunct aggravating factors for hypertriglyceridemia.

At present, the cause of decreased LPL and HTGL activities in gout remains undetermined, however, these decreased activities may contribute to increased remnant lipoproteins, decreased HDL concentration, and small dense LDL[17] and lead to susceptibility to atherogenesis, as it has been suggested that patients with coronary heart disease have lower HTGL activity.[18] Therefore, it seems necessary to raise LPL and HTGL activity by moderate physical activity to reduce the susceptibility of gout patients to atherosclerotic coronary artery disease.

REFERENCES

1. Wyngaarden JB, Kelley WN: Gout, in Stanbury JB, Wyngaarden JB, Fredrickson DS, et al (eds): The Metabolic Basis of Inherited Disease (ed 5). New York, NY, McGraw-Hill, 1983, p 1043

2. Gibson T, Grahame R: Gout and hyperlipidaemia. Ann Rheum Dis 33:298-303, 1974

3. Nishida Y, Akaoka I, Nishizawa T, et al: Hyperlipidemia in gout. Clin Chim Acta 62:103-106, 1975

4. Fox IH, John D, DeBruyne S, et al: Hyperuricemia and hypertriglyceridemia: Metabolic basis for the association. Metabolism 34:741-746, 1985

5. Jiao S, Kameda K, Matsuzawa Y, et al: Hyperlipoproteinaemia in primary gout: Hyperliporoteinaemic phenotype and influence of alcohol intake and obesity in Japan. Ann Rheum Dis 45:308-313, 1986

6. Ferns GAA, Lanham J, Dieppe P, et al: A DNA polymorphism of an apoprotein gene associates with the hypertriglyceridaemia of primary gout. Hum Genet 78:55-59, 1988

7. Takahashi S, Yamamoto T, Moriwaki Y, et al: Impaired lipoprotein metabolism in patients with primary gout—Influence of alcohol intake and body weight. Br J Rheumatol 33:731-734, 1994

8. Wallace SL, Robinson H, Masi A, et al: Preliminary criteria for the classification of the acute arthritis of primary gout. Arthritis Rheum 20:895-900, 1977

9. Thomson AD, Majumdar SK: The hazard to health from moderate drinking, in Turner MR (ed): Nutrition and Health. A perspective. Lancaster, UK, MTP Press, 1982, pp 87-107

10. Hatch FT, Lees RS: Practical methods for plasma lipoprotein analysis. Adv Lipid Res 6:1-33, 1968

11. Baginski ML, Brown WV: A new method for the measurement of lipoprotein lipase in postheparin plasma using sodium dodecyl sulfate for the inactivation of hepatic triglyceride lipase. J Lipid Res 20:548-556, 1979

12. Ikeda Y, Takagi A, Yamamoto A: A sandwich-enzyme immunoassay for the quantification of lipoprotein lipase and hepatic triglyceride lipase in human postheparin plasma using monoclonal antibodies to the corresponding enzymes. J Lipid Res 31:1911-1924, 1990

13. Matsubara K, Matsuzawa Y, Jiao S, et al: Relationship between hypertriglyceridemia and uric acid production in primary gout. Metabolism 38:698-701, 1989

14. Kodama T, Murase T, Itakura H, et al: Postheparin plasma lipoprotein lipase and hepatic triglyceride lipase activities in patients with primary asymptomatic gout. Clin Chem 29:2124-2125, 1983

15. Moriwaki Y, Yamamoto T, Takahashi S, et al: Apolipoprotein E phenotypes in patients with gout: Relation with hypertriglyceridaemia. Ann Rheum Dis 54:351-354, 1995

16. Bensadoun A: Lipoprotein lipase. Annu Rev Nutr 11:217-237, 1991

17. Lahdenpera S, Sane T, Vuorinen-Markkola H, et al: LDL particle size in mildly hypertriglyceridemic subjects: No relation to insulin resistance or diabetes. Atherosclerosis 113:227-236, 1995

18. Groot PH, van Stiphout WA, Krauss XH, et al: Postprandial lipoprotein metabolism in normolipidemic men with and without coronary artery disease. Arterioscler Thromb 11:653-662, 1991