Influences of the PPAR α -L162V polymorphism on plasma HDL₂-cholesterol response of abdominally obese men treated with gemfibrozil

Yohan Bossé, MSc¹, Agnès Pascot, MSc¹, Martine Dumont, MSc², Martin Brochu, PhD³, Denis Prud'homme, MD, MSc⁴, Jean Bergeron, MD, MSc¹, Jean-Pierre Després, PhD^{1,5,6}, and Marie-Claude Vohl, PhD^{1,6}

Purpose: The effect of gemfibrozil is mediated by the activation of peroxisome proliferator-activated receptor alpha (PPAR α). The objective of this study was to determine whether the lipid response to gemfibrozil therapy is influenced by the PPAR α -L162V polymorphism. **Methods:** Sixty-three abdominally obese men were randomly assigned to a 6-month-intervention program with either receiving a placebo ($N = 31$) or gemfibrozil ($N = 32$). **Results:** In response to gemfibrozil therapy, L162-homozygotes exhibited a 5.5% increase in high-density lipoprotein 2 cholesterol (HDL₂-C) levels compared with a 50.0% increase among carriers of the V162 allele ($P = 0.03$). **Conclusion:** These results suggest that the HDL₂-C response to gemfibrozil is modulated by the PPAR α -L162V polymorphism. *Genet Med* 2002;4(4):311–315.

Key Words: genetics, fibrates, lipid response, lipoproteins, high-density lipoprotein cholesterol

Alterations of the lipoprotein-lipid profile are associated with an increased risk of developing coronary heart disease, the leading cause of death in westernized societies. Many lipid-lowering agents have been developed for the treatment of dyslipidemia. In this regard, the clinical benefits of gemfibrozil, a fibrate agent, have been reported in both primary¹ and secondary² prevention trials. Although fibrates have been used in clinical practice for more than three decades, their molecular mechanism of action has just recently been elucidated³, and it is now recognized that their effects are mediated by the activation of a specific nuclear receptor termed peroxisome proliferator-activated receptor alpha (PPAR α). The activation of PPAR α by fibrates causes the transactivation of PPAR α -responsive genes, which include those encoding proteins that control lipid metabolism.³

Recently, a molecular scanning of the human PPAR α has revealed a L162V polymorphism associated with alterations of the lipoprotein-lipid profile.^{4,5} The frequency for the rare allele is established to be 0.062 in the healthy European population⁵ and reaches 0.128 in French-Canadians.⁴ To demonstrate the functional consequences of this polymorphism, transient transfection assays in Hepa-1 and HepG2 cell lines have been performed.^{5,6} Both studies have shown enhanced transactiva-

tion activity in cells containing vectors expressing PPAR α -V162 allele, compared with L162 allele, when treated with the PPAR α ligand WY-14,643. In this context, it was interesting to verify whether the L162V polymorphism in the PPAR α gene can modulate the plasma lipoprotein/lipid response to gemfibrozil.

MATERIALS AND METHODS

Subjects

Subjects were asymptomatic, nondiabetic volunteers who had to fulfill the following criteria. Men had to be between 25 and 55 years of age and willing to participate to a 6-month intervention program in which they were asked to follow the National Cholesterol Education Program Phase 1 dietary guidelines with or without gemfibrozil. Experience from the Helsinki Heart Study revealed that the effect of gemfibrozil was largely confined to overweight subjects.⁷ Men in the present study were weight stable obese (27 and 40 kg/m²) and characterized by a dyslipidemic state (1.7 mmol/L $\geq$ TG $\leq$ 5.7 mmol/L; high-density lipoprotein cholesterol (HDL-C) $\leq$ 1.2 mmol/L and total plasma cholesterol $<$ 6.7 mmol/L). The study was approved by the Medical Ethics Committee of Laval University. All subjects gave their informed written consent to participate in this study.

Study design

After having completed their baseline measurements, 71 subjects were selected to participate to the study and were randomly assigned to either receiving a placebo or gemfibrozil 600 mg bid. Dietary recommendations were given by a registered dietician on a voluntary basis for the duration (6 months) of the study. Drug safety was assessed every 4 weeks by the phy-

From the ¹Lipid Research Center, CHUL Research Center, ²Physical Activity Sciences Laboratory, Kinesiology Division, Department of Social and Preventive Medicine, Laval University, Quebec, Canada; ³Department of Kinesiology, University of Montreal, Montreal, Canada; ⁴Human Kinetics, Faculty of Health Sciences, University of Ottawa, Ontario, Canada; ⁵The Quebec Heart Institute; and ⁶Department of Food Sciences and Nutrition, Laval University, Quebec, Canada.

Marie-Claude Vohl, PhD, Lipid Research Center, CHUL Research Center, RC-9500 2705, Boulevard Laurier, Sainte-Foy, Québec, G1V 4G2, Canada.

Received: January 18, 2002.

Accepted: April 25, 2002.

sician in charge of the project. Subjects were tested at baseline and at the end of the 6-month intervention protocol. During that period, there were six dropouts, and genotype information was not available for two subjects. We thus ended-up with a total of 63 subjects who completed the trial.

Lipids and lipoproteins

Fasting blood samples were collected and plasma lipid, lipoprotein, and apolipoprotein (apo) levels were measured as previously described.⁸ HDL₂ was precipitated from the HDL fraction with dextran sulfate.⁹ The cholesterol content of the supernatant fraction (HDL₃) was determined, and HDL₂-C levels were derived by subtracting HDL₃-C from total HDL-C concentrations. Nondenaturing 4% to 30% polyacrylamide gel electrophoresis was performed for the measurement of the average HDL size using whole plasma kept at -80°C as recently described.¹⁰ The same plasma were also used to determine the low-density lipoprotein (LDL) peak particle size measured by electrophoresis on a 2% to 16% polyacrylamide gradient gel as previously described.^{11,12}

DNA analysis

The L162V polymorphism does not alter any restriction site. A mismatch polymerase chain reaction method previously described was used to genotype all the subjects participating in the study.⁴

Statistical analyses

Variables not normally distributed were $\log_{10}$ transformed before analysis. The difference in response between genotype groups was assessed by a two-tailed unpaired Student's *t*-test. To evaluate whether the PPAR α -L162V genotype may interact with gemfibrozil treatment, we performed an ANOVA in which the interaction term was included (two-way factorial ANOVA). The source of variation in lipoprotein-lipid profile was computed using the type III sum of squares. This sum of squares applies to unbalanced study designs and quantifies the effects of an independent variable after adjustment for all other variables included in the model. All statistical analyses were performed using the SAS package (SAS Institute, Cary, NC), and a statistically significant difference was defined as $P < 0.05$.

RESULTS

Among the 63 men who completed the study, 31 were assigned to the placebo group and 32 to the gemfibrozil group. The effects of the 6-month-intervention program on plasma lipoprotein/lipid profile have been published elsewhere.⁸ In the present study, the placebo and gemfibrozil groups were further subdivided on the basis of the PPAR α -L162V genotype (Fig. 1).

The baseline lipoprotein-lipid profile according to the PPAR α -L162V genotype is presented in Table 1. For these analyses, subjects in the placebo and gemfibrozil groups were combined because medication had not started. No statistically significant difference was observed between the two genotype

Fig. 1 Distribution of subjects into the intervention program. Men were randomly assigned to either receiving a placebo or gemfibrozil and then divided according to PPAR α -L162V genotype.

Table 1
Baseline lipoprotein-lipid concentrations between carriers and noncarriers of the PPAR α -L162V polymorphism

Variable (mmol/L)	PPAR α -L162V genotype		<i>P</i>
	L162 HMZ (<i>N</i> = 52)	V162 carriers (<i>N</i> = 11) ^a	
Triglycerides	2.66 $\pm$ 0.80	2.34 $\pm$ 0.59	0.20
LDL-TG	0.39 $\pm$ 0.10	0.34 $\pm$ 0.04	0.23
HDL-TG	0.23 $\pm$ 0.05	0.21 $\pm$ 0.03	0.15
Cholesterol	5.57 $\pm$ 0.61	5.75 $\pm$ 0.37	0.35
LDL-C	3.71 $\pm$ 0.60	4.03 $\pm$ 0.35	0.08
HDL-C	0.84 $\pm$ 0.12	0.90 $\pm$ 0.14	0.16
HDL ₂ -C	0.17 $\pm$ 0.06	0.19 $\pm$ 0.09	0.43
HDL ₃ -C	0.67 $\pm$ 0.10	0.71 $\pm$ 0.07	0.18
Plasma apo B (g/L)	1.22 $\pm$ 0.15	1.24 $\pm$ 0.12	0.63
LDL-apo B (g/L)	1.04 $\pm$ 0.16	1.10 $\pm$ 0.09	0.23
HDL-apo A1 (g/L)	1.16 $\pm$ 0.12	1.20 $\pm$ 0.09	0.28
Average HDL size (Å)	82.08 $\pm$ 2.03	82.18 $\pm$ 2.49	0.90
LDL size (Å)	248.99 $\pm$ 3.81	251.04 $\pm$ 3.63	0.15

Values are means $\pm$ SD; all values are mmol/L except when indicated.

^aTen L162/V162-heterozygotes and one V162-homozygote.

groups for baseline lipid and lipoprotein concentrations. However, there was a tendency toward higher LDL-C levels among carriers of the V162 allele compared with L162-homozygotes (HMZ) ($P = 0.08$).

The plasma lipoprotein-lipid changes of the 6-month gemfibrozil therapy was compared between carriers and noncarriers of the PPAR α -L162V polymorphism (Table 2). There was no statistically significant difference between the two genotype groups for all lipoprotein-lipid changes except for HDL₂-C levels. Indeed, V162 carriers exhibited a 50% increase in HDL₂-C concentrations compared with a 5.5% increase among L162-HMZ (Fig. 2). The same trend was observed with changes in HDL-C levels although it did not reach statistical significance ($P = 0.08$). These results were the same after adjustment for either baseline measurements or changes observed in body weight (data not shown). To corroborate the difference observed between the two genotype groups on

Table 2Changes in plasma lipid and lipoprotein levels in gemfibrozil-treated participants according to PPAR α -L162V genotypes

Variable (mmol/L)	PPAR α -L162V genotype		P
	L162 HMZ (N = 26)	V162 carriers (N = 6) ^a	
Triglycerides	-1.14 $\pm$ 0.84	-0.76 $\pm$ 1.09	0.35
LDL-TG	-0.07 $\pm$ 0.09	-0.10 $\pm$ 0.05	0.41
HDL-TG	-0.02 $\pm$ 0.06	-0.00 $\pm$ 0.06	0.67
Cholesterol	-0.51 $\pm$ 0.52	-0.67 $\pm$ 0.49	0.49
LDL-C	-0.05 $\pm$ 0.69	-0.48 $\pm$ 0.33	0.15
HDL-C	0.06 $\pm$ 0.09	0.14 $\pm$ 0.10	0.08
HDL ₂ -C	0.01 $\pm$ 0.07	0.09 $\pm$ 0.11	0.03
HDL ₃ -C	0.06 $\pm$ 0.11	0.05 $\pm$ 0.11	0.91
Plasma apo B (g/L)	-0.13 $\pm$ 0.21	-0.21 $\pm$ 0.08	0.38
LDL-apo B (g/L)	-0.04 $\pm$ 0.19	-0.18 $\pm$ 0.08	0.09
HDL-apo A1 (g/L)	0.03 $\pm$ 0.10	0.09 $\pm$ 0.12	0.27
Average HDL size (Å)	-0.43 $\pm$ 1.44	0.59 $\pm$ 1.12	0.13
LDL size (Å)	1.07 $\pm$ 3.67	1.28 $\pm$ 2.86	0.91

Values are means $\pm$ SD; all values are mmol/L except when indicated.^aSix L162/V162-heterozygotes. **Fig. 2** Changes in HDL-C and HDL-C subfractions in gemfibrozil-treated participants according to PPAR α -L162V genotype (N = 26 L162-HMZ and 6 V162 carriers). Each dot represents the change observed for one individual within his respective group. Relative change is indicated at the bottom of the graph. The bar chart illustrates mean HDL changes in the different groups.

HDL₂-C changes with gemfibrozil, we analyzed the average HDL size change with the therapy. Although not statistically significant, carriers of the V162 allele had a more favorable HDL size response compared with L162-HMZ (Table 2). There was also a tendency toward a greater decrease in LDL-apo B for those carrying the V162 allele ($P = 0.09$). This trend might be explained by a greater reduction in LDL-C and a

greater increase in LDL size observed in V162 allele carriers although the difference did not reach statistical significance.

When individual HDL-C responses to gemfibrozil were plotted (Fig. 3), two phenomena were noted. First, there was a large interindividual variation in HDL-C changes with gemfibrozil therapy. Second, there was a cluster of carriers of the V162-allele on the right side of the graph, indicating a greater increase of HDL-C levels among these individuals. In these individuals, the apparent difference in plasma HDL-C levels was mainly explained by changes in the HDL₂-C subfraction as no difference was observed between the genotype groups regarding the change in the HDL₃-C subfraction (Fig. 2). Subsequent analyses performed in men treated with gemfibrozil revealed that after the 6-month-intervention program, V162 carriers were characterized by higher levels of HDL₂-C compared with L162-HMZ (Fig. 4).

To test the potential interaction between the PPAR α -L162V polymorphism and gemfibrozil treatment on plasma lipoprotein-lipid concentrations, an analysis of variance was performed for each lipoprotein-lipid variable. The effects of the genotype and the treatment, as well as the potential interaction between these two independent variables, are presented in Table 3. As expected, the treatment (placebo versus gemfibrozil) had a statistically significant impact on plasma lipoprotein-lipid changes. On the other hand, the PPAR α -L162V genotype by itself did not have a significant impact on plasma lipoprotein-lipid responses. However, a significant genotype-by-treatment interaction was observed for changes in plasma HDL₂-C levels. This interaction explained 7.0% of the total variance of the change in plasma HDL₂-C concentrations. This finding suggests that the PPAR α -L162V polymorphism may influence plasma HDL₂-C responsiveness to gemfibrozil therapy. Additionally, it also implies that men carrying the PPAR α -V162 allele will experience a higher increase in HDL₂-C levels when treated with fibrates.

Fig. 3 Individual changes in HDL-C levels among gemfibrozil-treated subjects (N = 32). Each bar represents the after-before difference in HDL-cholesterol levels observed in response to gemfibrozil therapy. Individual responses of L162-homozygotes are illustrated by the gray bars, whereas the white bars show the responses of carriers of the V162 allele.

Fig. 4 Mean HDL₂-C values before and after gemfibrozil treatment according to PPAR α -L162V genotype ($N = 26$ L162-HMZ and 6 V162 carriers). Values are mean $\pm$ SE.

Table 3

Effects of PPAR α -L162V polymorphism, the treatment (placebo vs. gemfibrozil) and their interaction on plasma lipid-lipoprotein response to the intervention program

Phenotypes (mmol/L)	Mean changes ($N = 63$)	P		
		Genotypes	Treatment	Interaction
Triglycerides	-0.55 ± 0.94	0.52	0.0009	0.42
LDL-TG	-0.04 ± 0.08	0.83	0.001	0.35
HDL-TG	0.00 ± 0.06	0.70	0.18	0.80
Cholesterol	-0.36 ± 0.47	0.66	0.005	0.50
LDL-C	-0.13 ± 0.52	0.96	0.86	0.62
HDL-C	0.03 ± 0.10	0.21	0.0001	0.14
HDL ₂ -C	0.02 ± 0.08	0.32	0.15	0.04
HDL ₃ -C	0.01 ± 0.11	0.88	0.01	0.76
Plasma apo B (g/L)	-0.10 ± 0.16	0.69	0.02	0.29
LDL-apo B (g/L)	-0.05 ± 0.15	0.31	0.08	0.06
HDL-apo A1 (g/L)	0.03 ± 0.10	0.74	0.06	0.20
Average HDL size (Å)	0.03 ± 1.56	0.19	0.56	0.55
LDL size (Å)	0.15 ± 2.91	0.12	0.51	0.18

Values are means $\pm$ SD; all values are mmol/L except when indicated.

DISCUSSION

The documented interindividual variation in the response to fibrate therapy is clinically relevant. In this report, we demonstrated that a naturally occurring variation, L162V, in the PPAR α gene is associated with greater responsiveness of HDL₂-C levels after a 6-month-therapy with gemfibrozil. Before this study, Flavell et al.⁵ had reported a greater lowering effect of bezafibrate on total cholesterol and non-HDL-C in

V162 carriers. In the present study, there was a trend toward higher increase in HDL-C levels after the administration of gemfibrozil, but it did not reach statistical significance. Taken together, these results suggest potentially greater benefits of fibrate treatment among individuals carrying the PPAR α -V162 allele. This could be of great interest as previous work suggests that the PPAR α -L162V polymorphism seems to have a deleterious impact on plasma lipoprotein-lipid levels.^{4,5,13}

PPAR α regulates the expression of gene encoding proteins that control lipoprotein metabolism. In vitro studies performed on hepatic cells (HepG2 and Hepa-1) have demonstrated higher transactivation activity in PPAR α carrying the V162 allele when treated with a PPAR α agonist.^{5,6} Differences observed in HDL₂-C responsiveness in the present study could then be explained by a greater transcriptional regulation of PPAR α target genes in subjects carrying the V162 allele. Several proteins controlling HDL metabolism are regulated at the gene level by PPAR α activators such as fibrates. In fact, fibrate therapy has been shown to induce overexpression of apo A-I and apo A-II genes, leading to an increase in plasma HDL-C levels.^{14,15} Recently, other proteins such as ATP-binding cassette transporter 1 and CD36- and LIMPII-analogous 1/scavenger receptor class B type 1 involved in the reverse-cholesterol-transport pathway have been shown to be up-regulated by PPAR α activators in macrophages.^{16,17} Furthermore, it has been demonstrated in mice that fibrate treatment increases phospholipid transfer protein gene expression through a PPAR α -dependent mechanism, which accounts for a marked enlargement of HDL particles.¹⁸ The functional consequence of the PPAR α -L162V polymorphism could result in higher transcriptional regulation of genes controlling HDL metabolism and then explain the relationship between the PPAR α -L162V polymorphism and the HDL₂-C response to gemfibrozil observed in the present study.

This study reported associations that make biological sense and study genetic variations that affect the gene product in a physiologically meaningful way. However, certain limitations may give some uncertainty about our results. First, dealing with the low frequency of the polymorphism and with a small sample size impaired our ability to recognize association with a smaller effect. Second, considering the multiple comparisons made in the study, some of the significant findings may have occurred by chance. Consequently, further studies with a larger sample size are warranted to determine the interest of the PPAR α L162V polymorphism in the management of dyslipidemia.

In summary, we have demonstrated that subjects carrying the PPAR α -V162 allele showed higher increases in HDL₂-C levels in response to gemfibrozil therapy. This finding could be of clinical relevance for these individuals as a low concentration of the HDL₂ subfraction seems to be more closely related to the incidence of ischemic heart disease.^{19,20} We speculate that the greater HDL₂-C increase among gemfibrozil-treated men carrying the PPAR α -L162V polymorphism is mediated by a more pronounced transcriptional regulation of genes controlling HDL metabolism.

Acknowledgment

This study was supported by Parke Davis/Warner-Lambert Canada Inc. and by the Natural Sciences and Engineering Research Council of Canada. The authors would like to express their gratitude to the subjects for their excellent collaboration and to the staff of the Lipid Research Center and of the Physical Activity Sciences Laboratory for their contribution to the study. We especially want to thank the staff of the Physical Activity Sciences Laboratory, Laval University, for their help in the collection of the data. Y. Bossé is the recipient of a studentship from the "Fonds pour la formation de chercheurs et l'aide à la recherche (FCAR) et le Fonds de la recherche en santé du Québec (FRSQ)." M. C. Vohl and J. Bergeron are research and clinical scholars, respectively of the FCAR and the FRSQ. J. P. Després is chair professor of human nutrition, lipidology, and prevention of cardiovascular disease, supported by Provigo and Pfizer Canada.

References

1. Frick MH, Elo O, Haapa K, Heinonen OP, Heinsalmi P, Helo P, Huttunen JK, Kaitaniemi P, Koskinen P, Manninen V, et al. Helsinki Heart Study: primary-prevention trial with gemfibrozil in middle-aged men with dyslipidemia. Safety of treatment, changes in risk factors, and incidence of coronary heart disease. *N Engl J Med* 1987;317:1237–1245.
2. Rubins HB, Robins SJ, Collins D, Fye CL, Anderson JW, Elam MB, Faas FH, Linares E, Schaefer EJ, Schectman G, Wilt TJ, Wittes J. Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol: Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group. *N Engl J Med* 1999;341:410–418.
3. Staels B, Dallongeville J, Auwerx J, Schoonjans K, Leitersdorf E, Fruchart JC. Mechanism of action of fibrates on lipid and lipoprotein metabolism. *Circulation* 1998;98:2088–2093.
4. Vohl MC, Lepage P, Gaudet D, Brewer CG, Betard C, Perron P, Houde G, Cellier C, Faith JM, Després JP, Morgan K, Hudson TJ. Molecular scanning of the human PPAR α gene: association of the L162v mutation with hyperapobetalipoproteinemia. *J Lipid Res* 2000;41:945–952.
5. Flavell DM, Pineda Torra I, Jamshidi Y, Evans D, Diamond JR, Elkeles RS, Bujac SR, Miller G, Talmud PJ, Staels B, Humphries SE. Variation in the PPAR α gene is associated with altered function in vitro and plasma lipid concentrations in Type II diabetic subjects. *Diabetologia* 2000;43:673–680.
6. Sapone A, Peters JM, Sakai S, Tomita S, Papiha SS, Dai R, Friedman FK, Gonzalez FJ. The human peroxisome proliferator-activated receptor α gene: identification and functional characterization of two natural allelic variants. *Pharmacogenetics* 2000;10:321–333.
7. Tenkanen L, Manttari M, Manninen V. Some coronary risk factors related to the insulin resistance syndrome and treatment with gemfibrozil: experience from the Helsinki Heart Study. *Circulation* 1995;92:1779–1785.
8. Dumont M, Mauriege P, Bergeron J, Després JP, Prud'homme D. Effect of a six month gemfibrozil treatment and dietary recommendations on the metabolic risk profile of visceral obese men. *Int J Obes Relat Metab Disord* 2001;25:1136–1143.
9. Gidez LI, Miller GJ, Burstein M, Slagle S, Eder HA. Separation and quantitation of subclasses of human plasma high density lipoproteins by a simple precipitation procedure. *J Lipid Res* 1982;23:1206–1223.
10. Perusse M, Pascot A, Després JP, Couillard C, Lamarche B. A new method for HDL particle sizing by polyacrylamide gradient gel electrophoresis using whole plasma. *J Lipid Res* 2001;42:1331–1334.
11. Lamarche B, Tchernof A, Moorjani S, Cantin B, Dagenais GR, Lupien PJ, Després JP. Small, dense low-density lipoprotein particles as a predictor of the risk of ischemic heart disease in men: prospective results from the Quebec Cardiovascular Study. *Circulation* 1997;95:69–75.
12. Tchernof A, Lamarche B, Prud'Homme D, Nadeau A, Moorjani S, Labrie F, Lupien PJ, Després JP. The dense LDL phenotype: association with plasma lipoprotein levels, visceral obesity, and hyperinsulinemia in men. *Diabetes Care* 1996;19:629–637.
13. Lacquemant C, Lepretre F, Pineda Torra I, Manraj M, Charpentier G, Ruiz J, Staels B, Froguel P. Mutation screening of the PPAR α gene in type 2 diabetes associated with coronary heart disease. *Diabetes Metab* 2000;26:393–401.
14. Vu-Dac N, Schoonjans K, Kosykh V, Dallongeville J, Fruchart JC, Staels B, Auwerx J. Fibrates increase human apolipoprotein A-II expression through activation of the peroxisome proliferator-activated receptor. *J Clin Invest* 1995;96:741–750.
15. Berthou L, Duverger N, Emmanuel F, Langouet S, Auwerx J, Guillouzo A, Fruchart JC, Rubin E, Deneffe P, Staels B, Branellec D. Opposite regulation of human versus mouse apolipoprotein A-I by fibrates in human apolipoprotein A-I transgenic mice. *J Clin Invest* 1996;97:2408–2416.
16. Chinetti G, Gbaguidi FG, Griglio S, Mallat Z, Antonucci M, Poulain P, Chapman J, Fruchart JC, Tedgui A, Najib-Fruchart J, Staels B. CLA-1/SR-BI is expressed in atherosclerotic lesion macrophages and regulated by activators of peroxisome proliferator-activated receptors. *Circulation* 2000;101:2411–2417.
17. Chinetti G, Lestavel S, Bocher V, Remaley AT, Neve B, Torra IP, Teissier E, Minnich A, Jaye M, Duverger N, Brewer HB, Fruchart JC, Clavey V, Staels B. PPAR- α and PPAR- γ activators induce cholesterol removal from human macrophage foam cells through stimulation of the ABCA1 pathway. *Nat Med* 2001;7:53–58.
18. Bouly M, Masson D, Gross B, Jiang XC, Fievet C, Castro G, Tall AR, Fruchart JC, Staels B, Lagrost L, Luc G. Induction of the phospholipid transfer protein gene accounts for the high density lipoprotein enlargement in mice treated with fenofibrate. *J Biol Chem* 2001;276:25841–25847.
19. Lamarche B, Moorjani S, Cantin B, Dagenais GR, Lupien PJ, Després JP. Associations of HDL2 and HDL3 subfractions with ischemic heart disease in men: prospective results from the Quebec Cardiovascular Study. *Arterioscler Thromb Vasc Biol* 1997;17:1098–1105.
20. Johansson J, Carlson LA, Landou C, Hamsten A. High density lipoproteins and coronary atherosclerosis: a strong inverse relation with the largest particles is confined to normotriglyceridemic patients. *Arterioscler Thromb* 1991;11:174–182.