Original Article

Determinants of Bezafibrate-induced Improvements in LDL Cholesterol in Dyslipidemic Patients with Diabetes

A Subgroup Analysis of the J-BENEFIT Study

Takahisa Hirose¹, Tamio Teramoto², Kazunori Abe³, Takehiko Taneyama³ and the J-BENEFIT study group

¹Division of Diabetes, Metabolism, and Endocrinology, Department of Medicine, Toho University School of Medicine, Tokyo Japan

²Teikyo Academic Research Center, Teikyo University, Tokyo Japan

³Pharmacovigilance & Post-marketing Surveillance, KISSEI Pharmaceutical Co., Ltd., Tokyo Japan

Aim: Our previous “J-BENEFIT (Japan Bezafibrate cliNical EffectIveness and Tolerability)” study demonstrated that bezafibrate improves blood lipid profiles and glucose control in dyslipidemic patients with diabetes. However, bezafibrate did not significantly improve low-density lipoprotein cholesterol (LDL-C), although some patients showed decreases while others showed increases in the LDL-C levels. Therefore, a subgroup analysis of the J-BENEFIT study was conducted to identify factors influencing the bezafibrate-induced changes in the LDL-C levels.

Methods: Of the 3,316 patients in the J-BENEFIT study, 2,116 not treated with other lipid-lowering drugs were enrolled in the current study, and the effects of 24-week treatment with bezafibrate on the LDL-C levels were analyzed. A reduction in the LDL-C level of $\geq 25\%$ occurred in 253 patients, and a logistic-regression analysis was used to identify factors associated with this improvement.

Results: Among the 2,116 overall patients, bezafibrate treatment significantly increased the LDL-C levels from 123.9 ± 36.7 to 125.7 ± 31.3 mg/dL. The subanalysis showed that the treatment responses varied according to the baseline LDL-C level, with significant decreases in the ≥ 160 and ≥ 140 - < 160 mg/dL groups, no significant decrease in the ≥ 120 - < 140 mg/dL group and a significant increase in the < 120 mg/dL group. A multivariate logistic-regression analysis of the data for the patients with an LDL-C of $\geq 25\%$ identified a female sex, the use of anti-hypertensive and hypoglycemic agents and a high baseline LDL-C level to be significant determinants of the LDL-C response to bezafibrate.

Conclusions: Our results showed that treatment with bezafibrate improves the LDL-C levels and lipid profiles in dyslipidemic diabetic patients, especially women, subjects co-treated with anti-hypertensive or hypoglycemic agents and those with high baseline LDL-C levels.

See editorial vol. 22: 658-659

J Atheroscler Thromb, 2015; 22: 676-684.

Key words: Bezafibrate, LDL cholesterol, Diabetes, J-BENEFIT study

Introduction

Patients with type 2 diabetes mellitus tend to

Address for correspondence: Takahisa Hirose, Division of Diabetes, Metabolism, and Endocrinology, Department of Medicine, Toho University School of Medicine, 6-11-1, Omori-nishi, Ota-ku, Tokyo 143-8541, Japan

E-mail: takahisa.hirose@med.toho-u.ac.jp

Received: August 4, 2014

Accepted for publication: January 13, 2015

have high triglyceride (TG) and low high-density lipoprotein cholesterol (HDL-C) levels compared with non-diabetic patients¹⁾. In the prospective observational J-BENEFIT (Japan Bezafibrate cliNical EffectIveness and Tolerability) study, we reported that bezafibrate improved serum lipids, such as TG and HDL-C, in dyslipidemic patients with diabetes mellitus untreated or treated with hypoglycemic agents in association with a significant decrease in the glycosylated hemoglobin (HbA1c) levels correlating with the

rate of change in TG²). However, this treatment had no effect on the low-density lipoprotein cholesterol (LDL-C) levels overall, although some patients showed either a decrease or increase in LDL-C. The present study constitutes a follow-up assessment of the above findings and includes a J-BENEFIT subgroup analysis to identify factors that may explain the variable effects of bezafibrate on LDL-C.

Subjects and Methods

The patients were enrolled via a centralized registration system between June 2003 and March 2005, and the J-BENEFIT study was conducted from June 2003 to September 2005. All subjects were treated with bezafibrate (400 mg/day) for 24 weeks, and the efficacy and safety of this treatment were subsequently evaluated. In order to exclude confounding factors possibly influencing the effects of bezafibrate on LDL-C, 2,116 patients taking no other lipid-lowering agents were extracted from the efficacy analysis group of 3,316 patients. This study was conducted in accordance with the Good Post-marketing Study Practice of the Ministry of Health, Labour and Welfare, Japan.

The efficacy of bezafibrate was evaluated by measuring changes in serum lipids, changes in LDL-C based on the baseline level, the status of LDL-C treatment goal attainment based on the baseline the LDL-C level, changes in other serum lipid parameters in the LDL-C goal attainment population and factors influencing the marked improvements noted in the patients exhibiting a reduction in LDL-C of $\geq 25\%$. The LDL-C levels were calculated using Friedewald formula³) in patients with a TG level of < 400 mg/dL, and the non-HDL-C level was calculated using the total cholesterol (TC) and HDL-C values.

Statistical Analysis

The statistical analyses were carried out using the SAS version 9.2 software program (SAS Institute Inc., Cary, NC, USA). Differences between the baseline and post-treatment values were analyzed using the paired *t*-test. Univariate and stepwise multivariate logistic-regression analyses were performed to identify factors influencing an LDL-C reduction of $\geq 25\%$. The *p* value for the selection criteria was set at 10% for the univariate logistic-regression analysis and 25% for the stepwise multivariate logistic regression analysis. A two-sided *p* value of < 0.05 was considered to be statistically significant for within-group changes and in the multivariate logistic-regression analysis.

Results

Subjects

Table 1 lists the demographic and baseline features of the 2,116 patients in total and the 253 patients who showed a post-treatment LDL-C reduction of $\geq 25\%$. The mean age $\pm$ standard deviation of the total study population was 62.4 ± 11.6 years, comprising 1,238 (58.5%) men and 991 (46.8%) patients ≥ 65 years of age. The mean duration of dyslipidemia was 4.6 ± 4.1 years, and 337 (15.9%) patients had previously been treated for dyslipidemia.

Effects of Bezafibrate on Serum Lipids

Table 2 summarizes the serum lipid profiles at baseline and after the 24-week treatment period with bezafibrate. The HDL-C, TG and non-HDL-C levels improved significantly after the treatment (mean dose: 367 mg/day) ($p < 0.001$ for all parameters). On the other hand, the LDL-C levels increased significantly from 123.9 ± 36.7 at baseline to 125.7 ± 31.3 mg/dL after treatment ($p = 0.015$).

Effects of Bezafibrate on LDL-C

The above apparent effect of bezafibrate on LDL-C was further evaluated in more detail. **Table 3** shows the changes in LDL-C, non-HDL-C, TG and TG/HDL-C based on the baseline levels. The LDL-C levels decreased significantly in the patients with a baseline level of ≥ 140 mg/dL (" ≥ 140 to < 160 mg/dL" group and " ≥ 160 mg/dL" group, $p < 0.001$ for each group). In contrast, a significant increase was noted in the patients with a baseline level of < 120 mg/dL ($p < 0.001$). Interestingly, the latter group showed a significant decrease in the non-HDL-C levels from 148.1 ± 23.2 to 143.5 ± 31.5 mg/dL and the TG/HDL-C ratio from 6.7 ± 2.8 to 3.3 ± 2.0 mg/dL after treatment with bezafibrate ($p < 0.001$). The TG levels were highest in the patients with a baseline LDL-C level of < 120 mg/dL, and significant improvements were noted in all patient groups.

Table 4 shows the changes in LDL-C, non-HDL-C, TG and TG/HDL-C based on the baseline TG levels. The TG/HDL-C ratio decreased significantly in the patients with a baseline TG level of ≥ 200 to < 250 mg/dL ($p < 0.001$), whereas the LDL-C levels did not change. In the patients with a baseline TG level of ≥ 250 mg/dL (" ≥ 250 to < 300 mg/dL" group and " ≥ 300 mg/dL" group), bezafibrate increased the LDL-C levels, with a marked decrease in the TG/HDL-C ratio ($p < 0.001$ for each group).

Table 1. Demographic and baseline characteristics of the patients

	Entire study population	Patients with LDL-C reduction $\geq 25\%$
Number of patients (<i>n</i>)	2,116	253
Entry category		
Type 1 diabetes mellitus, <i>n</i> (%)	25 (1.2)	6 (2.4)
Type 2 diabetes mellitus, <i>n</i> (%)	2,069 (97.8)	243 (96.0)
Diabetes mellitus (unknown type), <i>n</i> (%)	20 (0.9)	4 (1.6)
Fasting blood glucose ≥ 110 mg/dL, <i>n</i> (%)	2 (0.1)	-
Outpatients, <i>n</i> (%)	2,046 (96.7)	242 (95.7)
Men, <i>n</i> (%)	1,238 (58.5)	107 (42.3)
Age, mean $\pm$ SD	62.4 $\pm$ 11.6	64.1 $\pm$ 11.5
≥ 65 years, <i>n</i> (%)	991 (46.8)	133 (52.6)
Body mass index (kg/m ²), mean $\pm$ SD	25.5 $\pm$ 3.5	25.2 $\pm$ 3.5
Smoking, <i>n</i> (%)	505 (23.9)	46 (18.2)
Alcohol use, <i>n</i> (%)	663 (31.3)	60 (23.7)
Complications, <i>n</i> (%)	1,532 (72.4)	187 (73.9)
Hepatic disease, <i>n</i> (%)	186 (8.8)	21 (8.3)
Renal disease, <i>n</i> (%)	29 (1.4)	3 (1.2)
Hypertension, <i>n</i> (%)	1,061 (50.1)	138 (54.5)
Cardiovascular disease, <i>n</i> (%)	196 (9.3)	26 (10.3)
Duration of dyslipidemia (years), mean $\pm$ SD	4.6 $\pm$ 4.1	4.5 $\pm$ 4.8
Duration of diabetes mellitus (years), mean $\pm$ SD	6.8 $\pm$ 6.0	7.3 $\pm$ 6.7
Previous treatment of dyslipidemia, <i>n</i> (%)	337 (15.9)	31 (12.3)
Previous treatment of diabetes, <i>n</i> (%)	1,263 (59.7)	151 (59.7)
Use of hypoglycemic agents, <i>n</i> (%)	1,501 (70.9)	191 (75.5)
Use of anti-hypertensive agents, <i>n</i> (%)	1,101 (52.0)	150 (59.3)
Dietary therapy, <i>n</i> (%)	1,672 (79.0)	201 (79.4)
Exercise therapy, <i>n</i> (%)	1,285 (60.7)	156 (61.7)
Total cholesterol (mg/dL), mean $\pm$ SD	219.9 $\pm$ 36.0	250.0 $\pm$ 39.8
LDL-C (mg/dL), mean $\pm$ SD	123.9 $\pm$ 36.7	156.8 $\pm$ 39.4
HDL-C (mg/dL), mean $\pm$ SD	46.6 $\pm$ 12.7	52.9 $\pm$ 13.4
Triglyceride (mg/dL), mean $\pm$ SD	249.5 $\pm$ 84.5	205.0 $\pm$ 79.3
non-HDL-C (mg/dL), mean $\pm$ SD	173.3 $\pm$ 34.0	197.2 $\pm$ 40.1
HbA1c (%), mean $\pm$ SD, <NGSP equivalent value>	7.55 $\pm$ 1.44	7.65 $\pm$ 1.51
Fasting blood glucose (mg/dL), mean $\pm$ SD	157.1 $\pm$ 58.0	158.5 $\pm$ 58.7
Systolic blood pressure (mmHg), mean $\pm$ SD	135.3 $\pm$ 15.5	137.4 $\pm$ 16.0
Diastolic blood pressure (mmHg), mean $\pm$ SD	78.6 $\pm$ 9.8	78.5 $\pm$ 9.3

SD: standard deviation

Table 2. Changes in serum lipids following the 24-week treatment period with bezafibrate

Serum lipids (mg/dL)	Number of patients	Baseline	Post-treatment	<i>p</i> value
LDL-C	2116	123.9 $\pm$ 36.7	125.7 $\pm$ 31.3	0.015
HDL-C	2116	46.6 $\pm$ 12.7	52.8 $\pm$ 13.7	<0.001
TG	2116	249.5 $\pm$ 84.5	149.5 $\pm$ 70.5	<0.001
non-HDL-C	2116	173.3 $\pm$ 34.0	155.4 $\pm$ 34.3	<0.001

The data are shown as the mean $\pm$ standard deviation.The paired *t*-test was used to examine the significance of within-group changes.

Table 3. Changes in serum lipids (based on the baseline LDL-C level) after treatment with bezafibrate

Baseline LDL-C level (mg/dL)	Number of patients		Baseline	Post-treatment	Change of value	<i>p</i> value
≥ 160	325	LDL-C	181.4 ± 21.9	148.1 ± 30.1	-33.2 ± 32.3	<0.001
		non-HDL-C	220.6 ± 26.4	175.3 ± 33.8	-45.3 ± 36.0	<0.001
		TG	198.4 ± 78.5	135.5 ± 64.0	-62.9 ± 73.5	<0.001
		TG/HDL-C	4.4 ± 2.3	2.8 ± 1.7	-1.6 ± 2.1	<0.001
≥ 140- < 160	372	LDL-C	149.1 ± 5.6	137.1 ± 26.1	-12.0 ± 26.2	<0.001
		non-HDL-C	193.5 ± 15.2	166.4 ± 31.5	-27.1 ± 29.1	<0.001
		TG	226.2 ± 84.8	146.5 ± 67.1	-79.7 ± 80.4	<0.001
		TG/HDL-C	5.1 ± 2.7	3.0 ± 1.8	-2.2 ± 2.4	<0.001
≥ 120- < 140	435	LDL-C	129.1 ± 5.5	129.2 ± 28.1	0.1 ± 28.1	0.968
		non-HDL-C	177.6 ± 14.5	158.2 ± 32.6	-19.4 ± 29.9	<0.001
		TG	244.4 ± 76.9	146.8 ± 73.7	-97.6 ± 87.9	<0.001
		TG/HDL-C	5.7 ± 2.7	3.1 ± 2.0	-2.6 ± 2.6	<0.001
< 120	984	LDL-C	93.1 ± 20.5	112.5 ± 28.3	19.4 ± 28.2	<0.001
		non-HDL-C	148.1 ± 23.2	143.5 ± 31.5	-4.6 ± 29.9	<0.001
		TG	277.6 ± 78.5	156.5 ± 71.6	-121.1 ± 88.2	<0.001
		TG/HDL-C	6.7 ± 2.8	3.3 ± 2.0	-3.4 ± 2.7	<0.001

The data are shown as the mean ± standard deviation.

The paired *t*-test was used to examine the significance of within-group changes.

Table 4. Changes in serum lipids (based on the baseline TG level) after treatment with bezafibrate

Baseline TG level (mg/dL)	Number of patients		Baseline	Post-treatment	Change of value	<i>p</i> value
≥ 300	586	LDL-C	109.0 ± 35.6	125.6 ± 31.9	16.6 ± 33.6	<0.001
		non-HDL-C	177.6 ± 35.2	161.4 ± 34.1	-16.2 ± 34.8	<0.001
		TG	352.3 ± 59.4	180.8 ± 76.5	-171.5 ± 90.3	<0.001
		TG/HDL-C	8.7 ± 2.8	3.9 ± 2.2	-4.8 ± 3.0	<0.001
≥ 250- < 300	413	LDL-C	117.5 ± 32.8	125.0 ± 31.3	7.5 ± 32.8	<0.001
		non-HDL-C	172.2 ± 32.9	157.0 ± 34.8	-15.2 ± 34.7	<0.001
		TG	273.8 ± 14.8	160.0 ± 65.2	-113.8 ± 64.9	<0.001
		TG/HDL-C	6.4 ± 1.5	3.4 ± 1.8	-3.1 ± 1.9	<0.001
≥ 200- < 250	487	LDL-C	125.8 ± 34.0	124.8 ± 32.7	-1.0 ± 31.6	0.478
		non-HDL-C	170.8 ± 33.7	154.0 ± 35.8	-16.8 ± 33.7	<0.001
		TG	224.8 ± 14.6	146.3 ± 64.2	-78.5 ± 65.5	<0.001
		TG/HDL-C	5.2 ± 1.4	3.0 ± 1.8	-2.2 ± 1.9	<0.001
< 200	630	LDL-C	140.5 ± 35.3	126.9 ± 29.6	-13.5 ± 31.1	<0.001
		non-HDL-C	172.0 ± 33.4	150.0 ± 32.1	-21.9 ± 32.8	<0.001
		TG	157.2 ± 34.8	116.0 ± 56.5	-41.2 ± 58.1	<0.001
		TG/HDL-C	3.3 ± 1.3	2.3 ± 1.6	-1.0 ± 1.6	<0.001

The data are shown as the mean ± standard deviation.

The paired *t*-test was used to examine the significance of within-group changes.

Status of LDL-C Treatment Goal Attainment

Of the 372 patients in the baseline LDL-C ≥ 140 to < 160 mg/dL group, 206 (55.4%) showed an

improvement to an LDL-C level of < 140 mg/dL after treatment and 91 (24.5%) showed an improvement to an LDL-C level of < 120 mg/dL. Furthermore, of the

Table 5. Status of LDL-C treatment goal attainment (classified by the baseline LDL-C level)

Baseline LDL-C level (mg/dL)	Number of patients (n)	Post-treatment LDL-C level (mg/dL)	
		< 140, n (%)	< 120, n (%)
≥ 160	325	134 (41.2)	57 (17.5)
≥ 140- < 160	372	206 (55.4)	91 (24.5)

Fig. 1. Changes in various serum lipid parameters in the LDL-C goal attainment population of patients with a baseline LDL-C level of ≥ 160 mg/dL ($n=325$)

The paired *t*-test was used to examine the significance of within-group changes. *: $p < 0.01$

325 patients in the baseline LDL-C level ≥ 160 mg/dL group, 134 (41.2%) showed an improvement to an LDL-C level of < 140 mg/dL after treatment and 57 (17.5%) showed an improvement to an LDL-C level of < 120 mg/dL (Table 5).

Changes in Serum Lipid Parameters in the LDL-C Goal Attainment Population

Changes in the various serum lipid parameters were analyzed in the patients with a baseline LDL-C level of ≥ 160 mg/dL. The 134 patients who showed a reduction in LDL-C to < 140 mg/dL after treatment also showed significant improvements in the LDL-C, TG, HDL-C and non-HDL-C levels, from baseline values of 175.9 ± 6.7 , 195.6 ± 86.0 , 51.9 ± 11.6 and 214.0 ± 23.5 to 119.8 ± 14.8 , 128.7 ± 65.9 , 56.5 ± 14.5 and 145.7 ± 19.1 mg/dL, respectively, after treatment ($p < 0.01$ for each parameter). The 57 patients who showed a reduction in the LDL-C level to < 120 mg/dL after treatment also showed significant improvements in the LDL-C, TG, HDL-C and non-HDL-C levels, from baseline levels of 178.3 ± 21.3 , 210.7 ± 92.3 ,

51.1 ± 12.3 and 217.9 ± 28.6 to 105.4 ± 10.6 , 132.0 ± 73.6 , 54.9 ± 12.7 and 132.0 ± 15.7 mg/dL, respectively, after treatment ($p < 0.01$, for each parameter) (Fig. 1).

Determinants of the LDL-C Response to Treatment with Bezafibrate

The effect of bezafibrate on LDL-C was evaluated using univariate and multivariate logistic-regression analyses. The following variables were analyzed for their effects on the LDL-C improvement ($\geq 25\%$): sex, age, baseline body mass index, smoking, alcohol use, dietary therapy, exercise therapy, treatment with anti-hypertensive agents, duration of dyslipidemia, duration of diabetes mellitus, treatment with hypoglycemic agents, previous treatment for dyslipidemia, baseline systolic blood pressure, baseline diastolic blood pressure, baseline LDL-C level, baseline HbA1c level and baseline fasting blood glucose level, using a univariate logistic-regression analysis. The results showed that the response to treatment correlated with sex, age, smoking, alcohol use, treatment with anti-hypertensive agents, treatment with hypoglycemic

Table 6. Multivariate logistic-regression analysis of factors influencing the LDL-C response to bezafibrate therapy

Factor	Regression coefficient	Standard error	<i>t</i> value	<i>p</i> value
Sex	-0.045	0.014	-3.27	0.001
Anti-hypertensive agents	0.045	0.013	3.41	<0.001
Hypoglycemic agents	0.030	0.015	2.04	0.042
Baseline LDL-C level	0.029	0.002	15.58	<0.001

$R^2=0.1200$.

The following factors were treated as dummy variables: Sex, 0 for female and 1 for male, anti-hypertensive or hypoglycemic agents, 0 for non-use and 1 for use.

The baseline LDL-C level reflects the influence each 10 mg/dL increment.

agents, previous treatment for dyslipidemia, baseline systolic blood pressure and the baseline LDL-C level. The above factors were entered into the multivariate logistic-regression analysis. The results of the latter analysis identified a female sex, treatment with anti-hypertensive drugs, treatment with hypoglycemic agents and the baseline LDL-C level as significant and independent determinants of the response of LDL-C to 24 weeks of treatment with bezafibrate (**Table 6**).

Discussion

Bezafibrate reportedly improves the TG, HDL-C, and HbA1c levels in dyslipidemic patients with diabetes²⁾ and inhibits both postprandial TG elevation and the development of vascular endothelial dysfunction in patients with metabolic syndrome⁴⁾. The first goal of cardiovascular event prevention in dyslipidemic patients is to control the LDL-C level; however, simply controlling the LDL-C level is insufficient to accomplish this goal. There are reports of the superior effect of bezafibrate when used in combination with a statin, as compared to statin therapy alone^{5, 6)}, in addition to improvements in all lipid parameters with the concurrent use of bezafibrate and ezetimibe⁷⁾. These combined therapies, however, also have safety concerns⁸⁾.

Bezafibrate can enhance the beneficial effects on lipids, especially in patients with diabetes mellitus or metabolic syndrome. However, its effect on the LDL-C level is unclear.

The present subanalysis demonstrated that bezafibrate significantly decreases the LDL-C levels in patients with a baseline LDL-C level of ≥ 140 mg/dL and significantly improves overall serum lipid parameters (TG, LDL-C, HDL-C and non-HDL-C) in the LDL-C goal attainment population. Furthermore, a multivariate logistic-regression analysis of the factors possibly influencing LDL-C responses in patients exhibiting a $\geq 25\%$ reduction in the LDL-C level indicated that the maximum beneficial effects of bezafibrate on LDL-C are seen in women, subjects co-

treated with anti-hypertensive or hypoglycemic agents and those with high baseline LDL-C levels.

Our results showed two contradictory effects of bezafibrate on the serum LDL-C level, and these effects were dependent on the baseline LDL-C level. Specifically, 24 weeks of treatment with bezafibrate significantly decreased the LDL-C levels in the patients with a baseline LDL-C level of ≥ 140 mg/dL (both the ≥ 140 to <160 mg/dL group and the ≥ 160 mg/dL group), whereas it significantly increased the levels in those with a baseline LDL-C level of <120 mg/dL.

The present results are similar to those reported by Ikewaki *et al.*⁹⁾, who studied hypertriglyceridemic patients treated with bezafibrate. They reported an 11% rise in the LDL-C level in their patients. What is the mechanism underlying the bezafibrate-induced increase in LDL-C? While this question was not investigated in the present study, such mechanisms may involve the activation by bezafibrate of lipoprotein lipase (LPL) and hepatic triglyceride lipase to promote the catabolism of TG-rich lipoproteins (e.g., very low-density lipoprotein). LDL-C catabolism mediated by LDL receptors may also be transiently delayed in some patients. In patients with diabetes mellitus, the HbA1c levels correlate closely with the TG and HDL-C levels^{10, 11)}, and high proportions of diabetics are also known to have hypertriglyceridemia and/or hypo-HDL cholesterolemia^{1, 12)}. In cases of diabetes mellitus complicated by hypertriglyceridemia, the small dense LDL level is frequently increased due to a reduced LDL-C particle size¹³⁾. Bezafibrate has been reported to increase the size and decrease the number of small dense LDL particles^{9, 14)}. As mentioned above, the patients in the present study who showed a rise in the LDL-C level in response to bezafibrate had low baseline LDL-C levels and high baseline TG levels. Therefore, in these patients, the high baseline TG levels promoted TG-rich lipoprotein catabolism, such that the post-treatment TG/HDL-C ratio decreased significantly despite the elevated post-treatment LDL-C levels, thus indicating that bezafibrate may change

LDL-C from small dense particles to normal size particles, thereby increasing the LDL-C level.

The size of lipoprotein particles was not measured in the J-BENEFIT study. Hence, it was not possible to investigate small dense LDL in the present study, although the levels of non-HDL-C, including small dense LDL, remnant lipoproteins and all other components of lipoproteins, could be calculated. Accordingly, we studied changes in the non-HDL-C fraction. This includes LDL-C, remnant lipoprotein, small dense LDL and all other atherogenic components of lipoproteins. These forms of cholesterol are considered to be more significant determinants of atherosclerotic disease than LDL-C^{15, 16}. Other studies have shown that the higher the non-HDL-C level, the higher the incidence of coronary artery disease¹⁷. The present study found that bezafibrate increased the LDL-C levels in the patients with a baseline level of <120 mg/dL and significantly decreased the non-HDL-C levels from 148.1 ± 23.2 to 143.5 ± 31.5 mg/dL. In addition, the entire group of 2,116 patients in this study showed improvements in the non-HDL-C levels after treatment (**Table 2**).

The present results demonstrate the overall effect of bezafibrate to be an improvement in serum lipids, as evidenced by a significant decrease in the LDL-C levels in the patients with a baseline LDL-C level of ≥ 140 mg/dL, together with significant decreases in the LDL-C, TG and non-HDL-C levels and a significant increase in the HDL-C levels in the LDL-C goal attainment population. Furthermore, the patients with a baseline LDL-C level of ≥ 160 mg/dL showed a significant decrease in the LDL-C level to <120 mg/dL and the TG level to <150 mg/dL after treatment with bezafibrate. In order to prevent atherosclerosis during treatment for dyslipidemia, it is important to implement a comprehensive therapeutic intervention, not only for the LDL-C level, but also the TG and HDL-C levels. The Japan Diabetes Complications Study (JDCS)¹⁸ considers the TG level, like the LDL-C level, to be a powerful risk factor for coronary artery disease in Japanese patients with type 2 diabetes mellitus. In addition, the authors of that trial reported that, unlike other factors, TG acts synergistically with LDL-C to increase cardiovascular risks. These findings suggest that the ideal treatment for dyslipidemia in patients with type 2 diabetes mellitus should include controlling the TG and HDL-C levels in addition to the LDL-C level. Statins have been widely used to treat dyslipidemia, and evidence supports their role in preventing cardiovascular events¹⁹⁻²³. On the other hand, fibrates, including bezafibrate, improve the TG and HDL-C levels much more effectively than do statins²⁴⁻²⁶. In

the J-BENEFIT study, bezafibrate decreased the TG levels by 45.4% and increased the HDL-C levels by 14.0%². Furthermore, previous studies have described the presence of hypertriglyceridemia and excess atherogenic lipoproteins, e.g., remnant lipoproteins and small dense LDL, in patients with type 2 diabetes mellitus^{12,13}. A comparison of statins and bezafibrate showed a mild improvement in remnant-like lipoprotein cholesterol (RLP-C) with statins versus bezafibrate²⁷. In another study of patients with coronary artery disease complicated by high remnant lipoproteinemia, bezafibrate, but not pravastatin, significantly decreased the RLP-C levels and, during the one-year follow-up period, significantly decreased the incidence of cardiovascular events²⁸. Bezafibrate is thus a therapeutic option in cases of dyslipidemia associated with diabetes mellitus, as statins normalize only the LDL-C level, leaving remnant lipoproteins and small dense LDL as residual risk factors.

Using a multivariate logistic-regression analysis, we analyzed factors responsible for the LDL-C improvements. Consequently, the LDL-C-lowering effect of bezafibrate was especially marked in women, patients co-treated with anti-hypertensive or hypoglycemic agents and the subjects with a high baseline LDL-C level. Previous studies have shown that the LDL-C and TG levels and the incidence of dyslipidemia tend to increase with age in women²⁹ and that menopause and the associated decrease in estrogen results in an increased number of LDL-C particles in the blood and a concomitant decrease in the hepatic uptake of LDL-C from LDL receptors³⁰, resulting in stagnant LDL-C in the blood. In addition, low estrogen levels lead to increased LPL activity, thereby enhancing LDL synthesis and the accumulation of LDL-C. As mentioned above, small dense LDL, the level of which increases in patients with hypertriglyceridemia, should offset the antioxidant effect of estrogen and change LDL-C to induce atherosclerosis. In the JDCS subanalysis³¹, the authors concluded that coronary artery disease may be more sensitively predicted in women by TG than by LDL-C. This finding suggests that it is important to take sex differences into consideration when devising treatment strategies.

As discussed above, compared with statins, bezafibrate is superior with respect to the treatment of dyslipidemia complicated by type 2 diabetes mellitus. Therefore, bezafibrate can potentially be considered as the first choice of drug treatment for responders, as shown in the present study; i.e., in women, patients co-treated with anti-hypertensive or hypoglycemic agents and those with high baseline LDL-C levels.

The present study is associated with several limi-

tations. First, the J-BENEFIT study was a prospective observational trial with no control arm, and all potential confounding factors cannot therefore be excluded. Accordingly, our findings must be interpreted with caution until further controlled interventions are conducted. Second, for logistic, economic and other reasons, laboratory measurements could only be performed in each of the 1,066 participating institutions and not in a central laboratory under standardized conditions. Therefore, differences in the laboratory data due to methodological and racial differences cannot be ruled out. Nevertheless, we believe that, in the clinical setting, the results of the present study allow for the identification of factors influencing the LDL-C response to treatment with bezafibrate.

Conclusions

We evaluated factors influencing the LDL-C responses of dyslipidemic patients with diabetes mellitus to treatment with bezafibrate. The results demonstrated that bezafibrate holds promise in reducing the LDL-C levels in women, patients co-treated with anti-hypertensive or hypoglycemic agents and those with high baseline LDL-C levels.

Acknowledgments

The authors express deep gratitude to the study participants and staff at the 1,066 institutions that provided the data for this study.

Conflicts of Interest

This work was sponsored by KISSEI Pharmaceutical Co., Ltd., Japan.

T. Hirose has received research funds from Sanofi K.K., Eli Lilly Japan K.K., Takeda Pharmaceutical Co., Ltd., Mitsubishi Tanabe Pharma Corporation, MSD K.K., Sumitomo Dainippon Pharma Co., Ltd., Novartis Pharma K.K., Nippon Boehringer Ingelheim Co., Ltd., Ono Pharmaceutical Co., Ltd., AstraZeneca K.K., Taisho Toyama Pharmaceutical Co., Ltd., and Astellas Pharma Inc., lecture fees from Sanofi K.K., Eli Lilly Japan K.K., Novo Nordisk Pharm Ltd., Daiichi-Sankyo Co., Ltd., Mitsubishi Tanabe Pharma Corporation, MSD K.K., Sumitomo Dainippon Pharma Co., Ltd., Novartis Pharma K.K., KISSEI Pharmaceutical Co., Ltd., Nippon Boehringer Ingelheim Co., Ltd. Takeda Pharmaceutical Co., Ltd., and AstraZeneca K.K. and manuscript fees from KISSEI Pharmaceutical Co., Ltd.

T. Teramoto has received research funds from Shionogi Co., Ltd., Astellas Pharma Inc., MSD K.K.

and Daiichi-Sankyo Co., Ltd. and consulting fees, lecture fees and/or honoraria from Astellas Pharma Inc., Takeda Pharmaceutical Co., Ltd., AstraZeneca K.K., MSD K.K., KISSEI Pharmaceutical Co., Ltd., Kowa Co., Ltd., Shionogi Co., Ltd., Daiichi-Sankyo Co., Ltd., Pfizer Inc. and Bayer Yakuhin, Ltd.

K. Abe and T. Taneyama are employees of KISSEI Pharmaceutical Co., Ltd., the sponsor of this study.

References

- 1) Uusitupa MI, Niskanen LK, Siitonen O, Voutilainen E, Pyörälä K: 5-year incidence of atherosclerotic vascular disease in relation to general risk factors, insulin level, and abnormalities in lipoprotein composition in non-insulin-dependent diabetic and nondiabetic subjects. *Circulation*, 1990; 82: 27-36
- 2) Teramoto T, Shirai K, Daida H, Yamada N: Effects of bezafibrate on lipid and glucose metabolism in dyslipidemic patients with diabetes: the J-BENEFIT study. *Cardiovasc Diabetol*, 2012; 11: 29
- 3) Friedewald WT, Levy RI, Fredrickson DS: Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem*, 1972; 18: 499-502
- 4) Ohno Y, Miyoshi T, Noda Y, Oe H, Toh N, Nakamura K, Kohno K, Morita H, Ito H: Bezafibrate improves postprandial hypertriglyceridemia and associated endothelial dysfunction in patients with metabolic syndrome: a randomized crossover study. *Cardiovasc Diabetol*, 2014; 13: 71
- 5) Klempfner R, Goldenberg I, Fisman EZ, Matetzky S, Amit U, Shemesh J, Tenenbaum A: Comparison of statin alone versus bezafibrate and statin combination in patients with diabetes mellitus and acute coronary syndrome. *Am J Cardiol*, 2014; 113: 12-16
- 6) Tenenbaum A, Medvedofsky D, Fisman EZ, Buby L, Matetzky S, Tanne D, Klempfner R, Shemesh J, Goldenberg I: Cardiovascular events in patients received combined fibrate/statin treatment versus statin monotherapy: Acute Coronary Syndrome Israeli Surveys data. *PLoS One*, 2012; 7: e35298
- 7) Teramoto T, Abe K, Taneyama T: Safety and efficacy of long-term combination therapy with bezafibrate and ezetimibe in patients with dyslipidemia in the prospective, observational J-COMPATIBLE study. *Cardiovasc Diabetol*, 2013; 12: 163
- 8) Amend KL, Landon J, Thyagarajan V, Niemcryk S, McAfee A: Incidence of hospitalized rhabdomyolysis with statin and fibrate use in an insured US population. *Ann Pharmacother*, 2011; 45: 1230-1239
- 9) Ikekawa K, Noma K, Tohyama J, Kido T, Mochizuki S: Effects of bezafibrate on lipoprotein subclasses and inflammatory markers in patients with hypertriglyceridemia—a nuclear magnetic resonance study. *Int J Cardiol*, 2005; 101: 441-447
- 10) Khan HA, Sobki SH, Khan SA: Association between glycaemic control and serum lipids profile in type 2 diabetic

- patients: HbA1c predicts dyslipidaemia. *Clin Exp Med*, 2007; 7: 24-29
- 11) Zhang L, Qiao Q, Tuomilehto J, Hammar N, Janus ED, Söderberg S, Mohan V, Ramachandran A, Dong YH, Lam TH, Pang ZC: Blood lipid levels in relation to glucose status in seven populations of Asian origin without a prior history of diabetes: the DECODA study. *Diabetes Metab Res Rev*, 2009; 25: 549-557
 - 12) Schaefer EJ, McNamara JR, Shah PK, Nakajima K, Cupples LA, Ordovas JM, Wilson PW: Elevated remnant-like particle cholesterol and triglyceride levels in diabetic men and women in the Framingham Offspring Study. *Diabetes Care*, 2002; 25: 989-994
 - 13) Hirano T, Ito Y, Koba S, Toyoda M, Ikejiri A, Saegusa H, Yamazaki J, Yoshino G: Clinical significance of small dense low-density lipoprotein cholesterol levels determined by the simple precipitation method. *Arterioscler Thromb Vasc Biol*, 2004; 24: 558-563
 - 14) Hirano T, Kazumi T, Yoshino G: Long-term efficacy of bezafibrate in reduction of small, dense low-density lipoprotein by hypotriglyceridemic action. *Curr Ther Res Clin Exp*, 2000; 61: 127-136
 - 15) Cui Y, Blumenthal RS, Flaws JA, Whiteman MK, Langenberg P, Bachorik PS, Bush TL: Non-high-density lipoprotein cholesterol level as a predictor of cardiovascular disease mortality. *Arch Intern Med*, 2001; 161: 1413-1419
 - 16) Pischon T, Girman CJ, Sacks FM, Rifai N, Stampfer MJ, Rimm EB: Non-high-density lipoprotein cholesterol and apolipoprotein B in the prediction of coronary heart disease in men. *Circulation*, 2005; 112: 3375-3383
 - 17) Kitamura A, Noda H, Nakamura M, Kiyama M, Okada T, Imano H, Ohira T, Sato S, Yamagishi K, Iso H: Association between non-high-density lipoprotein cholesterol levels and the incidence of coronary heart disease among Japanese: the Circulatory Risk in Communities Study (CIRCS). *J Atheroscler Thromb*, 2011; 18: 454-463
 - 18) Sone H, Tanaka S, Tanaka S, Iimuro S, Oida K, Yamasaki Y, Oikawa S, Ishibashi S, Katayama S, Ohashi Y, Akanuma Y, Yamada N; Japan Diabetes Complications Study Group: Serum level of triglycerides is a potent risk factor comparable to LDL cholesterol for coronary heart disease in Japanese patients with type 2 diabetes: subanalysis of the Japan Diabetes Complications Study (JDCS). *J Clin Endocrinol Metab*, 2011; 96: 3448-3456
 - 19) Baigent C, Keech A, Kearney PM, Blackwell L, Buck G, Pollicino C, Kirby A, Sourjina T, Peto R, Collins R, Simes R; Cholesterol Treatment Trialists' (CTT) Collaborators: Efficacy and safety of cholesterol-lowering treatment: prospective meta-analysis of data from 90,056 participants in 14 randomised trials of statins. *Lancet*, 2005; 366: 1267-1278
 - 20) Mills EJ, Rachlis B, Wu P, Devereaux PJ, Arora P, Perri D: Primary prevention of cardiovascular mortality and events with statin treatments: a network meta-analysis involving more than 65,000 patients. *J Am Coll Cardiol*, 2008; 52: 1769-1781
 - 21) Brugts JJ, Yetgin T, Hoeks SE, Gotto Am, Shepherd J, Westendorp RG, de Craen AJ, Knopp RH, Nakamura H, Ridker P, van Domburg R, Deckers JW: The benefits of statins in people without established cardiovascular disease but with cardiovascular risk factors: meta-analysis of randomised controlled trials. *BMJ*, 2009; 338: b2376
 - 22) Cholesterol Treatment Trialists' (CTT) Collaborators, Kearney PM, Blackwell L, Collins R, Keech A, Simes J, Peto R, Armitage J, Baigent C: Efficacy of cholesterol-lowering therapy in 18,686 people with diabetes in 14 randomised trials of statins: a meta-analysis. *Lancet*, 2008; 371: 117-125
 - 23) Cholesterol Treatment Trialists' (CTT) Collaboration, Baigent C, Blackwell L, Emberson J, Holland LE, Reith C, Bhala N, Peto R, Barnes EH, Keech A, Simes J, Collins R: Efficacy and safety of more intensive lowering of LDL cholesterol: a meta-analysis of data from 170,000 participants in 26 randomised trials. *Lancet*, 2010; 376: 1670-1681
 - 24) Saito Y, Yamada N, Shirai K, Sasaki J, Ebihara Y, Yanase T, Fox JC: Effect of rosuvastatin 5-20 mg on triglycerides and other lipid parameters in Japanese patients with hypertriglyceridemia. *Atherosclerosis*, 2007; 194: 505-511
 - 25) Malik J, Melenovsky V, Wichterle D, Haas T, Simek J, Ceska R, Hradec J: Both fenofibrate and atorvastatin improve vascular reactivity in combined hyperlipidaemia (fenofibrate versus atorvastatin trial--FAT). *Cardiovasc Res*, 2001; 52: 290-298
 - 26) Gómez-Gerique JA, Ros E, Oliván J, Mostaza JM, Vilardell M, Pintó X, Civeira F, Hernández A, da Silva PM, Rodríguez-Botero A, Zambón D, Lima J, Díaz C, Aristegui R, Sol JM, Chaves J, Hernández G; ATOMIX Investigators: Effect of atorvastatin and bezafibrate on plasma levels of C-reactive protein in combined (mixed) hyperlipidemia. *Atherosclerosis*, 2002; 162: 245-251
 - 27) Nagai T, Tomizawa T, Nakajima K, Mori M: Effect of bezafibrate or pravastatin on serum lipid levels and albuminuria in NIDDM patients. *J Atheroscler Thromb*, 2000; 7: 91-96
 - 28) Sano K, Nakamura T, Hirano M, Kitta Y, Kobayashi T, Fujioka D, Saito Y, Yano T, Watanabe K, Watanabe Y, Mishina H, Obata JE, Kawabata K, Kugiyama K: Comparative study of bezafibrate and pravastatin in patients with coronary artery disease and high levels of remnant lipoprotein. *Circ J*, 2010; 74: 1644-1650
 - 29) Arai H, Yamamoto A, Matsuzawa Y, Saito Y, Yamada N, Oikawa S, Mabuchi H, Teramoto T, Sasaki J, Nakaya N, Itakura H, Ishikawa Y, Ouchi Y, Horibe H, Kita T: Serum lipid survey and its recent trend in the general Japanese population in 2000. *J Atheroscler Thromb*, 2005; 12: 98-106
 - 30) Wakatsuki A, Okatani Y, Ikenoue N, Fukaya T: Different effects of oral conjugated equine estrogen and transdermal estrogen replacement therapy on size and oxidative susceptibility of low-density lipoprotein particles in postmenopausal women. *Circulation*, 2002; 106: 1771-1776
 - 31) Sone H, Tanaka S, Tanaka S, Iimuro S, Ishibashi S, Oikawa S, Shimano H, Katayama S, Ohashi Y, Akanuma Y, Yamada N; Japan Diabetes Complications Study Group: Comparison of various lipid variables as predictors of coronary heart disease in Japanese men and women with type 2 diabetes: Subanalysis of the Japan Diabetes Complications Study. *Diabetes Care*, 2012; 35: 1150-1157