

# CAVI-Lowering Effect of Pitavastatin May Be Involved in the Prevention of Cardiovascular Disease: Subgroup Analysis of the TOHO-LIP

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**Aim:** In the TOHO Lipid Intervention Trial Using Pitavastatin (TOHO-LIP), a multicenter randomized controlled trial, pitavastatin significantly reduced cardiovascular (CV) events compared to atorvastatin in patients with hypercholesterolemia. To investigate the mechanism by which pitavastatin preferentially prevents CV events, we investigated the relationship between CV events and cardio-ankle vascular index (CAVI) using the TOHO-LIP database.

**Methods:** For the subgroup analysis, we selected patients from a single center, Toho University Sakura Medical Center. After excluding those who had CV events at baseline or during the first year, 254 patients were enrolled. The primary end point was the same as that of TOHO-LIP, and three-point major cardiac adverse events (3P-MACE) was added as secondary end point.

**Results:** The cumulative 5-year incidence of 3P-MACE (pitavastatin 1.6%, atorvastatin 6.1%,  $P=0.038$ ) was significantly lower in pitavastatin group (2 mg/day) than in atorvastatin group (10 mg/day). CAVI significantly decreased only in pitavastatin group during the first year (9.50–9.34,  $P=0.042$ ), while the change in low-density lipoprotein cholesterol (LDL-C) did not differ between the two groups. The change in CAVI during the first year positively correlated with 3P-MACE and tended to be an independent predictor of 3P-MACE in Cox proportional hazards model (hazard ratio, 1.736;  $P=0.079$ ). The annual change in CAVI throughout the observation period was significantly higher in subjects with CV events compared to those without.

**Conclusions:** In this subgroup analysis, the reduction in CV events tended to be associated with the CAVI-lowering effect of pitavastatin, which was independent of the LDL-C-lowering effect.

**Key words:** Low-density lipoprotein cholesterol, Pitavastatin, Atorvastatin, Cardio-ankle vascular index, Cardiovascular disease

## Introduction

Clinical trials have shown that individuals with elevated low-density lipoprotein cholesterol (LDL-C) are at increased risk of cardiovascular disease (CVD), and LDL-C reduction with statin therapy reduces

CVD morbidity and mortality rates<sup>1, 2</sup>. Especially, a potent LDL-C-lowering agent atorvastatin has been shown to prevent cardiovascular (CV) events in various populations at high risk of CVD<sup>3, 4</sup>. On the other hand, more intensive LDL-C lowering by high-dose atorvastatin in the IDEAL study did not result in

a significant reduction in the primary outcome of major coronary events<sup>5</sup>). Pitavastatin is also one of the most potent LDL-C-lowering statins. Pitavastatin has different chemical structure and pharmacokinetic profile compared to atorvastatin. Pitavastatin is lipophilic, and unlike atorvastatin, which is metabolized by CYP3A4, pitavastatin metabolism is not dependent on cytochrome P450<sup>6</sup>). In CHIBA study, pitavastatin 2 mg and atorvastatin 10 mg were equally effective in improving the lipid profile in Japanese patients with hypercholesterolemia, but only pitavastatin showed consistent reduction of non-HDL-C regardless of the body size<sup>7</sup>). Furthermore, numerous studies have reported that pitavastatin has more favorable effects on glucose metabolism<sup>8-10</sup>), high-density lipoprotein cholesterol (HDL-C)<sup>11</sup>), and endothelial function<sup>12</sup>) compared with atorvastatin. However, there is no randomized prospective study of pitavastatin in a 1-to-1 fashion to atorvastatin. Therefore, we conducted a multicenter, open-label, randomized controlled, head-to-head trial, the TOHO Lipid Intervention Trial Using Pitavastatin (TOHO-LIP), to compare the effects of pitavastatin with atorvastatin therapy for CV event prevention in patients with hypercholesterolemia at high risk of CVD<sup>13</sup>). The primary end point was a composite of CV death, sudden death of unknown origin, nonfatal myocardial infarction (MI), nonfatal stroke, transient ischemic attack, and heart failure requiring hospitalization. Pitavastatin significantly reduced the risk of the primary end point compared to atorvastatin [pitavastatin, 2.9%; atorvastatin, 8.1%; hazard ratio (HR), 0.366], while there was no difference in the change in LDL-C level between the two groups. However, the mechanism by which pitavastatin preferentially prevents CV events is not fully understood.

Arterial stiffness is mainly recognized as an indicator of arteriosclerosis and a predictor of CV events. Cardio-ankle vascular index (CAVI) is a new arterial stiffness index, and the equation is essentially derived from the stiffness parameter  $\beta$ <sup>14, 15</sup>). Therefore, CAVI is independent of blood pressure at the time of measurement<sup>16</sup>) and indicates the intrinsic stiffness of the aortic wall. Increased CAVI is observed in persons with CVD and risk factors such as dyslipidemia, and the improvement of risk factors reduces CAVI<sup>15, 17</sup>). Miyashita *et al.*<sup>18</sup>) reported that pitavastatin treatment significantly decreased CAVI, as well as marker of

oxidative stress in patients with type 2 diabetes. Furthermore, many prospective studies have revealed that CAVI predicts future CV events<sup>15, 19</sup>). Therefore, in the context of TOHO-LIP, we are keenly interested in whether pitavastatin lowers CAVI and whether the change in CAVI is associated with CV events. In the present subgroup analysis using the TOHO-LIP database, we investigated the relationship between CV events and annual CAVI changes.

## Subjects and Methods

### Study Design and Subjects

The design and results of the TOHO-LIP have been previously reported<sup>13</sup>). The TOHO-LIP was a randomized, controlled, open-label, parallel-grouped, multi-center clinical trial to examine the effect of pitavastatin compared with atorvastatin on cardiovascular events in patients with hypercholesterolemia who had one or more cardiovascular risk factors. Pitavastatin 2 mg/day is known to have a LDL-C-lowering effect comparable to that of atorvastatin 10 mg/day<sup>20</sup>). Overall, 664 patients were enrolled from April 1, 2006, to May 31, 2011, and were randomized to receive either pitavastatin 2 mg/day or atorvastatin 10 mg/day. The duration of treatment with the statins was 240 weeks. The LDL-C of <100 mg/dl was set at the optimal target level in the TOHO-LIP. For the present subgroup analysis, we used the database of TOHO-LIP and selected 390 outpatients who attended Toho University Sakura Medical Center in Chiba, Japan. Patients who had any CV event at baseline or during the first year were excluded. To avoid inaccurate CAVI measurements, patients with low ankle-brachial index (<0.9) or with atrial fibrillation were excluded. Eventually, 254 patients were enrolled (pitavastatin group, 123 patients; atorvastatin group, 131 patients). Background characteristics of this subgroup analysis compared to the excluded subjects were shown in **Table 4 (supplemental material)**.

During the follow-up period, CAVI was measured at baseline, at 1 year, and yearly thereafter. The numbers of CAVI follow-up at 1, 2, 3, 4, and 5 years after randomization were 254, 195, 172, 145, and 127, respectively. CAVI was measured with a VaSera CAVI instrument (Fukuda Denshi Co Ltd, Tokyo, Japan) by methods previously described<sup>21</sup>).

The study was conducted in accordance with the

Declaration of Helsinki. The present retrospective observational study was approved by the Ethics Committee of Toho University Sakura Medical Center (S18069). Informed consent was obtained in the form of opt-out on the website.

### End Points

The primary end point was a composite of CV death, sudden death of unknown origin, nonfatal MI, nonfatal stroke, transient ischemic attack, and heart failure requiring hospitalization. Details of the end point were previously described<sup>13</sup>. In this subgroup analysis, a composite of three-point major cardiac adverse events (3P-MACE: CV death, nonfatal MI, nonfatal stroke) was added as the secondary endpoint.

### Statistical Analysis

The results are expressed as mean  $\pm$  S.D. SPSS 15.0 (SPSS Inc., Chicago, Ill, USA) was used in all statistical analyses. For two-group comparisons, all parametric data were analyzed using Student's *t*-test. All nonparametric data were analyzed using Mann-Whitney *U* test. Fisher's exact test was used to detect significant differences between proportions and categorical variables. Comparisons among multiple groups were conducted using one-way ANOVA, followed by Tukey's test as post hoc test. Kaplan-Meier survival analysis was employed to estimate the differences of the time to end point between the groups. Cox proportional hazards regression analysis was used to identify the predictors of cardiovascular events, and the result is expressed as hazard ratio with 95% confidence interval. A two-sided *p* value of 0.05 was considered statistically significant.

## Results

### Background Characteristics at Baseline and End Points in the Subgroup Analysis

At baseline, the mean age was 64.8 years, body mass index was 24.5 kg/m<sup>2</sup>, blood pressure was 134.1/78.1 mmHg, HbA1c was 7.4%, LDL-C was 157.2 mg/dl, and CAVI was 9.56. Other background parameters are shown in **Table 1**. There were no significant differences in all background parameters between pitavastatin and atorvastatin groups.

In TOHO-LIP, pitavastatin significantly reduced the risk of the primary end point compared to atorvastatin<sup>13</sup>. In this subgroup analysis of 254 patients, we performed Kaplan-Meier survival analysis to reaffirm the difference between the two groups. The cumulative 5-year incidence of the primary end point [pitavastatin, 3.3% (*n*=4); atorvastatin, 6.9% (*n*=9); *P*=0.078] (**Fig. 1a**) was not significant

between groups, and the incidence of 3P-MACE [pitavastatin, 1.6% (*n*=2); atorvastatin, 6.1% (*n*=8); *P*=0.038] (**Fig. 1b**) was significantly lower in the pitavastatin group than in the atorvastatin group. The total incidence of the primary end point was 5.1% in this subgroup analysis (*n*=254), whereas the incidence in the excluded subjects (*n*=136) was 5.9%, and there were no significant differences (**Table 4, supplemental material**). Since patients with any CV event at baseline or during the first year were excluded in this subgroup analysis, there were no CV events during the first year.

### Changes in CAVI and Difference Between Pitavastatin and Atorvastatin Groups

**Fig. 2** shows the trend of CAVI in pitavastatin and atorvastatin groups during the 5-year study period. There was no significant change in CAVI for 5 years in each group, and there was no difference in the amount of change between the two groups. The trend of LDL-C and the achievement rates in both groups during the 5-year study period were shown in **Table 5 (supplemental material)**, and there was no difference in the tendency between the groups. The changes in the estimated glomerular filtration rate over 5 years ranged from 74.2 to 70.2 ml/min/1.73 m<sup>2</sup> in pitavastatin group and from 70.9 to 74.8 ml/min/1.73 m<sup>2</sup> in atorvastatin group (data not shown). There was no significant change in each group, and there was no difference in the changes between the groups. There was no difference between groups in change of % patients using each medications during the 5-year study period (**Table 6, supplemental material**).

Next, the changes in CAVI during the first year in pitavastatin and atorvastatin groups were compared (**Fig. 3**). The change in CAVI in atorvastatin group was not significant (9.61–9.51; *P*=0.213). In contrast, CAVI significantly decreased in pitavastatin group (9.50–9.34; *P*=0.042). In addition, there was no difference in mean change in LDL-C during the first year between the pitavastatin (155.2–97.1 mg/dl) and atorvastatin (159.1–94.5 mg/dl) groups (**Table 5, supplemental material**). The percentage of using antihypertensive agents, which might affect the change in CAVI, did not change in both groups, and the changes were not significant between the groups during the first year (ACE-I and/or ARB, 42.7%–43.5% in the pitavastatin group and 52.8%–53.6% in the atorvastatin group; calcium channel blocker, 42.0%–42.3% in the pitavastatin group and unchanged in the atorvastatin group; diuretics, unchanged in the pitavastatin group and 10.6%–11.4% in the atorvastatin group;  $\beta$ -receptor antagonist, unchanged in both groups) (data not

**Table 1.** Background characteristics at baseline in the subgroup analysis

	All (n = 254) mean±SD	Pitavastatin (n = 123) mean±SD	Atorvastatin (n = 131) mean±SD	P-value between groups
Age (years)	64.8±9.3	64.2±9.8	65.5±8.7	NS
Gender (male/female)	118/136	57/66	61/10	NS
Height (cm)	158.8±8.7	160.1±9.3	158.0±8.4	NS
Body weight (kg)	61.7±11.5	62.4±12.4	61.2±10.8	NS
BMI (kg/m <sup>2</sup> )	24.5±3.4	24.6±3.8	24.4±3.2	NS
Systolic blood pressure (mmHg)	134.1±16.8	131.9±16.9	135.9±16.6	NS
Diastolic blood pressure (mmHg)	78.1±10.6	77.4±11.1	78.7±10.1	NS
Heart rate (beats per minute)	68.8±11.0	70.2±11.0	68.1±11.0	NS
FBG (mg/dl)	152.1±62.0	161.9±66.2	142.9±56.4	NS
HbA1c (%)	7.4±1.3	7.4±1.2	7.5±1.4	NS
AST (IU/L)	23.3±8.0	23.9±8.8	22.8±7.2	NS
ALT (IU/L)	24.7±13.0	26.1±14.3	23.4±11.6	NS
Cr (mg/dl)	0.8±0.2	0.8±0.2	0.8±0.2	NS
eGFR (ml/ min/ 1.73 m <sup>2</sup> )	72.1±20.1	74.2±19.0	70.9±15.4	NS
Uric acid (mg/dl)	5.2±1.5	5.1±1.4	5.2±1.5	NS
TC (mg/dl)	245.0±39.1	243.3±39.2	246.6±39.0	NS
TG (mg/dl)	166.1±95.4	175.7±107.8	157.0±81.5	NS
HDL-C (mg/dl)	55.1±13.1	54.4±12.9	55.8±13.2	NS
LDL-C (mg/dl)	157.2±34.8	155.2±34.6	159.1±35.0	NS
CAVI	9.56±1.26	9.50±1.19	9.61±1.33	NS
Prevalence of hypertension (%)	68.1	67.2	69.1	NS
Prevalence of diabetes (%)	92.1	91.6	92.6	NS
% Patients using medications				
ACE-I and/or ARB	47.6	42.7	52.8	NS
Calcium channel blocker	42.5	42.0	43.1	NS
Diuretics	12.2	13.7	10.6	NS
β receptor antagonist	13.0	10.7	15.4	NS
Insulin	18.5	17.6	19.5	NS
Sulfonylurea	50.0	50.4	49.6	NS
Biguanide	31.5	29.0	34.1	NS
αglucosidase inhibitor	22.0	18.3	26.0	NS
Thiazolidinedione	16.1	16.0	16.3	NS

Abbreviations: BMI, body mass index; FBG, fasting blood glucose; HbA1c, glycosylated hemoglobin; AST, aspartate aminotransferase; ALT, alanine transaminase; Cr, Creatinine; eGFR, estimated glomerular filtration rate; TC, total cholesterol; TG, triglyceride; HDL-C; high-density lipoprotein cholesterol; LDL-C; low-density lipoprotein cholesterol; CAVI, Cardio-ankle Vascular Index; ACE-I, angiotensin converting enzyme inhibitors, ARB, angiotensin receptor antagonists; NS, not significant.

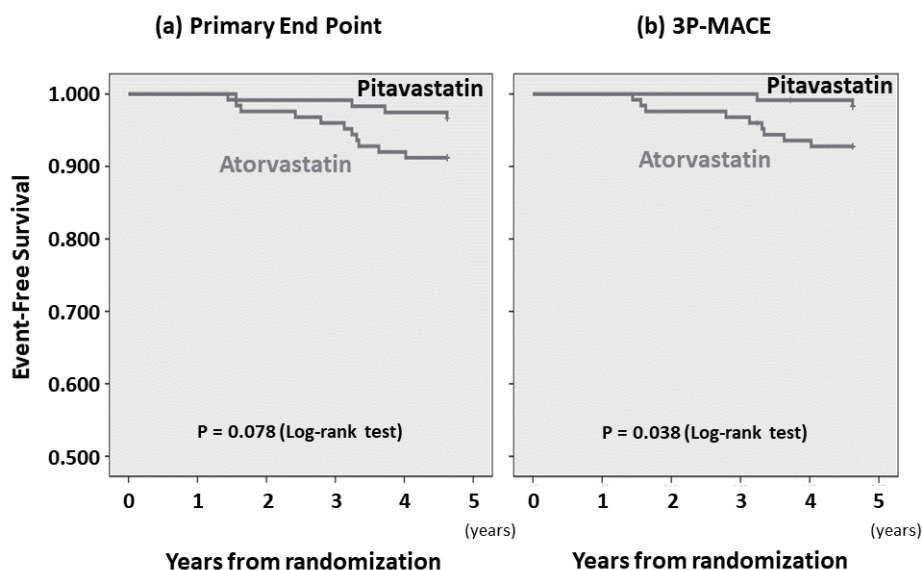
shown).

### Relationship Between CAVI and Future Cardiovascular Events

We examined whether the change in CAVI during the first year was related to future CV events. In univariate correlation analysis, 3P-MACE positively correlated only with the change in CAVI, but not with the change in body weight, blood pressure, HbA1c, or lipid parameters, including LDL-C (Table 2). On the other hand, there was no significant correlation between the primary end point and any of the tested variables.

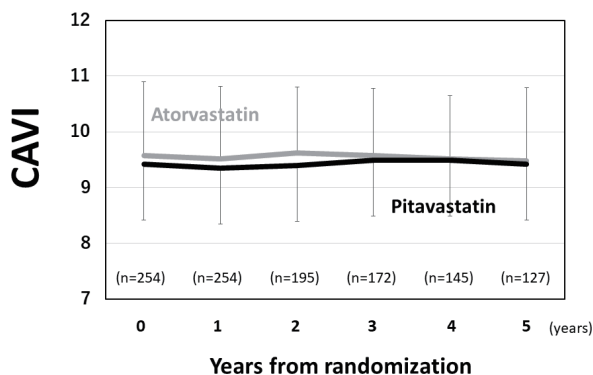
Next, we examined the factors associated with 3P-MACE using Cox proportional hazards regression analysis. To select variables for the regression model, the following were considered: LDL-C is the most important risk factor targeted by statins, CAVI was significantly associated with 3P-MACE in univariate analysis, and significant group difference of CV events was found in Kaplan-Meier analysis. Therefore, these three factors were included in model 1 (Table 3a). In addition, since gender and age are generally important risk factors, these two factors were added to model 2 (Table 3b). Both models 1 and 2 detected no significant independent predictor of future CV events.





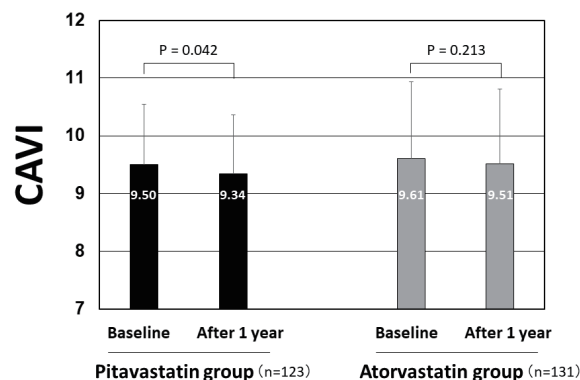
**Fig. 1.** Kaplan-Meier survival curves for the primary end point (a) and 3P-MACE (b)

Primary end point: composite of cardiovascular death, sudden death of unknown origin, nonfatal myocardial infarction, nonfatal stroke, transient ischemic attack, and heart failure requiring hospitalization. 3P-MACE: three-point major cardiac adverse events (cardiovascular death, nonfatal myocardial infarction, nonfatal stroke).



**Fig. 2.** Trend of CAVI in pitavastatin and atorvastatin groups during the 5-year study period

CAVI, cardio-ankle vascular index.



**Fig. 3.** Changes in CAVI during the first year in pitavastatin and atorvastatin groups

CAVI, cardio-ankle vascular index.

However, the change in CAVI during the first year tended to be an independent factor in models 1 (HR, 1.758;  $P=0.084$ ) and 2 (HR, 1.736;  $P=0.079$ ).

Furthermore, we examined whether the annual CAVI change until the occurrence of any CV event or the end of the 5-year study period was associated with future CV events. The annual CAVI change was significantly higher in subjects with CV events compared to subjects without CV events, both for the primary end point (Fig. 4a) and for 3P-MACE (Fig. 4b). In addition, the respective baseline CAVI in subjects without CV events and those with CV events

were 9.52 and 9.76 ( $P=0.307$ ) for primary end point and 9.52 and 9.78 ( $P=0.393$ ) for 3P-MACE, with no significant differences.

### Adverse Events and Laboratory Test Abnormalities

The rates of adverse events, including rhabdomyolysis and new onset of diabetes mellitus, were low and did not differ between the two groups. There was no difference also in the rate of laboratory test abnormalities between the two groups (Table 7, supplemental material).

**Table 2.** Univariate correlation analysis between the changes in clinical variables during the first year and the occurrence of future CV events

Variables	Primary End Point (no; 0, yes; 1)		3P-MACE (no; 0, yes; 1)	
	r	P-value	r	P-value
ΔBody Weight (kg)	0.034	0.652	0.017	0.818
ΔSystolic Blood Pressure (mmHg)	0.023	0.795	-0.037	0.672
ΔHbA1c (%)	0.053	0.413	0.052	0.419
Δlog TG (mg/ dl)	-0.037	0.559	-0.029	0.651
ΔHDL-C (mg/ dl)	0.018	0.771	-0.042	0.504
ΔLDL-C (mg/ dl)	0.002	0.971	0.074	0.245
ΔCAVI	0.179	0.210	0.219	0.049

Abbreviations: CV, cardiovascular; 3P-MACE, 3-point major cardiac adverse events; HbA1c, glycosylated hemoglobin; HCL-C, high-density lipoprotein cholesterol; LCL-C, low-density lipoprotein cholesterol; CAVI, Cardio-ankle Vascular Index.

**Table 3.** Cox proportional hazards regression analysis of the association between 3P-MACE and clinical variables

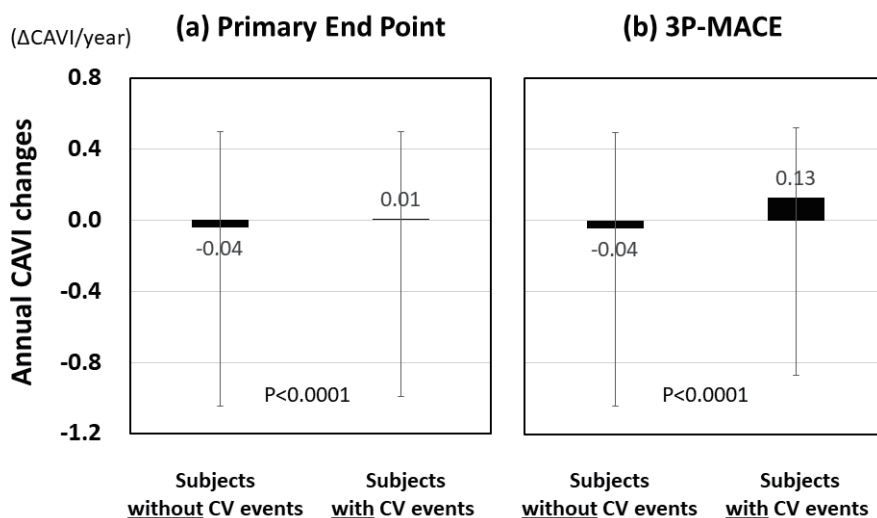
(a) Model 1			
Variables	Hazard Ratio	95% Confidential Interval	P-value
ΔLDL-C (Δmg/dl)	1.007	0.994 - 1.021	0.280
ΔCAVI	1.758	0.926 - 3.338	0.084
Group (Atorvastatin; 0, Pitavastatin; 1)	0.493	0.171 - 1.420	0.190
(b) Model 2			
Variables	Hazard Ratio	95% Confidential Interval	P-value
Gender (male; 0, female; 1)	0.666	0.245 - 1.813	0.426
Age (years old)	1.039	0.978 - 1.104	0.215
ΔLDL-C (Δmg/dl)	1.006	0.993 - 1.020	0.363
ΔCAVI	1.736	0.938 - 3.213	0.079
Group (Atorvastatin; 0, Pitavastatin; 1)	0.501	0.172 - 1.461	0.206

Abbreviations: 3P-MACE, 3-point major cardiac adverse events; LDL-C, low-density lipoprotein cholesterol; CAVI, cardio-ankle vascular index.

## Discussion

This subgroup analysis used the data of 254 patients selected from the study population of a primary prevention trial of CV events, the TOHO-LIP<sup>13</sup>. In this subgroup, the cumulative 5-year incidence of 3P-MACE was significantly lower in the pitavastatin group than in the atorvastatin group. On the other hand, despite excluding patients with any CV event at baseline in this subgroup, the total incidence of primary end point was 5.1%, which was equivalent to that of the main TOHO-LIP study. The average CAVI in this subgroup analysis was 9.56, which was more than 9 and was considered to be the high-risk group for future CV events<sup>15</sup>. The reason

why the baseline CAVI in this study is high, although there is no history of CV events, may be as follows. Shown in **Table 4 (supplemental material)**, the mean HbA1c and the prevalence of diabetes (92.1%) were higher in this subgroup compared with that of the excluded subjects. In a cross-sectional study in which Japanese urban residents participated, CAVI in the CVD risk-free group aged 60–69 were 8.73 for males and 8.26 for females, while they were 9.41 and 9.09 in the hyperglycemia group, respectively<sup>22</sup>. In the other studies in patients with type 2 diabetes and dyslipidemia, the baseline CAVI was approximately 9.5<sup>18, 23</sup>. In patients with diabetes, it is thought that CAVI is increased by mechanisms such as postprandial hyperglycemia, insulin resistance, and oxidative



**Fig. 4.** Differences in the annual CAVI changes in patients with or without CV events: (a) primary end points and (b) 3P-MACE

“Annual CAVI change” was defined as the annual change in CAVI until the occurrence of any CV event or the end of 5-year study period. Primary end point: composite of cardiovascular death, sudden death of unknown origin, nonfatal myocardial infarction, nonfatal stroke, transient ischemic attack, and heart failure requiring hospitalization. 3P-MACE: three-point major cardiac adverse events (cardiovascular death, nonfatal myocardial infarction, nonfatal stroke). CAVI, cardio-ankle vascular index; CV, cardiovascular.

stress<sup>15</sup>). The combination of diabetes and dyslipidemia may further increase CAVI and the risk of CVD.

In this subgroup study, the incidence of 3P-MACE was significantly lower in the pitavastatin group; however, there was no difference in the degree of LDL-C reduction between the two statins, similar to the main study. The reason why pitavastatin is superior to atorvastatin in CV event reduction despite comparable LDL-C-lowering effect remains unclear.

CAVI has been widely used in clinical medicine for the past 15 years as an index for the evaluation of CVD and risk factors. A number of studies have investigated the association between CAVI and the occurrence of CV events<sup>15</sup>. Overall, CAVI is high in patients with coronary risk factors. However, some conventional risk factors do not affect CAVI. Regarding lipid abnormalities, CAVI was higher in patients with dyslipidemia than in controls<sup>24</sup>, and a linear relation between CAVI and all the conventional lipid parameters was demonstrated by a trend test<sup>17</sup>. On the other hand, several researchers found no elevation of CAVI in patients with primary hypercholesterolemia<sup>24-27</sup>. Several studies have investigated the effect of cholesterol-lowering agents on CAVI. As mentioned above, Miyashita *et al.*<sup>18</sup> reported that pitavastatin 2 mg/day significantly reduced CAVI together with serum markers of oxidative stress, malondialdehyde-LDL, and urinary 8-hydroxy-2'-deoxyguanosine in patients with type 2 diabetes. Another lipid-lowering drug, bezafibrate, also reduced CAVI, accompanied by the improvement

of remnant-like particle cholesterol and diacron-reactive oxygen metabolites<sup>22</sup>. Pitavastatin may have reduced CAVI and the incidence of CV events associated with the serum markers of oxidative stress and remnant-like particle cholesterol; however, these data were not measured in this study. This is a limitation of this study. Furthermore, in the main TOHO-LIP study, C-reactive protein levels decreased after 1 year of pitavastatin therapy but did not change with atorvastatin therapy<sup>13</sup>. However, since there were very few cases with the data on both CAVI and C-reactive protein, examining the involvement of anti-inflammation effect of pitavastatin on arterial stiffness was impossible. This is also a limitation of this study. There is no doubt that LDL-C is an important risk factor; however, although the accumulation of LDL-C in blood vessels leads to lipidosis, the changes do not immediately affect the arterial stiffness<sup>26</sup>. These findings suggest that CAVI may only increase after the development of inflammation caused by oxidative stress, with subsequent formation of fibrous cap and complicated lesion. In other words, pitavastatin may lower CAVI by improving the oxidative stress and inflammation, independent of the LDL-C-lowering effects.

Furthermore, several studies have reported that pitavastatin is superior to other statins in improving triglyceride (TG) metabolism. In CHIBA study, which was a multicenter randomized study on drug intervention for hypercholesterolemia, pitavastatin, but not atorvastatin, significantly reduced TG and increased HDL-C in patients with metabolic

syndrome<sup>7</sup>). Saiki *et al.*<sup>28</sup>) reported that pitavastatin stimulated *in vitro* lipoprotein lipase (LPL) activity in 3T3-L1 preadipocytes more potently than atorvastatin, which may facilitate the increase in HDL through efficient metabolism of TG-rich lipoproteins. Nagayama *et al.*<sup>17</sup>) reported that all the conventional lipid parameters independently contributed to high CAVI in logistic regression models, although TG showed the largest area under the receiver operating characteristic curve for predicting high CAVI. Pavlovska *et al.*<sup>27</sup>) also concluded that high TG was associated with high CAVI independent of multiple cardiometabolic risk factors. In this study, TG metabolism related to LPL may be one of the mechanisms by which pitavastatin lowers CAVI and reduces CV events. However, there was no difference in the TG-lowering effect between pitavastatin and atorvastatin in the TOHO-LIP. This result may have been affected by the exclusion of patients with TG higher than 400 mg/dl due to the use of Friedewald formula for calculating LDL-C levels in the TOHO-LIP study.

In addition, **Fig. 4** shows that CAVI decreased in the subjects without CV events but increased in those with CV events, although the baseline CAVI in the two groups were similar. This finding might simply indicate that patients with improved arteriosclerosis have fewer CV events, whereas patients with advanced arteriosclerosis have more CV events. On the other hand, the improvement in CAVI may also reflect the effects of statins on vascular remodeling<sup>29</sup>), which may be associated with the suppression of CV events. Further investigation is required to elucidate the relationship between the changes in arterial stiffness and future CV events.

The dose of pitavastatin (2 mg/day) and atorvastatin (10 mg/day) in this study was determined by referring to phase IV Japanese studies, including the CHIBA study<sup>7</sup>) and LIVES study<sup>30</sup>). However, in studies such as REAL-CAD<sup>31</sup>) conducted after the TOHO-LIP study, the dose of pitavastatin and atorvastatin were applied at 4 and 20 mg/day, respectively. One limitation of this study is that the absence of high doses of statins may have affected the achievement rates of LDL-C and CV events. Furthermore, when this study was designed, the LDL-C target level was set at 100 mg/dl, which was not sufficient with the current guideline<sup>32</sup>).

There are several other limitations of this study. The original TOHO-LIP was conducted as an open-label trial, which has inherent limitations. However, to reduce the open-label effect, the independent event committee adjudicated all end point events while blinded to the randomization. This subgroup analysis

was a retrospective, single-center study using the TOHO-LIP database. The sample size was small, which could be a significant obstacle in showing a trend and significant relationship. The prevalence of diabetes mellitus and a history of CVD were different from the main TOHO-LIP study. The cumulative 5-year incidence of the primary end point was not significant between groups. These findings suggest that the population characteristics of this study is different from that of the main TOHO-LIP study. Change in CAVI during the first year tended to be an independent predictor of 3P-MACE in Cox proportional hazards model, but it was not significant. The percentage of patients using ACE-I and/or ARB was only 47.6%, although the prevalence of hypertension was 68.1%. This may affect the CAVI and CV events.

## Conclusions

In this subgroup analysis using the TOHO-LIP database, we retrospectively investigated the relationship between primary prevention of CV events and annual CAVI changes. The incidence of 3P-MACE was significantly lower in the pitavastatin group than in the atorvastatin group. CAVI significantly decreased in the pitavastatin group, but not in the atorvastatin group, while the change in LDL-C was not significantly different between the two groups. The change in CAVI during the first year positively correlated with 3P-MACE in a univariate correlation analysis and tended to be an independent predictor of 3P-MACE in Cox proportional hazards regression analysis. The annual CAVI changes were significantly higher in subjects with CV events compared to those without CV events, for both primary end point and 3P-MACE. This subgroup analysis indicates that the reduction in CV events by pitavastatin tends to be associated with the CAVI-lowering effect, which is independent of the LDL-C-lowering effect. The possibility that the CAVI-lowering effect is mediated by the improvement of oxidative stress, inflammation, and TG metabolism requires further investigations.

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**Table 4 (Supplemental).** Background characteristics of the subgroup analysis compared to excluded subjects

	Included subjects (n = 254)	Excluded subjects (n = 136)	P-value
Age (years)	64.8 ± 8.9	64.1 ± 9.9	NS
Gender (male/female)	118/136	70/66	NS
Height (cm)	158.8 ± 8.7	160.0 ± 10.5	NS
Body weight (kg)	61.7 ± 11.5	63.1 ± 12.3	NS
BMI (kg/m <sup>2</sup> )	24.5 ± 3.4	25.2 ± 4.0	NS
Systolic blood pressure (mmHg)	134.1 ± 16.8	136.4 ± 16.2	NS
Diastolic blood pressure (mmHg)	78.1 ± 10.5	79.3 ± 11.2	NS
FBG (mg/dl)	152.1 ± 62.0	124.9 ± 58.8	NS
HbA1c (%)	7.4 ± 1.3	6.9 ± 1.5	< 0.001
AST (IU/L)	23.3 ± 8.0	23.6 ± 8.8	NS
ALT (IU/L)	24.7 ± 13.0	24.2 ± 14.4	NS
Cr (mg/dl)	0.8 ± 0.2	0.8 ± 0.2	NS
Uric acid (mg/dl)	5.2 ± 1.5	5.2 ± 1.3	NS
TC (mg/dl)	245.0 ± 39.1	241.7 ± 41.8	NS
TG (mg/dl)	166.1 ± 95.4	166.2 ± 98.6	NS
HDL-C (mg/dl)	55.1 ± 13.1	53.9 ± 12.4	NS
LDL-C (mg/dl)	157.2 ± 34.8	155.0 ± 37.6	NS
Prevalence of hypertension (%)	68.1	70.6	NS
Prevalence of diabetes (%)	92.1	67.3	< 0.001
Prevalence of prior ACS and/or coronary revascularization (%)	0.0	26.2	< 0.001
Prevalence of stroke (%)	0.0	18.2	< 0.001
Prevalence of peripheral artery disease (%)	0.0	6.9	< 0.001
Primary End Point (%)	5.1	5.9	NS

**Table 5 (Supplemental).** Trend of LDL-C and the achievement rates in pitavastatin and atorvastatin groups during the 5-year study period

	Years from randomization					
	Baseline	1	2	3	4	5
<b>Pitavastatin</b>						
LDL-C (mg/dl)	155.2 ± 34.6	97.1 ± 27.3	96.2 ± 25.0	96.3 ± 29.6	95.8 ± 25.3	96.8 ± 27.6
Achievement rates of LDL-C < 100 mg/dl (%)	4.0	60.2	63.0	62.5	64.5	65.8
<b>Atorvastatin</b>						
LDL-C (mg/dl)	159.1 ± 35.0	94.5 ± 27.0	93.7 ± 25.2	93.1 ± 27.5	94.1 ± 25.9	94.7 ± 26.2
Achievement rates of LDL-C < 100 mg/dl (%)	3.5	63.0	62.1	63.3	66.1	68.3

Abbreviations: LDL-C, low-density lipoprotein cholesterol.

**Table 6 (Supplemental).** Change in the percentage of patients using medications

% Patients using medications	Pitavastatin (n = 123)			Atorvastatin (n = 131)			Difference in change between groups
	Baseline	After 5 years	P-value	Baseline	After 5 years	P-value	
ACE-I and/or ARB	42.7	49.2	P=0.004	52.8	57.2	P=0.048	NS
Calcium channel blocker	42.0	44.1	NS	43.1	44.9	NS	NS
Diuretics	13.7	15.1	NS	10.6	12.4	NS	NS
β receptor antagonist	10.7	12.8	NS	15.4	17.2	NS	NS
Insulin	17.6	20.7	NS	19.5	21.3	NS	NS
Sulfonylurea	50.4	55.8	P=0.016	49.6	55.4	P=0.020	NS
Biguanide	29.0	38.1	P<0.001	34.1	42.9	P=0.001	NS
αglucosidase inhibitor	18.3	21.5	NS	26.0	28.7	NS	NS
Thiazolidinedione	16.0	19.8	P=0.031	16.3	19.9	P=0.042	NS

Abbreviations: ACE-I, angiotensin converting enzyme inhibitors; ARB, angiotensin receptor antagonists; NS, not significant.

**Table 7 (Supplemental).** Adverse events and laboratory test abnormalities

Event	Pitavastatin (n = 123)	Atorvastatin (n = 131)	P value
	n (%)	n (%)	
Adverse events	0 (0.0)	0 (0.0)	NS
Rhabdomyolysis*	0 (0.0)	3 (2.3)	NS
Muscle complaints	1 (0.8)	1 (0.8)	NS
Gallbladder-related events	0 (0.0)	0 (0.0)	NS
Cholecystectomy	1 (0.8)	2 (1.5)	NS
New onset of diabetes mellitus**	0 (0.0)	0 (0.0)	NS
Psychiatric disorders			
Laboratory test abnormalities			
Elevation of alanine aminotransferase, aspartate aminotransferase, or both >3 upper limit of normal range	2 (1.6)	2 (1.5)	NS
Elevation of creatine kinase >5 upper limit of normal range	0 (0.0)	0 (0.0)	NS
Elevation of creatinine >150 % of baseline	8 (6.5)	9 (6.9)	NS
New-onset decrease of hemoglobin <11.0 g/dl	6 (4.9)	5 (3.8)	NS

\*Rhabdomyolysis was adjudicated as >10 times the elevation of creatine kinase compared to the upper limit of the normal range and/or a clinical course consistent with rhabdomyolysis.

\*\*New-onset diabetes mellitus was defined as a hemoglobin A1c of 6.4% at least once during follow-up in patients without a diagnosis of diabetes mellitus during the randomization assignment.

Fisher's exact test was used to determine P-values.