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Effect of Atorvastatin Therapy on Fibrous Cap Thickness in Coronary Atherosclerotic Plaque as Assessed by Optical Coherence Tomography

The EASY-FIT Study

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ABSTRACT

BACKGROUND The detailed mechanism of plaque stabilization by statin therapy is not fully understood.

OBJECTIVES The aim of this study was to assess the effect of lipid-lowering therapy with 20 mg/day of atorvastatin versus 5 mg/day of atorvastatin on fibrous cap thickness in coronary atherosclerotic plaques by using optical coherence tomography (OCT).

METHODS Seventy patients with unstable angina pectoris and untreated dyslipidemia were randomized to either 20 mg/day or 5 mg/day of atorvastatin therapy. OCT was performed to assess intermediate nonculprit lesions at baseline and 12-month follow-up.

RESULTS Serum low-density lipoprotein cholesterol level was significantly lower during therapy with 20 mg/day compared with 5 mg/day of atorvastatin (69 mg/dl vs. 78 mg/dl; $p = 0.039$). The increase in fibrous cap thickness was significantly greater with 20 mg/day compared with 5 mg/day of atorvastatin (69% vs. 17%; $p < 0.001$). The increase in fibrous cap thickness correlated with the decrease in serum levels of low-density lipoprotein cholesterol ($R = -0.450$; $p < 0.001$), malondialdehyde-modified low-density lipoprotein ($R = -0.283$; $p = 0.029$), high-sensitivity C-reactive protein ($R = -0.276$; $p = 0.033$), and matrix metalloproteinase-9 ($R = -0.502$; $p < 0.001$), and the decrease in grade of OCT-derived macrophages ($R = -0.415$; $p = 0.003$).

CONCLUSIONS Atorvastatin therapy at 20 mg/day provided a greater increase in fibrous cap thickness in coronary plaques compared with 5 mg/day of atorvastatin. The increase of fibrous cap was associated with the decrease in serum atherogenic lipoproteins and inflammatory biomarkers during atorvastatin therapy. (Effect of Atorvastatin Therapy on Fibrous Cap Thickness in Coronary Atherosclerotic Plaque as Assessed by Optical Coherence Tomography: The EASY-FIT Study; NCT00700037) (J Am Coll Cardiol 2014;64:2207–17) © 2014 by the American College of Cardiology Foundation.

Statins reduce circulating atherogenic lipoproteins and inflammatory biomarkers as well as cardiovascular morbidity and mortality (1-3). Clinical trials have shown that more intensive statin therapy provides incremental benefits beyond those of lower intensity statin therapy in the secondary prevention of acute coronary events (1-3).

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ABBREVIATIONS AND ACRONYMS

HDL-C = high-density lipoprotein cholesterol

hs-CRP = high-sensitivity C-reactive protein

ICC = intraclass correlation coefficient

IL = interleukin

IQR = interquartile range

IVUS = intravascular ultrasound

LDL-C = low-density lipoprotein cholesterol

MDA-LDL = malondialdehyde-modified low-density lipoprotein

MI = myocardial infarction

MMP = matrix metalloproteinase

OCT = optical coherence tomography

PCI = percutaneous coronary intervention

TCFA = thin-cap fibroatheroma

Intravascular imaging might provide useful approaches to assess the effects of statin therapy on coronary atherosclerosis. Intravascular ultrasound (IVUS) studies have revealed that statin therapy attenuates atherosclerosis progression (4–6). Angioscopy studies have demonstrated that statins decreased plaque yellow score, which is important given that coronary plaque color and morphology might reflect the lipid content of coronary plaques (7). However, the detailed mechanism of plaque stabilization by statin therapy is not fully understood.

Over the past decade, atorvastatin has been the most widely used and extensively studied statin. Using IVUS, the REVERSAL (Reversal of Atherosclerosis With Aggressive Lipid Lowering) trial demonstrated that high-intensity statin therapy (atorvastatin 80 mg/day) reduced serum low-density lipoprotein cholesterol (LDL-C) on average by 79 mg/dl (change from baseline: −46%) and decreased coronary plaque volume (4). In Japan, IVUS showed that moderate intensity statin therapy (atorvastatin 20 mg/day) also reduced serum LDL-C level on average by 70 mg/dl (change from baseline: −42%) and reversed the process of coronary atherosclerosis (5). Although the daily dose of statin might be a major factor for predicting the intensity of lipid-lowering therapy, drug effect could also be affected by the patient's body weight. Because Asians have lighter body weight, in general, than Caucasians, the effective statin dose for lipid lowering is thought to be lower in Asians. Consequently, 5 to 10 mg/day of atorvastatin is the standard dose in Japan and atorvastatin 20 mg/day the approved highest dose (6).

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Intravascular optical coherence tomography (OCT) is a high-resolution imaging technique for plaque characterization (8). OCT allows us to measure fibrous cap thickness, thought to be a major factor in plaque vulnerability (8). Recently, we conducted retrospective, nonrandomized OCT studies to demonstrate the increase in fibrous cap thickness with statin therapy (9,10). Therefore, we designed a prospective, randomized OCT study in Japan to assess the effect of 20 mg/day versus 5 mg/day of atorvastatin on fibrous cap thickness in coronary atherosclerotic plaques.

METHODS

STUDY DESIGN. The EASY-FIT (Effect of AtorvaStatin therapY on FIbrous cap Thickness in coronary atherosclerotic plaque as assessed by optical coherence tomography) study is a prospective, randomized, open-label, blind-endpoint evaluation, parallel-group, dual-center (Wakayama Medical University, Wakayama, Japan, and Social Insurance Kinan Hospital, Tanabe, Japan) study using OCT to compare change of fibrous cap thickness in coronary atherosclerotic plaque associated with 20 mg/day versus 5 mg/day of atorvastatin for 12 months. We performed an OCT examination in patients with successful percutaneous coronary intervention (PCI) for unstable angina pectoris and untreated dyslipidemia. The target lesion for this OCT examination was an intermediate nonculprit lesion in the PCI-treated coronary artery. Within 24 h after baseline OCT examination, patients were randomly assigned to 20 mg/day or 5 mg/day of atorvastatin therapy. Randomization was stratified by sex, presence of diabetes mellitus, and total cholesterol level via a Web response system. Clinical follow-up visits to assess adverse events were scheduled monthly for up to 12 months. Blood samples for lipid levels and inflammatory markers were collected at baseline and 12-month follow-up, with OCT also performed at both time points. The present study was approved by the institutional review board at each participating center, and all patients gave written informed consent before enrollment.

PATIENT POPULATION. Eligible patients were those who had the following: 1) successful PCI for unstable angina pectoris; 2) intermediate nonculprit lesion in the PCI-treated coronary artery; and 3) untreated dyslipidemia. Unstable angina pectoris was defined as new-onset (<2 months) exertional angina, recent (<2 months) acceleration of angina, or angina at rest (lasting >20 min) coinciding with objective evidence of coronary angiography (a >70% coronary stenosis but without significant elevation in troponin T [>0.1 ng/ml]). The intermediate nonculprit lesion had diameter stenosis percentage of 30% to 70% by visual estimation on angiogram and was located >10 mm from the PCI-treated lesion. When ≥2 intermediate nonculprit lesions were recognized in the PCI-treated coronary artery, the most severely stenotic lesion was selected for OCT examination. Dyslipidemia was defined as serum LDL-C levels >100 mg/dl. Exclusion criteria were secondary unstable angina pectoris, post-infarction angina, acute myocardial infarction (MI), left main coronary artery disease, recommended coronary artery bypass grafting, cardiogenic shock, renal insufficiency with a serum creatinine level >2.0 mg/dl, active systemic inflammation, and current use of any lipid-lowering therapy.

OCT IMAGE ACQUISITION. A time-domain OCT system (Model M2 Cardiology Imaging System, St. Jude Medical, St. Paul, Minnesota) was used, and images were acquired as previously reported (11). Briefly, a 0.016-inch OCT catheter was advanced to the distal end of the target lesion through a 3-F occlusion balloon catheter. To remove blood from the field of view, an occlusion balloon was inflated to 0.4 to 0.6 atm at the proximal site of the target lesion and lactated Ringer's solution was infused into the coronary artery from the distal tip of the occlusion balloon catheter at 0.5 ml/s. To evaluate the target lesion in proximal coronary arteries, a continuous-flushing (nonocclusive) technique of OCT imaging was used. To flush the vessel, a mixture of commercially available dextran 40 and lactated Ringer's solution was infused directly from the guiding catheter at a rate of 2.5 to 4.5 ml/s with an injector pump. Regardless of the OCT imaging technique used, in all cases, the whole target lesion was imaged with an automatic pullback device traveling at 1 mm/s. The OCT images were digitally stored for offline analysis.

OCT IMAGE ANALYSIS. The images obtained were analyzed in a blinded fashion using a dedicated off-line review system (St. Jude Medical) at the core laboratory (Wakayama Medical University). Serial OCT images at baseline and 12-month follow-up were reviewed side by side on the screen, and target lesions were matched based on the distance from landmarks, such as branches, calcifications, and stents. The Z offset was adjusted before the OCT analysis, which was performed in the segments causing a discrete focal narrowing of the lumen within the target lesions. Plaque characterization was performed by using previously validated criteria (8). The lipid core was characterized by a diffusely bordered, signal-poor region.

The fibrous cap was identified as a signal-rich band overlying the lipid core. By visual screening for all contiguous frames, 3 candidate frames were selected to measure minimum fibrous cap thickness based on the smallest fibrous cap thickness in the candidate frames. We compared minimum fibrous cap thickness at baseline with that at follow-up. Intraobserver and interobserver reproducibility of this method was assessed in 30 randomly selected plaques. The intraclass correlation coefficient (ICC) for the repeated measurements of fibrous cap thickness by the same observer was excellent (ICC [1,1] = 0.930), with an absolute difference of 14 ± 9 μm. Similarly, the ICC for measuring fibrous cap thickness by 2 different observers was good (ICC [2,1] = 0.897), with an absolute

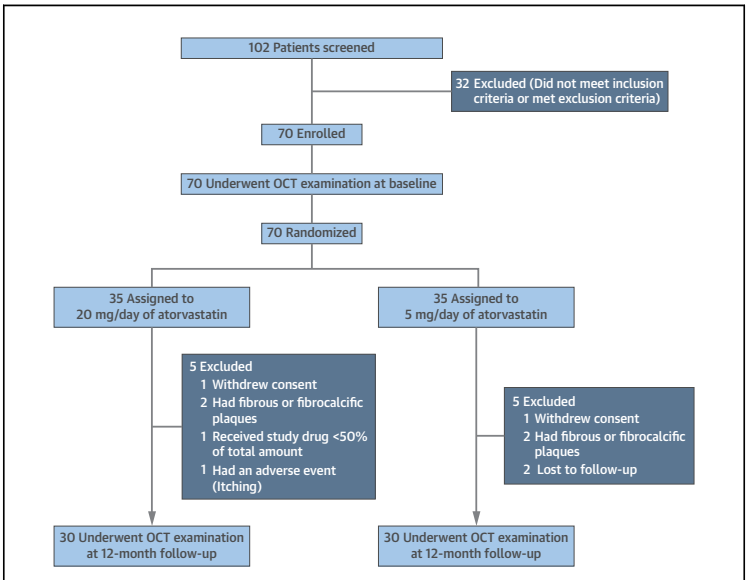


FIGURE 1 Disposition of Patients

After screening, eligibility, randomization, and later exclusion, 30 patients receiving 20 mg/day and 30 patients receiving 5 mg/day of atorvastatin who had successful optical coherence tomography (OCT) examinations at both baseline and 12-month follow-up constituted the final study population.

TABLE 1 Clinical Characteristics of Patients

	Atorvastatin 20 mg (n = 30)	Atorvastatin 5 mg (n = 30)	p Value
Age, yrs	63 (58–73)	69 (58–74)	0.446
Male	26 (87)	22 (73)	0.197
Diabetes mellitus	7 (23)	6 (20)	0.606
Hypertension	18 (60)	20 (67)	0.754
Current smoking	19 (63)	15 (50)	0.297
Family history of CAD	10 (33)	6 (20)	0.243
Target vessel			
LAD	14 (47)	10 (33)	0.274
LCX	8 (27)	6 (20)	
RCA	8 (27)	14 (47)	
Location of target plaque			
Proximal	14 (47)	16 (53)	0.776
Mid	13 (43)	13 (43)	
Distal	3 (10)	1 (3)	
Concomitant medications at follow-up			
Aspirin	30 (100)	30 (100)	1.000
Ticlopidine	1 (3)	1 (3)	1.000
Clopidogrel	21 (70)	25 (83)	0.360
Beta-blocker	20 (67)	18 (60)	0.789
ACE inhibitor or ARB	26 (87)	27 (90)	1.000
Calcium channel blocker	4 (13)	3 (10)	1.000
Oral hypoglycemic agents	5 (17)	3 (10)	0.353
Insulin	1 (3)	2 (7)	0.500

Values are median (interquartile range) or n (%).

ACE = angiotensin-converting enzyme; ARB = angiotensin II receptor blocker; CAD = coronary artery disease; LAD = left anterior descending coronary artery; LCX = left circumflex coronary artery; RCA = right coronary artery.

TABLE 2 Laboratory Results

	Atorvastatin 20 mg (n = 30)				Atorvastatin 5 mg (n = 30)			
	Baseline	Follow-Up	Nominal Change	p Value Compared With Baseline	Baseline	Follow-Up	Nominal Change	p Value Compared With Baseline
Total cholesterol, mg/dl	194 (176 to 220)	143 (128 to 156)	−51 (−73 to −39)	<0.001	195 (176 to 216)	159 (130 to 195)	−42 (−61 to −13)	<0.001
LDL cholesterol, mg/dl	127 (111 to 155)	69 (61 to 80)*	−58 (−74 to −45)*	<0.001	117 (110 to 138)	78 (66 to 108)	−44 (−63 to −29)	<0.001
MDA-LDL, U/l	119 (93 to 143)	87 (70 to 109)	−32 (−53 to 16)	0.026	102 (81 to 141)	99 (70 to 117)	−14 (−42 to 19)	0.206
HDL cholesterol, mg/dl	43 (34 to 49)	45 (36 to 49)	1 (−4 to 5)	0.310	42 (34 to 53)	43 (35 to 54)	3 (−4 to 6)	0.367
Triglyceride, mg/dl	109 (86 to 176)	96 (78 to 132)	−18 (−50 to 23)	0.211	138 (85 to 169)	128 (96 to 154)	−11 (−38 to 43)	0.918
hs-CRP, mg/l	2.1 (1.0 to 5.3)	0.5 (0.2 to 1.4)	−0.8 (−4.4 to −0.3)	<0.001	1.4 (0.9 to 5.1)	0.5 (0.3 to 1.1)	−0.8 (−3.9 to −0.3)	<0.001
IL-6, pg/ml	10.8 (7.7 to 20.1)	2.4 (1.5 to 5.7)	−8.1 (−16.7 to −3.4)	<0.001	9.3 (4.8 to 13.5)	2.4 (1.8 to 3.5)	−5.0 (−10.0 to −2.0)	<0.001
MMP-9, ng/ml	44 (34 to 53)	24 (18 to 30)*	−16 (−29 to −6)*	<0.001	33 (29 to 48)	32 (24 to 39)	−4 (−10 to 3)	0.023
HbA_{1c}, %	5.8 (5.3 to 7.2)	5.7 (5.3 to 6.5)	−0.1 (−0.4 to 0.1)	0.024	5.8 (5.3 to 7.2)	5.8 (5.2 to 6.3)	−0.1 (−0.3 to 0.2)	0.387

Values are median (interquartile range). *p < 0.05 versus 5 mg of atorvastatin. There was no significant difference in baseline laboratory results between the groups receiving 20 mg and 5 mg of atorvastatin.

HbA_{1c} = glycosylated hemoglobin; HDL = high-density lipoprotein; hs-CRP = high-sensitivity C-reactive protein; IL = interleukin; LDL = low-density lipoprotein; MDA-LDL = malondialdehyde-modified low-density lipoprotein; MMP = matrix metalloproteinase.

difference of 22 ± 11 μm. Thin-cap fibroatheroma (TCFA) was defined as a plaque with a minimal fibrous cap thickness <65 μm; thick-cap fibroatheroma was a plaque with a minimal fibrous cap thickness ≥65 μm.

The lipid arc was measured on the frame with the largest lipid core by visual screening. Lipid length was calculated from the number of frames with lipid core. Fibrous or fibrocalcific plaques, which lack a fibrous cap and lipid core, were excluded from analysis.

Macrophages were assessed using the previously reported technique (12). In short, macrophage accumulation was defined as high-intensity, signal-rich linear regions with sharp attenuation. Signals of macrophages were semiquantitatively graded as follows: grade 0, no macrophage; grade 1, localized macrophage accumulation; grade 2, clustered accumulation <1 quadrant; grade 3, clustered accumulation ≥1 quadrant but <3 quadrants; and grade 4, clustered accumulation ≥3 quadrants. Grading was performed in every 10 frames (i.e., every 0.66 mm) along the entire target lesion, and the summation of 0 to 4 grades was calculated. Reproducibility of measurements for calculating the macrophage grade was good (ICC [1,1] = 0.948, ICC [2,1] = 0.892), with absolute differences of 1.3 ± 1.0 and 2.2 ± 1.0, respectively. The minimal fibrous cap thickness, lipid arc, lipid length, and minimal lumen area were prespecified OCT parameters, but macrophage grade was not.

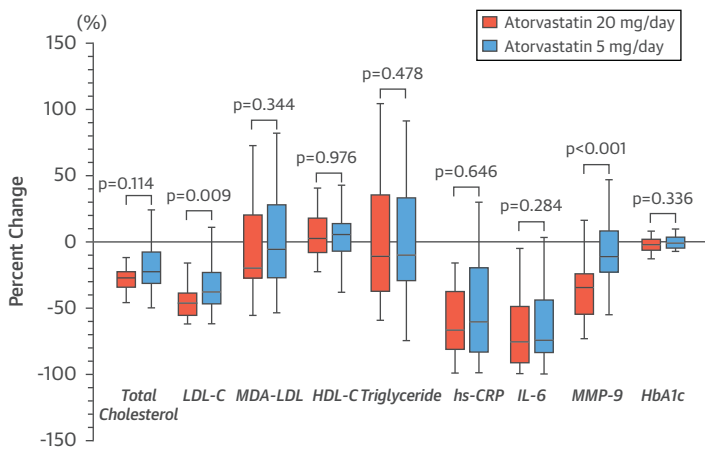


FIGURE 2 Percentage of Change in Laboratory Results Between Baseline and 12-Month Follow-Up

Although the percentages of decreases in serum LDL-C and MMP-9 levels during the follow-up period were significantly greater in the 20 mg/day than 5 mg/day of atorvastatin group, the percentage of changes in other markers were comparable between the 2 groups. Data are represented as boxplot with medians and 25th to 75th percentiles **(boxes)** and 10th to 90th percentiles **(whiskers)**. HbA_{1c} = glycosylated hemoglobin; HDL-C = high-density lipoprotein cholesterol; LDL-C = low-density lipoprotein cholesterol; MDA-LDL = malondialdehyde-modified low-density lipoprotein; MMP = matrix metalloproteinase.

BLOOD SAMPLES. Blood samples were collected in the fasting state. Serum samples were separated by centrifugation, stored at 4°C, and then analyzed (SRL Co., Ltd., Tokyo, Japan). Serum total cholesterol, high-density lipoprotein cholesterol (HDL-C), triglyceride, and hemoglobin (HbA_{1c}) levels were measured by enzymatic methods. Serum LDL-C level was calculated using the Friedwald equation. Serum level of malondialdehyde-modified LDL (MDA-LDL), an oxidized LDL, was determined by an enzyme-linked immunosorbent assay (13). Serum levels of the inflammatory biomarkers high-sensitivity

TABLE 3 OCT Measurements

	Atorvastatin 20 mg (n = 30)				Atorvastatin 5 mg (n = 30)			
	Baseline	Follow-Up	Nominal Change	p Value Compared With Baseline	Baseline	Follow-Up	Nominal Change	p Value Compared With Baseline
Minimal fibrous cap thickness, μm	105 (86 to 141)	174 (144 to 235)*	73 (28 to 113)*	<0.001	117 (78 to 153)	132 (82 to 165)	19 (−1 to 48)	0.002
Lipid arc, degree	145 (120 to 220)	110 (80 to 145)*	−50 (−60 to −30)*	<0.001	140 (105 to 225)	125 (100 to 203)	−10 (−20 to −5)	<0.001
Lipid length, mm	9.4 (7.4 to 11.0)	8.8 (6.4 to 10.1)	−0.6 (−1.1 to −0.2)	<0.001	8.3 (6.3 to 10.8)	8.1 (5.5 to 10.2)	−0.4 (−1.1 to −0.1)	<0.001
Minimal lumen area, mm^2	5.60 (3.89 to 6.65)	5.23 (4.00 to 6.38)	−0.05 (−0.58 to 0.30)	0.256	5.52 (4.49 to 6.50)	4.68 (4.18 to 6.01)	−0.09 (−0.87 to 0.19)	0.101
Macrophage grade	12.0 (10.0 to 14.0)	7.0 (6.0 to 9.0)	−4.5 (−6.0 to −3.0)*	<0.001	11.5 (6.0 to 15.0)	8.0 (4.5 to 10.0)	−2.0 (−5.0 to 0.0)	<0.001

Values are median (interquartile range). *p < 0.05 versus 5 mg of atorvastatin. There was no significant difference in baseline optical coherence tomography measurements between the groups receiving 20 mg and 5 mg of atorvastatin.

OCT = optical coherence tomography.

C-reactive protein (hs-CRP), interleukin (IL)-6, and matrix metalloproteinase (MMP)-9 were measured by a latex particle-enhanced turbidimetric immunoassay, a sandwich enzyme-linked immunosorbent assay system, and a chemiluminescent enzyme immunoassay, respectively (6).

CLINICAL OUTCOMES. Major adverse cardiac events such as cardiac death, MI, repeat revascularization, coronary artery bypass grafting, and other adverse events, including adverse drug reactions, were reported.

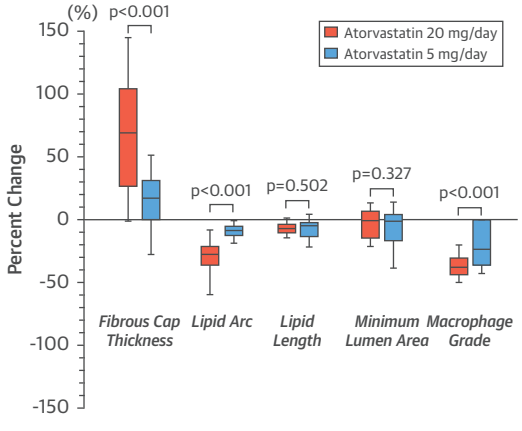


FIGURE 3 Percentage of Change in Optical Coherence Tomography Measurements Between Baseline and 12-Month Follow-Up

Percentage of increase in the fibrous cap thickness and percentage of decreases in the lipid arc and macrophage grade during the follow-up period were significantly greater in the group receiving 20 mg/day of atorvastatin. The percentage of changes in lipid length and minimal lumen area did not differ between the 2 groups.

STATISTICAL ANALYSIS. Sample size calculation was based on the assumption that the average difference in fibrous cap thickness between the groups receiving 20 mg/day of atorvastatin and 5 mg/day of atorvastatin is 100 μm, and the SD of fibrous cap thickness distribution for either group is 120 μm (9). With a 2-sided alpha level of 0.05 and a power of 80%, 23 patients were required in each group. To accommodate a maximal 15% for using nonparametric tests and 30% for possible missing investigations or withdrawals, sample size was increased to 35 patients per group.

Categorical variables were presented as frequencies, with comparison using chi-square or Fisher exact test (for an expected cell value <5). Continuous variables were presented as medians and interquartile ranges (IQRs) and compared using the Mann-Whitney *U* test (between-group comparison) or Wilcoxon signed rank test (if variables were compared between baseline and 12-month follow-up). The relationships between the percentage of changes in biomarkers and fibrous cap thickness during follow-up were investigated using a simple regression analysis. Computations were performed with SPSS version 20.0 (IBM, Armonk, New York). A p value <0.05 was considered statistically significant.

RESULTS

PATIENT POPULATION. Patient distribution is presented in Figure 1. Between August 2009 and February 2011, a total of 102 patients were screened; subsequently, 70 patients were enrolled, randomized, and received atorvastatin 20 mg/day or 5 mg/day. Of these, 60 patients (30 patients receiving 20 mg/day and 30 receiving 5 mg/day of atorvastatin) had

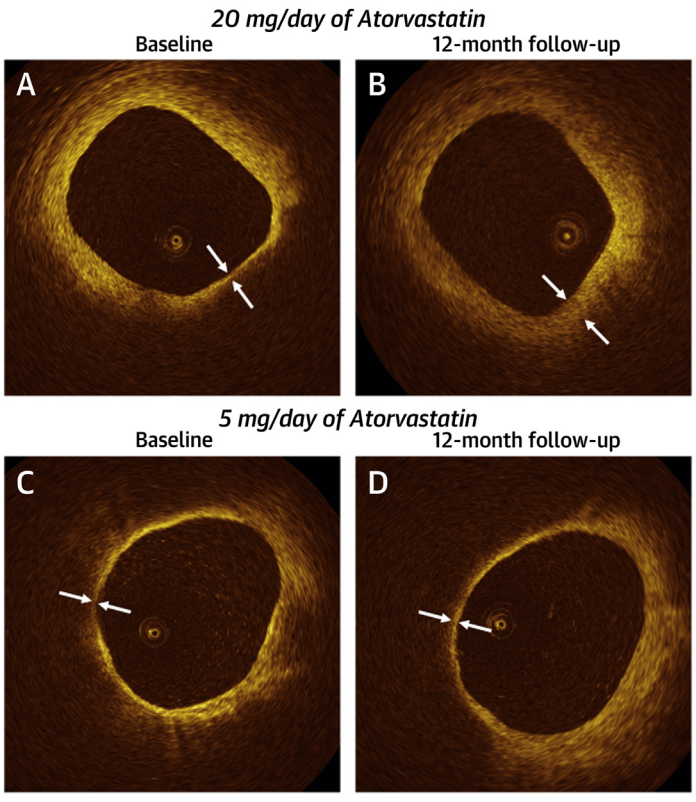


FIGURE 4 Representative Optical Coherence Tomography Images of Fibrous Cap Thickness Increase

The fibrous cap thickness dramatically increased from baseline **(A)** to 12-month follow-up **(B)** in a patient receiving 20 mg/day of atorvastatin, whereas the fibrous cap hardly changed from baseline **(C)** to 12-month follow-up **(D)** in a patient receiving 5 mg/day of atorvastatin. **Arrows** indicate fibrous cap thickness.

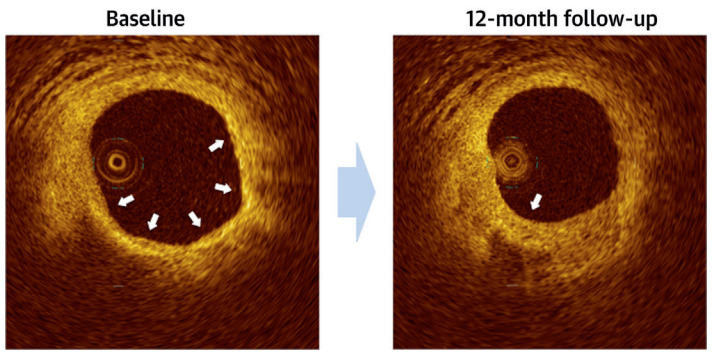


FIGURE 5 Representative Optical Coherence Tomography Images of Macrophage Signal Decrease

The macrophage signals decreased dramatically during follow-up in a patient receiving 20 mg/day of atorvastatin. **Arrows** indicate macrophage signals.

successful OCT examinations at both baseline and follow-up. Median follow-up time was 12.0 months (IQR: 11.6 to 12.2 months) in the group receiving 20 mg/day of atorvastatin and 11.9 months (IQR: 11.3 to 12.4 months) in the group receiving 5 mg/day of atorvastatin. Table 1 contains patient clinical characteristics. The 2 groups were well-matched at baseline, and their pattern of use of concomitant medications was similar.

LABORATORY RESULTS. Table 2 summarizes laboratory results at baseline and 12-month follow-up; Figure 2 shows the percentage of changes in laboratory results during follow-up. The serum LDL-C level decreased significantly in both groups. The percentage of decrease in serum LDL-C was significantly greater (−46% [IQR: −56% to −39%] vs. −38% [IQR: −47% to −23%]; $p = 0.009$) and serum LDL-C level at follow-up was significantly lower (69 mg/dl [IQR: 61 to 80 mg/dl] vs. 78 mg/dl [IQR: 66 to 108 mg/dl]; $p = 0.039$) in the group receiving 20 mg/day of atorvastatin as opposed to the group receiving 5 mg/day of atorvastatin. Serum MDA-LDL level decreased significantly in the 20 mg/day group (119 U/l [IQR: 93 to 143 U/l] to 87 U/l [IQR: 70 to 109 U/l]; $p = 0.026$) but not in the group receiving 5 mg/day of atorvastatin (102 U/l [IQR: 81 to 141 U/l] to 99 U/l [IQR: 70 to 117 U/l]; $p = 0.206$). Serum hs-CRP and IL-6 levels decreased comparably between the 2 groups. The serum MMP-9 level showed a more remarkable decrease in the group receiving atorvastatin 20 mg/day (−35% [IQR: −56% to −25%] vs. −12% [IQR: −23% to 9%]; $p < 0.001$).

OCT FINDINGS. Table 3 summarizes OCT measurements at baseline and 12-month follow-up; Figure 3 shows the percentage of changes in OCT measurements during follow-up. The analyzed lesion length was 12.2 mm (IQR: 9.7 to 13.6 mm) with 20 mg/day and 10.2 mm (IQR: 8.0 to 12.5 mm) with 5 mg/day of atorvastatin. Fibrous cap thickness increased significantly in both groups; the percentage of increase was significantly greater in the group receiving atorvastatin 20 mg/day (69% [IQR: 25% to 104%] vs. 17% [IQR: −1% to 34%]; $p < 0.001$).

Representative OCT images showing the increase of fibrous cap thickness by therapy are seen in Figure 4. The lipid arc decreased significantly in both groups with the 20 mg/day group demonstrating a significantly greater percentage of decrease (−27% [IQR: −37% to −20%] vs. −8% [IQR: −13% to −4%]; $p < 0.001$). Lipid length decreased comparably between the 2 groups. The minimal lumen area did not change during follow-up in either group. In the 20

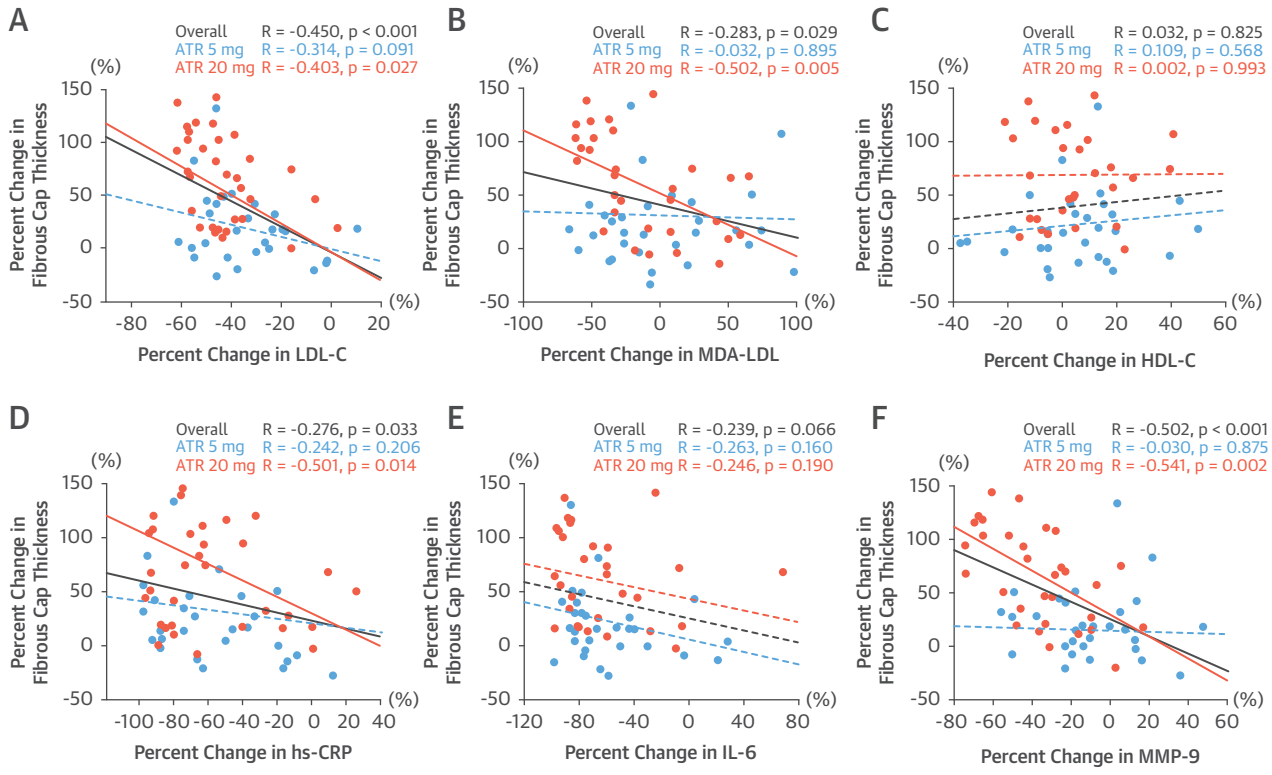


FIGURE 6 Relationships Between Percentage of Changes in Biomarkers and in Fibrous Cap Thickness

The correlations between serum LDL-C **(A)**, MDA-LDL **(B)**, HDL-C **(C)**, hs-CRP **(D)**, IL-6 **(E)**, and MMP-9 **(F)** levels and the percentage of change in fibrous cap thickness were analyzed in the group receiving atorvastatin 20 mg/day **(salmon)**, the group receiving atorvastatin 5 mg/day **(blue)**, and overall subjects **(black)**. ATR = atorvastatin; hs-CRP = high-sensitivity C-reactive protein; IL = interleukin; other abbreviations as in Figure 2.

mg/day group, there were 6 OCT-derived TCFAs at baseline, and all 6 evolved into thick-cap fibroatheroma at follow-up. In the 5 mg/day group, 2 (33%) OCT-derived TCFAs at baseline evolved into thick-cap fibroatheroma during follow-up and 4 (66%) remained unchanged. Neither group had newly evolved OCT-derived TCFA at follow-up. Macrophage accumulation was detected in 48 plaques (80%) at baseline: 25 plaques (83%) in the 20 mg/day group and 23 plaques (77%) in the 5 mg/day group ($p = 0.748$). There were no plaques newly acquiring or absolutely depleting signals of macrophages at follow-up. The macrophage grade decreased significantly in both groups, but the percentage of decrease in macrophage grade was significantly greater in the 20 mg/day group (−38% [IQR: −44% to −31%] vs. −24% [IQR: −33% to 0%]; $p < 0.001$). Representative OCT images showing the decrease in macrophage signals with atorvastatin therapy are shown in Figure 5.

FIBROUS CAP THICKNESS AND BIOMARKERS. The relationship between the percentage of changes in biomarkers and in fibrous cap thickness during follow-up can be seen in Figure 6. The percentage of change in the fibrous cap thickness was negatively correlated with the percentage of change in serum LDL-C ($R = −0.450$; $p < 0.001$), MDA-LDL ($R = −0.283$; $p = 0.029$), hs-CRP ($R = −0.276$; $p = 0.033$), and MMP-9 ($R = −0.502$; $p < 0.001$) levels but not with the percentage of change in the serum total cholesterol ($R = −0.238$; $p = 0.067$), HDL-C ($R = 0.032$; $p = 0.825$), triglyceride ($R = −0.059$; $p = 0.670$), IL-6 ($R = −0.239$; $p = 0.066$), and glycosylated hemoglobin ($R = −0.170$; $p = 0.195$) levels.

MACROPHAGE GRADE AND BIOMARKERS. The relationships between the percentage of changes in biomarkers and in macrophage grade are depicted in Figure 7. The percentage of change in macrophage grade was negatively correlated with the percentage

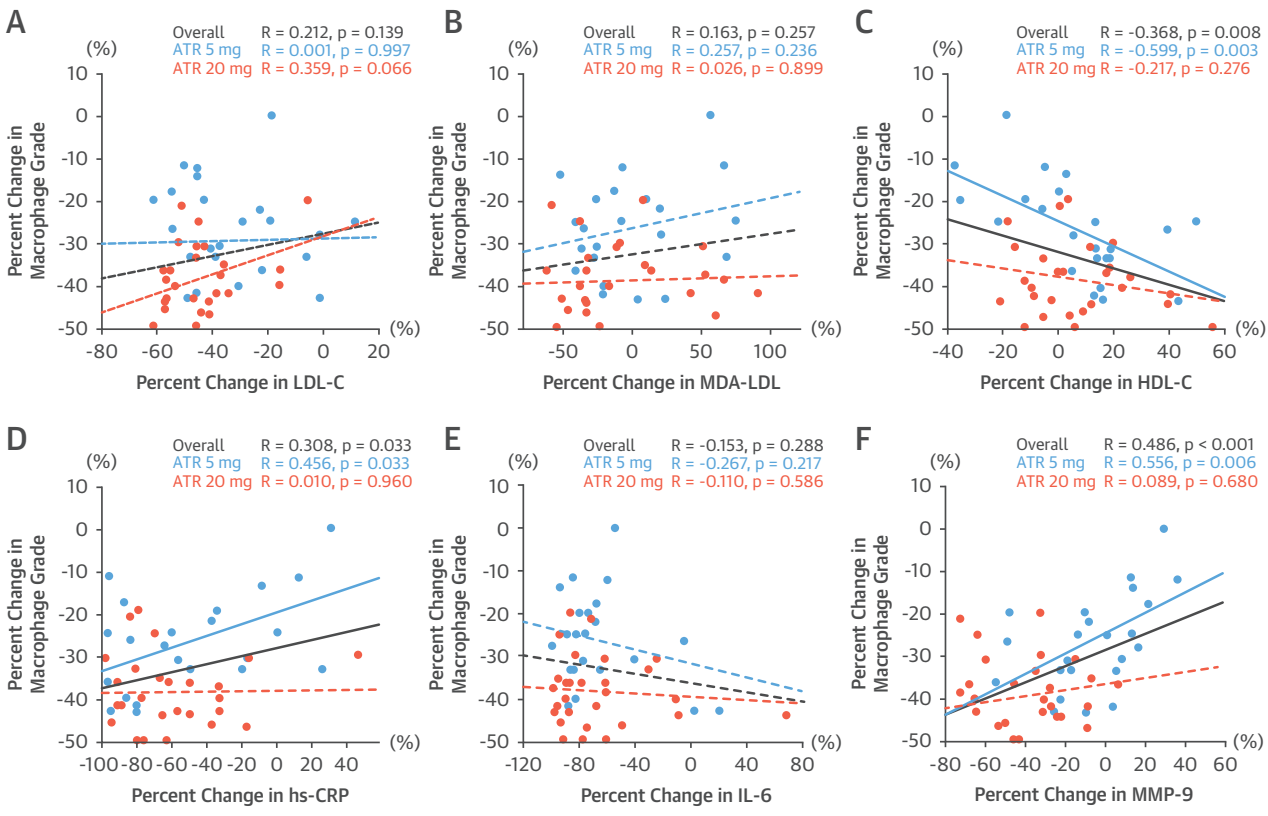


FIGURE 7 Relationships Between Percentage of Changes in Biomarkers and in Macrophage Grade

The correlations between serum LDL-C **(A)**, MDA-LDL **(B)**, HDL-C **(C)**, hs-CRP **(D)**, IL-6 **(E)**, and MMP-9 **(F)** levels and the percentage of change in macrophage grade were analyzed in the atorvastatin 20 mg/day group **(salmon)**, the atorvastatin 5 mg/day group **(blue)**, and overall subjects **(black)**. Abbreviations as in Figures 2 and 6.

of change in serum HDL-C levels (R = −0.368; p = 0.008) and positively correlated with the percentage of change in serum hs-CRP (R = 0.308; p = 0.033) and MMP-9 levels (R = 0.486; p < 0.001); however, it was not correlated with the percentage of change in serum total cholesterol (R = 0.038; p = 0.793), LDL-C (R = 0.212; p = 0.139), triglyceride (R = 0.070; p = 0.627), MDA-LDL (R = 0.163; p = 0.257), IL-6 (R = −0.153; p = 0.288), and glycosylated hemoglobin (R = 0.086; p = 0.551) levels. Additionally, the percentage of change in macrophage grade negatively correlated with the percentage of change in fibrous cap thickness (R = −0.415; p = 0.003) (Figure 8).

CLINICAL OUTCOMES. No patients experienced cardiac death or MI in either group. One patient receiving atorvastatin 20 mg/day underwent repeat revascularization due to in-stent restenosis and 1 patient on 5 mg/day of atorvastatin had coronary artery bypass grafting during follow-up.

DISCUSSION

Our main findings demonstrated that therapy with 20 mg/day of atorvastatin resulted in a greater increase in fibrous cap thickness and a decrease in the lipid arc and macrophage grade compared with 5 mg/day of atorvastatin, and the increase in fibrous cap thickness was associated with a decrease in serum LDL-C, MDA-LDL, hs-CRP, and MMP-9 levels (Central Illustration). The present study suggests that the therapy with 20 mg/day of atorvastatin might more reliably stabilize coronary atherosclerotic plaques compared with 5 mg/day.

CORONARY PLAQUE STABILIZATION WITH STATIN. TCFA is considered a precursor of plaque rupture, which accounts for most coronary thrombi (14). TCFA morphologic features include a large lipid core, a thin fibrous cap <65 μm, and numerous macrophages within the fibrous cap (14). Because catheter examination can be performed repeatedly over sequential

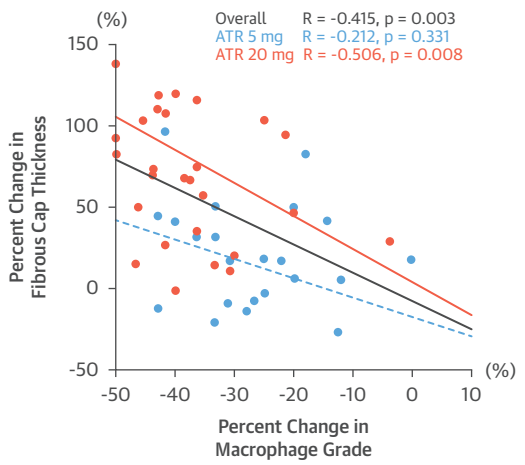


FIGURE 8 Relationship Between Percentage of Change in Macrophage Grade and in Fibrous Cap Thickness

The correlation of the percentage of change in macrophage grade with the percentage of changes in fibrous cap thickness was analyzed in the group receiving 20 mg/day of atorvastatin **(salmon)**, the group receiving 5 mg/day of atorvastatin **(blue)**, and overall subjects **(black)**. Target lesions without macrophage accumulation at baseline were excluded from the analysis. ATR = atorvastatin.

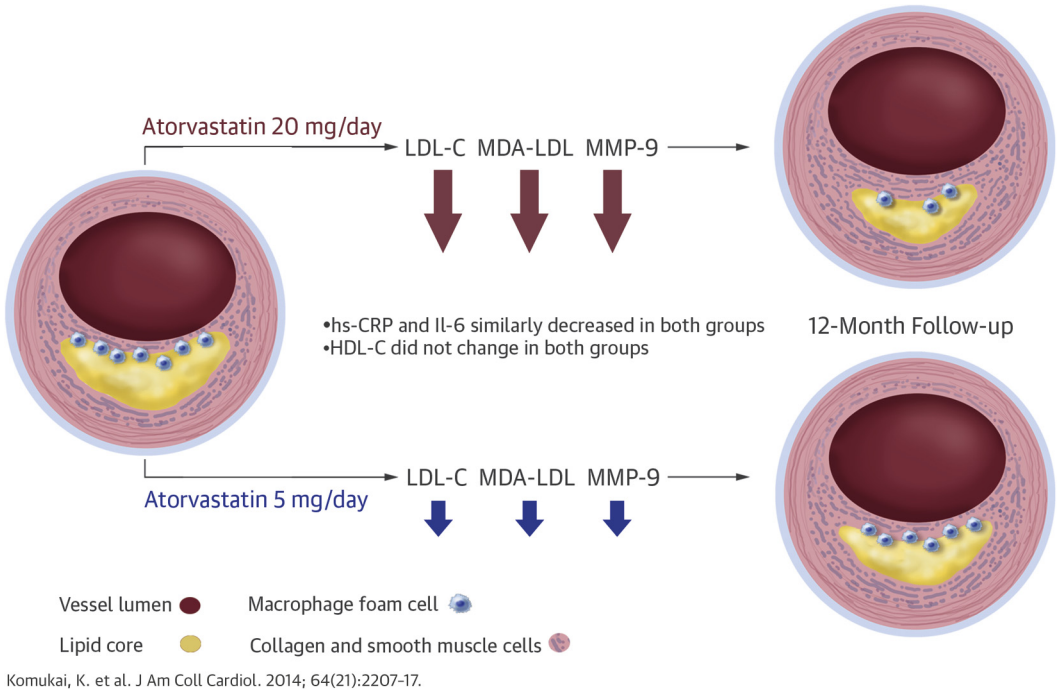
time points, intravascular imaging seems to be an optimal means of assessing serial changes in coronary plaque vulnerability and elucidating plaque-stabilizing mechanisms of lipid-lowering therapy with statins. Several IVUS studies have correlated coronary plaque volume reduction with the decrease in serum LDL levels with statin therapy (4–6). Recent angioscopy studies showed a statin-induced reduction of yellow color intensity of plaques, presumably reflecting a decrease in the plaques' lipid content (7). The present OCT study demonstrated an increase in the thickness of fibrous caps overlying lipid cores with statin therapy. Treatment with atorvastatin seemed to stabilize vulnerable plaques by both increasing fibrous cap thickness and decreasing the amount of atherosclerotic plaque.

INTENSITY OF STATIN THERAPY AND PLAQUE STABILIZATION. Many studies have investigated the effect of high-intensity statin therapy on coronary atherosclerosis. The TNT (Treating to New Targets) trial demonstrated that high-dose atorvastatin therapy (80 mg/day) versus moderate-intensity atorvastatin therapy (10 mg/day) reduced the risk of cardiovascular events by 22% during 4.9 years of follow-up (3). The REVERSAL used gray-scale IVUS and showed a significantly lower progression of plaque volume after 18 months of 80 mg/day of atorvastatin versus 40 mg/day of pravastatin therapy (4). A trial performing serial virtual histology-IVUS examinations at baseline and 12-month follow-up revealed a significant decrease in necrotic core volume with high-dose rosuvastatin (40 mg/day) compared with low-intensity rosuvastatin therapy (5 mg/day) (15). Furthermore, the ESTABLISH (Early Statin Treatment in Patients With Acute Coronary Syndrome) and the JAPAN-ACS (Japan Assessment of Pitavastatin and Atorvastatin in Acute Coronary Syndrome) trials showed that moderate-intensity atorvastatin (20 mg/day) therapy reduced plaque volume in patients with acute coronary syndrome (5,6).

Our study disclosed a greater increase of fibrous cap thickness after 12 months in the moderate-intensity atorvastatin (20 mg/day) therapy compared with low-intensity atorvastatin (5 mg/day) therapy. Overall, these findings provide strong evidence that high- and moderate-intensity statin therapy might provide greater benefit for plaque stabilization compared with a lower dose regimen.

FIBROUS CAP THICKENING AND BIOMARKERS. Vascular inflammation plays an important role in vulnerable plaque pathogenesis (16). Various biomarkers have been studied as candidates for monitoring plaque vulnerability. Induced by IL-6, the acute-phase protein CRP has predictive abilities for acute coronary events. MMP-9 is produced by activated macrophages and induces collagen breakdown in the fibrous cap. Oxidized LDL stimulates MMP-9 expression in human vascular endothelial cells and initiates the CD40/CD40L signaling pathway, resulting in atherosclerotic plaque development and prothrombotic actions. Previous studies have reported that statins could reduce inflammatory markers in patients with coronary artery disease (17). In the present study, the serum hs-CRP and IL-6 levels decreased with atorvastatin regardless of dose, but serum MMP-9 and MDA-LDL levels showed a greater decrease with higher intensity atorvastatin therapy. Additionally, the decreases in the serum hs-CRP, MMP-9, and MDA-LDL levels correlated with increased fibrous cap thickness. Our results suggest that statin therapy could induce fibrous cap thickening and stabilize coronary plaques through its anti-inflammatory properties.

STUDY LIMITATIONS. First, we used 5 mg/day of atorvastatin for lipid-lowering therapy. However, an Asian population receiving 5 mg/day is not critically undertreated as evidenced by the average 33%



CENTRAL ILLUSTRATION Vascular Responses to Different Atorvastatin Doses

Therapy with 20 mg/day of atorvastatin yielded significant decreases in serum LDL-C, MDA-LDL, and MMP-9 levels; a greater increase in fibrous cap thickness and decrease in lipid core; and a significant reduction of macrophage accumulation compared with therapy with 5 mg/day of atorvastatin in patients with unstable angina pectoris during 12-month follow-up. HDL-C = high-density lipoprotein cholesterol; hs-CRP = high-sensitivity C-reactive protein; IL = interleukin; LDL-C = low-density lipoprotein cholesterol; MDA-LDL = malondialdehyde-modified low-density lipoprotein; MMP = matrix metalloproteinase.

decrease in serum LDL-C in this study. Also, various factors in acute coronary syndrome may increase serum levels of inflammatory markers. Although the present study included patients with unstable angina pectoris, it did exclude patients with acute MI to eliminate any influence of myocardial necrosis on serum inflammatory markers.

This study did not use recently developed frequency-domain OCT, but rather a conventional time-domain OCT. However, it is unlikely that fibrous cap thickness measurement would be affected. Still, the complicated imaging procedure of time-domain OCT limited observation to a single plaque in the PCI-treated vessel. The simple imaging procedure and long scanning length of frequency-domain OCT would allow us to evaluate multiple plaques in the entire coronary tree. Also, the relatively shallow penetration depth of OCT does not allow quantitative assessment of lipid core size. Other techniques may be helpful for assessing the effect of statin on the lipid core size in coronary atherosclerotic plaques. Another issue is that statin-induced modification of the lipid core underlying a fibrous cap may obscure the delineation of the border between those tissues and thus decrease the accuracy of OCT measurements of fibrous cap thickness. Yet there was no plaque excluded from OCT analysis due to the inability to measure fibrous cap thickness, even at follow-up. Despite the data presented here, the direct relationship between increased fibrous cap thickness and risk reduction of coronary events remains unknown. Further studies are needed to elucidate the clinical implications of evidence of drug benefit derived from OCT.

Image artifacts inherent to OCT can lead to misclassification of plaque pathology. If the imaging beam strikes the tissue at a glancing angle, OCT images can appear like TCFA or macrophages on the coronary plaque surface. The present study assessed circumferential, but not axial, distribution of macrophages, both of which might contribute to plaque vulnerability (18). Finally, some caution might be required for interpreting correlation analyses because the groups receiving 20 mg/day and

5 mg/day of atorvastatin had mean differences in many of the variables, which may produce Simpson's paradox.

CONCLUSIONS

Therapy with 20 mg/day of atorvastatin for 12 months led to a greater increase of fibrous cap thickness in coronary atherosclerotic plaque compared with 5 mg/day. The increase in fibrous cap thickness was associated with a decrease in serum atherogenic lipoproteins and inflammatory biomarkers during atorvastatin therapy.

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PERSPECTIVES

COMPETENCY IN MEDICAL KNOWLEDGE 1: More intensive statin therapy reduces cardiovascular morbidity and mortality. However, the mechanism of the dose-dependent effect on plaque stabilization by statin therapy is not fully elucidated.

COMPETENCY IN MEDICAL KNOWLEDGE 2: Micron resolution of optical coherence tomography allows detailed assessment of fibrous cap thickness, which contributes to plaque instability.

COMPETENCY IN PATIENT CARE: Increase in fibrous cap thickness in coronary atherosclerotic plaque benefits from higher dose atorvastatin compared with a lower regimen in patients with unstable angina pectoris.

TRANSLATIONAL OUTLOOK: Larger and longer term studies are needed to determine the direct relationship between an increase in fibrous cap thickness and coronary event risk reduction.

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