

Effect of Atorvastatin on Regional Arterial Stiffness in Patients with Type 2 Diabetes Mellitus

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Objective: A statin, a potent lipid-lowering drug, improves pain-free walking distance in patients with peripheral arterial disease (PAD) without increasing the ankle-brachial pressure index (ABI). Arterial stiffness affects the blood flow of peripheral arteries. The purpose of this study was to evaluate the effect of cholesterol-lowering with atorvastatin on regional arterial stiffness in patients with type 2 diabetes mellitus.

Methods: The subjects were 22 type 2 diabetic patients with hypercholesterolemia, who received atorvastatin at a daily dose of 10 mg for 6 months. Before and after the treatment with atorvastatin, we measured pulse wave velocity (PWV) in the heart-brachial, heart-carotid, heart-femoral and femoral-ankle segments.

Results: Following treatment with atorvastatin, femoral-ankle PWV showed a significant reduction. The PWV of other arterial segments tended to decrease, although the changes were not statistically significant. We found no significant changes in blood pressure, heart rate, ABI, or plasma concentrations of glucose, L-arginine and asymmetric dimethylarginine (ADMA), an endogenous inhibitor of endothelial function.

Conclusions: Atorvastatin treatment was associated with an improvement in the stiffness of leg arteries in type 2 diabetes mellitus. This may partly explain the statin-mediated improvement of walking performance in those with PAD. *J Atheroscler Thromb, 2005; 12: 205-210.*

Key word: Pulse wave velocity (PWV)

Introduction

Peripheral arterial disease (PAD) is one of the major manifestations of diabetic macroangiopathy. The prevalence of PAD is higher in patients with diabetes mellitus than the general population (1-3). The presence of PAD predicts a higher risk of amputation (4) in type 2 diabetes mellitus. Mortality risk is particularly increased in diabetic patients with PAD (5). Strict lipid-lowering is rec-

ommended for those with PAD, because such patients often have coronary artery disease and are at an increased risk of mortality. Interestingly, some studies reported that lipid reduction with statins improves claudication, a symptom of peripheral arterial disease. In a post-hoc analysis of the Scandinavian Simvastatin Survival Study (4S) data (6), new or worsening of claudication was reduced in the group receiving simvastatin. Another study reported a similar beneficial effect of simvastatin on walking time in patients with PAD (7). Recently, Mohler et al (8) clearly showed that 12 months of atorvastatin therapy at a daily dose of 80 mg improved pain-free walking distance due to PAD in a study including 354 patients with claudication. In that study, treatment with atorvastatin did not increase ankle-brachial pressure index (ABI), an index for the presence and the severity of PAD in lower

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extremities. These studies suggest that statins improve the peripheral circulation by some mechanism other than the improvement of arterial stenosis.

Arterial stiffness is an important factor regulating blood flow. The contraction of the left ventricle and ejection of blood dilate the aorta and its major branches. The recoil of the large arteries generates blood flow during the diastole, and the large arteries are called "the second heart". Stiffening of the aorta impairs coronary perfusion, and reflects an increased risk of cardiovascular mortality in high-risk populations (9–12) including diabetes mellitus (13). We and others have shown that stiffness of the lower-limb arteries is associated with reduced diastolic blood flow of the popliteal artery (14), exercise-induced foot ischemia (15), and ischemic symptoms due to PAD (16) in patients with type 2 diabetes mellitus.

It is possible that statin treatment improves stiffness of the lower-limb arteries. So far, however, only a few studies (17, 18) have examined this possibility in nondiabetic individuals, and no such study in diabetic patients is available in the literature. In the present study, we examined the possible change in stiffness of the lower-limb arteries following treatment with atorvastatin in type 2 diabetes patients in comparison with the stiffness in other parts of the arterial tree.

Methods

Subjects

The subjects were 22 (14 men and 8 women) consecutive outpatients with type 2 diabetes mellitus who visited the Diabetes Center at the Osaka City University Hospital for an evaluation of dyslipidemia and vascular complications and started receiving treatment with atorvastatin. Their mean age was 61.7 ± 7.2 years, and their known duration of diabetes was 8.1 ± 5.4 years. Fifty-five percent of the patients were smokers. Table 1 gives other characteristics of the subjects. We excluded those with renal dysfunction, overt proteinuria, a history of cardiovascular disease or any signs of peripheral artery disease. The diagnosis of diabetes was made according to the American Diabetes Association criteria (19). Treatment for diabetes was with insulin ($n = 5$), sulfonylureas ($n = 7$), α -glucosidase inhibitors ($n = 3$), nateglinide ($n = 1$) and life-style modification alone ($n = 9$). Some patients received a combination of medications. Anti-hypertensive medications were calcium channel blockers ($n = 5$), angiotensin converting enzyme inhibitors ($n = 6$), β blockers ($n = 1$), and angiotensin 2 antagonists ($n = 1$). All the subjects gave written informed consent before participating in this study. After signing a consent form, patients received 10 mg of atorvastatin daily and were followed up for 6 months. They were asked not to change their medications, or eating, smoking, and exercise habits during the study.

Pulse wave velocity and blood pressure measurements

PWV and blood pressure were measured supine after 5 min bed rest using an automatic waveform analyzer BP-203RPE (Colin, Komaki, Japan) as previously described (20). Briefly, pressure waveforms of the brachial and tibial arteries were recorded by an oscillometric method using cuffs adapted to both arms and ankles, while those of the carotid and femoral arteries were recorded using multi-element tonometric sensors placed at the left carotid and femoral arteries. The electrocardiogram was monitored with electrodes placed on the both wrists. Heart sounds S1 and S2 were detected by a microphone set on the left edge of the sternum at the third intercostals space. The waveform analyzer measures time intervals between S2 and the notch of the carotid pulse wave (Thc), between S2 and the notch of the brachial pulse wave (Thb), between pulse waves of the carotid and femoral arteries (Tcf), and between pulse waves of the femoral and tibial arteries (Tfa). The sum of Thc and Tcf gives the time taken for pulse waves to travel from the heart to the femoral artery (Thf). Also, the path lengths of these segments were estimated using formulas described previously (20). The waveform analyzer reported averaged values during 30 seconds of stable measurements.

The coefficients of variation (CV) were 6.0%, 3.3%, 4.9% and 3.3% each for the heart-carotid, heart-brachial, heart-femoral, and femoral-ankle PWV, respectively, based on repeated measurements on two different occasions in 17 healthy subjects (20).

Blood sampling and biochemical assays

Blood was drawn in the morning after an overnight fast for at least 12 hours to measure glucose, insulin, total cholesterol, triglyceride, high-density lipoprotein (HDL) cholesterol, and HbA_{1c} levels. Among the insulin users, the evening dose of insulin was discontinued prior to the study. These measurements were made using routine laboratory methods. Non-HDL-cholesterol was calculated by subtracting HDL-cholesterol from total cholesterol. LDL-cholesterol was calculated with the Friedewald formula. Fasting serum arginine and ADMA concentrations were measured by high-performance liquid chromatography (HPLC) using the method of Boger *et al.* (21).

Statistics

Data are summarized as the mean \pm standard deviation (SD). The difference between means before and after the medication was assessed with the repeated measure analysis of variance (ANOVA). p values < 0.05 were considered statistically significant. These statistical analyses were performed using statistics software (StatView version 5.0, SAS Institute, Cary, NC).

Table 1. Metabolic parameters before and after lipid reduction with atorvastatin.

	Before	After 6 months	<i>p</i> -value
Body mass index (kg/m ²)	24.5 ± 2.9	24.5 ± 2.9	0.805
Fasting plasma glucose (mmol/l)	7.89 ± 1.56	8.00 ± 1.89	0.815
Hemoglobin A _{1c} (%)	7.4 ± 1.1	7.5 ± 1.0	0.499
HOMA-IR	2.88 ± 2.60	3.45 ± 4.81	0.471
Total cholesterol (mmol/l)	6.56 ± 0.88	4.68 ± 0.78	< 0.0001
Non-HDL cholesterol (mmol/l)	4.96 ± 0.85	3.10 ± 0.72	< 0.0001
LDL-cholesterol (mmol/l)	4.13 ± 0.65	2.56 ± 0.80	< 0.0001
HDL-cholesterol (mmol/l)	1.60 ± 0.74	1.60 ± 0.36	0.953
Triglycerides (mmol/l)	1.78 ± 0.99	1.41 ± 0.59	0.028
ADMA (μmol/l)	0.438 ± 0.064	0.440 ± 0.072	0.878
L-arginine (μmol/l)	107 ± 22	112 ± 20	0.314
L-arginine / ADMA ratio	248 ± 49	258 ± 49	0.099

Mean ± SD.

p-values are by the repeated measure analysis of variance.

Abbreviations are: HOMA-IR: insulin resistance by homeostasis model assessment, Non-HDL: non-high-density lipoprotein, LDL: low-density lipoprotein, HDL: high-density lipoprotein, ADMA: asymmetrical dimethyl arginine.

Table 2. Measurements of blood pressure and regional arterial stiffness before and after lipid reduction with atorvastatin.

	Before	After 6 months	<i>p</i> -value
Heart rate (beats/min)	66 ± 11	66 ± 10	0.953
Brachial SBP (mmHg)	137 ± 14	133 ± 12	0.145
Brachial DBP (mmHg)	81 ± 10	79 ± 6	0.251
Brachial PP (mmHg)	56 ± 11	54 ± 9	0.248
Ankle-brachial pressure index	1.12 ± 0.07	1.12 ± 0.06	0.902
Heart-carotid PWV (cm/s)	1,282 ± 334	1,195 ± 269	0.168
Heart-brachial PWV (cm/s)	666 ± 97	645 ± 84	0.268
Heart-femoral PWV (cm/s)	1,267 ± 225	1,209 ± 193	0.180
Femoral-ankle PWV (cm/s)	1,123 ± 162	1,073 ± 142	0.019

Mean ± SD.

p-values are by the repeated measure analysis of variance.

Abbreviations are: SBP: systolic blood pressure, DBP: diastolic blood pressure, PP: pulse pressure, PWV: pulse wave velocity.

Results

As shown in Table 1, total cholesterol, non-HDL-cholesterol and LDL-cholesterol levels were significantly decreased following treatment with atorvastatin. There was no significant change in other metabolic parameters such as body mass index, or fasting plasma glucose, HbA_{1c}, and HDL-cholesterol levels.

Table 2 shows the changes in regional PWV, pulse rate and blood pressure measurements. PWV was significantly decreased in the femoral-ankle segment. The PWV of the other arterial regions tended to decrease, although the changes were not statistically significant. Pulse rate was unchanged by atorvastatin. No significant change was observed in systolic, diastolic or pulse pressure of the brachial arteries, or of the tibial arteries.

L-arginine or ADMA levels did not change significantly, although the L-arginin/ADMA ratio tended to increase (Table 1). The percent reduction in LDL-cholesterol did not significantly correlate with the changes of PWV (data not shown).

Discussion

In the present study, we examined the possible change in regional arterial stiffness following atorvastatin treatment in patients with type 2 diabetes mellitus, and showed that the lipid-lowering treatment was associated with a reduction in PWV of the femoral-ankle segment more significantly than the changes in PWV of the other arterial regions. Since there was no significant change in the major factors affecting PWV such as blood pressure, heart rate (22), or ABI (23) following the treatment, we interpret the decrease in femoral-ankle PWV as indicating an improvement in the arterial stiffness of the leg. Although Ichihara *et al.* (24) found that fluvastatin reduced brachial-ankle PWV in diabetic patients with end-stage renal disease, the measurement of brachial-ankle PWV did not differentiate between the central and peripheral arteries. Therefore, the present study is the first to evaluate the effect of statin on regional arterial stiffness in diabetic patients.

Only a few previous studies evaluated the effect of the statins on stiffness of different arterial regions. Shige (17) showed that treatment with simvastatin for six months reduced the stiffness of peripheral arteries but not of central arteries in non-diabetic patients with hyperlipidemia. Leibovitz (18) found a similar effect of atorvastatin on peripheral arterial stiffness. These findings in nondiabetic subjects are in good agreement with the present observation in diabetic individuals, suggesting that the preferential decrease in peripheral arterial stiffness by statins occurs regardless of the presence of diabetes mellitus.

We detected a significant decrease in PWV of the leg arteries, whereas PWV values of the other arterial segments showed only a tendency to decrease. One of the explanations for this observation is that the leg arteries more rapidly respond to the atorvastatin treatment than do the other arteries. Otherwise, the change in arterial stiffness following treatment with atorvastatin may be greater in peripheral arteries than in central arteries. Peripheral arteries such as the femoral and brachial arteries are classified as muscular arteries composed mainly of smooth muscle cells with smaller amounts of extracellular matrix. Central arteries such as the aorta and carotid and subclavian arteries are examples of elastic arteries composed of smooth muscle cells with larger amounts of collagen, elastin and other extracellular matrix proteins forming multiple layers of elastic laminae (25). Therefore, the preferential improvement in peripheral arterial stiffness suggests that atorvastatin reduced vas-

cular smooth muscle tonus rather than changed the properties of the extracellular matrix. Statins are known to improve endothelial function and induce vasorelaxation (26, 27). Also, the effects of female gender (20) and estrogen (28), which improves endothelial function, on arterial stiffness were reported to be more significant in peripheral than central arteries.

We examined the possibility that the observed improvement in arterial stiffness following atorvastatin treatment may be associated with an increase in the serum level of L-arginine, the precursor of nitric oxide (NO), or decrease in the serum level of asymmetric dimethyl arginine (ADMA), a known endogenous inhibitor of NO synthase (NOS) (21). Although the ratio of L-arginine to ADMA tended to increase following atorvastatin treatment, neither the L-arginine nor ADMA level changed significantly as reported by Sasaki *et al.* (29), suggesting that atorvastatin reduces arterial stiffness without significantly changing the L-arginine or ADMA level.

What is the implication of the observed improvement in stiffness of the lower limb arteries following treatment with atorvastatin? Increased stiffness of the arteries in the lower limbs is associated with impaired arterial blood flow (14, 30), subclinical hypoxia of the foot (15), and ischemic symptoms of the lower extremities (16). Thus, the decreased PWV in the leg arteries following atorvastatin treatment may have beneficial effects on local blood flow, accounting for the favorable effects on walking performance (6–8).

There are a few limitations in this study. First, the number of subjects may be too small to draw a solid conclusion. Second, we performed post-treatment PWV measurements once. More frequent measurements would have provided more reliable results. Third, we cannot completely rule out the possibility that a change in blood pressure affected the results of PWV in some patients. However, such an effect of blood pressure would be an unlikely explanation for the preferential reduction in the femoral-ankle PWV, because no significant change was observed in blood pressure or PWV values of the other arterial segments. It may have been better to measure stiffness parameter β (31), a blood pressure-independent index of arterial stiffness, to avoid such problems.

In conclusion, following treatment with atorvastatin, PWV was decreased more significantly in the lower limb arteries than other arterial segments in diabetic patients. Although such an arterial change is expected to have potentially favorable influences on blood flow in the lower extremities and clinical outcome, further studies are needed to clarify the benefits and the mechanisms of the reduction of arterial stiffness by atorvastatin.

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