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Original Article

Pravastatin therapy attenuates left ventricular dysfunction and improves remodeling after myocardial infarction

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Abstract: Objective: The purpose of this study was to investigate the effect of pravastatin on cardiac function with myocardial infarction (MI) and reveal the possible mechanism involved in it. Methods: In the patients assay, 30 consecutive patients with MI (16 males and 14 females) were enrolled. Brain natriuretic peptide (BNP), atrial natriuretic peptide (ANP), and left ventricular ejection fraction (LVEF) function before and after pravastatin usage were compared. In the animal assay, two groups of rats were subjected to permanent coronary occlusion. Group II (n = 14) received oral pravastatin (10 mg/kg/d) daily at 3 wk before and 4 wk after MI, while group I (n = 12) received equivalent doses of vehicle. Results: The percentage of patients with high BNP level (BNP > 20 pg/mL) significantly decreased, from 2 D to 24 W in the pravastatin group, but not in the control group. Similar results occurred with respect to the ANP level (ANP > 40 pg/mL). LVEF in pravastatin group was significantly higher than that of the control group at 3 W and 24 W. In the animal assay, compared with group I, echocardiographic LVEF and fractional area change (FAC) were higher while left ventricular end-diastolic volume (LVEDV) and left ventricular end-systolic and end-diastolic diameters (LVESD and LVEDD) were lower in treated rats. Conclusion: Both patients and animal assays showed that pravastatin therapy during peri-infarct period significantly improved left ventricular (LV) function and limited adverse LV remodeling following MI independent of a reduction in infarct size.

Keywords: Pravastatin therapy, left ventricular dysfunction, heart remodeling, myocardial infarction

Introduction

Myocardial infarction (MI) frequently produces left ventricular (LV) dilatation associated with myocyte hypertrophy and interstitial fibrosis in non-infarcted myocardium. These changes in LV geometry, referred to as remodeling, contribute to the development of depressed cardiac performance [1]. Statin treatment has been suggested to exert beneficial effects on left ventricular (LV) function and remodeling after experimental myocardial infarction (MI) [2, 3], raising the possibility that statins may aid in the prevention and treatment of heart failure [4]. In patients with heart failure, statin therapy is associated with improved LV function [5] and survival [6]. The underlying mechanisms, however, of how statins may exert these beneficial effects remain to be determined.

Statins belong to 3-hydroxy-3-methylglutaryl coenzyme A (HMGCoA) reductase inhibitors. They can lower plasma cholesterol levels and have been associated with reduced morbidity and mortality in patients with coronary artery disease [7, 8]. It is presumed that statins may cause regression or stabilization of atherosclerotic plaques by lowering serum cholesterol levels [9]. However, the beneficial effects of statins on coronary artery disease are not limited to their ability of lowering plasma cholesterol [7]. Statins have various pleiotropic effects on atherosclerosis including reduction of plaque thrombogenicity, inhibition of cellular proliferation and migration, anti-inflammatory, and endothelial function improvement [10]. Despite the widespread clinical use of statins for hypercholesterolemia and prevention of coronary artery disease, data are lacking on the effects of statins on clinical outcome in heart failure secondary to MI. Although some clinical trials suggest that statins may attenuate remodeling and inflammation in patients with cardiomyopathy [11, 12], a recent randomized placebo-controlled study of high-dose statin in patients with sys-

tolic heart failureshows that statins have no effect on ventricular remodeling [13]. Thus, the role of statins in heart failure due to MI remains controversial. Therefore, the purpose of this study was to determine whether administration of a statin during the peri-infarct period could attenuate MI.

Materials and methods

Patients

This study included 30 male patients. All the patients gave their written informed consent prior to participation in the study. The Institutional Ethical Committee on Human Research approved the study protocol. Patients with prior myocardial infarction, liver (elevated activities of aminotransferases), kidney (elevated level of creatinine or urea) or lung dysfunction (restrictive or obstructive pattern in spirometry) were excluded from the study.

Treatment and surgical protocols

A total of 344 female Fischer rats (age 10-12 wk; body wt, 175-200 g, Harlan Sprague-Dawley, Inc.) received oral pravastatin (10 mg/kg/d by gavage, group II, n = 20) or an equivalent volume of vehicle (water, group I, n = 20) daily for 7 weeks starting at 3 wk before and continuing for 4 wk after MI. To adjust the daily dose of the drug, body weight was measured twice a week. The dose of pravastatin used in this study was considered safe and was similar to previously used doses shown to have cardioprotective effects in terms of limiting infarct size in rats. It has been reported that statins have cardio-protective effects in patients when initiated prior to the onset of MI and in the first few weeks after MI. The animals were treated before and after the onset of MI to mimic the common clinical situation in which patients are treated with statins chronically, i.e., during the entire peri-infarct period. Three weeks after the treatment started, the rats were anesthetized with ketamine (37 mg/kg) and xylazine (5 mg/kg), intubated, and ventilated with a rodent respirator (Harvard Apparatus). Under sterile conditions, a left thoracotomy was performed in the fourth intercostal space, and a 5-0 Prolene suture tied around the left anterior descending coronary artery at 2-3 millimeters from its origin. The chest was closed with a 3-0 silk suture. The animals continued to receive the assigned treatment and were euthanized at 4 wk after coronary artery ligation.

Echocardiography

Serial echocardiographic studies were performed 4 days before surgery (baseline, BSL) and at 48 h and 4 wk after infarction under light anesthesia (pentobarbital, 25 mg/kg, i.p.). The anterior chest was shaved and rats were placed in the left lateral decubitus position. Body temperature was maintained between 36.9 uC and 37.3 uC. Echocardiographic images were obtained using a Philips HDI 5000 SonoCT ultrasound system equipped with a 12-5 MHz phased-array probe fitted with a 0.3 cm standoff and a 15-7 MHz broadband linear probe. The heart was imaged in the para-sternal short axis view at the level of the papillary muscles to obtain LV wall thickness and ejection fraction (EF), and in the para-sternal long axis view to measure LV end-systolic and end-diastolic volumes (LVESV and LVEDV). All measurements were averaged in three consecutive cardiac cycles and analyzed off-line by a single blinded observer using the ProSolv image analysis software. All calculations were derived using standard formulas. LV end-systolic and end-diastolic diameters (LVESD and LVEDD) were measured from M-mode tracings obtained at the mid-papillary level and analyzed according to modified American Society for Echocardiography standards (posterior wall leading-edge to leading-edge and anterior wall trailing-edge to trailing-edge).

Hemodynamics

Hemodynamic studies were performed at 4 wk after MI, just before euthanasia. Rats were anesthetized with ketamine (37 mg/kg) and xylazine (5 mg/kg), intubated, and mechanically ventilated. Anesthesia was maintained with 1% isoflurane and the core temperature was kept at 37.0 uC with a heating pad throughout the study. A 2F microtip pressure-volume (PV) catheter (SPR-869, Millar Instruments) was inserted into the right carotid artery and advanced into the LV cavity. The right jugular vein was cannulated for fluid administration. After 20 min of stabilization, the PV signals were recorded continuously with an ARIA PV conductance system (Millar Instruments) coupled with a Powerlab/4SP A/D converter (AD Instruments), stored, and displayed on a personal computer. PV

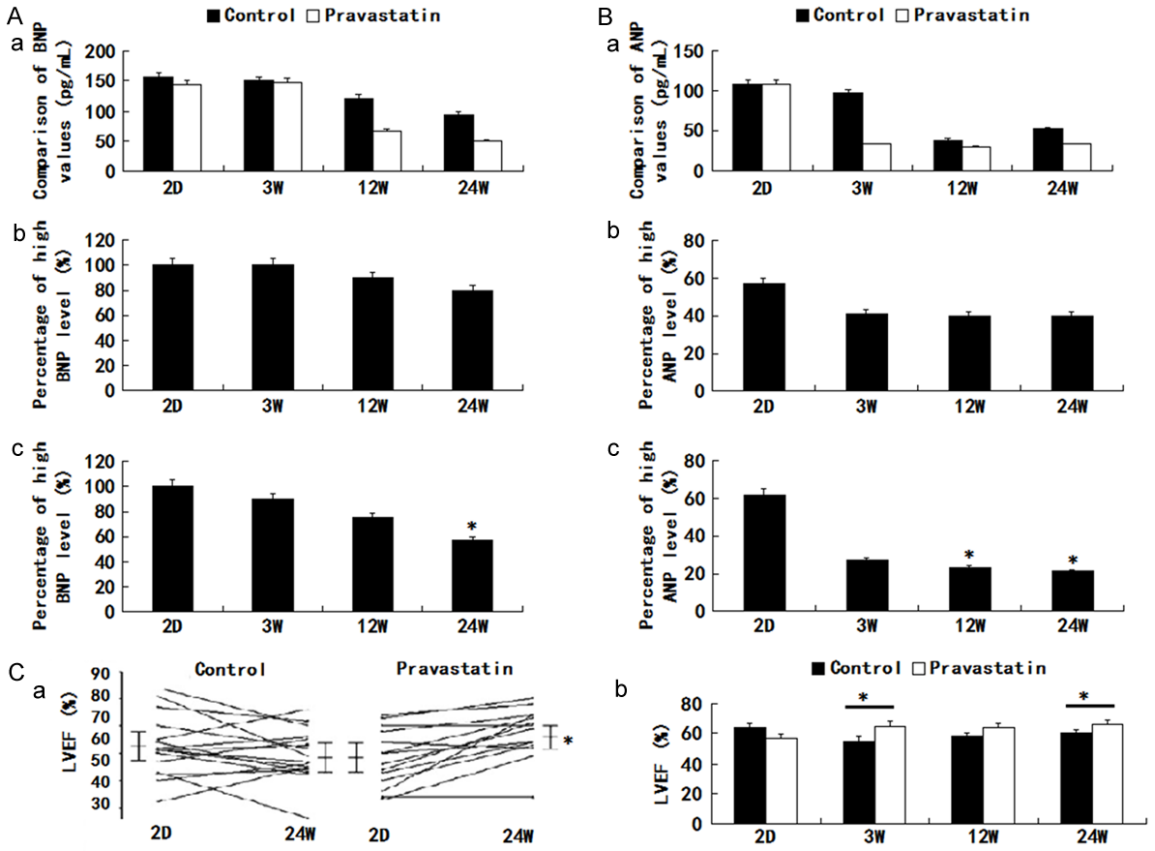

Figure 1. Effects of pravastatin on cardiac function (A) and plasma BNP level. (A) Comparison of mean ± SD values between the control and pravastatin groups at each sampling point. (B) The percentage of patients with an elevated BNP level (> 20 pg/mL) at various time points in the control group. (C) The percentage of patients with an elevated BNP level in the pravastatin group. *P < 0.05 vs. 2D.

relations were assessed by transiently compressing the inferior vena cava with a cotton swab. Parallel conductance from surrounding structures was calculated by injecting a small bolus of 15% NaCl through the jugular vein. LV end-diastolic pressure (LVEDP), dP/dtmax, end-systolic elastance (Ees), and preload recruitable stroke work (PRSW) were calculated using the PVAN software program (Millar).

Morphometry and histology

After hemodynamic measurements, a polyethylene catheter filled with phosphate buffer (0.2 M, pH 7.4) and heparin (100 IU/ml) was applied to the ascending aorta via the right carotid artery. In rapid succession, the heart was arrested in diastole by injecting 1.0 ml cadmium chloride (100 mM)/potassium chloride (3 M) mixture through the aortic catheter. The heart was then excised and perfused retrogradely with phosphate buffer for 3 min to flush out residual blood in the coronary circulation, followed by perfusion with 10% neutral buffered formalin solution for 15 min. Perfusion pressure was maintained between 60 and 80 mmHg while end-diastolic pressure was kept at 8 mmHg. After perfusion-fixation, the atria and right ventricle were dissected from the left ventricle. The LV weight was measured. The heart was cut into four transverse slices (2-mm thick), which were processed, embedded in paraffin, sectioned at 4-mm intervals, and stained with Masson's trichrome. Images were acquired digitally and analyzed using NIH ImageJ (1.37v). From the Masson's trichrome-stained images, morphometric parameters including LV chamber diameter and infarct wall thickness were measured in each section. All anatomical parameters were corrected according to a uniform sarcomere length.

Statistical analysis

Data are reported as mean ± SEM. Measurements were analyzed by ANOVA followed by

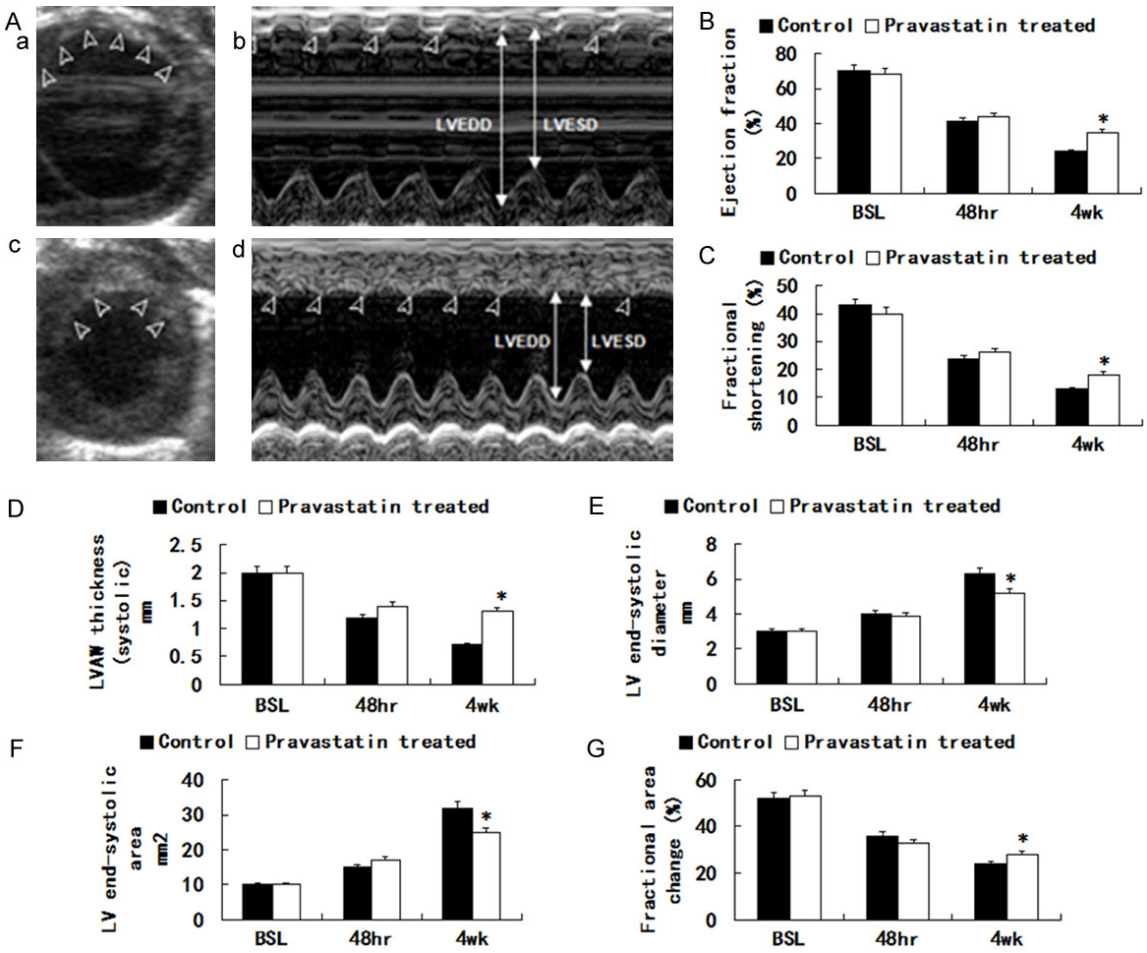

Figure 2. Echocardiographic assessment of LV function. Representative two-dimensional (A, C) and M-mode (B, D) images from vehicletreated (A, B), and pravastatin-treated (C, D) rats 4 wk after coronary occlusion. The infarct wall is delineated by arrowheads. Compared with the vehicle-treated heart, the pravastatin-treated heart exhibited a smaller LV cavity, a thicker infarct wall, and improved motion of the infarct wall. Panels (E-G) demonstrate that treatment with pravastatin improved echocardiographic measurements of LV systolic function at 4 wk after myocardial infarction. Data are mean ± SEM. N = 8-12 rats per group. *P < 0.05 versus vehicle-treated rats at 4 wk. BSL, baseline.

unpaired Student t-tests with the Bonferroni correction. A value of $P < 0.05$ was considered statistically significant. All statistical analyses were performed using the SPSS software (version 8, SPSS, Inc., Chicago, IL).

Results

Effects of pravastatin treatment on plasma BNP and ANP levels

Figure 1Aa shows the mean ± SD values of BNP level in the control and pravastatin groups at each sampling point. There were no significant differences between the two groups. To reduce the possible influence of variance of values, the subjects were classified into two groups by cut-off line (20 pg/mL). In the control group, the percentage of patients with a high BNP level was higher throughout the follow-up period and still 80% at 24 W (**Figure 1A**, **1B**). The percentage of patients with a high BNP level decreased gradually and was significantly lower at 24 W compared with that at 2 D in the pravastatin group (100 to 57%, $P < 0.05$) (**Figure 1A-C**). Similar results were also observed with respect to ANP levels. There were no significant differences between the two groups in ANP values at each sampling point (**Figure 1Ba**). The percentage of the patients with a high ANP level (> 40 pg/mL) significantly decreased at 12 W and 24 W compared to that at 2 D in the pravastatin group (62% at 2D vs. 23% at 12 W and 21% at 24 W, $P < 0.05$ for each) (**Figure 1Bc**), although no significant dif-

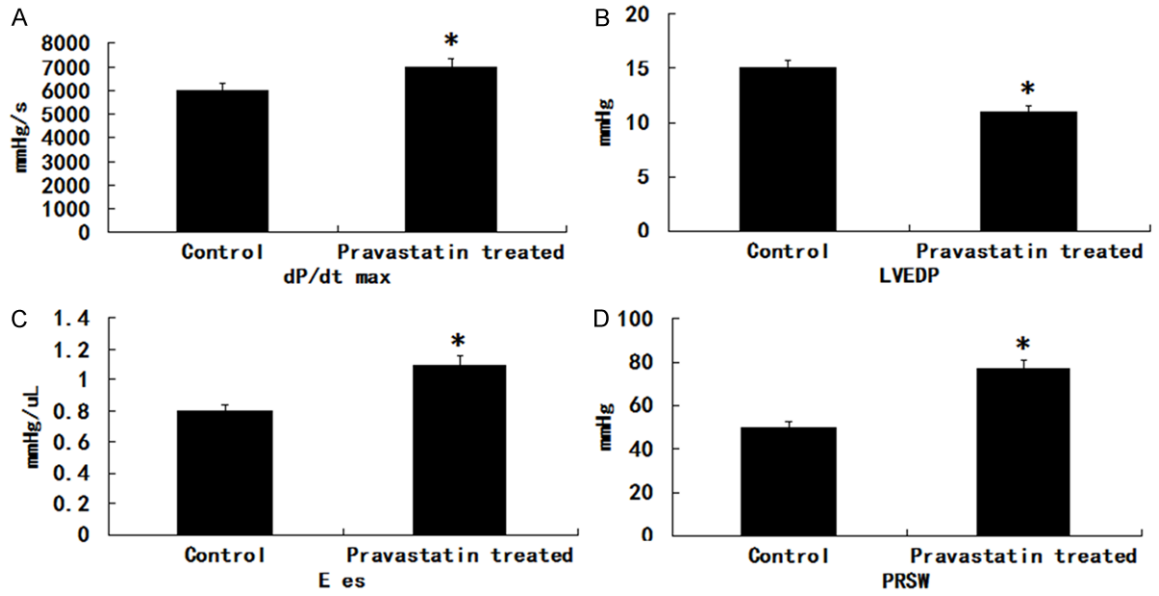

Figure 3. Hemodynamic assessment of LV function at 4 wk after pravastatin treatment. Panels (A-D) illustrate the quantitative analysis of hemodynamic variables including dP/dt (A), LV end-diastolic pressure (B), end-systolic elastance (C), and preload recruitable stroke work (D). Data are mean ± SEM. n = 8-12 rats per group. *P, 0.05 versus vehicle-treated rats at 4 wk. Abbreviations: Ees, end-systolic elastance; LVEDP, left ventricular end-diastolic pressure; PRSW, preload recruitable stroke work.

ference existed in the control group (**Figure 1Bb**).

Effects of pravastatin therapy on left ventricular ejection fraction

In **Figure 1C**, each line represents the change of left ventricular ejection fraction (LVEF) in each patient. Pravastatin significantly improved LVEF from 58.2 ± 10.5% to 67.5 ± 9.2% at 24 W ($P < 0.05$), while no difference was noted in the control group (**Figure 1Ca**). **Figure 1Cb** shows the comparison of LVEF between the two groups at each sampling point during follow-up. LVEF in the pravastatin group was significantly higher than that in the control group at 3 W (66.0 ± 7.8% vs. 56.5 ± 11.8%; $P < 0.05$) and 24 W (67.5 ± 9.2% vs. 59.7 ± 9.8%; $P < 0.05$).

Pravastatin attenuates LV dysfunction

At baseline (4 days before surgery), all parameters of LV function, measured by echocardiography, were similar in groups I and II (**Figure 2**). At 48 h after surgery, the degree of LV systolic functional impairment did not differ among the groups (**Figure 2B-G**), indicating that the extent of injury sustained during MI was comparable. As expected, in vehicle-treated rats the infarct wall thickness decreased (**Figure 2Aa**, **2Ab** and **2D**) and the LVESD increased at 4 wk of follow-up compared with baseline (**Figure 2Aa**, **2Ab** and **2D**). In rats treated with pravastatin, however, infarct wall thickness was greater (**Figure 2Ac**, **2Ad** and **2D**) and the LVESD was smaller (**Figure 2Ac**, **2Ad** and **2E**) while compared with vehicle-treated rats. Vehicle-treated rats exhibited a progressive deterioration in LVEF between 48 h and 4 wk after surgery; this worsening in LVEF was attenuated in rats treated with pravastatin, resulting in a markedly greater LVEF at 4 wk compared with vehicle-treated rats (34.1661.64% vs. 24.3063.39%; $P < 0.05$; **Figure 2B**). Furthermore, LV systolic area was smaller and fractional area change was larger in pravastatin-treated rats compared with vehicle-treated rats (**Figure 2F** and **2G**). Consistent with the echocardiographic data, invasive assessment of cardiac function using the conductance catheter showed improved LV function in pravastatin-treated animals. **Figure 3** shows representative P-V loops, dP/dt max, LVEDP, end-systolic elastance (Ees), and preload recruitable stroke work (PRSW) obtained during IVC occlusion. Both load-dependent (LV dP/dt max, LVEDP) and load-independent (Ees, PRSW) parameters of LV performance were improved in pravastatin-treated group.

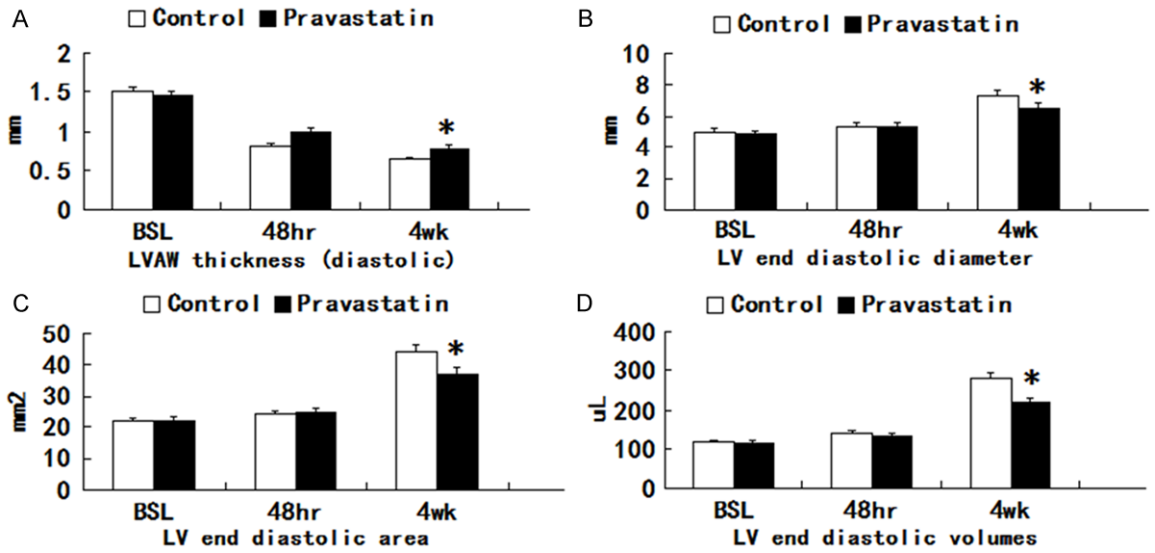

Figure 4. Quantitative assessment of LV remodeling. Panels (A-D) illustrates echocardiographic measurement of LV AW thickness in diastole (A), LV end-diastolic diameter (B), LV end-diastolic area (C) and LV end-diastolic volume (D) at baseline, 48 hr and 4 wk after myocardial infarction. Data are mean ± SEM. N = 8-12 rats per group. *P < 0.05 versus vehicle-treated rats at 4 wk. Abbreviations: AW, anterior wall; BSL, baseline; hr, hours; LV, left ventricular.

Pravastatin halts LV remodeling

At 4 wk after MI, echocardiographic assessment showed an increase in LVEDD, LV end-diastolic area, and LVEDV in vehicle-treated rats, consistent with postinfarction LV remodeling (**Figures 2Aa**, **2Ab** and **4B-D**). However, in pravastatin-treated rats, these variables were significantly smaller compared with vehicle treated rats, indicating improvement in LV remodeling (**Figures 2Ac**, **2Ad** and **4B-D**). Morphometric analysis confirmed the echocardiographic findings. Compared with the vehicle-treated group, the LV chamber diameter was smaller and the infarct wall thickness and infarct wall thickness/chamber diameter ratio were greater in the pravastatin treated group (**Figure 5**).

Discussion

Left ventricular (LV) remodeling after acute myocardial infarction (MI) is a process of progressive changes in LV chamber size, shape, composition and resulting function. Highly deleterious LV dilation, significantly related to poor outcome, is its most characteristic feature [14, 15]. The intensity of ventricular remodeling depends mainly on ventricular wall stress and neurohumoral factors, such as the renin-angiotensin system (RAS) and the sympathetic nervous system [16].

In the present study, we first showed that initiating pravastatin therapy early after the onset of acute MI decreased plasma ANP and BNP levels and improved LVEF by reducing LVDs. Because of the variance of values among each patient, we classified the subjects into two groups by cut-off line (20 pg/mL for BNP and 40 pg/mL for ANP). In the pravastatin group, the percentage of patients with an elevated ANP level tended to decrease at 3 W and significantly decreased at 12 W and 24 W. These short-term effects of pravastatin agree with the improved results of LVEF at 3 W. The effects on BNP were slower than ANP, and the percentage of patients with an elevated BNP level tended to decrease at 12 W and significantly decreased at 24 W after pravastatin treatment. We have no reasonable explanation for the delayed BNP response because generally, BNP is considered a more sensitive indicator of myocardial injury. The study population, a large part of whom had relatively mild cases of acute MI, may have influenced the present results. We found improvement in LVEF during the follow-up period in the pravastatin group. Besides, decreased LVDs predominantly contributed to an increase in LVEF. These results are consistent with previous reports that statin therapy for 3 months decreased left ventricular end-systolic volume in patients with idiopathic dilated cardiomyopathy [17] and that pitavastatin treatment for 6 months reduced LVDs in ischemic and nonisch-

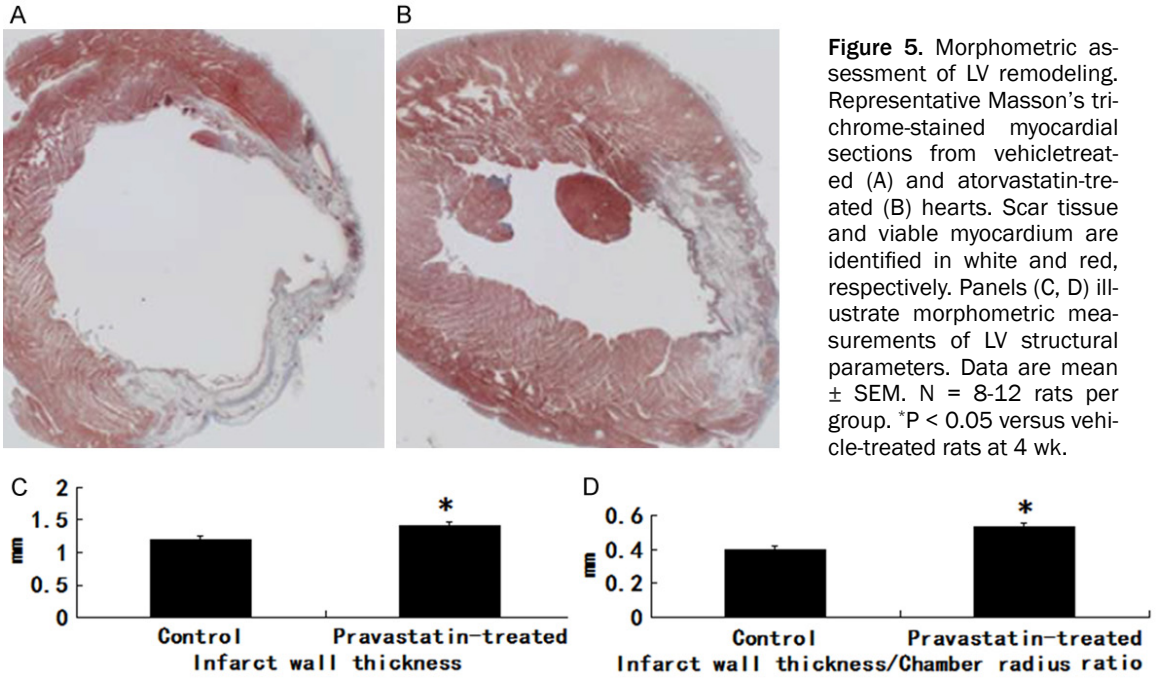

Figure 5. Morphometric assessment of LV remodeling. Representative Masson's trichrome-stained myocardial sections from vehicletreated (A) and atorvastatin-treated (B) hearts. Scar tissue and viable myocardium are identified in white and red, respectively. Panels (C, D) illustrate morphometric measurements of LV structural parameters. Data are mean ± SEM. N = 8-12 rats per group. *P < 0.05 versus vehicle-treated rats at 4 wk.

emic heart failure [18]. On the other hand, reports showed that both LVDd and LVDs were decreased by pravastatin treatment for 12 months [19]. Taking these results into account, statin treatment may initially decrease LVDs and subsequently decrease LVDd after 6 months to improve LVEF. Furthermore, we found that the effect of pravastatin treatment on LVEF appeared within 3 weeks and LVDs tended to decrease. This is the first report that shows a short duration of statin treatment (3 weeks) could improve left ventricular systolic function.

The mechanisms of cardio-protective effects by statins have not been fully established. In the present study, changes in total cholesterol and LDL cholesterol levels had no correlation with changes in LVEF and LVDs. This result indicates that the cardio-protective effect of pravastatin is independent of lipid-lowering and may be mediated by commonly called pleiotropic effects. It is reported [20] that pravastatin suppressed pro-inflammatory effects of vascular endothelial growth factor (VEGF) and placental growth factor (P1GF) by increasing soluble Fms-like tyrosine kinase 1 (sFlt-1), and improved LVEF at 6 months after AMI. In the present study, LVEF was significantly improved 3 weeks after acute MI onset. It is hard to explain the discrepancy of timing when pravastatin treatment showed improvement in LVEF. Other mechanisms of anti-inflammatory effects or increased coronary flow or reduced peripheral resistance by improvement of endothelial function [21] may be associated with the early effects of pravastatin observed in the present study.

Our observations of preserved LV function and mitigated LV remodeling are consistent with previous studies of statins in acute MI. In animal models of MI, cerivastatin (started on the 7th day after MI) [22] and fluvastatin (administered for 4 weeks after MI) [23] have been reported to increase survival and improve cardiac function along with an attenuation of LV remodeling. Furthermore, results from clinical studies in patients with idiopathic dilated cardiomyopathy [24] and nonischemic forms of heart failure [11, 25] also suggest that statins may improve the clinical status of patients by improving LV function and attenuating adverse remodeling in heart failure. Similarly, statin therapy has been shown to reduce the extent of periprocedural non-Q-wave MI [26], microembolization and microinfarction [27] when started before percutaneous coronary intervention.

The finding that pravastatin therapy led to a substantial reduction of myocardial collagen content is novel. Although reduction in collagen content by pravastatin has been demonstrated in animal models of diabetic cardiomyopathy [28] and in spontaneously hypertensive rats [29], such an effect of the drug has not been previously reported in the setting of MI. The end diastolic pressure-volume relationship, which indicates diastolic stiffness, was improved by pravastatin treatment. Unlike LVEDP, which

is load dependent, diastolic stiffness is independent of load.

In summary, our data have obvious translational implications. They suggest that the use of pravastatin during the peri-infarct period might be useful for limiting adverse LV remodeling and improving LV function after MI. This approach might have a potential use in clinical settings such as cardio-protection before surgical procedures (including cardiac and major non-cardiac vascular surgery) and patients with acute coronary syndromes. The exact mechanism(s) of this protective effect, the minimal effective dose, and the minimal duration of treatment required to achieve a beneficial effect need further elucidation.

Disclosure of conflict of interest

None.

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References

[1] Wang HW, Huang TS, Lo HH, Huang PH, Lin CC, Chang SJ, Liao KH, Tsai CH, Chan CH, Tsai CF, Cheng YC, Chiu YL, Tsai TN, Cheng CC, Cheng SM. Deficiency of the microRNA-31-microRNA-720 pathway in the plasma and endothelial progenitor cells from patients with coronary artery disease. Arterioscler Thromb Vasc Biol 2014; 34: 857-69.

[2] Bollati V, Angelici L, Rizzo G, Pergoli L, Rota F, Hoxha M, Nordio F, Bonzini M, Tarantini L, Cantone L, Pesatori AC, Apostoli P, Baccarelli AA, Bertazzi PA. Microvesicle-associated microRNA expression is altered upon particulate matter exposure in healthy workers and in A549 cells. J Appl Toxicol 2015; 35: 59-67.

[3] Xiong XD, Cho M, Cai XP, Cheng J, Jing X, Cen JM, Liu X, Yang XL, Suh Y. A common variant in pre-miR-146 is associated with coronary artery disease risk and its mature miRNA expression. Mutat Res Fundam Mol Mech Mutagen 2014; 761: 15-20.

[4] Slagsvold KH, Rognmo O, Høydal M, Wisløff U, Wahba A. Remote ischemic preconditioning preserves mitochondrial function and influences myocardial microRNA expression in atrial myocardium during coronary bypass surgery. Circ Res 2014; 114: 851-9.

[5] Stratz C, Nührenberg T, Fiebich BL, Amann M, Kumar A, Binder H, Hoffmann I, Valina C, Hochholzer W, Trenk D, Neumann FJ. Controlled type II diabetes mellitus has no major influence on platelet micro-RNA expression. Results from micro-array profiling in a cohort of 60 patients. Thromb Haemost 2014; 111: 902-11.

[6] D'Alessandra Y, Carena MC, Spazzafumo L, Martinelli F, Bassetti B, Devanna P, Rubino M, Marenzi G, Colombo GI, Achilli F, Maggiolini S, Capogrossi MC, Pompilio G. Diagnostic potential of plasmatic MicroRNA signatures in stable and unstable angina. PLoS One 2013; 8: e80345.

[7] Nabiałek E, Wańha W, Kula D, Jadczyk T, Krajewska M, Kowalówka A, Dworowy S, Hrycek E, Władarczyk W, Parma Z, Michalewska-Włudarczyk A, Pawłowski T, Ochała B, Jarząb B, Tendera M, Wojakowski W. Circulating microRNAs (miR-423-5p, miR-208a and miR-1) in acute myocardial infarction and stable coronary heart disease. Minerva Cardioangiol 2013; 61: 627-37.

[8] Vickers KC, Moore KJ. Small RNA overcomes the challenges of therapeutic targeting of microsomal triglyceride transfer protein. Circ Res 2013; 113: 1189-91.

[9] Sun X, He S, Wara AK, Icli B, Shvartz E, Tesmenitsky Y, Belkin N, Li D, Blackwell TS, Sukhova GK, Croce K, Feinberg MW. Systemic delivery of microRNA-181b inhibits nuclear factor-κB activation, vascular inflammation, and atherosclerosis in apolipoprotein E-deficient mice. Circ Res 2014; 114: 32-40.

[10] Wang F, Long G, Zhao C, Li H, Chaugai S, Wang Y, Chen C, Wang DW. Plasma microRNA-133a is a new marker for both acute myocardial infarction and underlying coronary artery stenosis. J Transl Med 2013; 11: 222.

[11] Zhang Y, Wang R, Du W, Wang S, Yang L, Pan Z, Li X, Xiong X, He H, Shi Y, Liu X, Yu S, Bi Z, Lu Y, Shan H. Downregulation of miR-151-5p contributes to increased susceptibility to arrhythmogenesis during myocardial infarction with estrogen deprivation. PLoS One 2013; 8: e72985.

[12] Jansen F, Yang X, Hoelscher M, Cattelan A, Schmitz T, Proebsting S, Wenzel D, Vosen S, Franklin BS, Fleischmann BK, Nickenig G, Werner N. Endothelial microparticle-mediated transfer of MicroRNA-126 promotes vascular endothelial cell repair via SPRED1 and is abrogated in glucose-damaged endothelial microparticles. Circulation 2013; 128: 2026-38.

[13] Tousoulis D, Papageorgiou N, Androulakis E, Siasos G, Latsios G, Tentolouris K, Stefanadis C. Diabetes mellitus-associated vascular impairment: novel circulating biomarkers and therapeutic approaches. J Am Coll Cardiol 2013; 62: 667-76.

[14] Liao XB, Zhang ZY, Yuan K, Liu Y, Feng X, Cui RR, Hu YR, Yuan ZS, Gu L, Li SJ, Mao DA, Lu Q, Zhou XM, de Jesus Perez VA, Yuan LQ. MiR-133a modulates osteogenic differentiation of vascular smooth muscle cells. Endocrinology 2013; 154: 3344-52.

[15] Danowski N, Manthey I, Jakob HG, Siffert W, Peters J, Frey UH. Decreased expression of miR-133a but not of miR-1 is associated with signs of heart failure in patients undergoing coronary bypass surgery. Cardiology 2013; 125: 125-30.

[16] Coleman CB, Lightell DJ Jr, Moss SC, Bates M, Parrino PE, Woods TC. Elevation of miR-221 and -222 in the internal mammary arteries of diabetic subjects and normalization with metformin. Mol Cell Endocrinol 2013; 374: 125-9.

[17] Huang F, Li ML, Fang ZF, Hu XQ, Liu QM, Liu ZJ, Tang L, Zhao YS, Zhou SH. Overexpression of MicroRNA-1 improves the efficacy of mesenchymal stem cell transplantation after myocardial infarction. Cardiology 2013; 125: 18-30.

[18] Lu HQ, Liang C, He ZQ, Fan M, Wu ZG. Circulating miR-214 is associated with the severity of coronary artery disease. J Geriatr Cardiol 2013; 10: 34-8.

[19] Wagner J, Riwanto M, Besler C, Knau A, Fichtlscherer S, Röxe T, Zeiher AM, Landmesser U, Dimmeler S. Characterization of levels and cellular transfer of circulating lipoprotein-bound microRNAs. Arterioscler Thromb Vasc Biol 2013; 33: 1392-400.

[20] Tomé-Carneiro J, Larrosa M, Yáñez-Gascón MJ, Dávalos A, Gil-Zamorano J, Gonzálvez M, García-Almagro FJ, Ruiz Ros JA, Tomás-Barberán FA, Espín JC, García-Conesa MT. One-year supplementation with a grape extract containing resveratrol modulates inflammatory-related microRNAs and cytokines expression in peripheral blood mononuclear cells of type 2 diabetes and hypertensive patients with coronary artery disease. Pharmacol Res 2013; 72: 69-82.

[21] Wang J, Yan CH, Li Y, Xu K, Tian XX, Peng CF, Tao J, Sun MY, Han YL. MicroRNA-31 controls phenotypic modulation of human vascular smooth muscle cells by regulating its target gene cellular repressor of E1A-stimulated genes. Exp Cell Res 2013; 319: 1165-75.

[22] Hutcheson R, Terry R, Chaplin J, Smith E, Musiyenko A, Russell JC, Lincoln T, Rocic P. MicroRNA-145 restores contractile vascular smooth muscle phenotype and coronary collateral growth in the metabolic syndrome. Arterioscler Thromb Vasc Biol 2013; 33: 727-36.

[23] Zhang X, Mao H, Chen JY, Wen S, Li D, Ye M, Lv Z. Increased expression of microRNA-221 inhibits PAK1 in endothelial progenitor cells and impairs its function via c-Raf/MEK/ERK pathway. Biochem Biophys Res Commun 2013; 431: 404-8.

[24] Balderman JA, Lee HY, Mahoney CE, Handy DE, White K, Annis S, Lebeche D, Hajjar RJ, Loscalzo J, Leopold JA. Bone morphogenetic protein-2 decreases microRNA-30b and microRNA-30c to promote vascular smooth muscle cell calcification. J Am Heart Assoc 2012; 1: e003905.

[25] Kin K, Miyagawa S, Fukushima S, Shirakawa Y, Torikai K, Shimamura K, Daimon T, Kawahara Y, Kuratani T, Sawa Y. Tissue- and plasma-specific MicroRNA signatures for atherosclerotic abdominal aortic aneurysm. J Am Heart Assoc 2012; 1: e000745.

[26] Zhu J, Chen T, Yang L, Li Z, Wong MM, Zheng X, Pan X, Zhang L, Yan H. Regulation of microRNA-155 in atherosclerotic inflammatory responses by targeting MAP3K10. PLoS One 2012; 7.

[27] Barjaktarovic Z, Anastasov N, Azimzadeh O, Sriharshan A, Sarioglu H, Ueffing M, Tammio H, Hakanen A, Leszczynski D, Atkinson MJ, Tapio S. Integrative proteomic and microRNA analysis of primary human coronary artery endothelial cells exposed to low-dose gamma radiation. Radiat Environ Biophys 2013; 52: 87-98.

[28] Greliche N, Zeller T, Wild PS, Rotival M, Schillert A, Ziegler A, Deloukas P, Erdmann J, Hengstenberg C, Ouwehand WH, Samani NJ, Schunkert H, Munzel T, Lackner KJ, Cambien F, Goodall AH, Tiret L, Blankenberg S, Trégouët DA; Cardiogenics Consortium. Comprehensive exploration of the effects of miRNA SNPs on monocyte gene expression. PLoS One 2012; 7: e45863.

[29] Sun X, Zhang M, Sanagawa A, Mori C, Ito S, Iwaki S, Satoh H, Fujii S. Circulating microRNA-126 in patients with coronary artery disease: correlation with LDL cholesterol. Thromb J 2012; 10: 16.