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Original Article

Effects of rosuvastatin on atrial nerve sprouting and electrical remodeling in rabbits with myocardial infarction

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Abstract: Objective: This study is to investigate the effect of rosuvastatin on atrial nerve sprouting and electrical remodeling after myocardial infarction (MI). Methods: Rabbit MI model was established by anterior descending branch ligation. These models were divided into the sham (n = 9), MI model (n = 7), and rosuvastatin intervention (n = 8) groups. Immunohistochemistry was used to detect the autonomic atrial nerve distribution. Real-time PCR and Western blot analysis were performed to evaluate the mRNA and protein expression levels, respectively. Results: Our results from immunohistochemistry showed that, compared with the sham group, the densities of tyrosine hydroxylase (TH)- and choline acetyltransferase (CHAT)-positive nerve fibers were significantly elevated in the MI model group. However, TH- and CHAT-positive nerve fibers were significantly decreased by rosuvastatin treatment, suggesting that rosuvastatin could reduce autonomic nerve sprouting in acute MI. Moreover, rosuvastatin decreased the mRNA and protein expression levels of TH in atrial tissues following MI. Compared with the sham group, the mRNA expression level of KCND3 was significantly down-regulated in the MI model group. And, this down-regulation was restored by rosuvastatin treatment. These results suggested that rosuvastatin could inhibit the electrical remodeling in atrium after acute MI. Conclusion: Atrial nerve sprouting and electrical remodeling occur following MI, which could be suppressed by rosuvastatin treatment. Our findings provide insights into the understanding of the mechanism through which statins decrease the risk of atrium arrhythmia after MI.

Keywords: Rosuvastatin, myocardial infarction, atrial nerve sprouting, electrical remodeling

Introduction

Statins, also known as hydroxy-methylglutaryl-CoA (HMG-CoA) reductase inhibitors, are the most effective and widely used lipid-regulating drugs in clinic. It has been well accepted that, statins could exert many biological effects independent of the lipid-lowering activity, including anti-inflammation, anti-oxidative stress, and anti-thrombus effects, as well as myocardium protecting and anti-arrhythmia activities [1, 2]. In recent years, increasing attention has been given to the preventive effects of statins against arrhythmia and cardiac remodeling.

Currently, there are several studies concerning the ventricular remodeling after myocardial infarction (MI) and the effects of statins on the remodeling process [3-5]. However, investigations on atrial remodeling after MI and the involvement of statins have been rarely reported. Moreover, in addition to ventricular arrhythmia, atrial arrhythmia (especially atrial fibrillation, AF) has always been observed following the occurrence of MI [6]. Statins have been shown to have protective effects, and could be used in the treatment of atrial arrhythmia [7], even though the underlying mechanism has not yet been clearly elucidated.

In the present study, rabbit MI models were established, and the effects of rosuvastatin on atrial nerve sproutingand electrical remodeling following MI were investigated. Our findings might contribute to a better understanding of effects of statins on atrial arrhythmia following MI.

Materials and methods

Animal modeling and grouping

Totally 28 male New Zealand rabbits, weighing 2.2 ± 0.3 kg, were purchased from Xi Lin Jiao Animal Center (Jinan, Shandong, China). MI modeling was performed as previously described [8]. Briefly, 3% pentobarbital sodium was slowly injected via ear vein, and 100,000 U/kg penicillin was administrated 30 min before surgery. After anesthesia, rabbits were fixed on a board, followed by surgical skin preparation and disinfection (from supraclavicular fossa to pubic symphysis). Routine ECG was conducted before surgery. A vertical cut was made on the left chest, between the 3rd and 4thribs, and the rib cartilages were cut off, leaving a 3-4-cm vertical incision. The mediastinal pleura were removed, and the parietal pericardium was cut off. The heart was softly lifted up, and the front descending branch of the left coronary artery was fully exposed. Then the front descending branch was ligated at upper 1/3 position with 6-0 silk suture. MI was confirmed by the observation that the left ventricular anterior wall and apex myocardium gradually turned grey-white (with weak pulse), and the ECG results indicated ST-segment elevation in I and aVL leads. Then the thorax was closed, without sewing the pericardium. For the sham group, the same procedures were conducted, in which the left front descending branch was not ligated. After surgery, all rabbits were injected with 100,000 U/kg penicillin for 3 days. These rabbits were then divided into the MI model group (n = 7) and the intervention group (n = 8). For the intervention group, from the first day after surgery, rosuvastatin (10 mg/kg; Astrazeneca, Lund, Sweden) were daily administered by gavage, for 8 weeks. Rabbits from the MI model group and the sham group (n = 9) were subjected to saline administration instead. In addition, four rabbits died during the procedures, due to infection or pneumothorax.

Sample collection

After drug administration, rabbits were anesthetized by intraperitoneal injection of 10% chloral hydrate (2 ml/kg). The heart was removed under sterile conditions, and washed with saline. Large vessels, right atrium, left ventricle, and right ventricle were cut off. Left atrium was divided into two parts: one was kept in liquid nitrogen, and the other was fixed in 4% paraformaldehyde.

Immunohistochemistry

Expressions of tyrosine hydroxylase (TH) and choline acetyltransferase (CHAT) were assessed by immunohistochemistry. Samples were dehydrated and embedded with paraffin, and then cut into 3-4-μm serial sections. After dewaxing, hydration, and antigen retrieval, sections were incubated with mouse anti-rabbit anti-TH (1:125 dilution; PL Laboratories, Milwaukee, WI, USA) or anti-CHAT (1:1000 dilution; Abcam, Cambridge, MA, USA) antibodies, respectively, at 4°C overnight. Horseradish peroxidase-conjugated secondary antibodies (ZS-GB-BIO, Beijing, China) were added for incubation at 37°C for 1 h. Coloration was developed with DAB reagent kit (Beyotime, Shanghai, China), and the images were analyzed by Image Pro Plus software (Media Cybernetics, Inc., Rockville, MD, USA). Nerve density was calculated accordingly.

Quantitative real-time PCR

Total RNA was extracted with Trizol (Invitrogen, Carlsbad, CA, USA), and cDNA was synthesized. The mRNA expression levels of TH and KCND3were detected with the RT-PCR reagent kit (Takara, Dalian, Liaoning, China). The 20-ml reaction system was prepared according to the manufacturer's instructions. Primers for TH and KCND3were synthesized by Sangon Biotech, Shanghai, China. Primer sequences were as follows: TH, forward 5'-GGATGTGGTCCTCGCTGTAG-3' and reverse 5'-GGATGTGGTCCTCGCTGTAG-3'; KCND3, forward 5'-CACTGGGTTGTCCTATCTTGTG-3' and reverse 5'-GGCAGGTGTGTGGTCTTCTTAC-3'. GAPDH was used as the internal control, and the primer sequence was synthesized by Takara: forward 5'-CCACTTTGTGAAGCTCATTTCCT-3' and reverse 5'-TCGTCCTCCTCTGGTGCTCT-3'. The reaction condition consisted of pre-denaturation at 95°C for 10 min, and then 40 cycles of 95°C for 30 s, 65°C for 34 s, and 72°C for 1 min. The relative mRNA expression levels were calculated with the $2^{-\Delta\Delta Ct}$ method.

Western blot analysis

Tissue samples were lysed with the lysis buffer, and the total protein concentration was deter-

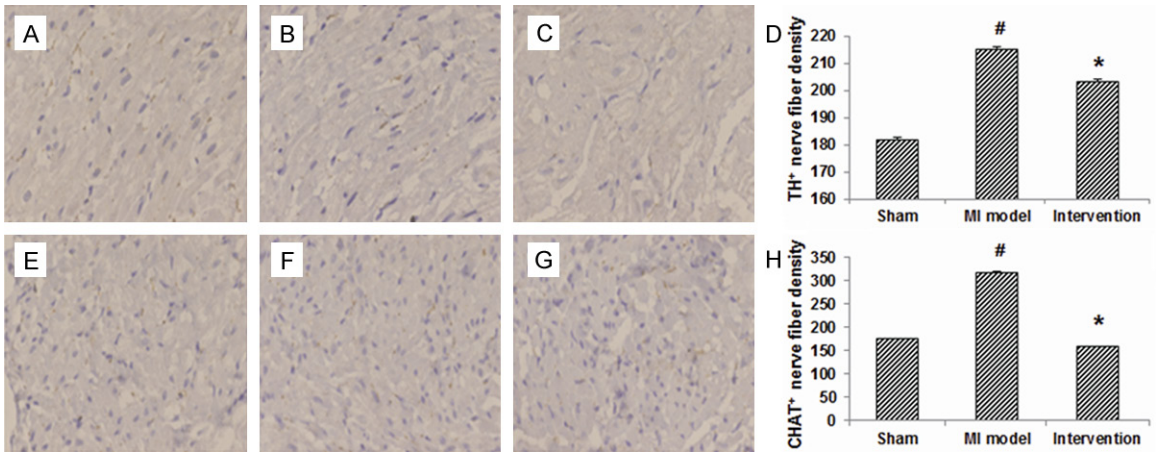

Figure 1. Effects of rosuvastatin on TH- and CHAT-positive nerve fiber regeneration after acute MI. Atrial TH-and CHAT-positive nerve fibers were detected by immunohistochemistry. (A-D) TH-positive nerve fibers were detected in the sham (A), MI model (B), and intervention (C) groups (×40). (D) Statistical analysis of the density of TH-positive nerve fibers. (E-H) CHAT-positive nerve fibers were detected in the sham (E), MI model (F), and intervention (G) groups (×40). (H) Statistical analysis of the density of CHAT-positive nerve fibers. Compared with the sham group, $^{\#}P < 0.05$, $^{\#\#}P < 0.01$; compared with the model group, $^{*}P < 0.05$, $^{**}P < 0.01$.

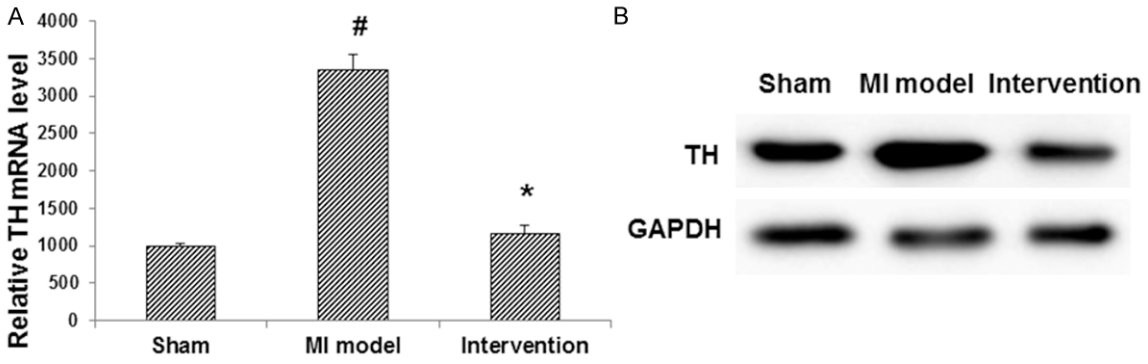

Figure 2. Effects of rosuvastatin on TH expression level in atrial tissues after acute MI. The mRNA and protein expression levels of TH in rabbit atrial tissues were detected by real-Time PCR (A) and Western blot analysis (B), respectively. Compared with the sham group, $^{\#}P < 0.05$, $^{\#\#}P < 0.01$; compared with the model group, $^{*}P < 0.05$, $^{**}P < 0.01$.

mined by the BCA method (Beyotime). The samples were subjected to SDS-PAGE, and then electronically transferred onto a PVDF membrane. After blocked with 5% skim milk for 1h, the membrane was incubated with the mouse anti-rabbit anti-TH primary antibody (1:200 dilution; PL Laboratories) at 4°C overnight. Then secondary antibody was added to incubate the membrane at 37°C for 1 h. The blot was developed using the chemiluminescence (ECL) reagent, and the bands were analyzed with the Alpha Imager gel imaging systems (Santa Clara, California, USA). GAPDH was used as the internal control.

Statistical analysis

Data were expressed as mean ± SD. SPSS17.0 software was used for statistical analysis. One-way ANOVA was performed for the comparison of the difference between groups. $P < 0.05$ was considered statistically significant.

Results

Rosuvastatin suppresses atrial nerve sprouting after acute MI

To analyze the autonomic nerve sprouting after MI and the effects of rosuvastatin on the pro-

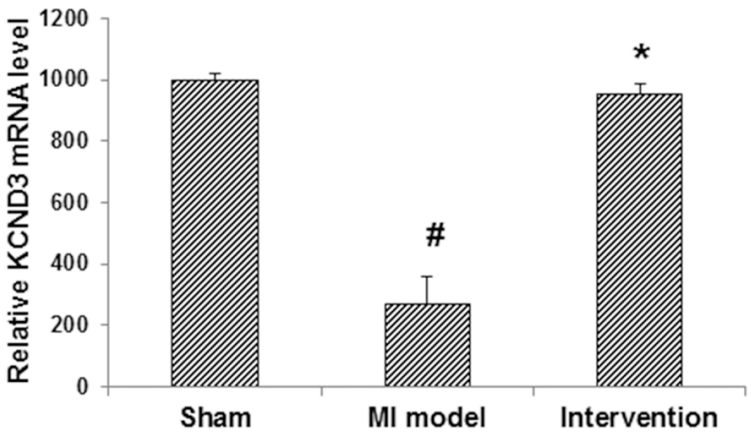

Figure 3. Effects of rosuvastatin on electrical remodeling after acute MI. The mRNA expression level of KCND3 potassium ion channel was detected by real-time PCR in atrial tissues following MI, in the sham, MI model, and intervention groups. Compared with the sham group, $^{\#}P < 0.05$, $^{\#\#}P < 0.01$; compared with the model group, $^{*}P < 0.05$, $^{**}P < 0.01$.

cess, immunohistochemistry was performed to assess TH- and CHAT-positive nerve fiber regeneration in rabbit atrial tissues. Our results showed that, compared with the sham group, the densities of TH- and CHAT-positive nerve fibers were significantly elevated in the model group ($P < 0.05$). However, the TH- and CHAT-positive nerve fibers were significantly decreased by rosuvastatin treatment ($P < 0.05$) (**Figure 1**). These results suggest that rosuvastatin could reduce autonomic nerve sprouting process in atrial tissues in acute MI rabbit models.

Rosuvastatin decreases TH expression levels in atrial tissues after acute MI

The mRNA and protein expression levels of TH were investigated by real-time PCR and Western blot analysis, respectively, in rabbit atrial tissues after surgery and drug administration. Our results from real-time PCR showed that, compared with the sham group, the mRNA expression level of TH was significantly elevated in the model group ($P < 0.05$) (**Figure 2A**). However, the mRNA expression level of TH was significantly declined in the intervention group compared with the model group ($P < 0.05$) (**Figure 2A**). Similar results were obtained from the Western blot analysis. The protein expression level of TH was significantly increased in the model group ($P < 0.05$), which was restored by the treatment of rosuvastatin ($P < 0.05$) (**Figure 2B**). These results suggest that rosuvastatin could decrease the mRNA and protein expression levels of TH in atrial tissues following acute MI.

Rosuvastatin inhibits electrical remodeling after acute MI

To investigate the effects of rosuvastatin on electrical remodeling after acute MI, the mRNA expression level of KCND3 potassium ion channel was detected by real-time PCR after surgery and drug administration. Our results showed that, compared with the sham group, the mRNA expression level of KCND3 was significantly down-regulated in the model group ($P < 0.05$). However, after rosuvastatin treatment, the altered KCND3 mRNA expression level following MI was restored ($P < 0.05$) (**Figure 3**). These results suggest that rosuvastatin could inhibit the electrical remodeling in atrium after acute MI.

Discussion

Autonomic nerve sprouting is sensitive to myocardial infarction (MI). Therefore, when acute MI occurs, coronary occlusion-induced myocardial ischemic necrosis might result in the damage, necrosis, regeneration, and remodeling of autonomic nerves. Cao *et al.* [9] claim that, in the healing phase of MI, cardiac autonomic nerve regeneration is closely linked with ventricular arrhythmia and sudden cardiac death. This viewpoint provides new insights into the further investigation of malignant arrhythmia, including ventricular arrhythmia. Thereafter, numerous studies based on animal models have confirmed the association of ventricular arrhythmia with cardiac nerve sprouting following MI [10-12]. In addition to ventricular arrhythmia, atrial arrhythmia is also commonly observed after MI (e.g., atrial fibrillation), and risk factors include hypertension, heart failure, valvulopathy, and old age (especially for elder people suffering from coronary diseases). It has been demonstrated that atrial fibrillation after MI may be related to the atrial sprouting process [13]. However, detailed underlying mechanisms remain to be elucidated.

The performance of cardiac structural remodeling in arrhythmia has been extensively investigated in recent studies. However, it could not provide satisfactory explanation for the arrhythmia cases caused by other partial heart diseases. We tried to establish the relationship between neural remodeling and arrhythmia, to elucidate the mechanisms of arrhythmia. A previous study has shown that, in rabbit acute MI models, cardiac reconstruction occurred not only in ventricle but also in atrium following MI [14]. Moreover, the cardiac structural remodeling process could be facilitated by rosuvastatin treatment. Based on these results, we hypothesized that nerve and electrical remodeling might also occur in atrium following MI, which could be improved by the treatment of rosuvastatin.

In this study, we investigated the expression levels of TH and CHAT in atrium in rabbit MI models. Our results indicated autonomic nerve sprouting in atrium following MI. TH is a rate-limiting enzyme of biological synthesis of noradrenaline (NE), which is mainly expressed in the cytoplasm of adrenergic nervous fibers. TH expression could affect NE distribution in myocardium, so as to influence the electrophysiological characteristics of the heart. TH is widely used as markers for sympathetic nerves to study the mechanism of arrhythmia [12, 15]. On the other hand, CHAT is considered as the specific marker for cholinesterases, which indicates the distribution and redistribution of vagal nerves. Accordingly, the distribution and activity of cardiac sympathetic and vagal nerves could be reflected by the expression levels and distribution pattern of TH and CHAT.

Heart is dominated by sympathetic and parasympathetic nerves, which form complicated nerve network, controlling cardiac electrical rhythm and myocardium contractile force. Cao *et al.* [9] have suggested that spontaneous ventricular arrhythmia is closely related to the increased density of sympathetic nerves. Moreover, decreased cardiac nerve regeneration can reduce the risk of ventricle arrhythmia [16, 17]. Nerve sprouting after MI is not limited in ventricle, but could be also observed in atrium [18]. The mechanism of axonal fiber regeneration is still unclear. Some studies have demonstrated that oxidative stress [19-21] and inflammation [22] are involved in sympathetic nerve sprouting. Damaged myocardium [23] and inflammatory tissue [24] synthesize and release nerve growth factor (NGF), and macrophages can secrete IL-1 to enhance the expression of NGF. Clearance of macrophages after acute MI can obviously reduce the expression of NGF, improving the remodeling process of sympathetic nerves. NGF [25] is one of the most important nerve nutrition factors in the body, which could modulate the growth and differentiation of sympathetic nerves, and enhance the regeneration of damaged nerves. When MI occurs, cardiac automatic nerve fibers are damaged due to myocardial ischemic necrosis. Meanwhile, surrounding tissues could secret NGF and initiate the repairing process, resulting in nerve redistribution. Our results showed that, compared with the MI model group, the density of cardiac autonomic nerve fibers were significantly decreased in the intervention group, demonstrating that rosuvastatin suppressed atrial autonomic nerve sprouting process, which was in line with the previous findings [14, 26].

Atrial fibrillation (AF) is the most common persistent arrhythmia in clinic, which leads to serious thromboembolic diseases, with unsatisfactory treatment outcomes. Atrial electrical remodeling has been found to play an important role in the development and maintenance of atrial fibrillation, and various ion channels represent the pathological basis for atrial electrical remodeling. Deng *et al.* [27] have found that the down-regulated mRNA expression level of Kv1.5 channel might greatly contribute to the initiation and maintenance of atrial electrical remodeling in patients with AF. Recently, AF has been shown to cause atrial electrophysiological changes (i.e., electrical remodeling), including alterations in atrial action potential duration (APD), effective refractory period (ERP), effective refractory period rate adaptability (ERP-RA), and atrial conduction rate [28, 29]. In turn, atrial electrical remodeling can aggravate the disease development, forming a vicious circle. Shorten ERP and decreased ERP-RA have been associated with the reduced current of L-type calcium channels on the cell membrane, or the enhanced outward potassium current during myocardial repolarization [29-31]. In the case of chronic AF, the current of the L-type calcium channel could be reduced by 70%, and the current of the atrium-specific potassium channel would be reduced by 50% [32, 33]. At present, there are only few studies

concerning the molecular basis of the changed potassium current. In this study, the expression of KCND3 potassium ion channel was detected in rabbit MI models. According to our results, we suppose that electrical remodeling happen in atrium after acute MI.

The treatment outcome for arrhythmia after MI is not satisfactory, while the implantable cardioverter defibrillator is rather expensive. Recent studies have shown that induced pluripotent stem (iPS) cells can differentiate into cardiovascular stem cells, and could be used in the treatment of cardiovascular diseases (including MI) [34, 35]. In animal studies, remaining iPS cells could be detected in the infarction area even at 6 w after injection, which still possess the ability of differentiating into vessels. Meanwhile, these iPS cells improve the nerve regeneration and change the gap junction, indicating that iPS cells could inhibit arrhythmia after MI [36]. However, the efficiency of iPS cells in the treatment of arrhythmia and its involvement in nerve regeneration are still needed to be further confirmed.

Statins are wildly used in the clinical treatment of cardiovascular diseases. In addition to the effective and safe lipid-lowering effects, these drugs also exert beneficial effects to improve the prognosis of patients with MI. In recent years, statins have been found to play versatile roles under various pathophysiological conditions. Particularly, the anti-arrhythmia and cardiac remodeling-preventing effects of statins have been attracting increasing attention in recent years [1, 2, 37]. There are several researches focusing on the ventricular remodeling process following MI and the involvement of statins. In addition to ventricular arrhythmia, atrial arrhythmia (especially AF) is also commonly found in the case of MI [6]. Few studies have concerned the role of statins in the treatment of atrial arrhythmia [7, 38, 39]. Moreover, the atrial nerve and electrical remodeling following MI has also been rarely reported. It has been found that MI is implicated in cardiac nerve sprouting. Moreover, MI could also dramatically reduce the anti-oxidative stress ability, contributing to cardiac autonomic nerve remodeling process. Atorvastatin suppresses nerve remodeling via suppressing oxidative stress. In addition, statins have been shown to decrease the over-proliferation of sympathetic nerves and improve myocardiac electrical remodeling in rabbits with abnormal blood lipid levels [26]. Meanwhile, statins inhibit the infiltration of inflammatory cells, clear the oxygen radicals [40], and alleviate oxidative stress [41].

In conclusion, atrial nerve sprouting and electrical remodeling occur after MI, which could be suppressed by rosuvastatin treatment. These findings provide insights into the understanding of the mechanism through which statins decrease the risk of atrium arrhythmia after MI, and provide theoretical support to the application of statins in the clinical treatment of MI.

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Disclosure of conflict of interest

None.

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References

[1] Abuissa H, O'Keefe JH and Bybee KA. Statins as anti-arrhythmics: a systematic review part II: effects on risk of ventricular arrhythmias. Clin Cardiol 2009; 32: 549-52.

[2] Pedersen TR, Faergeman O, Kastelein JJP, Olsson AG, Tikkanen MJ, Holme I, Larsen ML, Bendiksen FS, Lindahl C, Szarek M and Tsai J. High-dose atorvastatin vs usual-dose simvastatin for secondary prevention after myocardial infarction: the IDEAL study: a randomized controlled trial. JAMA 2005; 294: 2437-45.

[3] Tai G, Fu L, Wang Y and Zhang Y. Research progress on the association between connective tissue growth factor and ventricular remodeling after acute myocardial infarction. Zhonghua Xin Xue Guan Bing Za Zhi 2014; 42: 447-8.

[4] Porter KE and Turner NA. Statins and myocardial remodelling: cell and molecular pathways. Expert Rev Mol Med 2011; 13: e22.

[5] Latet SC, Hoymans VY, Van Herck PL and Vrints CJ. The cellular immune system in the post-myocardial infarction repair process. Int J Cardiol 2015; 179C: 240-247.

[6] Pedersen OD, Abildstrøm SZ, Ottesen MM, Rask-Madsen C, Bagger H, Køber L and Torp-Pedersen C. Increased risk of sudden and non-sudden cardiovascular death in patients with atrial fibrillation/flutter following acute myocardial infarction. Eur Heart J 2006; 27: 290-5.
[7] Kumagai K, Nakashima H and Saku K. The HMG-CoA reductase inhibitor atorvastatin prevents atrial fibrillation by inhibiting inflammation in a canine sterile pericarditis model. Cardiovasc Res 2004; 62: 105-11.
[8] Fujita M, Morimoto Y, Ishihara M, Shimizu M, Takase B, Maehara T and Kikuchi M. A new rabbit model of myocardial infarction without endotracheal intubation. J Surg Res 2004; 116: 124-8.
[9] Cao JM, Fishbein MC, Han JB, Lai WW, Lai AC, Wu TJ, Czer L, Wolf PL, Denton TA, Shintaku IP, Chen PS and Chen LS. Relationship between regional cardiac hyperinnervation and ventricular arrhythmia. Circulation 2000; 101: 1960-9.
[10] Cao JM, Chen LS, KenKnight BH, Ohara T, Lee MH, Tsai J, Lai WW, Karagueuzian HS, Wolf PL, Fishbein MC and Chen PS. Nerve sprouting and sudden cardiac death. Circ Res 2000; 86: 816-21.
[11] Swissa M, Zhou S, Gonzalez-Gomez I, Chang CM, Lai AC, Cates AW, Fishbein MC, Karagueuzian HS, Chen PS and Chen LS. Long-term subthreshold electrical stimulation of the left stellate ganglion and a canine model of sudden cardiac death. J Am Coll Cardiol 2004; 43: 858-64.
[12] Zhou S, Cao JM, Tebb ZD, Ohara T, Huang HL, Omichi C, Lee MH, Kenknight BH, Chen LS, Fishbein MC, Karagueuzian HS and Chen PS. Modulation of QT interval by cardiac sympathetic nerve sprouting and the mechanisms of ventricular arrhythmia in a canine model of sudden cardiac death. J Cardiovasc Electrophysiol 2001; 12: 1068-73.
[13] Hwang HJ, Ha JW, Joung B, Choi EH, Kim J, Ahn MS, Lee MH, Jang Y, Chung N and Kim SS. Relation of inflammation and left atrial remodeling in atrial fibrillation occurring in early phase of acute myocardial infarction. Int J Cardiol 2011; 146: 28-31.
[14] Wang W, Zhang Y, Gao M, Wang J and Hou Y. Effect of rosuvastatin on atrial structural remodeling in rabbits with myocardial infarction. Biomed Rep 2015; 3: 78-82.
[15] Chen PS, Chen LS, Cao JM, Sharifi B, Karagueuzian HS and Fishbein MC. Sympathetic nerve sprouting, electrical remodeling and the mechanisms of sudden cardiac death. Cardiovasc Res 2001; 50: 409-16.
[16] Lee TM, Lin MS and Chang NC. Effect of pravastatin on sympathetic reinnervation in postinfarcted rats. Am J Physiol Heart Circ Physiol 2007; 293: H3617-26.
[17] Liu YB, Wu CC, Lu LS, Su MJ, Lin CW, Lin SF, Chen LS, Fishbein MC, Chen PS and Lee YT. Sympathetic nerve sprouting, electrical remodeling, and increased vulnerability to ventricular fibrillation in hypercholesterolemic rabbits. Circ Res 2003; 92: 1145-52.
[18] Miyauchi Y, Zhou S, Okuyama Y, Miyauchi M, Hayashi H, Hamabe A, Fishbein MC, Mandel WJ, Chen LS, Chen PS and Karagueuzian HS. Altered atrial electrical restitution and heterogeneous sympathetic hyperinnervation in hearts with chronic left ventricular myocardial infarction: implications for atrial fibrillation. Circulation 2003; 108: 360-6.
[19] Gong Y, Li W, Li Y, Yang S, Sheng L, Yang N, Shan H, Xue H, Liu W, Yang B, Dong D and Li B. Probucol attenuates atrial autonomic remodeling in a canine model of atrial fibrillation produced by prolonged atrial pacing. Chin Med J (Engl) 2009; 122: 74-82.
[20] Olivieri G, Otten U, Meier F, Baysang G, Dimitriades-Schmutz B, Müller-Spahn F and Savaskan E. Oxidative stress modulates tyrosine kinase receptor A and p75 receptor (low-affinity nerve growth factor receptor) expression in SHSY5Y neuroblastoma cells. Neurol Clin Neurophysiol 2002; 2002: 2-10.
[21] Hu JL, Zhou QN, Yang SL, Chen H, Zhang L, Yan Y and Hou YM. Metal stress-induced arrhythmia and thoracic spinal cord 1-5 nerve remodeling and myocardial electrophysiological remodeling in rats. Zhonghua Xin Xue Guan Bing Za Zhi 2011; 39: 1094-100.
[22] von Boyen GB, Steinkamp M, Reinshagen M, Schäfer KH, Adler G and Kirsch J. Nerve growth factor secretion in cultured enteric glia cells is modulated by proinflammatory cytokines. J Neuroendocrinol 2006; 18: 820-5.
[23] Oh YS, Jong AY, Kim DT, Li H, Wang C, Zemljic-Harpf A, Ross RS, Fishbein MC, Chen PS and Chen LS. Spatial distribution of nerve sprouting after myocardial infarction in mice. Heart Rhythm 2006; 3: 728-36.
[24] Xin P, Pan Y, Zhu W, Huang S, Wei M and Chen C. Favorable effects of resveratrol on sympathetic neural remodeling in rats following myocardial infarction. Eur J Pharmacol 2010; 649: 293-300.
[25] Chen XF, Lin N, Zhang ZH, Gao JL and He ZJ. Expression of Ghrelin and PYY mRNA and protein in gastrointetinal tract and their receptors mRNA in hypothalamus in neonatal rats. J Shanghai Jiaotong Univ Sci 2010; 30: 550-553.
[26] Liu YB, Lee YT, Pak HN, Lin SF, Fishbein MC, Chen LS, Merz CNB and Chen PS. Effects of simvastatin on cardiac neural and electrophys-

iologic remodeling in rabbits with hypercholesterolemia. Heart Rhythm 2009; 6: 69-75.
[27] Deng YL, Gao F, Xu CX, Zhang JC and Chen L. Expression of delayed rectifier potassium channels in patients with atrial fibrillation. Chinese J Card Arrhythm 2005; 9: 445-448.
[28] Wijffels MC, Kirchhof CJ, Dorland R and Allessie MA. Atrial fibrillation begets atrial fibrillation. A study in awake chronically instrumented goats. Circulation 1995; 92: 1954-68.
[29] Bosch RF, Zeng X, Grammer JB, Popovic K, Mewis C and Kühlkamp V. Ionic mechanisms of electrical remodeling in human atrial fibrillation. Cardiovasc Res 1999; 44: 121-31.
[30] Yue L, Melnyk P, Gaspo R, Wang Z and Nattel S. Molecular mechanisms underlying ionic remodeling in a dog model of atrial fibrillation. Circ Res 1999; 84: 776-84.
[31] Van Wagoner DR, Pond AL, Lamorgese M, Rossie SS, McCarthy PM and Nerbonne JM. Atrial L-type Ca2+ currents and human atrial fibrillation. Circ Res 1999; 85: 428-36.
[32] Schotten U, Verheule S, Kirchhof P and Goette A. Pathophysiological mechanisms of atrial fibrillation: a translational appraisal. Physiol Rev 2011; 91: 265-325.
[33] Krogh-Madsen T, Abbott GW and Christini DJ. Effects of electrical and structural remodeling on atrial fibrillation maintenance: a simulation study. PLoS Comput Biol 2012; 8: e1002390.
[34] Narazaki G, Uosaki H, Teranishi M, Okita K, Kim B, Matsuoka S, Yamanaka S and Yamashita JK. Directed and systematic differentiation of cardiovascular cells from mouse induced pluripotent stem cells. Circulation 2008; 118: 498-506.
[35] Seki T, Yuasa S, Oda M, Egashira T, Yae K, Kusumoto D, Nakata H, Tohyama S, Hashimoto H, Kodaira M, Okada Y, Seimiya H, Fusaki N, Hasegawa M and Fukuda K. Generation of induced pluripotent stem cells from human terminally differentiated circulating T cells. Cell Stem Cell 2010; 7: 11-4.
[36] Zhang F, Song G, Li X, Gu W, Shen Y, Chen M, Yang B, Qian L and Cao K. Transplantation of iPSc ameliorates neural remodeling and reduces ventricular arrhythmias in a post-infarcted swine model. J Cell Biochem 2014; 115: 531-9.
[37] Zheng YY, Guo XH, Ma JW, Li JP, Sun SY and Liu JF. Atorvastatin impacts the serum MCP-1, IL-10 and hs-CRP levels in patients with ACS after PCI. J Clin Cardiol 2009; 25: 491-493.
[38] Mitchell LB, Powell JL, Gillis AM, Kehl V and Hallstrom AP. Are lipid-lowering drugs also antiarrhythmic drugs? An analysis of the Antiarrhythmics versus Implantable Defibrillators (AVID) trial. J Am Coll Cardiol 2003; 42: 81-7.
[39] Vyas AK, Guo H, Moss AJ, Olshansky B, McNitt SA, Hall WJ, Zareba W, Steinberg JS, Fischer A, Ruskin J and Andrews ML. Reduction in ventricular tachyarrhythmias with statins in the Multicenter Automatic Defibrillator Implantation Trial (MADIT)-II. J Am Coll Cardiol 2006; 47: 769-73.
[40] Whaley-Connell A, Habibi J, Nistala R, Cooper SA, Karuparthi PR, Hayden MR, Rehmer N, DeMarco VG, Andresen BT, Wei Y, Ferrario C and Sowers JR. Attenuation of NADPH oxidase activation and glomerular filtration barrier remodeling with statin treatment. Hypertension 2008; 51: 474-80.
[41] Zhou MS. Upregulation of nitric oxide, inhibition of oxidative stress, and antihypertensive effects of statins. Hypertension 2007; 49: e43.