e0038 EFFECTS OF ANGIOTENSIN II ON ATPASES IN AORTIC VASCULAR SMOOTH MUSCLE CELLS FROM WISTAR-KYOTO RATS AND SPONTANEOUSLY HYPERTENSIVE RATS

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Aim To explore the effects of Angiotensin II on the activities of Ca^{2+} -ATPase, Na^{+} , K^{+} -ATPase and mRNA expression levels of the plasma membrane Ca^{2+} -ATPase isoform 1 (PMCA1) and Na^{+} , K^{+} -ATPase α_1 -subunit in cultured thoracic aortic vascular smooth muscle cells (AMSCs) from Wistar-Kyoto (WKY) rats and spontaneously hypertensive rats (SHR).

Methods AMSCs isolated from 14-week-old male WKY rats and SHR were cultured and treated with different concentrations (1×10^{-9} , 1×10^{-8} , 1×10^{-7} mol/l) of Angiotensin II. The activities of Ca^{2+} -ATPase, Na^{+} , K^{+} -ATPase were measured by biochemistry and enzymology. RT-PCR assay was employed to determine the relative levels of PMCA1 and Na^{+} , K^{+} -ATPase α_1 -subunit mRNA in AMSCs.

Results Low and moderate concentration of Angiotensin II significantly increased the activity of Ca^{2+} -ATPase and up-regulated PMCA1 mRNA level in AMSCs from Wistar-Kyoto rats, while high concentration of Angiotensin II inhibited Ca^{2+} -ATPase activity and down-regulated PMCA1 mRNA level. Three different concentration of Angiotensin II significantly decreased the activity of Ca^{2+} -ATPase and PMCA1 mRNA level in AMSCs from SHR. Three different concentrations of Angiotensin II stimulated the activity of Na^{+} , K^{+} -ATPase and increased its α_1 -subunit mRNA expression in AMSCs from WKY rats. Low and moderate concentration of Angiotensin II did not affect the activity of Na^{+} , K^{+} -ATPase in SHR, while high concentration of Angiotensin II significantly suppressed the activity and α_1 -subunit mRNA level of Na^{+} , K^{+} -ATPase.

Conclusions In WKY rats, Angiotensin II may have biphasic effects on Ca^{2+} -ATPase activity and PMCA1 mRNA expression, and may promote the activity and α_1 subunit mRNA expression of Na^{+} , K^{+} -ATPase in a dose-dependent manner in AMSCs. In SHR, Angiotensin II can inhibit Ca^{2+} -ATPase activity and PMCA1 mRNA expression, and only high dose of Angiotensin II can suppress the activity and α_1 subunit mRNA expression of Na^{+} , K^{+} -ATPase in AMSCs.

e0039 EFFECTS OF LISINAPRIL ON THE ACTIVITIES AND MRNA EXPRESSION OF ION PUMPS IN AORTIC SMOOTH MUSCLE CELLS FROM SPONTANEOUSLY HYPERTENSIVE RATS

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Objectives To explore the effects of Lisinopril upon the activities of Na^{+} , K^{+} -ATPase and Ca^{2+} -ATPase and mRNA expression levels of Na^{+} , K^{+} -ATPase α_1 -subunit and plasma membrane Ca^{2+} -ATPase isoform 1 (PMCA1) in cultured thoracic aorta vascular smooth muscle cells (AMSCs) isolated from spontaneously hypertensive rats (SHR).

Methods AMSCs were divided into four groups: Wistar-Kyoto (WKY) control, SHR control, Lisinopril (1×10^{-5}) intervened SHR group and Lisinopril (1×10^{-6}) intervened SHR group. The activities of ion pumps were detected by spectrophotography and mRNA expressions were measured by real time PCR. The content of Angiotensin II (Ang II) in cells-cultured medium were detected by radioimmunoassay.

Results The activities of Na^{+} , K^{+} -ATPase, Ca^{2+} -ATPase and the mRNA expression levels of Na^{+} , K^{+} -ATPase α_1 -subunit and PMCA1 in AMSCs from SHR were significantly lower than those from WKY control ($p < 0.01$). Lisinopril significantly increased the activities of Na^{+} , K^{+} -ATPase and Ca^{2+} -ATPase and mRNA expression levels of Na^{+} , K^{+} -ATPase α_1 -subunit and PMCA1 in AMSCs from SHR ($p < 0.01$). Ang II content of culture medium in AMSCs from SHR was significantly more than those from WKY control ($p < 0.05$), Lisinopril attenuated Ang II content of AMSCs culture medium from SHR ($p < 0.05$).

Conclusions The decreased activities of Na^{+} , K^{+} -ATPase and Ca^{2+} -ATPase may be related to their lower expression of the mRNA in AMSCs from SHR. The Lisinopril may increase the activities of two ion pumps and upregulate the mRNA expression of Na^{+} , K^{+} -ATPase α_1 -subunit and PMCA1 in AMSCs from SHR through blocking the generation of Ang II.

e0040 THE EXPRESSION OF SIGNAL TRANSDUCTION PATHWAY OF TGF1SMAD IN THE RAT CARDIAC TISSUE WITH TYPE 2 DIABETES AND INTERVENTION BY ATORVASTATIN

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Objective Observation on changes of signal transduction pathway of TGF- β_1 /Smad in the course of myocardial fibrosis in the rat with type 2 diabetes and preventive effect of atorvastatin.

Methods The experimental type 2 diabetic rats were yielded by injecting streptozotocin (STZ, 30 mg/kg) and fed with high fat and glucose food, then intervention by atorvastatin ($20 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$) for 12 weeks. Collagen content was observed by Masson staining. RT-PCR was used to observe the gene expression of TGF- β_1 in experiment rat hearts. The protein expression and tissue localisation of TGF- β_1 , Smad2/3, Smad7 and were observed with the immunohistochemistry.

Results The interstitial collagen accumulation and thickened capillary basement membrane in the atorvastatin (Masson stain: 0.80 ± 0.16) administration group was obviously relieved compared with that of the DM (1.36 ± 0.16) group ($p < 0.01$). The expression levels of TGF- β_1 mRNA in the DM group was obviously increased compared with that of the control group (1.39 ± 0.10 vs 0.16 ± 0.02 , $p < 0.01$). The expression levels of TGF- β_1 mRNA in the atorvastatin administration groups was obviously reduced compared with that of the DM group (0.57 ± 0.04 vs 1.39 ± 0.10 , $p < 0.01$). Immunohistochemistry: the positive expressions of Smad2/3 and TGF- β_1 were present in fibroblasts, vascular endothelial and myocardial cells in the control group. The positive expressions of Smad2/3 (10.02 ± 2.32 vs 1.12 ± 0.11 , $p < 0.01$) and TGF- β_1 (18.19 ± 1.39 vs 3.93 ± 0.46 , $p < 0.01$) were even darker and larger in size compared with that in the control group in view of their optic density. There were only the vascular endothelial cells and a few of fibroblasts in the atorvastatin groups were the Smad2/3 (10.0 ± 2.32 vs 5.16 ± 0.17 , $p < 0.01$) and TGF- β_1 (18.19 ± 1.39 vs 10.21 ± 1.08 , $p < 0.01$) positive, but shallow in colour, the positive optic density was reduced obviously compared with that of DM group. In DM group the positive cells of Smad7 mainly distributed in the vascular endothelial cells, fibroblasts, myocardial cells, the numbers of the positive cells was reduced compared with that of the control group (1.26 ± 0.31 vs 10.16 ± 0.64 , $p < 0.01$). In atorvastatin group the positive cells of Smad7 was increased in such cells as those mentioned above, especially in the vascular endothelial cells (1.26 ± 0.31 vs 4.00 ± 0.20 , $p < 0.01$).

Conclusion By way of inhibition activations of pathway of TGF- β_1 /Smad, atorvastatin may obviously relieve the collagen accumulation

and fibrosis in myocardium, thus delay the progress of the diabetic cardiomyopathy.

e0041 VASOMOTOR FUNCTION FOLLOWING NEWER GENERATION OF BARE METAL STENT OVERSTRETCH IN A PORCINE CORONARY MODEL

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Backgrounds Overstretch damage after bare metal stent (BMS) placement could trigger cell proliferation and in-stent restenosis (ISR). Newer Co-Cr BMS has thinner stent struts, which designs to minimise cellular response to injury. We aimed to investigate neointimal growth, as well as vasomotor function after overstretch using Co-Cr BMS in a pig coronary model.

Methods 15 vessels in five pigs were assigned to receive BMS (stent struts 91 μ m) implantation with either S/A ratio 1.3 (group I, n=7) or 1.5 (group II, n=8). Quantitative coronary angiography (QCA) and optical coherence tomography (OCT) were performed at 14 days after stent implantation. Coronary vasomotor function was evaluated by incremental acetylcholine (Ach) (10^{-7} and 10^{-6} M) and nitroglycerin (NTG, 400 μ g) infusion before stent implantation and at 14 days. Endothelial response to Ach was measured at 5–10 mm distal to the stent edge.

Results Both QCA and OCT showed that in-stent stenosis of group I were significantly smaller than group II at 14 days (QCA-late loss (LL), 1.22 ± 0.21 mm vs 1.79 ± 0.17 mm; OCT % AS, $17.0 \pm 7.9\%$ vs $26.9 \pm 10.7\%$ at 14 days, $p < 0.05$ and 0.001 , respectively). Liner regression analysis QCA-LL is proportional to obtained S/A ratio ($r = 0.60$, $p < 0.05$). Endothelium-dependent vasomotion at distal non-stented reference segments was no difference between groups. The mean coronary diameter changes at Ach 10^{-7} M and 10^{-6} M was $2.1\% \pm 0.2\%$ and $2.1\% \pm 0.2\%$ in group I; $2.2\% \pm 0.2\%$ and $2.1\% \pm 0.2\%$ in group II ($p > 0.05$, accordingly). There was also no difference before and at 14 days after stent implantation.

Conclusion The progression of neointimal hyperplasia after BMS implantation is positively associated with the extent of coronary artery injury. Coronary endothelial function is preserved after BMS implantation at 14 days, which is independently of overstretch degree.

e0042 THE EFFECT OF ISCHAEMIC POSTCONDITIONING ON THE STRUCTURE, FUNCTION AND CX43 OF MITOCHONDRIA IN RABBIT MYOCARDIAL ISEHEMIA/REPERFUSION INJURY

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Objective To investigate the effects of ischaemic postconditioning on structural and functional and connexin 43(Cx43) changes of mitochondria induced by myocardial ischaemia reperfusion (IR) injury of rabbits in vivo and potential mechanism.

Methods In anaesthetised open-chest rabbits, the left anterior descending artery (LAD) was occluded for 30 min and reperfused for 4 h. Sixty-four rabbits were randomly divided into four groups (n=16 in each group): Sham operation group (Group Sham), ischaemic reperfusion group (Group IR), ischaemic preconditioning group (Group IP) and ischaemic postconditioning group (Group PC) with sixteen rabbits in each. All rabbits in the four groups were killed 4 h after reperfusion. Myocardial infarct size were determined at the end of the experiment. Mitochondria were isolated with different centrifugations. Ultrastructural changes of mitochondria

were observed under electronmicroscope and mitochondrial membrane potential, Ca^{2+} concentration, MDA content and SOD activity of myocardial mitochondria were examined. The content of the mitochondria Cx43 were detected with Western Blot.

Results Myocardial infarct size was significantly reduced in IP ($18.9 \pm 2.8\%$) and PC ($19.1 \pm 3.9\%$) groups as compared to IR groups ($35.7 \pm 5.8\%$), $p < 0.01$. Compared with group IR, the damage of mitochondrial ultrastructure were milder and Ca^{2+} concentration and MDA content were much lower in group IP and group PC ($p < 0.05$). Mitochondrial membrane potential ($p < 0.01$) and SOD activity of myocardial mitochondria in group IP and group PC was significantly higher than that in group IR ($p < 0.05$). Compared with sham group, the mitochondria Cx43 expression is distinctly decreased compared group IR ($p < 0.05$) and no significant difference was found between Group IP and Group PC.

Conclusion PC can protect mitochondrial ultrastructure by increasing mitochondrial membrane potential and SOD activity, and by alleviating Ca^{2+} overload, and by decreasing MDA content in myocardial mitochondria. The mechanism of PC protection to mitochondria may be concerned with PC attenuating the decrease the mitochondria Cx43 expression induced by ischaemia/reperfusion injury.

e0043 EFFECTS OF SIMVASTATIN ON ANGIOGENESIS AND THE EXPRESSION OF ANG1 AFTER MYOCARDIAL INFARCTION IN RATS

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Objective To investigate the effects of simvastatin on myocardial angiogenesis and the expression of angiopoietin-1 after experimental myocardial infarction (MI) in rats.

Methods 60 healthy adult SD rats were randomly divided into the sham operated group, the control group, low dose of simvastatin ($1 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$) group, medium dose of simvastatin ($10 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$) group, high dose of simvastatin ($40 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$) group. Left anterior descending coronary underwent permanent occlusion to establish the MI model. Rats were administered simvastatin respectively via oral gavage for four consecutive weeks starting at the next day. Density of new microvessels in the ischaemic area, LVMI, protein and mRNA expression of Ang-1 were detected 4 weeks after operation.

Results (1) Compared with the control group, the Density of new microvessels in low and medium dose of simvastatin group increased significantly ($p < 0.05$); and those did not changed significantly in high dose of simvastatin group ($p > 0.05$) (2) LVMI in low and medium dose of simvastatin group decreased significantly compared with that in control group ($p < 0.05$), and further decreased in high dose of simvastatin group. (3) The protein and mRNA expression of Ang-1 in all simvastatin group increased significantly compared with that in control group ($p < 0.05$).

Conclusion (1) Low and medium dose of simvastatin can stimulate myocardial angiogenesis after MI, whereas high dose of simvastatin have no pro-angiogenic effect. (2) the pro-angiogenic effect of simvastatin may be associated with upregulated expression of Ang-1.

e0044 THE ROLE OF ANG1 AND ENOS IN THE PROANGIOGENIC EFFECT OF SIMVASTATIN AFTER MYOCARDIAL INFARCTION IN RATS

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Objective To investigate the roles of angiopoietin-1 (Ang-1) and endothelial nitric oxide synthase (eNOS) in pro-angiogenic