Conclusions Direct adherent and modicum medium changing method is the best one for MSC isolation and culture. 11% is the most suitable serum concentration for MSC growth.

e0174 EFFECT OF HIF1A ON PROLIFERATION AND DIFFERENTIATION OF MSC UNDER HYPOXIA CONDITION IN VITRO

doi:10.1136/hrt.2010.208967.174

[1]Li Xueyuan, [2]Zhang Yang. *[1]The First Hospital of China Medical University, Shenyang, Liaoning, China; [2]The Centrol Affiliated Hospital of Shenyang Medical College, Shenyang, Liaoning, China*

Aims To investigate the effect of HIF-1a on MSC under hypoxia condition.
Materials and methods We transfected HIF-1a into MSC of P3 generation through liposome 2000, and observed the expression of green fluorescence protein in order to assess transfecting efficiency. G418 was used to screen stable transfected cells, and limited dilution method used for monoclone culture of screened cells. We identified the stable HIF-1a transfected MSC through the cell surface antigen testing. We compared the growth state among stable transfected MSC with HIF-1a, vacant plasmid transfected MSC and untransfected MSC under hypoxia condition, and the expression of HIF-1a mRNA, VEGF mRNA, HIF-1a protein and VEGF protein was tested.
Results pcDNA3.0-HIF-1a-eGFP can be successfully transfected into MSC mediated by liposome 2000, with efficiency of 21%. Stable monoclone of transfected MSC can be obtained by G418 screening and limited dilution method. Stable transfected MSCs still reserve the ability of differentiating to chondrocyte and lipocyte. MSCs transfected with pcDNA3.0-HIF-1a-eGFP had lower apoptosis ($p<0.05$), greater proliferation ($p<0.05$), and more expression of HIF-1a mRNA, VEGF mRNA, HIF-1a protein, VEGF protein than MSCs transfected with vacant plasmid pcDNA3.0- eGFP and untransfected ones under hypoxia condition.
Conclusions Stable transfected MSC with HIF-1a has a significant high expression of HIF-1a protein, HIF-1a mRNA, VEGF protein and VEGF mRNA under hypoxia condition. HIF-1a could reduce MSC apoptosis and enhance its proliferation under hypoxia condition.

e0175 THE EFFECT OF GHRELIN ON THE REGRESSION OF ATHEROSCLEROSIS PLAQUE IN APOE-/- MICE AORTA

doi:10.1136/hrt.2010.208967.175

Deng Bin, Xie Xiumei, Fang Li, Chen Xiaobin, Chen Meifang. *Department of Cardiology, Xiangya Hospital, Central South University, Changsha, Hunan, China*

Objective To observe the effect of ghrelin on reducing the apoE$^{-/-}$ mice plasma IL-8, MCP-1, TNFα level and the NFκBp65 expression in vascular wall and the regression of atherosclerotic plaque.
Method 8 week ApoE$^{-/-}$ mice were fed with Western style meals, and the same age mice C57BL/6J fed with the same meals as control. In the eighth week, ApoE$^{-/-}$ mice were assigned to ghrelin intraperitoneal injection and saline injection group randomly in the twelfth week.All of the groups had blood drawn from eye sockets, with isolated plasma used to measure IL-8, MCP-1, TNFα by ELISA. Mice were killed for examination with stereomicroscopy and paraffin imbedding for HE and immunohistochemistry, and frozen section for red oil stain.
Result 1. On stereomicroscopy, HE, oil red stain and image analysis equipment measurement demonstrated no plaque at C57BL/6J mice vessels, and both apoE$^{-/-}$ group and ApoE$^{-/-}$ +ghrelin groups had atherosclerosis plaque at vessels (22.56±2.2 vs 32.37±3.2 $p<0.01$) 2. Contrast to C57BL/6J mice, apoE$^{-/-}$ mice has higher plasma TNFα, IL-8, MCP-1level (28.81±1.8 vs 11.5±0.6, $p<0.05$; 335±16.7 vs 25.0±2.0, $p<0.05$; 78±5.6 vs 15.8±2.0, $p<0.05$), but apoE$^{-/-}$+ghrelin mice has lower TNFα, IL-8, MCP-1 level than ApoE$^{-/-}$ mice (15.45±0.98 vs 24.5±1.68, $p<0.05$; 168.32±8.78 vs 335±16.7 $p<0.05$; 45.5±4.5 vs 78.5±5.6, $p<0.05$). 3. Contrast to C57BL/6J mice, apoE$^{-/-}$mice NFκBp65 immunohistochemistry positive cell integral calculus value were increase (1424.23±167.80 vs 6859.68±675.34, $p<0.01$); ghrelin+ apoE$^{-/-}$ mice NFκBp65 immunohistochemistry positive cell integral calculus value was lower than apoE$^{-/-}$ mice (3424.78±321.6 vs 6859.68±675.34, $p<0.01$), ghrelin can decrease the expression of NFκBp65 in apoE$^{-/-}$mice aorta.
Conclusion Ghrelin can inhibit the inflammatory response to decrease ApoE$^{-/-}$ mice atherosclerosis plaque formation.

e0176 THE EFFECTS OF ROSUVASTATIN ON THE EXPRESSION OF HOMOCYSTEINE-INDUCED EXPRESSION OF MATRIX METALLOPROTEINASE-2 (MMP-2) AND CELL MIGRATION IN RAT VASCULAR SMOOTH MUSCLE CELLS

doi:10.1136/hrt.2010.208967.176

Yangbo Xing, Hangyuan Guo, Yafei Shi. *Department of Cardiology, Shaoxing People' Hospital, Shaoxing, China*

Objective The aim of this study was to investigate the effects of rosuvastatin on the expression of homocysteine-induced expression of matrix metalloproteinase-2 (MMP-2) and cell migration in rat vascular smooth muscle cells (VSMC).
Methods Cultured rat VSMC were incubated with different concentrations of Hcy and rosuvastatin (Hcy 1000 μmol/l) in vitro for 24, 48 and 72 h. The expression of MMP-2 was determined by using the methods of gelatin zymography and western blotting. Cultured rat VSMC was incubated with different concentrations of Hcy and rosuvastatin (Hcy 1000 μmol/l) in transwell for 24, 48 and 72 h. The number of VSMC which transited the membrane represented the aggressivity of VSMC.
Results Hcy (50～1000 μmol/l) increased the expression and activity of MMP-2 significantly. Incubated with the same concentration of Hcy the expression and activity of MMP-2 of 72 h was higher than that of 24 h and 48 h. Hcy reduced the expression of MMP-2 at the concentration of 5000 μmol/l. Rosuvastatin could inhibit the augmentation of homocysteine-induced expression and activity of MMP-2. Hcy (50～5000 μmol/l) could stimulate the migration of VSMC. Rosuvastatin could decrease the stimulation of homocysteine-induced migration of VSMC.
Conclusions These data suggested that Hcy can increase the MMP-2 expression/activity and the migration of VSMC. It may be one of the roles in the pathogenesis of atherosclerosis induced by Hcy. Rosuvastatin can inhibit the augmentation of homocysteine-induced MMP-2 expression/activity and migration of VSMC. This may be one of the pleiotropic of rosuvastatin besides lipid-lowering and benefit the therapy of CHD.

e0177 EXPLORATION NEW METHODS FOR ESTABLISHMENT OF PORCINE MODEL OF ACUTE MYOCARDIAL INFARCTION

doi:10.1136/hrt.2010.208967.177

Siming Tao, Tao Guo, Shunhua Pu, Zhuo Yu. *The First Affiliated Hospital of Kunming Medical College, Kunming, China*

Objective To explore and develop one optimise method that it could establish the porcine model of acute myocardial infraction more safer, quicker, convenient than routine methods.
Methods 30 animals with health condition, mean weight 26.5±4.8 kg; The pigs were divided into two groups randomly, group A (n=13) and group B (n=17), according to different method. Angioplasty balloon was positioned in the mid-distal of left anterior

descending (LAD). The full balloon was inflated and occluded the LAD for 60 min after ischaemia precondition in animals of group A, and then the balloon embolism was positioned in the target vessel; but in group B, the balloon embolism was positioned in the target vessel directly. Intervention operation times and success rate were observed and compared with two groups.

Results 27 pigs underwent induction of AMI successful. Three pigs died of ventricular fibrillation and shock. Success rate of group A was 84.6%, and that of group B was 94%, there was not statistic significant compared with two groups. But mean operation time of group B (28.4±9.4 min)was shorter than that of group A (105.8±27.6 min), furthermore, compared with two groups, there was statistic significant.

Conclusions The method of establishment closed chest porcine model of AMI by implantation balloon embolism in target vessel is feasible, safe, quick and relatively effective.

e0178 DIFFERENTIAL EXPRESSION OF N-MYC DOWNSTREAM REGULATED GENE 2 (NDRG2) IN THE RAT HEART AFTER ISCHAEMIA/REPERFUSION INJURY

doi:10.1136/hrt.2010.208967.178

Zhongchan Sun, Haichang Wang, Guang Tong, Dongdong Sun, Feng Cao. *Department of Cardiovascular medicine, Xijing Hospital, Fourth Military Medical University, Xi'an, China*

Aims It has been shown that Ndrg2 (N-Myc downstream-regulated gene 2), a Myc-repressed gene, is markedly expressed in heart. Ndrg2 can act as a stress responsor under hypoxia and is necessary for hypoxia-induced apoptosis in certain tumour cell lines. In the present study, we investigated whether ischaemia/reperfusion (I/R) injury played a role in the regulation of Ndrg2 expression in rat heart and further explored the possible relationship between Ndrg2 expression and cardiomyocyte apoptosis induced by I/R injury.

Methods Rats were subjected to open chest surgery coronary artery ligation for ischaemia only or followed by reperfusion. Immunostaining and Western blot were applied to test the expression of Ndrg2, c-Myc, cleaved-caspase3 from myocardium, and TUNEL (terminal dUTP nick end labelling)-staining for apoptosis determination of myocardium.

Results The immunostaining confirmed Ndrg2 distribution in cardiomyocytes. The Ndrg2 expression in myocardial tissue after I/R injury was significantly reduced at both mRNA and protein levels. We also observed that expression of c-Myc can be increased by I/R injury and was significantly inversely correlated with Ndrg2 expression. Furthermore, the rapid apoptotic rate at the early phase of reperfusion was ameliorated in the late phase. Some results in vivo were further confirmed by ex vivo study in cultured cardiomyocytes subjected to simulated I/R.

Conclusions Our data suggests that up-regulation of pro-apoptotic c-Myc expression induced by I/R injury in rat myocardium may contribute to the down-regulation of also pro-apoptotic Ndrg2. Such stress response may be involved in the post I/R anti-apoptosis mechanism and myocardial repair in rat.

e0179 IN ORDER TO INVESTIGATE THE POTENTIAL MECHANISM OF PIPERINE, WHICH IS THE ACTIVE SUBSTANCE FROM RHODOBRYUM ROSEUM LIMPR

doi:10.1136/hrt.2010.208967.179

Kai REN, Zhao-liang SHAN. *Department of Cardiology, Chinese PLA General Hospital, Beijing, China*

Objectives In order to investigate the potential mechanism of Piperine, which is the active substance from Rhodobryum roseum Limpr., on acute atrial electrical remodelling in atrial fibrillation by inducing of rapid atrial pacing, as well as its protective effect on injury of oxidative stress in myocardium.

Methods 24 healthy rabbits were collected, and randomly assigned to four groups as follows: normal saline (NS), normal saline+rapid atrial pacing (NS+RAP), piperine (PI), piperine+ rapid atrial pacing (PI+RAP). In the study, acute electrical remodelling was conducted by rapid atrial pacing. In pacing group, right atrium was paced with a frequency of 500–600 bpm for 3 h, atrial effective refractory period was measured at 0 h, 0.5 h, 1 h, 1.5 h, 2 h, 2.5 h and 3 h after pacing, respectively. Then we calculated the rate adaptation of atrial effective refractory periods in different basic pacing cycle lengths. Soon after the experiment, we dissected the atrium of rabbits, the left atrium, right atrium and pulmonary veins were dissected, consequently the levels of MDA, SOD, XOD and Calcium were measured with special kits. All the results were analysed with SPSS17.0.

Results 1. In the experiment, paroxysmal atrial fibrillation or atrial tachycardia can be induced only in NS+RAP group, whereas no similar phenomenon was observed in the other three groups. 2. AERP was markedly shorter in NS+RAP group but it was not changed in NS and PI+RAP group. The rate adaptation of AERP was reduced in NS+RAP, but got lowest point (−0.24±0.59) 1 h after pacing, while the rate adaptation of AERP presented no significant changes in NS and PI group. 3. MDA of PI+RAP group in left atrium and pulmonary vein was lower than that of NS+RAP group ($p<0.01$), but no significant difference of MDA in RA was observed between the two groups. 4. SOD activity in PV is higher in PI+RAP than that in NS+RAP, but no significant difference was observed in other locations between PI+RAP group and NS+RAP group. 5. XOD activity in LA and PV is lower in PI+RAP than that in NS +RAP ($p<0.05$), but XOD activity in RA presented no difference between the two groups. 6. Calcium level in LA, RA and PV, presented lower in PI+RAP compared with that in NS+RAP group.

Conclusion 1. Piperine can help reduce incidence of AF, prevent the shortening of AERP and the rate adaptation of AERP, in other words, piperine can alleviate acute electrical remodelling in acute phase of AF. 2. Piperine can alleviate injury of oxidative stress in AF through suppression of MDA overproduction, reducing the consumption of SOD, suppression of XOD activity as well as Calcium overload, consequently develops the protective effect on myocardium during AF. 3. When AF is present, PV has the most serious injury of oxidative stress but RA suffer the slightest injury. Meanwhile, antioxidant effect of piperine is the most conspicuous in PV.

e0180 THE ACUTE PROARRHYTHMIC EFFECTS OF LOW CONCENTRATION BPA ON FEMALE ADULT RAT AND THE ELECTROPHYSIOLOGIC MECHANISMS

doi:10.1136/hrt.2010.208967.180

[1]Sujuan Yan, [1]Xiaoshu Cheng, [1]Kui Hong, [2]Weizhong Song, [2]Yamei Chen. *[1]Department of Cardiology, The Second Affiliated Hospital of Nanchang University, Nanchang, Jiangxi, China; [2]Department of Pharmacology, College of Medicine, University of Cincinnati, Cincinnati, Ohio, USA*

Objective To investigate the acute proarrhythmic effects of low concentration BPA on adult rat and to demonstrate the electrophysiologic mechanisms.

Methods and results Acute exposure to BPA increased the contractility of cardiac myocytes from female rat heart with inverted U-shaped dose-response curve, these effects were female specific. After-contraction or after-transient rate of female rat cardiac myocytes increased in BPA group, and increased much more by exposure to the mixture of BPA and 10^{-9} M E_2. Increasing BPA or E_2 from 10^{-9} M to 2X 10^{-9} M did not increase the effects induced responses. Although BPA combined with E2 did not induce the