Objective of this work was to identify new biomarkers and establish serum protein fingerprint models for early diagnosis and preoperative evaluation of luminal stenosis severity of CAD.

Methods 48 eligible case subjects, having >50% luminal stenosis in at least one major coronary artery (defined as coronary artery disease (CAD) by angiography), 39 patients with luminal stenosis between 25% and 50% (defined as coronary atherosclerosis), and 52 eligible health individuals, were recruited randomly in this study. The above 139 samples were analysed by Surface-enhanced laser desorption/ionization time-of-flight mass spectrometry (SELDI-TOF-MS).

Results The clinical and biochemical characteristics in three groups matched well. To detect CAD, thirty samples from patients with CAD (stenosis ≥50%) and thirty-two samples from the control group were analysed and designated as pattern 1. This pattern 1 model comprised ten potential biomarkers with m/z of 4276, 5326, 4481, 4320, 4205, 5814, 5551, 3689, 5344 and 6134 Da, respectively. The peaks at 5326, 5814, 5551, 5344, 6134 Da were expressed greatly in samples from patients with CAD, but weakly expressed in healthy individuals; and the other five peaks were expressed greatly in healthy individuals but weakly expressed in patients with CAD. Pattern 1 had a specificity of 78.1% and a sensitivity of 80.0%, as evaluated by leave-one-out crossvalidation. The remaining 38 serum samples, as a blind test set, were analysed on the second day to test pattern 1. The specificity and sensitivity of the blind test were 83.3 and 72.2%, respectively. To distinguish different severities of coronary artery stenosis, twenty-seven samples from patients with coronary atherosclerosis (between 25% and 50%) were analysed and compared with thirty CAD (stenosis ≥50%) patient samples. Pattern 2 was established similarly to distinguish coronary atherosclerosis (stenosis between 25% and 50%) from CAD. Pattern 2 comprised seven potential biomarkers with m/z of 3280, 3689, 5326, 4289, 6135, 3164 and 6439 Da, respectively. While the peaks with m/z 5326 and 6135 Da, were expressed more significantly in CAD samples than in coronary atherosclerosis (stenosis between 25% and 50%), the other five biomarkers appeared to be expressed in the opposite manner. This model had a specificity of 92.6% and a sensitivity of 93.3%, as evaluated by leave-one-out crossvalidation. Pattern 2 was tested blindly in another 30 serum samples on the second day. The specificity and sensitivity of the blind test were 75.0% and 77.8%, respectively.

Conclusion Taken together, the SELDI-TOF-MS technique combined with bioinformatics approaches can not only facilitate the discovery of better biomarkers for CAD and its severity, but also provide a useful tool for molecular diagnosis.

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e0088 MOUSE MODEL CARRYING LMNAE82K MUTATION IN MYOCARDIUM DEVELOPS DILATED CARDIOMYOPATHY AND APOPTOSIS

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Objective To analyse the effects of LMNA[E82K] mutation on the transgenic mice heart.

Methods The transgenic mice were created by microinjection. Pathological changes in the heart of transgenic mice were observed by analyses from histologic, transmission electron microscopic, echocardiographic and ECG measurements. The expression of atrial natriuretic peptide (ANP), B-type natriuretic peptide (BNP), Col3α1, lamin A/C, emerin, fas, caspase-3, caspase-8, caspase-9 and cytochrome c in the myocardium of transgenic mice were determined by Reverse transcription PCR, Western Blot or immunofluorescence. Apoptotic cells were detected by In situ terminal dUTP nick end-labelling (TUNEL) analyses.

Results Two lines, indicating cardiac-specific over-expression of h-LMNA[E82K], were established amongst the transgenic mice. All the changes of the LMNA[E82K] transgenic mice heart displayed a similar pathological phenotype to dilated cardiomyopathy. To sum up, the LMNA[E82K] hearts exhibit thin-walled, dilated left and right ventricles, and had increased heart to body weight ratios as compared to WT hearts. Interstitial fibrosis, slight disarray of myofibrils, swollen mitochondria and loss of cristae as well as the loss of nuclear envelope integrity were observed in the myocardium of the LMNA[E82K] transgenic mice. The expression of fetal gene, BNP, was elevated in the LMNA[E82K] transgenic mice. Apoptosis in myocytes of LMNA[E82K] transgenic mice was detectable by TUNEL assay, apoptosis-related molecular signalling, especially in the Fas pathway, were activated by using Western Blot and immunofluorescence analysis.

Conclusions LMNA[E82K] transgenic mice developed DCM similar to the clinical features of human laminopathies and the most noteworthy was the apoptosis mechanism found in this transgenic mice. It may be a regulatory pathway providing an attractive therapeutic for the treatment of cardiomyopathy.

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e0089 THE ROLE OF PRENATAL CHRONIC HYPOXIA ON MYOCARDIAL ISCHAEMIA REPERFUSION INJURY IN ADULT RABBITS OFFSPRING

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Objective To evaluate the role of prenatal chronic hypoxia on myocardial ischaemia /reperfusion injury in adult rabbits' offspring and explore the relevant mechanism.

Methods The pregnant New Zealand rabbits were divided randomly into normoxic (n=8) and hypoxic (12% O_2 from days 10 to 28 of gestation, n=8) groups. One male offspring of each maternal rabbit was randomly selected to study. The offspring rabbits were subjected to heat stress (42°C for 15 min) at 6 months of age. After 24 h, left anterior descending branches were excised and subjected to ischaemia for 30 min and reperfusion for 120 min. Cardiac histopathological observation was performed by light microscope and electron microscope. The expression of heat shock protein 70 (HSP_{70}) in myocardium was detected by immunohistochemistry. Myocardial enzyme activity, apoptotic index and caspase-3 activity in myocardium were examined as well.

Results Ischemia-reperfusion after heat stress pretreatment increased myocardial enzyme activity, apoptotic index and caspase-3 activity in prenatal chronic hypoxia rabbits (4720.31±744.39 IU/l, 1849.13±416.58 IU/l, 40.43±5.03%, 12.43±1.77 unit, respectively) when compared with control (3388.95±532.43 IU/l, 1435.13±92.08 IU/l, 34.40±4.66%, 10.58±1.42 unit, respectively). Heat stress pretreatment induced HSP_{70} significant expression in left ventricular myocardium was not observed in prenatal chronic hypoxia rabbits but in normoxic control rabbits.

Conclusions Prenatal chronic hypoxia inhibits HSP_{70} synthesis in the heart of adult offspring in response to body heat stress, which might insult cardioprotection against ischaemia-reperfusion injury.

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e0090 EFFECT OF GRANULOCYTEMACROPHAGE COLONY STIMULATING FACTOR ON COAGULATION AND FIBRINOLYSIS IN RABBIT MODEL WITH ILIAC DAMAGED BY BALLOON

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Objective Endothelial dysfunction after percutaneous coronary intervention (PCI) played a key role in perturbations of haemostatic equilibrium. Granulocyte-macrophage colony stimulating factor

(GM-CSF) could enhance the restoration of cells' normal phenotype. However, previous studies suggested GM-CSF might induce hypercoagulability. In this study, we discussed the effect of GM-CSF on coagulation and fibrinolysis after artery deendothelialization.

Methods 24 male New Zealand White rabbits underwent primary iliac artery deendothelialization were randomised to two groups (GM-CSF group and control group). GM-CSF group animals received a subcutaneous injection of GM-CSF, the control group animals were given a subcutaneous injection of equivalent saline. The iliac arteries of all animals were damaged by balloon after 7 days. The plasma levels of tissue-type plasminogen activator (t-PA), plasminogen activator inhibitor-1 (PAI-1) and nitric oxide (NO) were detected before, 1 week, 2 weeks and 4 weeks after angioplasty. The repair and hyperplasia of intima were observed by microscope.

Results The NO levels of the GM-CSF group were higher than that of the control group 2 weeks and 4 weeks after operation [(91.9 ± 11.6) µmol/l vs (81.7 ± 12.2) µmol/l]; [(97.7 ± 10.1) µmol/l vs (83.2 ± 12.6) µmol/l]. Although there was no difference between the two groups in both activities of t-PA and PAI-1 in each specific time point, in 1, 2, 4 weeks after operation, the activities of t-PA became higher than the ones before operation, while the activities of PAI-1 stayed almost the same. Pathological examination showed that the level of intima hyperplasia. VSMC and fibrous tissue of neointima were much lower in GM-CSF group and endothelium was more integrated and smooth.

Conclusion GM-CSF could facilitate the repair of intima, better the function of endothelium without disturbing the balance of coagulation and fibrinolysis.

POTENTIAL EFFECT OF GRANULOCYTEMACROPHAGE COLONYSTIMULATING FACTOR ON THE ARTERIAL REMODELLING AT THE REFERENCE SITE AFTER ANGIOPLASTY

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Objective Arterial remodelling may also occur at the reference site. Resent studies showed that granulocyte-macrophage colony-stimulating factor (GM-CSF) could facilitate the repair of endothelium and reduce vascular inflammation. To observe the effect of granulocyte-macrophage colony-stimulating factor on arterial remodelling, we evaluated the remodelling at the reference site and the lesion site in rabbit model.

Methods 28 healthy New Zealand White rabbits were randomised to two groups (GM-CSF group and control group). GM-CSF group received a subcutaneous injection of GM-CSF (10 ug/kg/d), the control group were given a subcutaneous injection of equivalent saline. The iliac arteries of all animals were damaged by balloon after 7d. The levels of nitrogen monoxide (NO) were detected before and 4 weeks after angioplasty. Histological sections of iliac from rabbits killed 4 weeks after angioplasty were analysed. Luminal area (LA), external elastic lamina area (EEL), and intimal plus medial areas (I+M) were measured at the lesion ($_L$) and reference ($_R$) sites.

Results The NO level of the GM-CSF group was significantly higher than that of the control group after 4 weeks (97.69 ± 10.21 vs 83.18 ± 12.57 µmol/l). Morphometric analysis showed that the LA ($_L$) of control group was smaller than that of the GM-CSF group (0.87 ± 0.40 vs 1.34 ± 0.52 mm^2, $p<0.05$) and I+M ($_L$) was greater than that of the GM-CSF group (2.62 ± 0.48 vs 2.26 ± 0.43 mm^2, $p<0.05$). There was no difference in EEL ($_L$) between the two groups (3.48 ± 0.80 vs 3.60 ± 0.91, $p>0.05$). Morphometric analysis also showed a smaller LA ($_R$) in the control group (1.60 ± 0.48 vs 1.99 ± 0.54 mm^2, $p<0.05$), whereas there was no difference in I+M ($_R$) between the two groups. EEL ($_R$) significantly correlated with LA ($_R$), EEL ($_L$), and I+M ($_R$) in both groups combined (r=0.91,

$p<0.0001$; r=0.909, $p<0.0001$; and r=0.685; $p<0.0001$, respectively). LA ($_R$) also correlated with LA ($_L$) (r=0.919; $p<0.0001$).

Conclusion Remodelling can affect both the lesion and the reference sites and appears to occur in parallel and proportionately at both sites. GM-CSF treatment increased reendothelialization of the injured artery and inhibited unfavourable remodelling.

AVE0991 IMPROVES LEFT VENTRICULAR REMODELLING AND CARDIAC FUNCTION INDUCED BY MYOCARDIAL INFARCTION IN RATS

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Objective To evaluate the effects of 4-week treatment with the nonpeptide angiotensin-(1-7) analogue AVE 0991 on heart function and left ventricular remodelling induced by myocardial infarction (MI).

Methods In this study, we evaluated the cardiac effects of AVE 0991 on normal and infracted male Sprague-Dawley (SD) rats. MI was induced by left coronary artery ligation. After 4 weeks of treatment, transthoracic echocardiography (TTE) was used to evaluate cardiac function. The heart wet weight was recorded, normalised for body weight. Left ventricle serial sections were dyed with triphenyltetrazolium chloride (TTC) stain to quantify the infarct size, with Masson's trichrome stain to quantify collagen volume fraction (CVF), and with haematoxylin-eosin (HE) stain for diameter measurement of cardiomyocytes.

Results Infarct size was slightly reduced in AVE 0991 group compared to control group ($42.6\pm3.6\%$ vs $50.9\pm4.4\%$, $p<0.01$). In addition, AVE 0991 treatment attenuated the decrease in LVFS ($25.54\pm7.33\%$ vs $18.41\pm3.32\%$, $p<0.05$) and LVEF ($54.82\pm11.63\%$ vs $42.7\pm6.5\%$, $p<0.05$) compared to control group. AVE 0991 also reduced MI-induced hypertrophy as quantified by diameter measurements of cardiomyocytes (vs. control group 25.49 ± 4.37 µm vs 32.06 ± 6.85 µm, $p<0.05$).

Conclusion The nonpeptide angiotensin-(1-7) analogue AVE 0991 has a cardioprotective effect on impairment of heart function and ventricular remodelling induced by MI.

MYOCARDIAL INFARCTION-INDUCED REMODELLING AND INFLAMMATORY CYTOKINES IN RATS ARE ATTENUATED BY AVE0991

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Objective To investigate the effects of 4-week treatment with the nonpeptide angiotensin-(1-7) analogue AVE 0991 on expression of inflammatory cytokines and remodelling induced by myocardial infarction (MI).

Methods In this study, MI was induced by left coronary artery ligation in male Sprague-Dawley (SD) rats. After 4 weeks of treatment, the heart wet weight was recorded, normalised for body weight. Left ventricle serial sections were dyed with triphenyltetrazolium chloride (TTC) stain to quantify the infarct size, with Masson's trichrome stain to quantify collagen volume fraction (CVF), and with haematoxylin-eosin (HE) stain for diameter measurement of cardiomyocytes. RT-PCR was used to investigate the synthesis of TGF-β1, TNF-α, collagen type I and III.