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denervation, n=10), RD-3d+MI group (RD performed three days before MI, n=15), Metoprolol-3d+MI group (Metoprolol treated three days before MI, n=15), ACEI-3d+MI group (Perindopril treated three days before MI, n=15), and ARB-3d+MI group (Losartan treated three days before MI, n=15). Cardiac function, autonomic nervous system parameters (HRV), and neuroendocrine activities (plasma renin, angiotensin II, aldosterone and norepinephrine Levels) were evaluated 8 weeks post MI.
Results: Ten of 20 animals in the MI group, 5 of 15 in the RD-3d+MI group, 5 of 15 in the metoprolol-3d+MI group, 7 of 15 in the ACEI-3d+MI group and 8 of 15 in the ARB-3d+MI group died within the eight week period after coronary artery ligation. The death rates of the RD-3d+MI group and the metoprolol group were the same and much less than the MI, ACEI, or ARB groups ($P<0.05$). The death rate did not differ between the latter three groups. None in the control and RD group died during the experiment. There were no significant differences in body weight or the infarct size among all experimental groups eight weeks post-MI. The results showed that the physiologic benefits of RD on improving cardiac remodeling and function, water and sodium excretion, autonomic modulation and suppression of RAAS activation were significantly better than any of the three drugs alone and had no effect on normal controls.
Conclusions: In this post-MI HF animal model, surgical RD provides effective autonomic modulation, inhibition of the RAAS, improved cardiac remodeling, and preserved renal function, without affecting normal circulation and cardiopulmonary function in normal rats. Compared to metoprolol, ACEI, and ARB single drug therapies, RD alone is more efficacious. These results suggest that RD may be an effective treatment option for HF, especially in patients who have contraindications to drug therapy.

GW25-e0746

Technique of synchronous culture of endothelial progenitor cells and Smooth muscle cell derived rabbit bone narrow

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Objectives: To isolate rabbit bone marrow-derived mononuclear cell then synchronous culture rabbit endothelial progenitor cells (EPCs) and smooth muscle progenitor cells (SPCs), study their biological properties and assess the possibility as the seed cells for tissue-engineered venous valves.
Methods: Density gradient centrifugation was used to obtain bone marrow blood mononuclear cells, which were separately cultured with EGM-2 complete medium containing 5% FBS to be induced to EPC and with EBM-2 medium without VEGF containing 5% FBS, 20ng/ml PDGF-BB for SPC induction.
Results: EPCs were cultured for 10 days and the cells fused as monolayer, showing a "stepping stone" appearance and expressing VEGFR-2, vWF and weakly expressing CD133. Under the transmission electron microscope, W-P bodies could be seen within the EPCs cytoplasm. Biological functions showed visible EPC grew on the matrigel in a blood wessel -like form. SPCs was cultured for 14 days and showed specific features of the vascular smooth muscle growth, namely, "peak-valley" growth way. SPCs expressed CD34 and SMA without vWF and VEGF-2 expression myofilaments, paralleled with the longitudinal axis, could be seen under the electron microscope. SPCs could not form vessel-like structures on the Matrigel.
Conclusions: Mononuclear cells could be obtained through density gradient centrifugation of the bone marrow blood, which could be synchronous cultured to EPCs and SPCs with high purity, provided seed cells for Venous valve tissue engineering economical and simply.

GW25-e0838

eNOS modified endothelial progenitor cells inhibit efficiently neointima formation and enhancement of vascular function

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Objectives: Loss of endothelial NO production after arterial injury may contribute to restenosis, characterized by neointima formation and elastic recoil. Previous studies have established that bone marrow-derived endothelial progenitor cells (EPCs) play an important role in vascular repair. In this study, we investigated that hypothesis that overexpression of vasculoprotective gene endothelial nitric oxide synthase (eNOS) in EPCs may restore NO production and inhibit neointimal hyperplasia.
Methods: EPCs obtained from rat bone marrow were isolated using a Ficoll density gradient centrifugation, and expanded in endothelial basal medium. The endothelial characteristics of EPCs were identified by immunologic cell chemical staining and fluorescent labeling. EPCs were transduced with pseudotyped retroviral vectors expressing human eNOS (eNOS-EPCs) or green fluorescent protein (GFP-EPCs). eNOS or GFP modified EPCs were injected directly by intravenous tail vein after arterial injury and again 24 hours later. Two weeks after transplantation, eNOS proteins in the rat vessels were assayed by western blot. The morphology of arterial intima and media was studied by optical microscopy and image analysis system.
Results: The adherent cells were considered EPCs which had spindle shape and form blood-island-like structures during development. The adherent cells had many endothelial characteristics. Transduction efficiency of EPCs ex vivo was above 90%. eNOS gene transfer augmented EPCs proliferative activity. eNOS proteins were detected in the rat vessels. Transfused EPCs may home to the injury site and enhanced reendothelialization associated with decreased neointima formation. The antiproliferative effect of EPCs is further enhanced by overexpression of eNOS. Furthermore, eNOS overexpressed EPCs could increase significantly endothelium- dependent vasodilation function (EDVR).
Conclusions: In vitro, eNOS gene transfer enhanced EPCs proliferative activity. In vivo, eNOS overexpressed EPCs could accelerated reendothelialization and inhibit neointimal hyperplasia. The results show that gene modified EPCs facilitate the strategy of cell transplantation for vascular dysfunction and prevention of restenosis after angioplasty.

GW25-e0843

c-Met overexpression promote reendothelialization and inhibit neointimal formation after balloon injury

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Objectives: to explore the effect of c-met overexpression in EPCs on reendothelialization after balloon injury.
Methods: EPCs derived from mouse bone marrow were isolated and cultured. 3- (4, 5-dimethylthiazol-2-yl) -2, 5-diphenyltetrazolium bromide assays were used to evaluate EPC proliferation. Adenoviral vector expressing c-Met was generated using the AdEasy system. To evaluate the role of HGF/Met in vascular repair in vivo, we used balloon-injured rat carotid artery model. Evans Blue dye was administered to evaluate reendothelialization after 10 days injury, and the neointimal formation was assessed at 21 days following vascular injury.
Results: The effect of HGF on EPC proliferation was examined 48 h after exposure to different quantities of HGF (range 2-20 ng/ml). The proliferation effect was strongly dose-dependent and significantly increased in c-met-EPCs group compared with EPCs group. After transfusion of c-met-EPCs or EPCs to balloon-injured rat via vessel, Evans Blue dye was administered to evaluate reendothelialization after balloon injury. reendothelialized area was significantly larger in c-met-EPCs group than in EPCs group ($64.25\pm8.90\%$ vs. $43.21\pm7.24\%$, n=5, $P<0.01$). A marked decrease in the neointimal area and I/M ratio was found in c-met–EPCs compared with EPCs group at day 21 (0.29 ± 0.06 vs. 0.63 ± 0.13, n=5, $P<0.01$).
Conclusions: c-Met overexpression improve EPCs proliferation, promote reendothelialization and inhibit neointimal formation after balloon injury.

GW25-e0845

Baseline serum uric acid level as a predictor of cardiovascular disease related mortality and all-cause mortality: A meta-analysis of prospective studies.

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Objectives: Serum uric acid (SUA) levels have been used to predict cardiovascular and all-cause mortality event, but the data have yielded conflicting results. We investigated whether SUA was an independent predictor for cardiovascular or all-cause mortality with prospective studies by meta-analysis. Serum uric acid (SUA) levels have been used to predict cardiovascular and all-cause mortality event, but the data have yielded conflicting results. We investigated whether SUA was an independent predictor for cardiovascular or all-cause mortality with prospective studies by meta-analysis.
Methods: Pubmed and Embase were searched without language restrictions for publications available till April 2013. Only prospective studies on cardiovascular or all-cause mortality related to SUA levels were included. Pooled adjust relative risk (RR) and corresponding 95% CI were calculated separately for the highest vs. lowest category or the lowest vs. middle category.
Results: For the highest SUA, eleven studies with 172, 123 participants were identified and analyzed. Elevated SUA increased risk of all-cause mortality (RR 1.24; 95% CI 1.09-1.42) and cardiovascular mortality (RR 1.37; 95% CI 1.19-1.57). Subgroup analyses showed that elevated SUA significantly increase the risk of all-cause mortality among men (RR 1.23; 95% CI 1.08-1.42), but not in women (RR 1.05; 95% CI 0.79-1.39). Risk of cardiovascular mortality appeared to be more pronounced among women (RR 1.35; 95% CI 1.06-1.72). The association between extremely low SUA and mortality was reported in three studies; we did not perform a pooled analysis because of high degree of heterogeneity in these studies.
Conclusions: Baseline SUA level is an independent predictor for future cardiovascular mortality. Elevated SUA appears to significantly increase the risk of all-cause mortality in men, but not in women. Whether low SUA levels are predictors of mortality is still inconclusive.

GW25-e0877

Relaxin-2 and relaxin-3 inhibit high glucose-induced apoptosis in neonatal rat ventricular myocytes

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Objectives: High concentrations of glucose induce apoptosis in cardiomyocytes, and contribute to diabetic cardiomyopathy. Relaxin-2 and relaxin-3 are two members of the relaxin peptide family that are cardioprotective. In the present study, we

investigated the effect of either relaxin-2 or relaxin-3 on high glucose-induced apoptosis in neonatal rat ventricular myocytes and its molecular mechanisms.
Methods: Primary neonatal rat ventricular myocytes (NRVMs) were treated with 33 mM high glucose (HG) for 0, 6, 12, 24, 48 and 72 h, and were evaluated for relaxin-1, relaxin-3 and relaxin family peptide receptor 1 (RXFP1) mRNA using real-time PCR. Moreover, cells were treated with/without 100 ng/ml relaxin-2 or 100 ng/ml relaxin-3 prior to HG treatment. Hoechst staining and flow cytometric analyses were performed to determine the level of apoptosis induced by HG in NRVMs. The expression levels of cleaved caspase-9, -8, -3, -12, GRP94, CHOP, LC3 and Beclin-1 were examined using western immunoblot analyses, and autophagosome formation was observed by transmission electron microscopy.
Results: HG-treated NRVMs showed lower relaxin-1 and RXFP1 mRNA, the receptor of relaxin-1 and relaxin-3, along with increased apoptosis. Furthermore, HG increased the protein expression of cleaved caspase-8 and -9, two initiators of the extrinsic and intrinsic pathways of apoptosis, which activated caspase-3. Treatment of NRVMs with HG activated endoplasmic reticulum stress (ERS), but reduced autophagy. However, the pre-administration of either relaxin-2 (HG+R2 group) or relaxin-3 (HG+R3 group) to NRVMs in the presence of HG, resulted in reduced apoptotic cells and protein expression of cleaved caspase-8, -9 and -3. In addition, the pre-administration of relaxin-2 and relaxin-3 blocked ERS, but did not affect autophagy.
Conclusions: HG-induced apoptosis in NRVMs was mediated, in part, by the activation of the extrinsic and intrinsic pathways of apoptosis and ERS. Both relaxin-2 and relaxin-3 inhibited NRVMs apoptosis induced by HG not through autophagy, but by suppressing the extrinsic and intrinsic pathways of apoptosis and ERS.

GW25-e1696

HDAC1 regulation of the transdifferentiation of mesenchymal stem cells to cardiomyocyte lineages in a myocardial microenvironment

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Objectives: In a myocardial microenvironment, bone marrow-derived mesenchymal stem cells (MSCs) can transdifferentiate to cardiomyocytes (CMs). However, the role of histone deacetylase 1 (HDAC1) in this differentiation process remains unclear. This study provided preliminary insights into the acetylation regulatory mechanisms that are involved in the transdifferentiation of adult stem cells by examining HDAC1 expression during the differentiation of MSCs to CMs.
Methods: MSCs were isolated and cultured from male Sprague-Dawley (SD) rats, transfected with Ad-EGFP, and then mixed and co-cultured with CMs at a 1:1 ratio. Subsequently, flow cytometry was used to sort EGFP-positive (EGFP+) MSCs from the co-culture system, the expression of cardiac troponin T (cTnT) in these MSCs was detected by immunofluorescence assay, and HDAC1 expression levels at different co-culture times were measured by quantitative real-time polymerase chain reaction (QT-PCR) and Western blotting.
Results: MSCs began expressing detectable levels of cTnT after 4 days of co-culture with CMs; however, no spontaneous beating was observed in these MSCs. Low levels of HDAC1 expression were observed in the examined CMs, and HDAC1 expression in co-cultured MSCs decreased with increasing co-culture duration. The HDAC1 mRNA expression in MSCs was significantly greater than the HDAC1 mRNA expression in either the CM group or the groups that had been co-cultured for 3, 6, 9, and 12 days. In particular, the 3-day co-culture, 6-day co-culture, 9-day co-culture, 12-day co-culture, and CM groups exhibited 0.69±0.04, 0.49±0.04, 0.35±0.05, 0.20±0.02, and 0.07±0.02 of the HDAC1 mRNA expression in the MSC group, respectively ($P<0.05$ in each case).
Conclusions: HDAC1 negatively regulates the trans differentiation of MSCs to CMs in a myocardial microenvironment.

GW25-e1730

AVE 3085, a novel endothelial nitric oxide synthase enhancer, attenuates cardiomyocyte hypertrophy through reducing oxidative stress

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Objectives: AVE3085 is a novel endothelial nitric oxide synthase enhancer. Although AVE3085 treatment has been shown to be effective for endothelial function, little is known about the effects of AVE 3085 in cardiomyocyte hypertrophy and the mechanisms. The aim of the present study was to investigate the effects of AVE 3085 on cardiomyocyte hypertrophy and the mechanisms involved.
Methods: Cardiomyocyte hypertrophy model was induced by angiotensin II (AngII). Cardiomyocyte were incubated with AngII (10^{-6} mol/L) for 24 h after pretreatment with AVE3085 (10^{-5} mol/L) for 1 h. Cell protein content was determined by Coomassie brilliant blue, cell surface area was measured by Image analysis system and protein synthesis rate was measured by [^{3}H] -Leucine incorporation. NADPH oxidase (NOX) 2 and Nox4 mRNA and protein expression was detected using Real-Time PCR and western blot analysis.
Results: Cell protein content, cell surface area and protein synthesis rate stimulated by AngII in cardiomyocytes were increased significantly, which was effectively attenuated by AVE3085 ($P<0.05$). Moreover, AngII significantly increased the mRNA and protein expression of Nox2 and Nox4, meaning that AngII significantly elevated oxidative stress in cardiomyocytes ($P<0.01$). AVE3085 markedly reversed the elevated T Nox2 and Nox4 mRNA and protein levels ($P<0.01$).
Conclusions: AVE3085 could inhibit cardiomyocyte hypertrophy induced by AngII. The antihypertrophic effect may be associated to reducing oxidative stress.

GW25-e0294

Heme oxygenase-1 protects against protoporphyrin IX-induced cytotoxic effects on macrophages through inhibiting ROS production

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Objectives: Protoporphyrin IX (PpIX) and its derivatives are widely used as a photosensitizer in photodynamic therapy, however, excessive PpIX accumulation are shown to be toxic to cells. Our previous study indicates that PpIX induces macrophage death by reactive oxygen species (ROS) production. As heme oxygenase-1 (HO-1) participates in PpIX metabolism and HO-1 has both anti-inflammatory and antioxidant effects, we aim to investigate the role of HO-1 in the toxicity of PpIX on macrophages.
Methods: THP-1 macrophages were exposed to PpIX and toxic effects were evaluated by CCK-8 assays as well as LDH releasing tests. HO-1 expression was evaluated by western blotting. Intracellular ROS production was analyzed using 2', 7'-Dichlorodihydrofluorescin diacetate (DCFH-DA) by flow cytometry.
Results: PpIX decreased macrophage viability accompanied with the induction of HO-1 expression in a time and dose-dependent way (0-24 hours and 0-20 μM). Pretreatment of cells with zinc protoporphyrin IX (ZnPP), a specific HO-1 inhibitor, for 4 hours enhanced the PpIX-induced cytotoxicity and ROS production. In contrast, pretreatment of cells with cobalt protoporphyrin IX (CoPP), a specific HO-1 inducer, for 4 hours induced elevated HO-1 expression and significantly alleviated the toxic effects of PpIX as well as ROS generation. Treatment with ZnPP or CoPP alone did not influence cell viability. Moreover, inhibition of ROS by N-acetylcysteine (NAC) did not block PpIX-induced HO-1 expression and treatment of cells with H_2O_2 had no effect on HO-1 induction.
Conclusions: HO-1 protects against PpIX-induced cell death via inhibiting ROS production and the HO-1 expression is independent of ROS levels. Therefore, upregulation of HO-1 expression could possibly reduce adverse reactions associated with the application of porphyrin derivatives.

GW25-e0297

Role of Caveolin-1 in Atrial Fibrillation as an Anti-Fibrotic Signaling Molecule in Human Atrial Fibroblasts

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Objectives: This report examines the hypothesis that Cav-1 confers an anti-AF effect by mediating atrial structural remodeling through its anti-fibrotic action.
Methods: We evaluated the expression of Cav-1, transforming growth factor-b1 (TGF-b1), and fibrosis in atrial specimens of 13 patients with AF and 10 subjects with sinus rhythm. Subsquently the role of caveolin-1 in human atrial fibroblasts was studied.
Results: The resuts showed that the expression of Cav-1 was significantly downregulated, whereas TGF-b1 level, collagens I/III contents and atrial fibrosis were markedly increased, in AF. Western blot analysis demonstrated that treatment of human atrial fibroblasts (HAFs) with TGF-b1 resulted in a concentration- and time-dependent repression of Cav-1. Downregulation of Cav-1 with siRNA increased the TGF-b1-induced activation of Smad signal pathway and collagens production in HAFs. Furthermore, incubation of HAFs with the peptides derived from Cav-1 to achieve Cav-1 gain-of function abolished the TGF-b1-induced production of collagens I/III and decreases of MMP-2/-9 expression.
Conclusions: Therefore it was concluded that Cav-1 is an important anti-AF signaling mediator by conferring its anti-fibrotic effects in atrium.

GW25-e0554

Changes of blood pressure and neural damage factors in hypertensive dogs after renal sympathetic denervation

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Objectives: To observe the changes of blood pressure and S-100B, Neuron Specific Enolase Protein in hypertensive dogs used high fat diet after catheter-based renal sympathetic denervation.
Methods: Beagles (n=12) were devided into an interventional group (n=6) and a sham-operation group (n=6). After baseline measurements, Beagles were fed with lard oil for three months. After three months, interventional group plus renal sympathetic denervation by percutaneous catheter-based radiofrequency ablation and control group plus renal angiography .Blood pressure, plasma S-100B, and neuron