Results: The recombinant lentivirus plasmid carrying ZO-1 was correctly constructed, it was identified correct by double enzyme digestion and DNA sequencing. The lentiviral packaging was succeed, and the titer of lentivirus was 1.42×10^8IU/ml.
Conclusion: Lenti-EF1α-EGFP-TRE-ZO1 lentiviral vector has been successfully constructed. The titer of lentivirus was 1.42×10^8IU/ml, it was used to infect iPS cells, which will be useful for further study in vivo, to understand its function and its unique effects under different pathological status of heart, and open new doors for the treatment of cardiovascular diseases.

GW25-e4428

Exogenous Hydrogen Sulfide Delays Nicotinamide-Induced Premature Senescence via SIRT1 pathway in Human Umbilical Vein Endothelial Cells

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Objectives: The present study was designed to observe the effect of hydrogen sulfide (H_2S) on cellular senescence of human umbilical vascular endothelial cells (HUVECs CC-2517) and its underlying mechanism.
Methods: The premature senescence-like phenotype HUVECs (the fourth passage) was induced by nicotinamide (NAM, an inhibitor of SIRT1) at the concentration of 5 mmol/L for 12 hours. Cells were cultured with sodium hydrosulfide (NaHS, 12.5, 25, 50, 100μmol/L) added into the fresh medium for 48 hours on the basis of premature senescence-like phenotyoe HUVECs. The fourth passage of HUVECs was considered as young group. Senescence associated (SA) -β-galactosidase activities were detected to evaluate cell senescence, and the expression of senescence associated heterochromatin foci (SAHF) was visualized by DAPI DNA staining. The mRNA and protein levels of SIRT1 were detected using RT-PCR and western blotting analysis, respectively.
Results: The results showed that β-galactosidase positive cells and the formation of SAHF were markedly increased after treatment with NAM (5mmol/L) for 12h. We also found that NaHS (12.5μmol/L) had no effect on the percentage of SA β-gal positive cells and the expression of SAHF, and the hallmarks decreased at the concentration of 25 and 50μmol/L, reaching the minimum at 50 μmol/L, while the percentage of SA β-gal positive cells and the expression of SAHF increased at the concentration of 100μmol/L. Furthermore we found that SIRT1 expression both on protein and mRNA in the Y+N+S50 group was significantly increased compared with that in Y+N group.
Conclusions: NaHS delays senescence of HUVECs induced by NAM via up-regulation of SIRT1 expression.

GW25-e4464

MicroRNAs and cardiac channelopathies

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Objectives: Cardiac channelopathies are primary arrhythmias caused by variations in the genes encoding cardiac ion channels. It is one of the major causes which could lead to sudden cardiac death.
Methods: The pathological change leading to cardiac channelopathies is intimately linked to the geno-variations. MicroRNAs (miRNAs) are one of the participants that regulate the target gene expression.
Results: Burgeoning evidences indicated that miRNAs are closely associated with myocardial development, proliferation, differentiation, apoptosis and ion channel remodeling, especially involved in arrhythmias, congenital heart disease, hypertension, and myocardial ischemia. In vivo and in vitro studies suggested that miRNAs may serve as a diagnostic biomarker for cardiac channelopathies. They can modulate the function of cardiac ion channel genes and would be a good choice to act as a miRNA-based therapy method.
Conclusions: We review the mechanisms and functions of miRNAs related to cardiac ion channels and focus on the potential perspective of miRNAs as a novel therapeutic target for cardiac channelopathies.

GW25-e4495

ZP2495 improves cardiomyocyte energy metabolism post myocardial ischemia-reperfusion injury in db/db diabetic mice

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Objectives: This study was designed to evaluate the impact of a glucagon-GLP-1 dual-agonist (ZP2495) on cardiac function and energy metabolism after myocardial ischemia-reperfusion injury in db/db mice, and the underlying mechanisms involved.
Methods: The peptides of glucagon, GLP-1 receptor agonist (ZP131) and glucagon-GLP-1 dual-agonist (ZP2495) were synthesized and purified by ChinaPeptides. Wild-type (WT) and db/db mice received 4-weeks treatment of glucagon, ZP131 or ZP2495 followed by left anterior descending coronary artery (LAD) ligation and reperfusion. Cardiac function, hemodynamic parameters, glucose metabolism, myocardial infarct size, cardiomyocyte apoptosis, mitochondrial ultrastructural morphology, mitochondrial respiration and ATP synthesis were measured. Mitochondrial membrane potential, mitochondrial cytochrome c release and mitochondria reactive oxygen species (ROS) generation were assessed in high-glucose-induced neonatal rat ventricular myocytes (NRVMs) after simulated ischemia/reperfusion (SI/R) injury. The expressions of FoxO3a, phospho-FoxO3a (Thr32, Ser413), Akt, phospho-Akt (Ser473, Thr308) AMPK, phospho-AMPK (Thr172), Bim, Bax, Bcl-2, Bad, phospho-Bad, MnSOD, Catalase,Sirt1, PGC1α, Acetylated-Lysine, Nrf-1, Tfam of the heart tissue were detected by Western blot.
Results: ZP2495 treatment improved cardiac function and cardiac glucose metabolism, decreased apoptotic cardiomyocytes, improved mitochondrial ultrastructural morphology and mitochondrial energetic transition of db/db mice after cardiac I/R injury. ZP2495 treatment prevented mitochondrial depolarization and reduced the release of cytochrome c and mitochondria reactive oxygen species (ROS) generation in high-glucose-induced NRVMs after SI/R injury. Western blot revealed that ZP2495 exerted anti-apoptotic and anti-oxidation effect on cardiomyocyte of db/db mice after cardiac I/R injury via Akt/FoxO3a-dependent mechanism. In addition, ZP2495 improved energy metabolism and increased mitochondrial biogenesis in hearts of db/db mice subjected to I/R injury through AMPK/FoxO3a and AMPK-SIRT1-PGC-1α pathway.
Conclusions: Glucagon and GLP-1 dual-agonist (ZP2495) protects against myocardial I/R injury in db/db mice, which may contribute to the improvement of cardiac function and cardiac energy metabolism. The protective effects of ZP2495 may be associated with the Akt/FoxO3a, AMPK/FoxO3a and AMPK-SIRT1-PGC-1α pathways activation.

GW25-e4531

Statin attenuate cardiac hypertrophy by regulating autophagy via AKT/mTOR Pathway in Spontaneously Hypertensive Rats

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Objectives: Autophagy is activated in hypertension-induced cardiac hypertrophy, in which the mechanisms are not clear. Our previous study has demonstrated that atorvastatin inhibites heart remodeling spontaneously hypertensive rats (SHR). This study was designed to determine the regulation of atorvastatin on cardiac autophagy in SHRs.
Methods: Twenty-four male SHRs at 8 weeks of age were randomly divided into four groups receiving vehicle (SHR+V) or atorstatin at a dose of 50 mg/kg/day (SHR+ATO) until ages of 6 months or 12 months old. WKY rats were used as controls. Cardiac magnetic resonance (CMR) was used to evaluate heart functions. Autophagy was demonstrated by the up-regulated expression of microtubule-associated protein-1 light chain 3-II (LC3-II), Beclin-1, caspase-3, the formation of autophagosomes, and by the electron microscopic findings. Western blot analysis shows the expression of the proteins.
Results: Atorstatin treatments for both 4 and 10 months significantly attenuated cardiac dysfunction, remodeling, and atrial natriuretic peptide expression in SHRs, which were increased compared with WKY rats. At 12 months of age, autophagy was found to be activated in the heart of SHRs treated with vehicle or atorstatin. The activated autophagy was especially increased in the cardiomyocytes of SHRs with atorstatin treatment for 10 months. P-Akt and P-mTor decreased in the heart of SHR versus WKY rats, while atorstatin further decreased cardiac akt and mTor phosphorylation.
Conclusions: These findings suggest that atorvastatin attenuated cardiac remodeling and heart dysfunction in SHR probably through its regulation on cardiac autophagy via akt/mTor pathways.

GW25-e4532

Effects of EPC-derived Microvesicles on Ang II-induced Cardiomyocyte Hypertrophy and Apoptosis

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Objectives: Cell-released microvesicles (MVs) are implicated in mediating cell-to-cell communication and are able to modulate target cell functions. Previous evidence indicates that endothelial progenitor cells (EPCs) -derived MVs can modulate endothelial cell survival and proliferation. In this study, we evaluated whether EPC-MVs could protect cardiomyocytes (CMs) against angiotensin II (Ang II) -induced hypertrophy and apoptosis.
Methods: The cell line H9c2 CMs were exposed to Ang II in the presence or absence of EPC-MVs. Cell viability, apoptosis, surface area and β-myosin heavy chain (β-MHC) expression were analyzed. To investigate the underlying mechanisms, reactive oxygen species (ROS) generation, serine/threonine kinase (Akt), phospho-Akt (P-Akt), endothelial nitric oxide synthase (eNOS) and phospho-eNOS (P-eNOS) expression levels were measured. The inhibitors of phosphatidylinositol-3-kinase (PI3K) and NOS were used for pathway verification. The role of MV carried RNAs in mediating these effects was also explored.
Results: Results showed: (1) EPC-MVs were able to protect CM against Ang II-induced changes in cell viability, apoptosis, surface area and β-MHC expression;