diabetic cardiomyopathy (DCM), but it remains largely unknown whether and how MIF regulates GLUT4 expression in cardiomyocytes. The present study aims to investigate the mechanism underlying the modulation of GLUT4 by MIF in cardiomyocytes.

**METHODS**

Activations of AKT and AMPK signaling, and expressions of MIF, GLUT4 and the candidate GLUT4 regulation associated transcription factors in the diabetic mouse myocardium were determined. The screened transcription factors mediating MIF-promoted GLUT4 expression were verified by RNA interference (RNAi) and electrophoretic mobility shift assay (EMSA), respectively.

**RESULTS**

MIF was increased, but GLUT4 was decreased in the diabetic mouse myocardium. MIF could enhance glucose uptake and up-regulate GLUT4 expression in NMVCs. Expressions of transcription factor MEF2A, -2C, -2D and Zac1 were significantly up-regulated in MIF-treated neonatal mouse ventricular cardiomyocytes (NMVCs), and markedly reduced in the diabetic myocardium. Knockdown of MEF2A, -2C, -2D and Zac1 could significantly inhibit glucose uptake and GLUT4 expression in cardiomyocytes. Moreover, EMSA results revealed that transcriptional activities of MEF2 and Zac1 were significantly increased in MIF-treated NMVCs. Additionally, MIF effects were inhibited by an AMPK inhibitor compound C and siRNA targeting MIF receptor CD74, suggesting the involvement of CD74-dependent AMPK activation.

**CONCLUSIONS**

Transcription factor MEF2 and Zac1 mediate MIF-induced GLUT4 expression through CD74-dependent AMPK activation in cardiomyocytes.

**GW26-e1024**

Mechanism of QSYQ on Anti-Apoptosis Mediated by Different Subtypes of Cyclooxygenase in AMI Induced Heart Failure Rats

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**OBJECTIVES**

Qi-shen-yi-qi (QSYQ), one of the most well-known TCM formulas, has been demonstrated to improve cardiac function by its anti-mycocardial apoptosis efficacy in the acute myocardial infarction (AMI) heart failure (HF) rats. However, the mechanisms governing its therapeutic effects remain unclear. In this study, we aim to demonstrate that QSYQ treatment can prevent left ventricular remodeling in HF by inhibiting myocardial apoptosis and inflammation mainly through the PGE2/EPI-PG2/P53 pathway.

**METHODS**

Sprague-Dawley (SD) rats were randomly divided into 4 groups: sham group, model group (left anterior descending (LAD) coronary artery ligation), QSYQ treatment group and aspirin group. 28 days after surgery, hemodynamic measurements were performed. The immunohistochemical studies were used to assess myocardial apoptosis rate, COX-1, COX-2, Fas and MDM2 protein expressions were measured by western-blot. RT-PCR was used to detect mRNA expressions of P53 and 4 subtype receptors of PGE2 (EPI, 2, 3, and 4).

**RESULTS**

Ultrasonic testing results showed that EF and FS values were significantly decreased and abnormal hemodynamic alterations were observed in model group compared to sham group. These findings indicated that HF models were successful induced. Levels of pro-inflammatory cytokines (TNF-α and IL-6) in myocardial tissue were up-regulated in model group compared to sham group. Western-blot results showed that cyclooxygenase 2, which is highly inducible by pro-inflammatory cytokines, increased significantly. Moreover, RT-PCR showed that expressions of EP2 and EP4, which are the receptors of PGE2, were also up-regulated. Increased expressions of apoptotic pathway factors, including P53 and Fas, might be induced by the binding of PGE2 with EP2/4. MDM2, the inhibitor of P53, decreased in model group. TUNEL assay manifested that apoptosis rates of myocardial cells increased in model group. After treatment with QSYQ, expressions of inflammation factors, including TNF-α, IL-6 and COX-2, were reduced. Expressions of EP2 and EP4 receptors also decreased, suggesting that PGE2-mediated apoptosis was inhibited by QSYQ. MDM2 was up-regulated and p53 and Fas in the apoptotic pathway were down-regulated. Apoptosis rates in myocardial tissue in QSYQ group decreased compared with model group.

**CONCLUSIONS**

QSYQ has definite cardiac protective efficacy in improving cardiac function mainly through inhibiting the inflammatory response and down-regulating apoptosis. Its anti-inflammatory and anti-apoptosis efficacies are probably related to inhibition of COXs-induced P53/Fas pathway. These findings provide evidence for cardiac protective efficacy of QSYQ and validate the beneficial effects of QSYQ in the clinical application for HF.

**GW26-e1037**

Tanshinones IIA Attenuated Oxidative Stress and Restored Balance between Pro- and Anti-inflammatory Cytokines in Hypothalamic Paraventricular Nucleus Contribute to Sympathoexcitation in Chronic High-Salt Intake Induced Hypertension

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**OBJECTIVES**

In this study, arterial blood pressure (BP) of the rats, renal sympathetic nerve activity (RSNA) and the expression of gp91phox, angiotensin-converting enzyme (ACE), interleukin (IL)-beta (IL-1β), interleukin (IL)-6 (IL-6) in the PVN had been determined. We conducted to assess whether Tan IIA attenuates oxidative stress in the hypothalamic paraventricular nucleus (PVN) and contributes to the high-salt induced hypertensive response. The results from this study will lead to a better understanding of the disease process and aid in designing new therapeutic strategies for the treatment of hypertension.

**METHODS**

Thirty two Sprague-Dawley rats which were 7 weeks old with baseline body weights between 150 and 170 grams fed with a normal salt (0.3% NaCl) (NS) or a high salt (6% NaCl) (HS) diet for 16 weeks. Meanwhile half of each team received Tan IIA administration or vehicle by intragastric administration. Arterial pressure was determined every 2 weeks by a tail-cuff occlusion and their recording system which was noninvasively. After 16 weeks, the rats were decapitated while were under anesthesia. The PVN tissue samples were collected and stored at –80 °C for later analyses. We performed the following experimental procedures: Western blot analysis, immunofluorescence, immunofluorescence and statistical analysis.

**RESULTS**

There was a blunted increase in arterial blood pressure (BP) of high-salt fed also induced an increase in renal sympathetic nerve activity (RSNA) compared with control rats. Reductions in MAP and RSNA values elicited by Tan IIA administration. Expression of gp91phox and DHE were markedly higher in high-salt fed groups when compared to normal-salt fed groups. In addition, angiotensin-converting enzyme (ACE), interleukin (IL)-beta (IL-1β), interleukin (IL)-6 (IL-6) in the PVN were also increased but interleukin-10(IL-10) was decreased in high-salt induced hypertensive rats. However, Tan IIA administration attenuated above changes in hypertensive rats in the immunohistochemical studies.

**CONCLUSIONS**

High-salt administration with concomitant Tan IIA treatment acts upon neurons in PVN to reduce arterial pressure and renal sympathetic nerve activity, perhaps by reducing the influence of reactive oxygen species in the region. Tan IIA may be used as a nature antioxidant to attenuated Oxidative Stress via modulating proinflammatory cytokines and reducing component of RAS in the PVN.

**GW26-e1078**

A Molecular Switch Regulating Heart and Pancreatic Development

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**OBJECTIVES**

Mesoderm is thought to be a population of the intermediate and transient common progenitors prior to the emergence of the mesoderm (Me, the parental progenitor population of heart) and the endoderm (En, the parental progenitor population of pancreas) in vitro; however, it is unclear whether it exists in vivo, and if so, what are the molecular mechanisms that establish the subsequent cell fate differentiation. The purpose of this study is to delineate the stem cell origin of the heart and the pancreas in vivo, and the
role of basic Helix-Loop-Helix transcription factor (TF) Mesp1 in the common progenitors in vivo regulating heart and pancreas formation.

**METHODS** We did in vivo cell lineage tracing and conditional spatial-temporal gene knockout by using genetically engineered Cre/loxp mouse model, in which the loxp-site flanked genes were ablated from the genome by DNA recombination Cre. We generated chimeric embryos and traced the progeny of EF1α-MCM6-GFP (Mesp1-expressing cells) in the presence or absence of Mesp1. We used double/triple immunohistochemistry staining on mouse embryos and tissues for characterizing the progeny of Mesp1+ cells in vivo. We generated a doxycyclin (Dox)-inducible Mesp1 overexpression embryonic stem cell (ESC) line to determine the role of Mesp1 in mesendoderm formation. We also used RNA-Seq and chromatin-immunoprecipitation Sequencing (ChiP-sequencing) to identify the direct molecular targets of Mesp1 regulating the mesendoderm formation.

**RESULTS** We showed in mouse that Mesp1 is transiently expressed in a subset of the epiblast. Lineage tracing revealed the contribution of the Mesp1+ progenitors to both the mesoderm (heart) and the endoderm (pancreas), suggesting that Mesp1 marked the bio-potent mesendodermal progenitors. Mesp1-fated En exclusively gave rise to the Pdx1+/Foxa2+ progenitors in the mesendoderm, suggesting that Mesp1 marked the bi-potent mesoderm by inhibiting the endodermal transcription program in a cell-autonomous manner.

**CONCLUSIONS** Our results demonstrated that Mesp1 marks the bi-potent mesendoderm progenitors and acts as a molecular switch between the heart and the pancreas formation via an orchestrating dual molecular mechanism.

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**GW26-e1516 The Effects of Nox4 on Myocardial Apoptosis in Coxsackievirus B3 Induced Myocarditis**

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**OBJECTIVES** Viral myocarditis is a serious disease but whose pathogenesis has not been completely elucidated. Some studies have shown that Nox4 possibly involves in cardiovascular disease through mediating apoptosis. Therefore, we aim to research the effects of Nox4 in myocardial apoptosis in Coxsackievirus B3 infected mice.

**METHODS** Seventy male BALB/c mice were randomly divided into control, CVB3, and DPI group. Control group (n=10) were intraperitoneally injected with 0.1ml blank saline. CVB3 group (n=30) were intraperitoneal injected with 10^{7}TCID_{50} CVB3 0.1ml only for one day. DPI group(n=30) were injected 1mg/kg DPI 0.1ml firstly, injected with CVB3 0.1ml 1h later, and then injected with DPI for every day. Each CVB3 and DPI group was raised in single cage (n=10). To test serum content of TGF-β1 in patients with acute myocardial infarction (AMI), 20 cases (group C) healthy normal, from September 2012 to February 2012 in our hospital. The blood from the above disease attacks 6 h, 24 h, 48 h, using enzyme linked immunosorbent assay (ELISA) respectively to detect the concentration of TGF-β1.

**RESULTS**

1. Pathological changes of DPI group were alleviative that compared with the CVB3 group. The difference of pathological score among the three groups had statistical significance (p < 0.05).

2. Results of Western blot: compared with the control group, the expression of Nox4 was increased significantly. That is the expression of Nox4 began to increase on the 4thd and reached its crest on the 7thd (p<0.05). While decreasing in DPI group but was still higher than that in the normal group. The trend of expression of Cyt C was the same to Nox4. Compared with the CVB3 group, the expression of both Nox4 and Cyt C in DPI group had the same trend(p<0.05).

3. Results of scanning electron microscope: CVB3, 4thd group: mitochondrial were swelling and whose density was increased. CVB3, 7thd group: mitochondrial got more swelling and there appeared karyopyknosis, karyolysis and apoptotic bodies.

4. Results of flow cytometry: in control group, there was no myocardial apoptosis. CVB3 group had significant myocardial apoptosis. The 7thd group mainly showed early apoptosis and the 14thd group mainly late apoptosis. The amount of myocardial apoptosis in DPI group was significantly reduced compared with the CVB3 group (p<0.05).

**CONCLUSIONS** (1) The high expression of Nox4 was associated with viral myocarditis. (2) Nox4 possibly involved in viral myocarditis through mitochondrial/ Cyt C pathway mediated cardiomyocyte apoptosis.