were upregulated in the model group, indicating the downstream activation of RAAS system.

After intervention of QSKL, renin and angiotensin II level in the QSKL group decreased dramatically compared with model group, suggesting the inhibition of RAAS system. Pathological changes detected by Sirius red and TUNEL staining in QSKL group were not as significant as those in model group. Compared with model group, the expressions of a-SMA and MMP9 were decreased while TIMP-1 were upregulated in QSKL group, indicating QSKL can prohibit the fibrotic process. The mRNA expressions of LIMK1, LIMK2, NHE, moesin and cofilin in QSKL group were down regulated compared with model group, which showed the downstream inhibition of key molecules in RAAS system.

CONCLUSIONS QSKL has definite anti-fibrotic effect in inhibiting RAAS system mainly through attenuating the serum renin and angiotensin II, down-regulating myocyte apoptosis and collagen genesis, besides inhibiting the downstream key molecules of RAAS system. These findings provide evidence for anti-fibrotic effect of QSKL and validate the benefit of QSKL in the clinical application for myocardial fibrosis.

GW28-e0628

Implication of peripheral blood miRNA-124 in predicting acute myocardial infarction

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OBJECTIVES This study aimed to determine the expression of miR-124 in the patients with acute myocardial infarction (AMI) and elucidated the role of miR-124 on early diagnosis of AMI.

METHODS A total of 90 AMI patients were recruited, along with 45 healthy individuals as the control group. Blood samples were collected at different time points (0 h at admission, 6 h, 12 h and 24 h of disease onset). Real-time PCR was used to test miRNA-124 level. ELISA was used to test serum troponin (cTnI) and creatine kinase-MB isoenzyme (CK-MB) levels. The correlation between miRNA-124, cTnI and CK-MB was analyzed. Receiver operating characteristic curve (ROC) was used to analyze sensitivity and specificity of AMI.

RESULTS MiRNA-124 expression in experimental group was significantly elevated in peripheral blood of AMI patients. It can reach the peak at 6h after onset. AMI patients had significantly elevated cTnI and CK-MB expression level ($p<0.05$ compared to control group). The expression of miRNA-124 reached the peak earlier than cTnI and CK-MB. miRNA-124 was positively correlated with cTnI and CK-MB ($p<0.05$). The area under the curve of ROC of miRNA-124 was 0.86 (95% CI: 0.815-0.937), with 52% sensitivity and 91% specificity.

CONCLUSIONS AMI patients presented a significantly elevated level of miRNA-124 in peripheral blood. Our data suggested that miR-124 contributed to an earlier detection than other diagnostic markers for AMI. Therefore, peripheral miRNA-124 can serve as a novel biological marker for early diagnosis of AMI.

GW28-e0629

Interleukin 37 attenuates osteo-fibrotic responses mediated by the advanced glycation end-products modified low density lipoprotein in human aortic valve intersitial cells

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OBJECTIVES Calcific aortic valve disease is one of leading cardiovascular diseases, and pharmacological intervention is currently unavailable. Increasing number of studies implicate aortic valve interstitial cells (AVICs) play a critical role in the pathogenesis of calcific aortic valve disease. IL-37 is recently found to be a novel anti-inflammatory cytokine. The preliminary experiments showed that the accumulation of advanced glycation end-products modified low density lipoprotein (AGE-LDL) within the human stenotic aortic valve, mainly surrounding calcified areas, were associated with the severity of aortic valve stenosis. It is unknown whether IL-37 suppresses the osteo-fibrotic responses mediated by AGE-LDL in human AVICs. Whether IL-37 promotes osteo-fibrotic responses mediated by AGE-LDL in human AVICs remains unknown. We aim to investigate the effect of IL-37 on the modulation of osteo-fibrotic responses mediated by AGE-LDL in human AVICs.

METHODS A total of 10 patients were enrolled in our study. Interleukin 37, AGEs production and TLR4/RAGE receptors were detected with immunohistochemistry in the collected aortic valve specimens. Furthermore, AVICs were seeded and subjected to AGE-LDL stimulation for 48 hours. Alkaline phosphatase (ALP), bone morphogenetic protein-2 (BMP-2), α-smooth muscle actin (α-SMA), human transforming growth factor-β1 (TGF-β1) and collagen type I (Collagen I) expression were examined by immunoblotting. The involved signals pathway was also detected via immunoblotting. Finally, we investigate the osteo-fibrotic responses induced by AGE-LDL after pre-treatment recombinant interleukin 37 in human AVICs.

RESULTS Human diseased aortic valves exhibit greater levels of the advanced glycation end-products modified low density lipoprotein (AGE-LDL) and lower levels of interleukin 37(IL-37). Stimulation of HAVICs with AGE-LDL up-regulates the expression of BMP-2, ALP, TGF-β1, α-SMA, as well as activation of NF-kB signaling pathway. Inhibition of NF-kB suppressed the expression of osteo-fibrotic proteins induced by AGE-LDL. Also, TLR4 and RAGE were detected by immunohistochemistry and immunoblots in human AVICs, and knockdown of TLR4 and RAGE reduced osteo-fibrotic responses induced by AGE-LDL. Moreover, pre-treatment of human AVICs with recombinant human IL-37 suppresses the levels of osteo-fibrotic responses as well as calcium deposit formation mediated by AGE-LDL. IL-37 suppresses AVICs osteo-fibrotic responses through inhibition of NF-κB and TLR4/RAGE.

CONCLUSIONS Our results revealed that AGE-LDL induced osteo-fibrotic response through activation of NF-kB pathway. Furthermore, IL-37 attenuates the osteo-fibrotic responses mediated by AGE-LDL in human AVICs through modulation of NF-kB activation. Recombinant IL-37 might have therapeutic potential for prevention of progression in calcific aortic valve disease.

GW28-e0631

X-box binding protein 1 blocks FFA-induced NF-κB pathway in 3T3-L1 adipocytes

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OBJECTIVES Adipose tissue inflammation is closely linked with the development of insulin resistance, heart disease, diabetes and cancer. The nuclear factor-κB (NF-κB) p65 pathway is one of the most inflammation signaling pathway, moreover, free fatty acid (FFA) is one of the most important inducements to NF-κB pathway in adipose tissue inflammation. It is well known that exposure of 3T3-L1 adipocytes to FFA is a strong inducement to NF-κB pathway. Activation of IκB kinase (IKK) is a critical step that lead to NF-κB activation. IKK phosphorylates the NF-κB regulatory protein IκB, resulting in the release of NF-κB for nuclear translocation. In addition, IKK directly phosphorylates p65 subunit of NF-κB Ser^{536}, Ser^{276}, and Ser^{468}, enhancing the transcriptional activity of NF-κB. Activated X-box binding protein 1 (XBP1) is a key transcription factor which protects injured cells by modulate the function of unfolded protein response (UPR). This study is to investigate whether spliced XBP1 blocks FFA-induced NF-κB pathway in 3T3-L1 adipocytes.

METHODS Small RNA interference (siRNA) was used to achieve knock-down of XBP1. The knock-down efficiency was tested by measuring the protein level of spliced XBP1 using western blotting. Briefly, 3T3-L1 adipocytes were transfected with scramble siRNA or XBP1 siRNA using Lipofectamine 2000 for 6 h. Overexpression of spliced XBP1 was achieved by using adenovirus encoding mouse spliced XBP1 (Ad-XBP1s) in cultured 3T3-L1 adipocytes. The effect of overexpression by spliced XBP1 was examined by western blot analysis. Adenoviruses encoding green fluorescent protein (Ad-GFP) were used as negative control. 3T3-L1 adipocytes were infected with Ad-GFP or Ad-XBP1 for 48 h, and then were stimulated in the presence or absence of FFA (0.5 mM) for 4 h.

Phos-phorylation of IKK, phos-phorylation IκB at Ser^{32}, and a phosphorylation of NF-κB p65 at Ser^{536} induced by FFA were determined at the protein levels by western blot analysis to measure the activation of NF-κB pathway.

RESULTS Down-regulation of XBP1 by transfection with XBP1 siRNA increased phos-phorylation of IKK, phos-phorylation IκB at Ser32, and phosphorylation of NF-κB p65 subunit at Ser^{536} levels in 3T3-L1 adipocytes. In parallel, up-regulation of XBP1 by infection with Ad-XBP1 inhibited phosphorylation of IKK, phosphorylation IκB at Ser32, and