level of CHOP, GRP78, GRP94, IRE1α, and ATF6) were assessed by both Western Blot and immunohistochemical staining. SIRT1 signal and its substrates were also detected by Western Blot

RESULTS Our data showed that LXR agonist protected against CLP-induced myocardial injury and dysfunction, as evidenced by improved myocardial morphological changes, improved hypotension, reduced serum concentration of biochemical indices of heart injury(AST, LDH, CK and CK-MB), and elevation of cardiac functional parameters(LVEF 55.6±2.2% vs. 64.1±3.4%, respectively, N=11-14, p=0.0005). Furthermore, we found that LXR agonist reduced the level of inflammatory cytokines, such as TNF-α, IL-1β, IL-6 and HMGB1, the overproduction of ROS and MDA, and the accumulation of endoplasmic-reticulum stress. Additionally, LXR agonist increases the activity of antioxidant enzymes, such as SOD, GPx, and CAT. However, these aforementioned beneficial actions of LXR agonist administration following CLP was abolished in cardiac specific SIRT1 KO mice, suggesting the protective effects of LXR agonist is associated with SIRT1 signaling activation. Western Blot analysis of SIRT1 signal and its substrates further revealed that T0901317 enhanced SIRT1 signaling and subsequent deacetylation and activation of anti-oxidative FoxO1 and anti-ER stress HSF1, deacetylation and inhibition of pro-inflammatory NF-KB and pro-apoptotic P53, thereby alleviating sepsis-induced myocardial injury and dysfunction.

CONCLUSIONS In conclusion, our current work demonstrated a beneficial role of LXR agonist in septic murine heart mainly through attenuating oxidative stress, endoplasmic-reticulum stress, inflammation, and apoptosis. Furthermore, by employing SIRT1-/- mice, we demonstrated that the beneficial actions of LXR agonist in septic heart were possibly associated with activation of SIRT1 signaling and subsequent deacetylation and activation of FoxO1 and HSF1, as well as deacetylation and inhibition of NF-KB and P53. Those findings may guide the prospective clinical trial to evaluate the latent therapeutic effect of LXR agonist for patients with sepsis.

GW28-e0617

Angiotensin II increases the small conductance calcium-activated potassium channels in rat atrial myocytes

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OBJECTIVES Renin-angiotensin system is known to induce the development of atrial fibrillation through atrial structural and electrical remodeling. small conductance calcium-activated potassium channels (SK channels) are close association with development and progression of atrial fibrillation. SK2 is the main subunits of SK channels. However, the effect of Angiotensin II on SK channels remain unknown.

METHODS Male rats were subcutaneously infused with AngII 200ng/kg/min using Alzet Osmotic minipumps. After administration for 14 days to measure the mRNA and protein expression levels of SK2 by Western blotting and qRT-PCR. SK2 expression levels were measured by administration with Ang II, Valsartan or PD123319 for 72 hours in neonatal rat myocytes. SK2 was measured under conditions SK2 co-transfected with AT1R or AT2R in HEK293T. SK2 interaction with AT1R or AT2R was detected by CO-IP. SK2 trafficking were measured by flow cytometry in HEK293T.

RESULTS AngII concentration dependently upregulated SK2 mRNA expression level and SK2 protein expression level in adult rat atrial and neonatal rat atrial myocytes. SK2 exists a close protein-protein interaction with and AT2R in HEK293T cells. AngII increased SK2 expression through AT1R. Angiotensin II induced SK channels trafficking from intracellular to cell-surface membrane expression.

CONCLUSIONS The findings revealed that Ang II increases SK2 expression through AT1R by SK2 trafficking from intracellular to the plasma membrane in atrial myocytes, which involving in electrical remodeling and provides arrhythmic substrate.

GW28-e0620

Late diagnosis of Severe Delayed Postpartum Preeclampsia with a Rare CACNA1D Mutation

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OBJECTIVES Preeclampsia (PE) is the most representative type of hypertensive disorders complicating pregnancy. It affects approximately 5%–9% of pregnancies and it is responsible for major maternal and perinatal morbidity and mortality. Since the etiology and pathogenesis of PE is still unknown, further research is needed. L-type calcium channels (LTCCs; Cav1) are one of the three major classes (Cav1-Cav3) of voltage-gated calcium channels. Blocking of LTCCs in vascular smooth muscle and the heart has been therapeutically used for decades to treat elevated blood pressure and cardiac ischemia. The human wild-type Cav1.3 channel α1-subunit (CACNA1D gene) mutation was reported in aldosterone-producing adenoma.

METHODS We delayed diagnosed severe postpartum PE in an early-onset hypertension Chinese female. To screen for the responsible gene mutation, 43 monogenic hypertension related pathogenic gene encoding area and flank area were tested with the sequencing technology in the proband, 5 other hypertension patients, as well as 4 normal individuals from the family.

RESULTS A 29-years-old Chinese female who were diagnosed hypertension 10 years ago readmitted 2 days after the caesarean surgery. Her blood pressure was controlled by methyldopa 0.25g*3 /day during pregnancy and her blood and urine test results were within normal ranges during pregnancy. Her blood pressure and lab results showed no abnormal before and during the caesarean surgery. Two days after delivered a health boy, she complained of chest pain and had hypertension (160/100mmHg). Blood laboratory tests revealed significant raised Troponin I(2.00/mL), serum creatinine(511umol/L), blood uric acid(823umol/L), serum cystatin c, white blood cell count, C-reactive protein, prohormone brain natriuretic peptide (proBNP: 8516.00pg/ml) levels with urine protein 2+ and Urine red blood cells 3+. Renal biopsy showed mild mesangial proliferative glomerulonephritis with acute tubular injury. After treatment and undergoing oliguria stage and diuretic phase, her creatinine as well as other blood and urine test results returned to normal and blood pressure was controlled to 135-125/75-85mmHg using methyldopa 0.25g*3 /day. After reviewing this case, we delayed diagnosed her chronic hypertension with severe delayed postpartum PE which was not discovered by her obstetricians, cardiologists or nephrologists. We found a rare CACNA1D gene p.D307G missense mutation in the proband, so we tested 5 other hypertension patients, as well as 4 normal individuals from the family. We found among these 10 family members, nine are carrying this gene mutation with 6 hypertension patients, who were the proband, her mother, twin sister, younger brother and her mother's two brothers.

CONCLUSIONS Delayed Postpartum PE is rare but potentially life-threatening, it needs attention from medical professionals, not just obstetricians. CACNA1D gene p.D307G mutation could be a risk mutation of early-onset hypertension or preeclampsia, it might lead to new diagnostic or therapeutic approach of these diseases.

GW28-e0627

QSKL attenuates myocardial fibrosis in rats by inhibiting key molecules of RAAS system

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OBJECTIVES In this study, we aimed to explore if QSKL could exert an anti-fibrotic effect by attenuating key molecules of RAAS system.

METHODS Sprague-Dawley (SD) rats were randomly divided into 4 groups: sham group, model group (left anterior descending coronary artery ligation), positive control group (LAD ligation and treated with fosinopril) and QSKL group (LAD ligation and treated with QSKL). 28 days after surgery, radioimmunoassay was used to detect the renin and angiotensin II level in serum. Sirius red staining were performed to assess the degree of myocardial fibrosis. Transferase-mediated deoxyuridine triphosphate-biotin nick end labeling (TUNEL) staining were performed to observe the degree of myocyte apoptosis. Alpha smooth muscle actin (a-SMA), matrix metalloproteinase 9 (MMP-9) and tissue inhibitor of metalloproteinase 1 (TIMP-1) were assessed by immunohistochemically method. Real-time PCR was used to detect the mRNA expressions of LIMK1, LIMK2, NHE, moesin and cofilin.

RESULTS At 28 days after surgery, radioimmunoassay showed that renin and angiotensin II level in the model group increased significantly, indicating the activation of RAAS system. Sirius red and TUNEL staining results demonstrated pathological changes of myocyte apoptosis and myocardial fibrosis. Expressions of fibrotic markers, such as a-SMA and MMP9, were elevated while TIMP-1 were downregulated in model group. LIMK1, LIMK2, moesin and cofilin

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