ABSTRACTS

Abstracts for Chaired Posters:

CP1.

C-REACTIVE PROTEIN TRENDS AND ASSOCIATIONS WITH TOTAL MORTALITY IN UNITED STATES ADULTS BETWEEN 1999-2010

KL Ong,[1,2] MA Allison,[3] BM Cheung,[4] BJ Wu,[1] PJ Barter,[1,2] KA Rye[1,2,5]
[1]Lipid Research Group, Heart Research Institute; [2]Faculty of Medicine, University of Sydney, Sydney, Australia; [3]Department of Family and Preventive Medicine, University of California San Diego, La Jolla, CA, United States; [4]Department of Medicine, The University of Hong Kong, Hong Kong; [5]Department of Medicine, University of Melbourne, Melbourne, Australia

Introduction: C-reactive protein (CRP) is a well-known biomarker of systemic inflammation. Elevation of CRP levels is predictive of incident cardiovascular disease morbidity and mortality. We investigated the trends in prevalence of elevated CRP levels (>3.0 mg/L), as well as mean CRP levels, in a general population of United States adults and then determine the associations with mortality.
Methods: Data from the 27,214 subjects aged ≥20 years in the National Health and Nutrition Examination Survey (NHANES) 1999-2010 were analyzed. Among them, 12,259 subjects from NHANES 1999-2004 had mortality data followed up through December 31, 2006.
Results: After adjusting for age, sex, race/ethnicity, body mass index, and medications for lowering blood pressure, glucose and lipids, the prevalence of elevated CRP decreased significantly from 36.7% in 1999-2002 to 32.0% in 2007-2010, corresponding to a decrease in mean CRP levels from 1.92 to 1.66 mg/L (both P<0.001). The trend remained significant after additional adjustment for several traditional cardiovascular risk factors and use of medications, including statins, angiotensin-converting enzyme inhibitors, metformin, aspirin and clopidogrel. However, the decreasing trends were attenuated after additional adjustment for total bilirubin (P=0.076 and 0.024), which increased from 0.62 to 0.73 mg/dL over 12 years (P<0.001). Of note, higher CRP levels were modestly associated with a higher risk of total mortality (adjusted hazard ratio: 1.07 [95%CI, 1.03-1.12] per 10 mg/L increase, P=0.001), and this association tended to be stronger in subjects with higher total bilirubin levels (P for interaction <0.001).
Conclusion: The decreasing trend of CRP levels is encouraging and may be related to the increase in total bilirubin levels. Such trends may be explained in part by the increasing use of some medications such as statins that can increase bilirubin levels and decrease CRP levels. Moreover, the prediction of total mortality by CRP levels seems to be stronger in subjects with higher total bilirubin levels.

CP2.

ACTIVATION OF ANGIOTENSIN CONVERTING ENZYME-2 RESCUES ENDOTHELIAL FUNCTION BY OXIDATIVE STRESS REDUCTION IN DB/DB DIABETIC MICE

Y Zhang, J Liu, XY Tian, L Wang, WT Wong, CW Lau, Y Huang
Institute of Vascular Medicine, School of Biomedical Sciences, The Chinese University of Hong Kong, Hong Kong

Objective: Angiotensin converting enzyme-2 (ACE2), a new member of the vasoprotective axis of the renin-angiotensin system, is a pleiotropic exopeptidase that plays a regulatory role in cardiac and vascular physiology. The present study investigates whether ACE2 activation improves endothelial function in type 2 diabetic mice and explores the potential participating cellular events.
Methods: 12-week-old male db/db mice were orally administered with ACE2 activator diminazene aceturate (DIZE, 15 mg/kg/day) for 2 weeks. Wire myograph was utilized to measure isometric force of isolated aortae, while DHE and CM-H2DCFDA fluorescence were used to determine the reactive oxidative species (ROS) generation in aortae and endothelial cells. In addition, ACE2 activity was assayed by Mc-Ala/Dnp fluorescence resonance energy transfer (FRET) assay.
Results: Chronic treatments with DIZE in db/db mice were able to reduce systolic blood pressure and increase ACE2 activity without affecting the insulin and glucose tolerance. In vivo DIZE treatment restored the impaired endothelium-dependent relaxations (EDRs) in response to acetylcholine and lowered oxidative stress in aortae of diabetic mice. In addition, ex vivo experiments show that 24-hour treatment with DIZE (100 μgmol/L) not only improved EDRs in db/db mouse aortae, but also prevented the impairment of EDR by high glucose (30 mmol/L) or angiotensin II (Ang II, 500 nmol/L) in normal C57BL/6 mouse aortae. The beneficial effects of DIZE were counteracted by ACE2 inhibitor DX600 and Mas receptor blocker A779, and absent in ACE2 knockout mice. Furthermore, both ex vivo and in vitro treatment with DIZE normalized ROS over-production in the endothelium of db/db mouse aortae and in high glucose- or Ang II-treated human umbilical vein endothelial cells. Such effects of DIZE were also inhibited by ACE2 inhibitor and ACE2 siRNA transfection.
Conclusions: This study provides novel evidence that ACE2 activator can rescue the impaired endothelial function in diabetic mice largely through reducing the elevated oxidative stress in the endothelium. Our results suggest that activating ACE2-Ang(1-7)-Mas signaling could be another promising therapeutic option against diabetic vascular dysfunction. (Supported by Hong Kong GRF).

ABSTRACTS

Abstracts for Chaired Posters:

CP3.

ENDOTHELIN-1 CONTRIBUTES TO ENDOTHELIUM-DEPENDENT CONTRACTIONS IN ZUCKER FATTY RATS

J Liu, XY Tian, WT Wong, LM Liu, Y Zhang, L Wang, Y Huang
Institute of Vascular Medicine, School of Biomedical Sciences, The Chinese University of Hong Kong, Hong Kong

Objective: Cardiovascular disease is the leading cause of patients with metabolic syndrome and endothelin-1 (ET-1) is shown to be closely associated with cardiovascular disease. However, whether ET-1 participates in endothelial dysfunction in metabolic syndrome is not well established. The present study aims at investigating the pathological role of ET-1 in vasculopathy in metabolic syndrome.
Methods: Six-month old Zucker lean rats and Zucker fatty rats were orally treated with or without ET-1 receptor antagonist bosentan for one month and then sacrificed. Renal arteries were dissected out and endothelium-dependent contractions (EDCs) were measured in wire myograph. Some renal arteries were kept for Western blotting to detect the expression of ET type A receptor (ETAR), p38 AMPK and cyclooxygenase-2 (COX-2).
Results: EDCs in response to acetylcholine in L-NAME-treated arteries were augmented in Zucker fatty rats compared with Zucker lean rats and were suppressed following chronic bosentan treatment. Overnight incubation with bosentan, ETAR antagonist ABT627, p38 MAPK inhibitor SB203580 and COX-2 inhibitor NS398 all reduced EDCs in Zucker fatty rats. Moreover, overnight exposure to ET-1 exaggerated EDCs in arteries from Zucker lean rats, which was inhibited by ABT627, SB203580 or NS398. Western blotting showed an increased phosphorylation of p38 MAPK and up-regulation of COX-2 upon ET-1 treatment.
Conclusion: The present study provides showed a contributory role of ET-1 in endothelial dysfunction in Zucker fatty rats and the effect of ET-1 is likely mediated by ETAR/p38 MAPK/COX-2 signaling cascade. Our results highlight potential efficacy of ET receptor blockers in preserving endothelial function in metabolic syndrome (Supported by Hong Kong GRF).

CP4.

L*YCIUM BARBARUM* POLYSACCHARIDES MITIGATE CHRONIC INERMITTENT HYPOXIA INDUCED HIPPOCAMPAL OXIDATIVE STRESS, INFLAMMATION AND APOPTOSIS IN RATS

CS Lam,[1] GL Tipoe,[2] RCC Chang,[2] KF So,[2] KH Cheung,[1] ML Fung[1]
Departments of [1]Physiology and [2]Anatomy, The University of Hong Kong, Hong Kong

Background: Chronic intermittent hypoxia (CIH), mimicking severe conditions of obstructive sleeping apnea, induces oxidative stress, inflammation and apoptosis in the hippocampus. *Lycium barbarum* polysaccharides (LBP) are biological significant portions of traditional Chinese herbal medicine Goji, which possess anti-oxidative and anti-inflammatory properties.
Objectives: Our study aims to investigate the neuroprotective effect of LBP in CIH rats. We hypothesize that LBP ameliorate oxidative stress, inflammation and apoptosis in the hippocampus of rats exposed to CIH.
Methods: MDA assay was used to measure the lipid peroxidation extent, a marker of oxidative stress. Western blot was employed to examine the expressions level of antioxidant enzymes (SOD-1, SOD-2); inflammatory mediators (IL-1β], TNFα, COX-2); caspase-dependent extrinsic (FADD, caspase 8, Bid) and intrinsic apoptotic (Bax, Bcl2, cytochrome C) cell death (cleaved caspase 3) cascades; proliferative marker (PCNA) and trophic factor (BDNF). In situ cell death staining (TUNEL) was utilized to reveal the apoptotic situation of hippocampal subfields (DG, CA1 and CA3).
Results: LBP administrations remarkably attenuated the level of oxidative stress, inflammation and apoptosis in the hypoxic group. In addition, LBP significantly mitigated caspase-dependent apoptotic extrinsic and intrinsic (Bax, Bcl2, cytochrome C) cascades in the hypoxic group. Surprisingly, LBP increased the expression of proliferative marker (PCNA) and trophic factor (BDNF) in the hypoxic group.
Conclusion: LBP are neuroprotective against CIH-induced hippocampal injury and may facilitate CIH-induced neurogenesis.

ABSTRACTS

Abstracts for Chaired Posters:

CP5.

CARDIOPROTECTIVE EFFECTS OF TOTAL FLAVONES OF ELSHOLTZIA SPLENDENS ON ANOXIA/REOXYGENATION INDUCED INJURY IN H9c2 RAT CARDIOMYOBLAST CELLS

Z Li, L Zhao, LB Qian, ZG Ye, Q Xia, HP Wang
Department of Physiology, Zhejiang University School of Medicine, Hangzhou, China

Objective: The present study was performed to investigate the effect of total flavones of Elsholtzia splendens (TFES) in the H9c2 rat cardiomyoblast cell line subjected to anoxia/reoxygenation and the underlying mechanism.
Methods: H9c2 cells were pretreated with or without TFES (0.001, 0.01, 0.1 mg/ml) for 4 h, and then were cultured in the medium pre-saturated with 5% CO_2 + 95% N_2 for 6 h to simulate anoxia. The incubator chamber was ventilated with 5% CO_2 + 95% N_2 during the whole anoxia. After that, cells were re-oxygenated in normal medium with 95% O_2 + 5% CO_2 for 2 h. At the end of reoxygenation, the cell viability was assayed using MTT method. Cell lactate dehydrogenase (LDH) release, total-superoxide dismutase (T-SOD) activity and malondialdehyde (MDA) level were determined using commercial kits.
Results: Administration with TFES (0.001, 0.01, 0.1 mg/ml) produced a dose-dependent reduction in the cell death, LDH release and MDA production induced by anoxia/reoxygenation in H9c2 cells. In addition, the inhibition of T-SOD activity induced by hypoxia/reoxygenation was markedly reversed by TFES (0.01, 0.1 mg/ml) but not by 0.001 mg/ml TFES.
Conclusion: Treatment with TFES (0.01, 0.1 mg/ml) for 4 h protected H9c2 cells against anoxia/reoxygenation induced injuries, which may be related to enhancing SOD activity and reducing oxidative stress.

CP6.

HAY FEVER AND HYPERTENSION IN THE US ADULT POPULATION

C Li, CL Cheung, TT Cheung, NR Samaranayake, BMY Cheung
Department of Medicine, The University of Hong Kong, Hong Kong

Purpose: Hypertension is associated with inflammation. Whether the inflammation caused by allergic diseases such as allergic rhinitis can predispose to hypertension is controversial. Therefore, we studied the association between hay fever and hypertension in the United States National Health and Nutrition Examination Survey (NHANES).
Methods: We analyzed data on 1883 men and 2029 women in NHANES 2005-2006. We included participants aged 20 years or older who had valid data on hay fever and hypertension.
Results: 13.5% of the participants had a previous diagnosis of hay fever and 36.2% of them had hypertension. There were ethnic differences in the prevalence of previous hay fever diagnosis (P<0.001) and hypertension (P<0.001). There was no significant association between previous hay fever diagnosis and hypertension in men in any age group. The association between previous hay fever diagnosis and hypertension in women was significant in those aged 20-39 years (OR=2.59, 95%CI=1.26-5.30, P=0.013). The association between previous hay fever diagnosis and hypertension in women aged form 20-39 years remained significant after adjustment for age, race and body mass index (OR=2.74, 95%CI=1.48-5.06, P=0.003). After further adjustment for physical activity, alcohol consumption and smoking, the association was not attenuated (OR= 2.68, 95%CI=1.38-5.18, P=0.006). Further adjustment for liver enzymes, C-reactive protein and IgE level attenuated the association slightly (OR=2.72, 95%CI=1.19-6.22, P=0.021).
Conclusions: In this nationally representative population-based survey, previous hay fever diagnosis is not significantly associated with hypertension in adults except for young women aged 20-39. Further work is needed to confirm that this is a true association.

CP7.

TELMISARTAN AMELIORATES OXIDATIVE STRESS AND APOPTOSIS IN THE LIVER OF RATS EXPOSED TO CHRONIC INTERMITTENT HYPOXIA

JF Pan,[1] J Xiao,[2] GL Tipoe,[2] Y Huang,[3] ML Fung[1]
Departments of [1]Physiology and [2]Anatomy, Li Ka Shing Faculty of Medicine, The University of Hong Kong; [3]School of Biomedical Sciences, The Chinese University of Hong Kong, Hong Kong

Background and Hypothesis: Recurrent cycles of severe hypoxia and reoxygenation are a hallmark feature of obstructive sleep apnea (OSA) syndrome, resulting in chronic intermittent hypoxia (IH) that causes excessive production of reactive oxygen species and oxidative stress in tissues and organs. It has been shown that blockade of angiotensin (AT) receptors alleviated the IH-induced systemic hypertension. However, the mechanistic involvement of AT receptors in the pathogenesis of liver injury under chronic IH condition is currently unknown. We hypothesized that the expression of NADPH oxidase induced by activation of AT receptor type 1 plays a role in the IH-induced oxidative stress and apoptosis in the rat liver.
Methodology: Adult Sprague-Dawley rats were exposed to air for normoxic (Nx) control or to normobaric oxygen levels alternating between 5-21% 8 hr/day for IH treatment for 2 weeks. Rats were fed with an AT1 receptor blocker telmisartan (10 mg/kg body weight), or vehicle daily before the hypoxic treatment. The mRNA levels of NADPH oxidase subunits (p22-phox and NOX-4), the pro-apoptotic genes (BAX and FAS) and anti-apoptotic gene (Bcl-2) were examined by RT-PCR; the cell apoptosis was detected by TUNEL assay; also the level of oxidative stress with malondialdehyde (MDA) in the liver.
Results: Our results showed that the MDA level and the mRNA levels of p22-phox, NOX-4, BAX, FAS and also the amount of TUNEL-positive cells were significantly increased in the hypoxic group, when compared with the Nx and telmisartan-treated hypoxic (TIH) groups. In addition, the expression of anti-apoptotic gene Bcl-2 was decreased significantly in the hypoxic group than those of the Nx and TIH groups.
Summary and Conclusion: Blockade of the AT1 receptor with telmisartan mitigates oxidative stress and apoptosis in the liver of rats exposed to chronic IH, thus supporting a pathogenic role of NADPH oxidase induced by AT1 receptor activation in the hepatic injury in chronic IH resembling a severe OSA condition.