Proceedings of the British Pharmacological Society at http://www.pA2online.org/abstracts/Vol12Issue3abst055P.pdf

Pre-existing chronic kidney disease worsens the myocardial dysfunction caused by sepsis in mice

J Chen[1], J Kieswich[1], F Chiazza[2], N Patel[1], M Collino[2], M Yaqoob[1], C Thiemermann[1].
[1]Queen Mary University of London, London, UK, [2]University of Turin, Turin, Italy

Patients with chronic kidney disease (CKD) requiring dialysis have a higher risk of infection and sepsis (1) due to uremia-induced immune deficiency, significant co-morbidities and the dialysis procedure itself. Dialysis patients with sepsis have a 100-fold higher mortality than other patients with sepsis (2). The severity of myocardial dysfunction is an important predictor of morbidity and mortality among patients with sepsis (3). However, it is unknown whether pre-existing CKD worsens the cardiac outcome in animals or man with sepsis. This study was designed to investigate the effects of lipopolysaccharide (LPS) administration or cecum ligation and puncture (CLP) on cardiac performance in mice with CKD.

Male C57BL/6 mice (age: 4-6 weeks) were subjected to a two-stage, subtotal (5/6th) nephrectomy (SNX). After 8 weeks, mice with SNX/CKD were subjected to either low dose LPS (2mg/kg i.p.) administration or CLP. At 18 hours after LPS injection or 24 hours after CLP, cardiac function was evaluated by echocardiography. The following signal transduction events were evaluated in the heart of all animals by Western blot analysis: (a) Phosphorylation of the inhibitor of κBα (IκBα), (b) the nuclear translocation of the nuclear factor κB (NF-κB) subunit p65, (c) expression of iNOS, (d) phosphorylation of Akt and (e) phosphorylation of ERK1/2. All data is represented as mean ± standard error of the mean (SEM). Data was analyzed either by using unpaired Student's t-test (for comparisons between two groups) or by one-way ANOVA followed by Bonferroni's post hoc test for multiple comparisons. A p-value of $p<0.05$ was considered significant.

When compared to age-matched sham mice, SNX for 8 weeks resulted in a significant rise in urea (from 8.17 ± 0.41, n=12; to 17.43 ± 0.61 mmol/L, n=14; $p<0.05$) and creatinine (from 29.72 ± 0.38, n=12; to 45.96 ± 1.76 μmol/L, n=14; $p<0.05$), indicating development of CKD. Moreover, when compared to sham mice, mice with CKD showed a slight, but significant, decrease in ejection fraction (EF) (from 72.78 ± 0.56, n=11; to 65.25 ± 0.89%, n=23; $p<0.05$). In sham mice without CKD, the intraperitoneal administration of low dose LPS (2 mg/kg) for 18 hours or CLP for 24 hours had no effect on EF (70.69 ± 1.24 and 72.26 ± 0.89%, respectively, n=6-7; $p>0.05$). In contrast, administration of low dose LPS (2 mg/kg) or CLP in mice with CKD caused profound reductions in EF (to 40.38 ± 2.77 and 41.74 ± 1.31%, respectively, n=7 per group; both $p<0.05$). Interestingly, compared with sham mice, western blot analysis of the myocardium from mice with CKD revealed profound increases in phosphorylation of IκBα, nuclear translocation of the NF-κB subunit p65, increased expression of iNOS protein, and significant increases in phosphorylation of Akt and ERK1/2. Moreover in mice with CKD, administration of low dose LPS or CLP further increased the phosphorylation of IκBα, the nuclear translocation of the NF-κB subunit p65 and the expression of iNOS. In contrast, administration of LPS or

CLP to mice, which did not have CKD, had no significant effect on any of the above signalling pathways.

In conclusion, the presence of CKD aggravates the cardiac dysfunction caused by LPS or CLP in the mouse. The observed increase in the severity of cardiac dysfunction in CKD mice with sepsis was associated with and may (at least in part) be due to increased cardiac activation of NF-κB and increased expression of iNOS, both of which are known to contribute to the cardiac dysfunction caused by sepsis (4, 5).

(1) Collins AJ *et al*. (2011). *Am J Kidney Dis* **57**: A8, e1-526.

(2) Sarnak MJ and Jaber BL (2000). *Kidney Int* **58 (4)**: 1758-1764.

(3) Blanco J *et al*. (2008). *Crit Care* **12 (6)**: R158.

(4) Khan AI *et al.* (2013). *Dis Model Mech* **6 (4)**: 1021-1030.

(5) Joe EK *et al.* (1998). *J Mol Cell Cardiol* **30 (2)**: 303-315.