THE EFFECT OF ERYTHROPOIETIN ON THE EXPRESSION OF CYTOCHROME C AND FAS/FASL IN AN EXPERIMENTAL ACUTE RENAL ISCHEMIA/REPERFUSION MODEL

K. Xanthopoulou[1], I. Tsouchnikas[1], G. Ouzounidis[1], G. Kokaraki[2], R. Lagoudaki[2], C. Simeonidou[2], G. Karkavelas[3], E. Spandou[2], D. Tsakiris[1].

[1]Department of Nephrology, General Hospital of Veria, [2]Department of Physiology and [3]Department of Pathology, Aristotle University of Thessaloniki, Greece.

INTRODUCTION AND AIMS

The role of cytochrome c (cyt c) and Fas/FasL in apoptosis are well established. However, their participation in apoptotic signaling pathways in acute kidney ischemia-reperfusion (I/R) injury are not completely understood. Erythropoietin (EPO), through its antiapototic action, was found to be renoprotective in experimental models of I/R-induced acute kidney injury (AKI).

The aim of the study was to investigate the effect of EPO on the expression of cyt c and Fas/FasL in an experimental model of I/R AKI at different time points of reperfusion.

METHODS

Male Wistar rats, randomly divided into two groups: the I/R group (n=12) and the EPO group (500 IU/Kg, i.p. 20min prior to ischemia, n=15), were subjected to bilateral renal ischemia for 45 min. Each group was allocated in three subgroups according to the timing the animals were sacrificed at 6, 24 and 48 hrs of reperfusion. Rats subjected to identical surgical procedure without occlusion of renal pedicles were used as sham-operated group (n=6).

Renal injury was assessed by measurement of serum biochemical markers (urea and creatinine) and histological grading (stages 1 to 4 according to renal tubular damage). Expression of cyt c and Fas/FasL mRNA was evaluated by RT-PCR and protein expression of cyt c by immunohistochemistry.

RESULTS

EPO group had significantly lower serum urea and creatinine levels compared to I/R group at 6, 24 and 48 hours of reperfusion ($p<0.05$), (Figure 1). Histological evaluation revealed significantly less tubular damage in the EPO-treated group compared to I/R group ($p<0.05$), (Figure 2).

Fas/FasL and cyt c were detected in normal kidneys (Figures 3-5). Cyt c mRNA were significantly increased in I/R group compared to sham group at all time points ($p<0.05$), (Figure 4). EPO administration caused significant reduction of cyt c mRNA expression compared to I/R group at 48hrs ($p<0.05$), (Figure 4). A pronounced upregulation of protein cyt c expression was observed in I/R group, localized mainly in renal cortical tubules, compared to sham group (Figure 6). EPO administration markedly reduced cyt c staining compared to I/R group. In both groups immunoreactivity became gradually weaker from 6 to 48hrs (Figure 6).

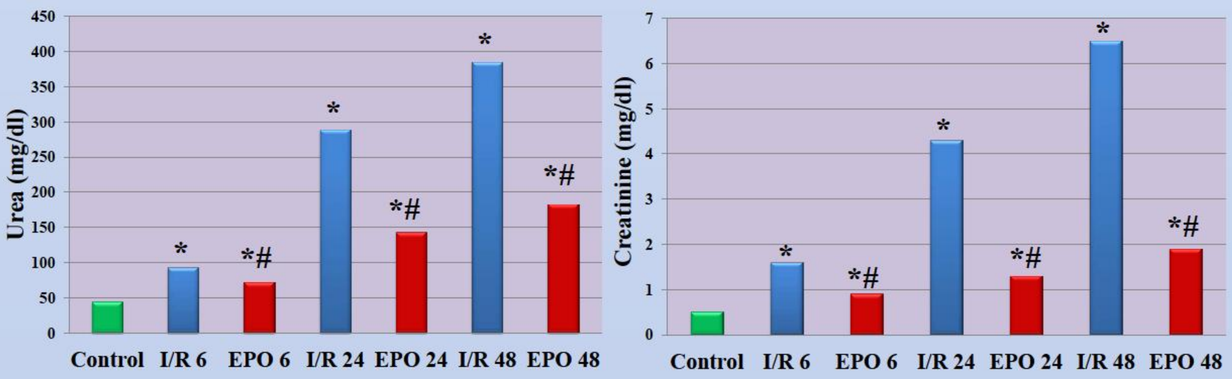

Figure 1. Serum levels of urea and creatinine at 6, 24 and 48h in I/R and EPO groups.

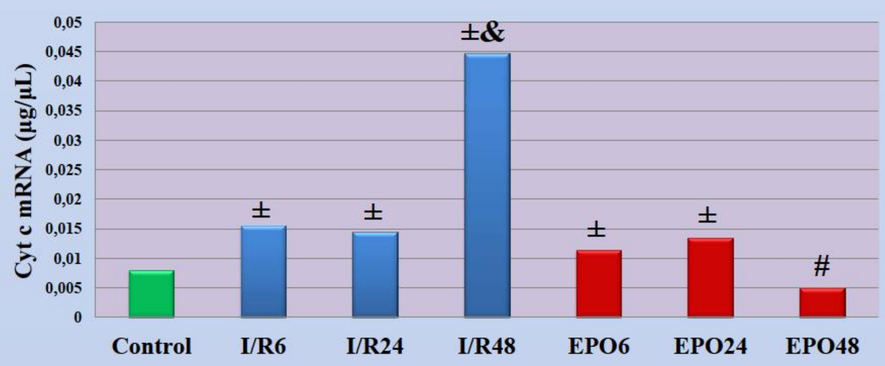

Figure 4. The expression of Cyt c mRNA in I/R and EPO groups.

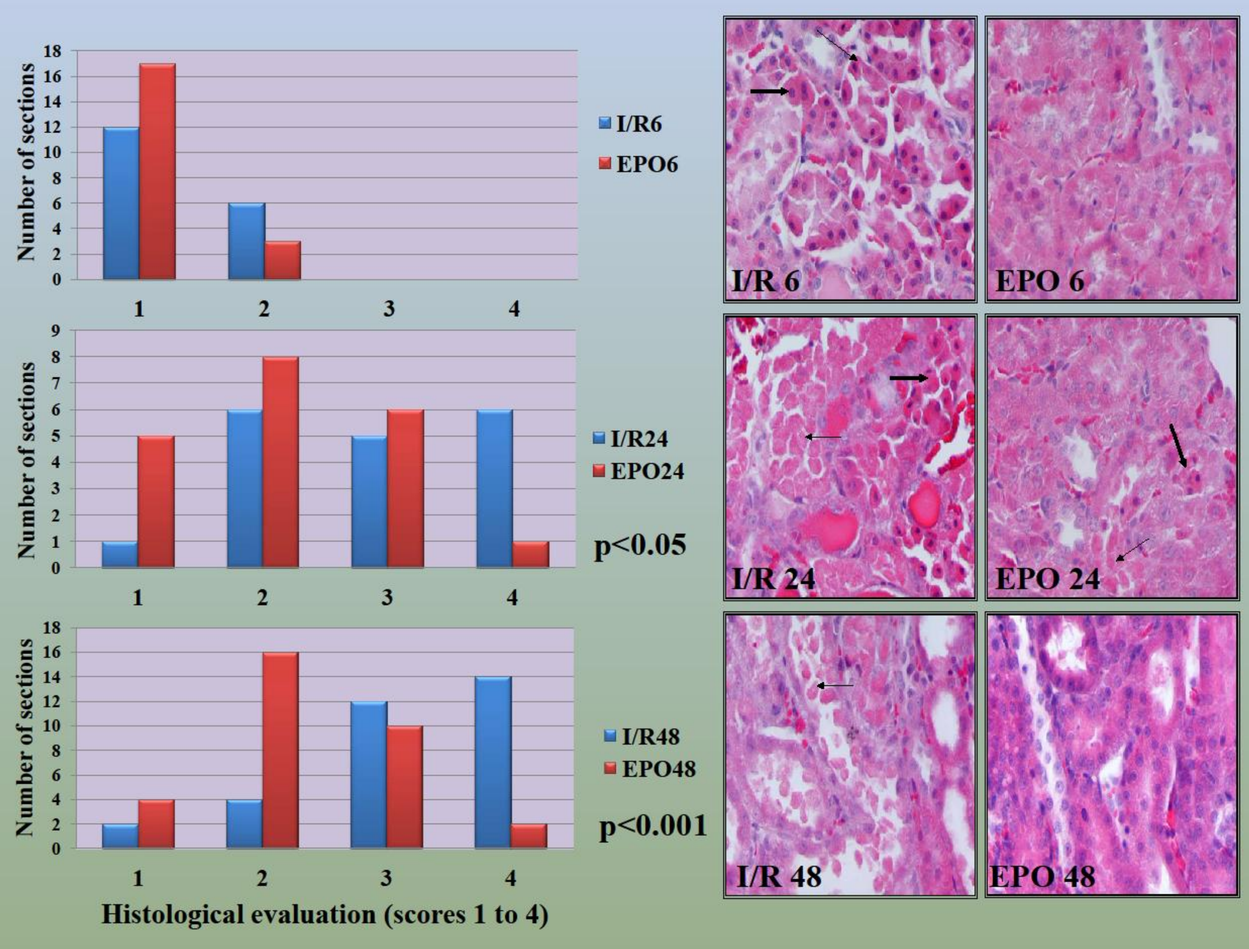

Figure 2. Histological scoring in the I/R and EPO groups.

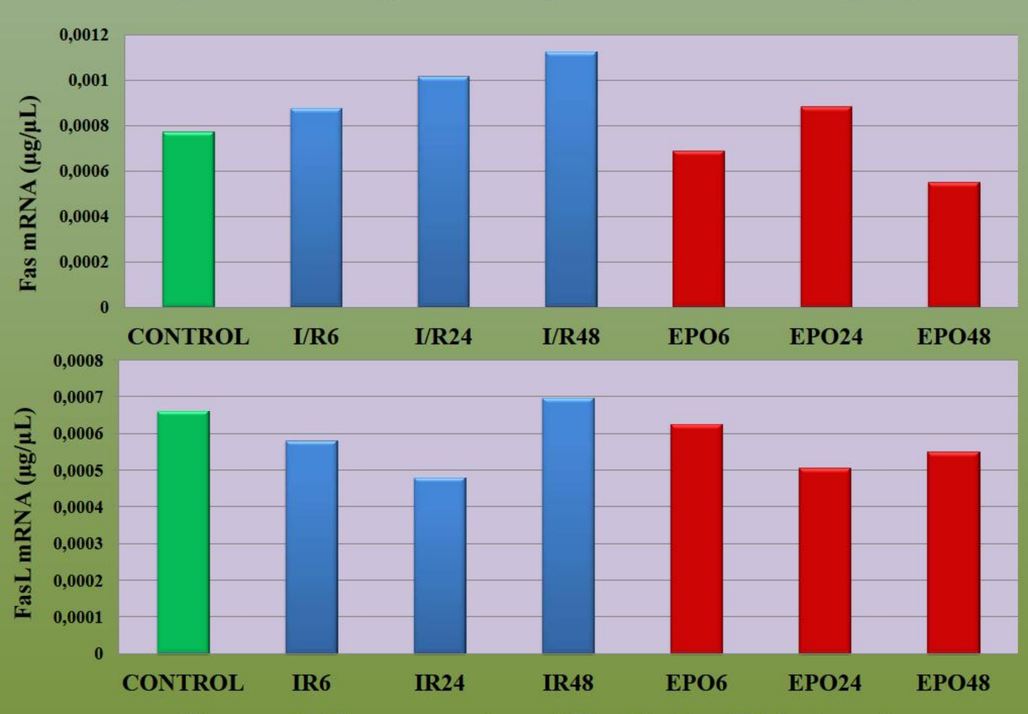

Figure 3. The expression of Fas/FasL mRNA (p=ns).

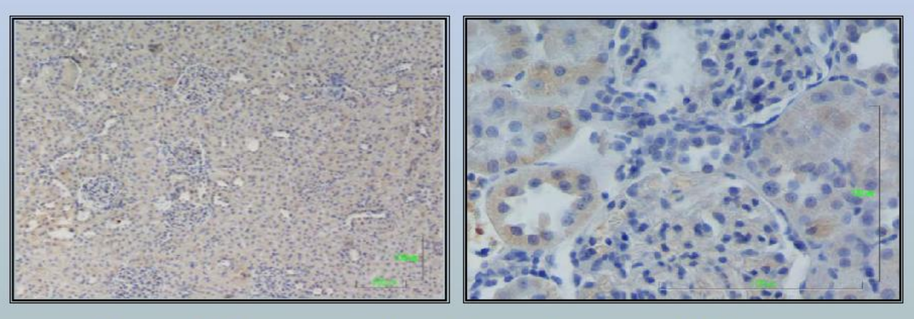

Figure 5. The expression of cyt c in normal kidneys.

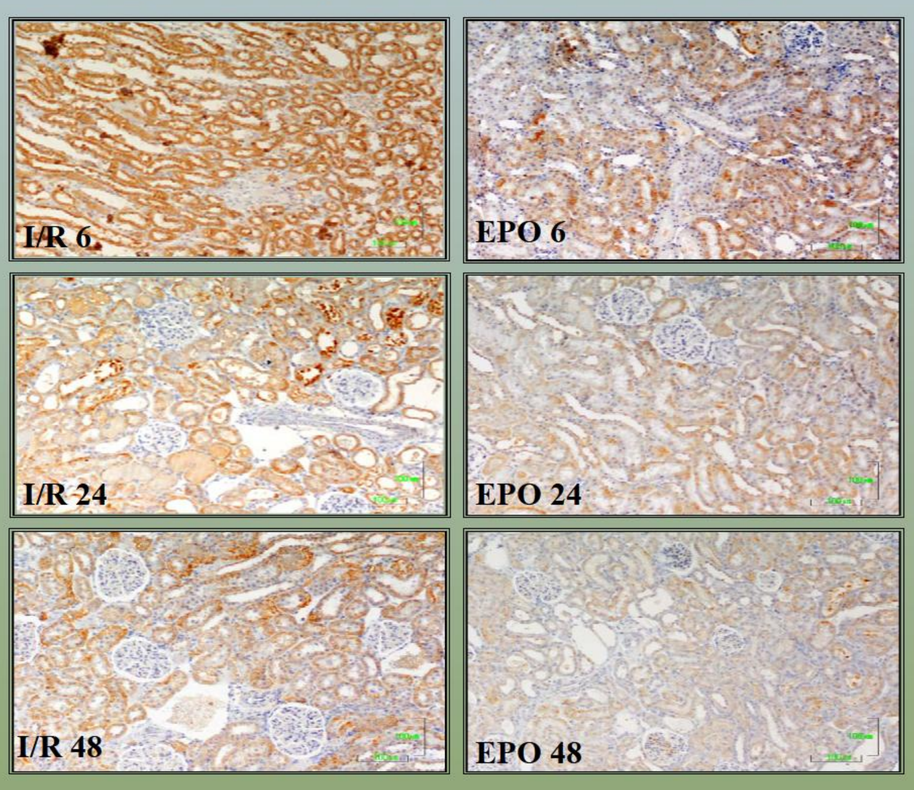

Figure 6. The expression of Cyt c in renal I/R and EPO groups.

CONCLUSIONS

- ✓ EPO pretreatment reduced I/R-induced tubular injury and ameliorated renal function
- ✓ Acute renal I/R injury and renoprotective action of EPO were not observed to correlate directly with changes in gene expression of Fas and FasL
- ✓ Early induction of cyt c expression following I/R might contribute to severe tissue damage observed at later time points
- ✓ EPO reduced cyt c immunostaining throughout reperfusion and gene expression at later stages suggesting a potential antiapoptotic action in I/R-induced renal injury through the endogenous pathway of apoptosis

51ERA-EDTA

71--SP

G) Acute Renal Failure. G1) Experimental models.

Kyriaki Xanthopoulou

DOI: 10.3252/pso.eu.51era.2014

Poster presented at: 51 ERA-EDTA CONGRESS AMSTERDAM 2014