Antioxidant and antiinflammatory strategies to prevent endothelial dysfunction in chronic kidney disease: The role of N-acetylcysteine

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Objectives:

Accelerated atherosclerosis in chronic kidney disease (CKD) is preceded by the development of an endothelial dysfunction (ED), which is characterized by a proinflammatory and a prothrombotic phenotype, as well as an enhanced oxidative stress. N-acetylcysteine (NAC) is a drug with potential interest in diseases characterized by increased oxidative stress or decreased glutathione (GSH) level, such as CKD.

NAC acts mainly on the supply of cysteine for GSH synthesis. In previous studies it has been shown that mimetics of glutathione peroxidase, like ebselen, better improved the ED in uremia than antioxidant mimetics of the superoxide dysmutase (EUK 134 or EUK 118) in an *in vitro* model of uremic ED. The possible protective effect of a NAC-based antioxidant strategy has been evaluated in an *in vitro* model of ED in uremia.

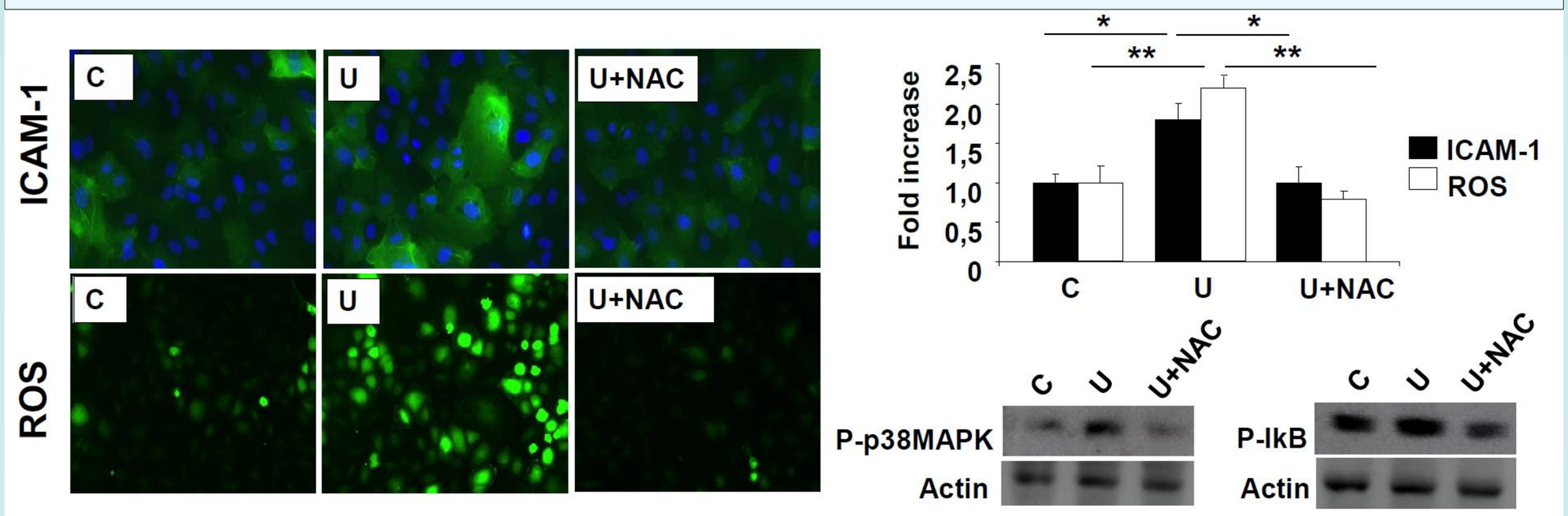
Methods:

Endothelial cells (ECs) were pretreated with NAC. ECs were exposed to a medium containing a pool of serum from patients on dialysis (n=10) or from healthy donors (n=15). ECs were isolated from human umbilical veins and maintained in a medium 199 supplemented with 100 U/mL penicillin,100 g/mL streptomycin and 20% human serum at 37ºC under a humidified atmosphere with 5% of CO_2. Changes in the expression of the adhesion receptor ICAM-1 and the production of intracellular reactive oxygen species (ROS) were assessed. Activation of inflammation-related proteins p38 MAPK and NFkappaB (NFκB) was also evaluated. ROS production was explored by using 5-(and 6)-chloromethyl-2',7' -dichlorodihydrofluorescein diacetate, acetyl ester (CM-H_2DCFDA). The activation of the signal transduction protein p38MAPK and NFκB by ELISA and Western-blot techniques.

Results:

Exposure of ECs to uremic media resulted in a significantly increased expression of ICAM-1 (fold increase 1.9±0.2 vs control, p< 0.01), overproduction of ROS (2.2±0.1 fold-increase) and activation of p38MAPK (1.9±0.2 fold-increase) and NFκB (1.8±0.1 fold-increase), compared to control ECs (p<0.05 for all).

NAC inhibited ICAM-1 expression (49±4% decrease, p<0.05) and ROS generation (62±6% decrease, p<0.05) in the uremic condition. Furthermore, it reduced p38MAPK activation (47±6% decrease, p<0.05) and the activation of NFκB induced by the uremic media (31±4% decrease, p<0.05).



Conclusions:

Endothelial dysfunction associated with CKD is considered to be the first step in the progression of atherosclerosis. Inflammation and oxidative stress seem to be involved in this ED. Our results indicate that NAC exhibit not only antioxidant but also anti-inflammatory effects on ED in uremia. Therefore, further research on the protective effects of NAC may provide further evidence for its use in the prevention of the cardiovascular complications in uremia.

References:

1. Serradell M, et al: Uremic medium disturbs the hemostatic balance of cultured human endothelial cells. Thromb Haemost 2001; 86:1099-105-. **2.** Serradell M, et al: Uremic medium causes expression, redistribution and shedding of adhesion molecules in cultured endothelial cells. Haematologica 2002; 87:1053-61. **3.** Caballo C, et al. NFκB in the development of endothelial activation and damage in uremia: an in vitro approach. PLoS One. 2012;7(8):e43374. **4.** Carbó C, et al Differential expression of proteins from cultured endothelial cells exposed to uremic versus normal serum. Am J Kidney Dis. 2008; 51: 603-12. **5**. Martin-Rodriguez S, et al. TLR4 and NALP3 inflammasome in the development of endothelial dysfunction in uraemia. Eur J Clin Invest. 2015; 45: 160-9.

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