



UNIVERSITÀ DEGLI STUDI DI SALERNO



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***Tesi di Dottorato in
Farmacologia e Farmacoterapia***

***Neurodegeneration in Chronic Kidney
Disease: role of neuroinflammation and
uremic toxins***

Dottorando

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ABSTRACT

Neuroinflammation and oxidative stress have been recognized as common aspects in neurodegenerative diseases. Although the inflammatory process may induce beneficial effects, such as the elimination of the pathogen, uncontrolled inflammation can lead to adverse outcomes through the production of neurotoxic factors that exacerbate neurodegenerative disease. Living cells continually generate reactive oxygen species (ROS) during energetic metabolism. ROS play an important physiological role, however, imbalanced defence mechanism of antioxidants, overproduction or incorporation of free radicals from environment to living system could be extremely deleterious, especially for the central nervous system (CNS). CNS is particularly vulnerable to oxidative stress and ROS production has been associated with different neurodegenerative diseases. Microglia and astrocytes, as the immune cells in the brain, are primary involved in different forms of neurodegeneration. These cells in response to a variety of stimuli and pathological events become activated and promote the release of inflammatory mediators and ROS. Neurodegenerative complications often occur in chronic kidney disease (CKD), a condition characterized by a progressive loss of renal metabolic activities and glomerular filtration, resulting in retention of solutes, normally excreted by healthy kidneys. These compounds, called uremic toxins, accumulate in patients with CKD and may have deleterious effects in various physiological functions in these patients, such as neurological complications associated to uremic syndrome. Indoxyl sulphate (IS) is a protein-bound uremic toxin, poorly eliminated by dialytic process, recognized as an uremic nephrovascular-toxin. IS has been reported to have effects on different type of cells such as renal tubular cells, vascular smooth muscle cells, vascular endothelial cells and osteoblasts, but no data regards CNS cells. Because of the mechanism/s involved in uremic-toxins induced neurological complications are not understood, in this project it has been examined the effects of IS on

neuroinflammation and oxidative stress in CNS cells. IS (15-60 μ M) treatment in C6 astrocyte cells increased inducible nitric oxide synthase (iNOS) and cyclooxygenase-2 (COX-2) expression, tumor necrosis factor (TNF- α). Moreover IS increased Aryl hydrocarbon Receptor (AhR), Nuclear Factor-kB (NF-kB) and inflammasome activation in these cells. In the same experimental condition, IS enhanced ROS release and decreased nuclear factor (erythroid-derived 2)-like 2 (Nrf2) activation, and heme oxygenase-1 (HO-1) and NAD(P)H dehydrogenase quinone 1 (NQO1) expression. Similar observations are made in primary mouse astrocytes and mixed glial cells. It has been also observed that IS can also affect cell viability and cycle distribution, increasing the G0/G1 and S phases and decreasing G2 phase in C6 cells and in astrocytes and in mixed glial cells. Moreover neurons incubation with IS (15-60 μ M) induced cell death in a dose dependent fashion. In vivo data indicate that IS (800 mg/kg, i.p) induced histological changes and an increase in COX-2 expression and in nitrotyrosine formation in mice brain. Furthermore preliminary experiments on the effect of the human serum on ROS release in C6 cells, indicate that IS significantly contributes to oxidative stress in astrocytes. This study will be a step towards elucidating if this toxin could be not only a biomarker of disease progression in CKD patients, but also a potential pharmacological therapeutic target.