

## EFFECTS OF A METHANOLIC EXTRACT OF SEDUM TELEPHIUM IN LPS-ACTIVATED RAT MACROPHAGES

<u>Polito Francesca<sup>1</sup></u>, Bitto Alessandra<sup>1</sup>, Minutoli Letteria<sup>1</sup>, Fiumara Tiziana<sup>1</sup>, Giachetti Daniela<sup>2</sup>, Miraldi Elisabetta<sup>2</sup>, Biagi Marco<sup>2</sup> Squadrito Francesco<sup>1</sup>, Caputi Achille P.<sup>1</sup>, Altavilla Domenica<sup>1</sup>

<sup>1</sup>Dept of Exp and Clinical Medicine and Pharmacology and <sup>2</sup>Dept of Environmental Sci, Section of Pharm Biol, University of Siena, Italy

Sedum telephium subsp. maximum (Crassulaceae) is a medicinal plant used in antiquity to cure many types of inflammatory disease. We investigated whether a methanolic extract obtained from sedum telephium (ST) may inhibit endotoxin-induced activation of the inflammatory phenotype in macrophages (M $\Phi$ ) culture. Rat peritoneal M $\Phi$  were stimulated with 50 µg/ml of Salmonella enteritidis lipopolysaccharide (LPS). Stimulated M $\Phi$  were coincubated with different doses of ST extract (8-16-32 µg/ml) or RPMI alone using different times of incubation. M $\Phi$  cultures were used to evaluate (1hr incubation) extracellular regulated kinase (ERK 1/2) and c-jun-N-terminal kinae (JNK) by western blot analysis, to study (4hrs incubation) tumor necrosis factor alpha (TNF- $\alpha$ ) gene expression by real time polymerase chain reaction and to analyze the levels of the mature protein by ELISA determination. Furthermore after 24 hours of incubation cell lysates were used for western blot analysis of inducible nitric oxide synthase (iNOS) and nitrite content was evaluated in supernatants. ST significantly blunted LPS-induced ERK (LPS= 9.8±1 integrated intensity; LPS+ST= 1.7±0.8 integrated intensity), JNK (LPS= 7.7±1.3 integrated intensity; LPS+ST= 2.2±0.6 integrated intensity) and iNOS (LPS= 8.1±1.1 integrated intensity; LPS+ST= 0.8±0.3 integrated intensity) activation. Furthermore, this compound inhibited TNF- $\alpha$  gene expression (LPS= 7.5 $\pm$ 0.8 n-fold/ $\beta$ -actin; LPS+ST= 1.9 $\pm$ 0.3 n-fold/ $\beta$ -actin) and markedly reduced the release of the inflammatory cytokine and nitrite content (LPS=  $25\pm2$  µM; LPS+ST=  $4\pm1.3$  µM). Our data indicate that the methanolic extract of ST possess a high anti-inflammatory potential and could have important therapeutic application in inflammatory diseases.