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Evaluating the capacity of several antioxidants to attenuate the renal toxicity induced by methotrexate therapy

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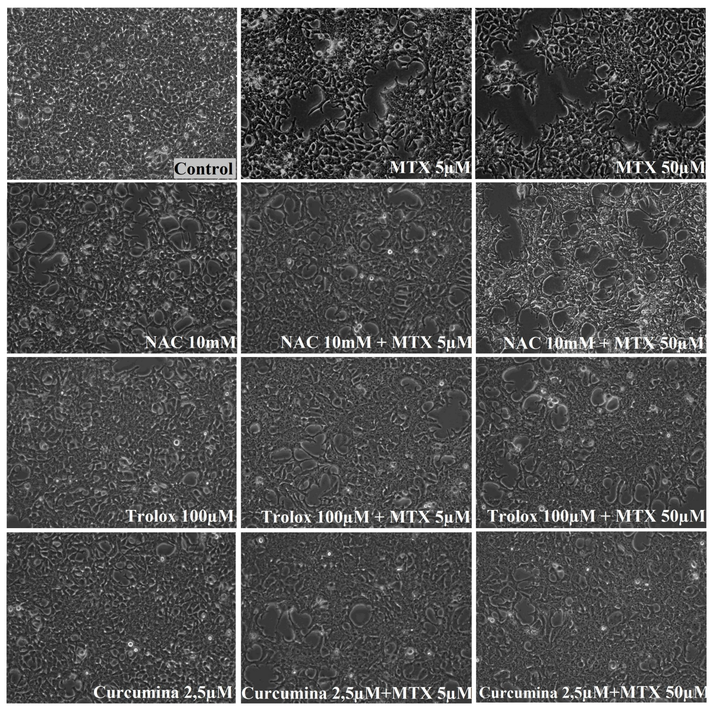
INTRODUCTION

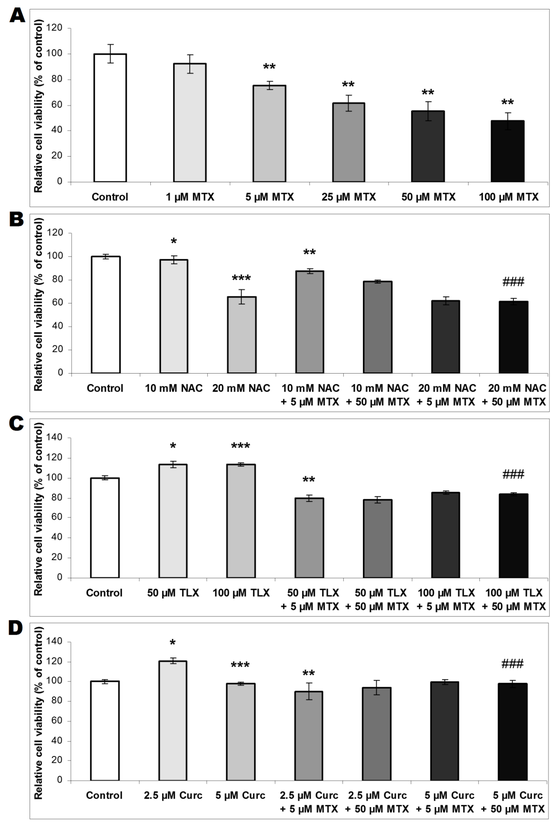
- **Methotrexate (MTX) chemotherapy is often limited by its severe side effects which include nephrotoxicity.**
- **In the continuous search of efficient antioxidants that could ameliorate this toxic condition of MTX, our study aimed to evaluate the efficiency of N-acetyl cysteine (NAC), Trolox methyl ether (Trolox-Me), and curcumin as potent antioxidants using an *in vitro* model of MTX-induced toxicity.**

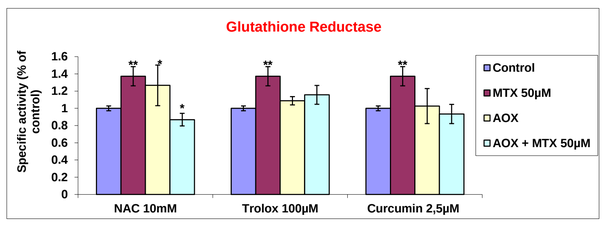
MATERIALS AND METHODS

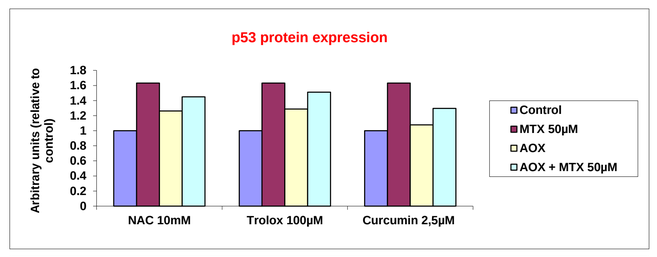
- **Human embryonic kidney (HEK293) cells were pre-exposed to different antioxidants for 2 hours prior to MTX (5 μM and 50 μM).**
- **After 24 hours of exposure to MTX, the cell viability and morphology were assessed, activities of antioxidant enzymes and levels of lipid peroxidation were measured by spectrophotometrically methods, and protein expression was determined by Western blotting.**

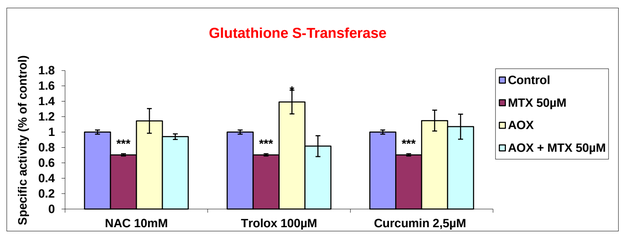
MTT assay

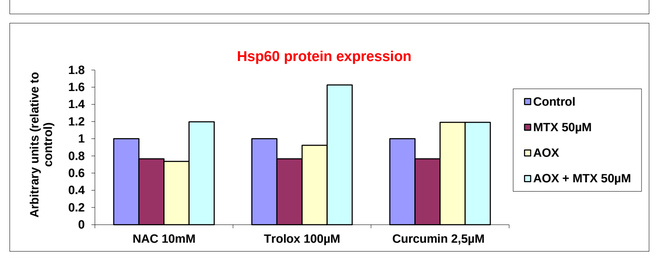


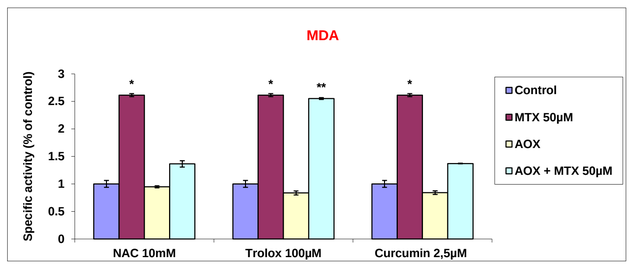


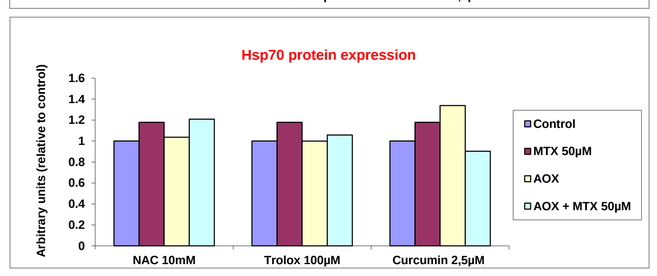












RESULTS AND DISCUSSIONS

- **Exposure to MTX at concentrations between 1 and 100 μM for 24 hours decreased cell viability in a dose-dependent manner and was correlated with the increase of p53 protein expression.**
- **All three antioxidants tested have proved that can inhibit the apoptosis induced by MTX, as revealed by the expression of heat shock proteins (Hsp27, Hsp60, Hsp70 and Hsp90).**
- **Pre-treatment of cells with 50 μM of Trolox-Me succeeded to significantly decrease the MTX-induced cell death.**
- **The reduction in the activities of glutathione reductase and glutathione S-transferase after MTX incubation was correlated with a low level of GSH, and was attenuated by the pre-incubation with Trolox-Me or curcumin, these antioxidants being able to maintain enough GSH for the reactions of conjugation with MTX metabolites in order to decrease its toxicity.**

CONCLUSIONS

- **The pre-treatment with curcumin, Trolox-Me or NAC proved extremely effective at blocking MTX toxicity at the concentration investigated in vitro on kidney cells.**
- **The results of our study encourage further clinical assessments in order to use these antioxidants in dietary prevention of renal side effect of MTX.**

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