Research in Pharmaceutical Sciences, 2012;7(5)
School of Pharmacy and Pharmaceutical Sciences
Isfahan University of Medical Sciences

Effect of Osthole and oxypeucedanin on doxorubicin -induced apoptosis in PC12 cells

M. Moieni-Arya[1,*], Y. Shookohinia[2], L. Hosseinzadeh[3], E. Alizadeh[4]

[1]Students research committee, School of Pharmacy, Kermanshah University of Medical Sciences, Kermanshah, Iran.
[2]Department of Pharmacognosy and Biotechnology, School of Pharmacy, Kermanshah University of Medical Sciences, Kermanshah, Iran.
[3]Department of Pharmacology and Toxicology, School of Pharmacy, Kermanshah University of Medical Sciences, Kermanshah, Iran.
[4]School of Pharmacy, Kermanshah University of Medical Sciences, Kermanshah, Iran.

Background and Amis: Doxorubicin is a potent, broad-spectrum chemotherapeutic drug with toxic effects on normal tissues, including brain tissue. Coumarins are bioactive secondary metabolites could be found in roots of Umbelliferae. In the current study, we investigated the protective effects of Osthole and oxypeucedanin isolated from Prangos ferulacea roots on apoptosis induced by doxorubicin in PC12 as a neuronal model cell line.
Methods: Air-dried roots of P. ferulacea were extracted with acetone. Repeated open column chromatography in normal phase chromatographies using n-heptane and ethyl acetate, as mobile phases resulted in isolation of coumarins. PC12 cells were cultured in DMEM medium containing 10% (v/v) fetal bovine serum, 100 units/ml penicillin, and100 µg/ml streptomycin. Cell viability was determined by MTT assay. Quantitative Real Time RT-PCR was used to evaluate the expression of Bax and Bcl-2. Activation of caspase-3 was evaluated by spectrophotometer.
Results: Osthole and oxypeucedanin were purified using column chromatography. Treatment of cells with doxorubicin reduced PC12 viability in a dose dependent manner. DOX- induced cytotoxicity in a concentration and time – dependent manner. The IC50 value was 5µM.For evaluation of effect of Osthole and oxypeucedanin pretreatment on DOX- induced cytotoxicity, PC12 Cells were pretreated for different time interval with Osthole and oxypeucedanin then cells were treated with doxorubicin. Pretreatment of cells with Osthole and oxypeucedanin decreased doxorubicin cytotoxicity. We wished to characterize the type of cell death involved in our experiments. Real time RT PCR Results clearly showed osthole (7 µg/cc)and oxypeucedanin(80 µg/cc)were able to decrease significantly mRNA gene expression of Bax and increase mRNA gene expression of Bcl2. Moreover, pre-treatment of cells with osthole and oxypeucedanin decreased significantly the caspase-3 activation to 46.8 (8.2) %and 59.78(3.65) %, respectively.
Conclusions: Our observation indicated that subtoxic concentration of osthole and oxypeucedanin has protective effect on doxorubicin induced apoptosis in PC12 cells.

Keywords: Doxorubicin; Osthole; Oxypeucedanin; PC12 cells; Apoptosis