

Structure elucidation was done using NMR and IR spectrum. Yields of production of thiazolidone in both method is less than previous steps (80-90%). The yield of two stage method was about 10% more than one step method and microwave only decreased the time of reaction the yield was unchanged. As the reaction is reversible, the water that is produced during the synthesis, takes the reaction to left side thus without using the dean stark and drying solvent, the yield decrease.

**Keywords:** Cyclooxygenase Inhibitor, Derivatives Of Thiazolidine 4-One, Anti Cancer.

---

#### Abstract No.196

##### **Study on p53R2 (small Subunit of Ribonucleotide Reductase) Function Via Exploring its Expression Level After Different Doses of Irradiation in Human Mouth Epitelial ,KB, Cells**

*Farideh Habibi, Reza Rofugaran,  
Bahram Goliae\**

Institute of Biochemistry and Biophysics, University of Tehran, Tehran, IR  
(E-mail: frd.habibi@gmail.com)

Ribonucleotide reductase (RNR), is the enzyme that provides deoxyribonucleoside triphosphates (dNTP) for DNA synthesis. The recently discovered p53-inducible small subunit of ribonucleotide reductase (p53R2) plays essential roles in DNA repair and mitochondrial DNA synthesis. It is believed to provide deoxyribonucleotides for DNA repair since the protein is expressed via p53 which is involved in DNA damage checkpoints. Because of late expression of p53R2 (12h) in response to DNA damage to repair, it was suggested that this protein has other functions in the cell. Taken together, in this study, we are investigating the expression level changes of p53R2 after different doses of irradiation treatment of KB cells. This project hopefully will be able to shed light on the function of p53R2 in DNA repair and mitochondrial DNA synthesis. We did KB cell culture, and obtained doubling time (20.5 h) and clonogenic assay for obtaining plating efficiency (38%) to investigate cell survival. In final step, will do western blotting to probe the protein level in different doses of x-ray. The detailed results will be discussed.

**Keywords:** RNR, p53R2, DNA repair, Radiation.

#### Abstract No.197

##### **Dynamics and Flexibility of Mnemiopsin in the Presence of Calcium**

*Shima Tarahomi<sup>1</sup>, Reza Hassan Sajedi\*<sup>2</sup>, Bijan Ranjbar<sup>2</sup>,  
Majid Taghdir<sup>1</sup>*

1. Dept. of Biology, Faculty of Sciences, University of Guilan, Rasht, IR

2. Dept. of Biochemistry, Faculty of Biological Sciences, Tarbiat

Modares University, Tehran, IR

(E-mail: shima\_ta83@yahoo.com)

Mnemiopsin is a photoprotein that belongs to the EF-hand calcium-binding proteins (EFCaBP).  $Ca^{2+}$  is involved in specific and selective regulation of cellular processes. Many of these functions are accomplished through the interaction of  $Ca^{2+}$  with specific proteins. EFCaBPs have eminent properties which change upon interacting with calcium ions. It is generally believed that  $Ca^{2+}$  confers on proteins an increased thermal stability, affect flexibility and rigidity, interface reversible unfolding, protein-protein interaction and etc. Our previous spectroscopic (CD and fluorescence) studies indicate that the apo-form ( $Ca^{2+}$ -depleted) of the protein adopt a closed conformation when compared to the  $Ca^{2+}$ -loaded once. In this study, flexibility and dynamics of mnemiopsin were investigated for apo-mnemiopsin and  $Ca^{2+}$ -loaded-mnemiopsin with fluorescence acrylamide quenching and limited proteolysis. Calcium is removed by TCA (trichloroacetic acid) precipitation to obtain apo-mnemiopsin. Fluorescence quenching was carried out by the addition of 1M acrylamide to protein solutions at an excitation wavelength of 295 nm and an emission wavelength of 337 nm in a Perkin-Elmer luminescence spectrometer. Limited proteolysis was performed by incubation of protein with thermolysin at 37 °C. Acrylamide quenching of tryptophan fluorescence is widely used to monitor tryptophan environments in proteins. Dynamic quenching analysis revealed that  $Ca^{2+}$ -loaded- mnemiopsin was quenched more than the apo-form. Molecular modeling indicates microenvironment of Trp21 is important to explain this experimental data. Limited proteolysis experiments can be used to identify the sites of enhanced flexibility or of local unfolding of a polypeptide chain. Although thermolysin has been classified as proteases with broad specificity, it should be considered that thermolysin cleaves mostly the amino side of bulky and hydrophobic amino acids. Mnemiopsin proteolysis results indicate that  $Ca^{2+}$ -loaded form is more sensitive to proteolysis than apo-form. Therefore, the present results from limited proteolysis and digestion experiments as well as quenching analysis can be explained by the fact that  $Ca^{2+}$ -loaded-mnemiopsin is more flexible in some regions when compared to apo-mnemiopsin.

**Keywords:** Mnemiopsin, Flexibility, Quenching, Proteolysis.