PROTECTIVE EFFECT OF MN(III) TETRAKIS (4-BENZOIC ACID) PORPHYRIN ON THE INACTIVATION OF PRIMARY ANTIOXIDANT ENZYMES IN KERATINOCYTES BY PHOTODYNAMICALLY GENERATED FREE RADICALS

Carolina Muscoli[1], Steven Nistico'[2], Rosaria Arcone[1], Carmen Colica[1], Valeria Visalli[1], Domenica Vetrice[1], Francesca Casale[1], Immacolata Del Vecchio[3], Maria Concetta Strongoli[3], Vincenzo Mollace[1] and Domenicantonio Rotiroti[3]

[1]Dept of Pharmacobiol. Sciences, University of Catanzaro Magna Graecia, Italy; [2]School of Medicine, Univesity of Rome "Tor Vergata"; [3]ISN-CNR

Cellular antioxidant enzymes protect against damage caused by exposure to endogenous or exogenous prooxidants. Super oxide is a reactive form of oxygen that can be produced in vivo either in normal and under pathophysiologic conditions or by photosensitizing chemicals, as during photodynamic treatment. We hypothesized that over production of free radicals would decrease the enzymatic activities of endogenous cellular antioxidants. To test this hypothesis, we treated cultured epidermal keratinocytes with the photosensitizer Photofrin plus visible light to produce free radicals, and then measured CuZnSOD and MnSOD activities. Our results demonstrated that photodinamical treatment of keratincocytes increases malonildyaldeide production, nitrotyrosine staining and superoxide production. Furthermore, the enzymatic activities of cellular CuZnSOD and MnSOD were significantly decreased after keratinocytes were treated with Photofrin plus visible light. By contrast, the enzymatic activities of cellular CuZnSOD, MnSOD, and catalase
were unaffected in control cells. Despite the decreased levels of enzymatic activities, the protein levels of all three primary antioxidant enzymes remained constant after photodynamic treatment, as determined by Western blotting. Removal of free radicals Pretreatment of keratinocytes with SOD mimics such as Mn(III) tetrakis (4-benzoic acid) porphyrin (MnTBAP) were able to restore the endogenous antioxidant system activities inhibiting the MDA formation, nitrotyrosine staining and superoxide formation. The conclusion from these experiments is that the primary cellular antioxidant enzymes CuZnSOD and MnSOD can be inactivated by photodynamically generated free radicals in nucleated mammalian cells. These findings may be useful in the future development of new targets for therapies that use photodynamic generation of free radicals to inactivate antioxidant defenses with a goal of sensitizing tumor cells to prooxidant-generating drugs.