

## **SNAM1-2**

### **Cytochrome c Oxidation by the phthalocyanine Pc 4 and light: Detection of a unique singlet oxygen derivate product**

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Singlet oxygen ( $^1O_2$ ) has long been proposed as the primary reactive oxygen species in photodynamic therapy (PDT). Once generated,  $^1O_2$  may undergo radiative decay at 1270 nm and its emission is habitually measured in solutions where the lifetime is on the order of  $\mu s$ . The lifetime in  $H_2O$  is 4  $\mu s$  but drops dramatically in vitro- in vivo. The measurement of  $^1O_2$  luminescence in biological media is still a challenge because of this short lifetime, limited detector sensitivity, and/or temporal response. However, some research teams are working in vivo monitoring singlet oxygen with interesting results.

One target of  $^1O_2$  in biological systems may be proteins, and numerous studies have shown that  $^1O_2$  can inactivate enzymes such as cytochrome c (Cyt-c). It is known that PDT induces apoptosis via the release of Cyt-c from mitochondria into the cytosol, followed by activation of caspases. Because Cyt-c resides in the mitochondrial intermembrane space, associated with the inner membrane, it could be exposed to  $^1O_2$  generated by photoactivation of photosensitizers like Pc 4 with mitochondrial localization.

Here, we used MALDI-TOF-MS and LC-ESI-MS to study the reaction of  $^1O_2$  with amino acid residues within two model peptides in homogeneous medium, and within Cyt-c in homogeneous medium and in liposomes. The analyses revealed multiple oxidation products, including at least one His-derived product that is unique to singlet oxygen and not found following reaction with other reactive oxygen species. This product may serve as a marker of the mitochondrial photodynamic damage derived from  $^1O_2$ .