Quenching of Singlet Oxygen by Double Fluorescent and Spin Sensors, Consisting of Fluorophore Moiety and Heterocyclic Amine Oxidizable to Stable Nitroxide

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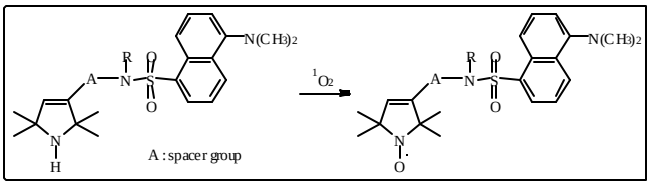
Double fluorescent and spin sensors were recently used to detect transient oxidants *via* simultaneous fluorescence change and production of nitroxide observed by electron paramagnetic resonance spectrum. One such oxidant, singlet molecular oxygen ($^1O_2$), was detected in thylakoid membrane using these probes. In the present study, we investigated the total (physical and chemical) quenching of $^1O_2$ phosphorescence by



sensors composed of either 2,5-dihydro-2,2,5,5-tetramethyl-1*H*-pyrrole or 2,2,6,6-tetramethyl-piperidine attached to the xanthene or dansyl fluorophores. We found that the quenching rate constants were in the range (2-7)x10^7 $M^{-1}s^{-1}$ in acetonitrile or D_2O. Quenching of $^1O_2$ is usually an additive process in which different functional groups may contribute to the quenching. We estimated that the $^1O_2$ quenching by the amine fragments was *ca*. one to two orders of magnitude lower than that for the complete molecules. Our data suggests that the incorporation of a fluorescent chromophore induces an additional strong quenching of $^1O_2$, which may in turn decrease the nitroxide yield *via* $^1O_2$ path, and may affect quantitative interpretations. We hope that our results will contribute to a better characterization and wider use of these novel double sensors.