Modulation of the Efficiency of Phenothiazinium Photosensitizers by Interaction with Membranes

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INTRODUCTION: Many studies show that aiming for specific subcellular localization, rather than high singlet oxygen ($^1O_2$) quantum yield, may be a better strategy for searching for more efficient photosensitizers (PS) for Photodynamic Therapy (PDT). Specifically, PS binding to membranes is recognized as precondition to membrane damage, and probably to cell death. **OBJECTIVES:** The properties of the ground and excited states of phenothiazinium salts in membrane environment were studied to clarify the parameters that affect their ability to damage membranes. **MATERIALS AND METHODS:** Four PS were employed: methylene blue (MB), toluidine blue (TBO), 1,9-dimethyl methylene blue (DMMB) and DO15. Molar absorptivity coefficient and $^1O_2$ quantum yield were determined in ethanol, whereas PS aggregation was studied in NaCl or SDS solutions, these latter being also used to access the effect of aggregation on triplet excited state lifetime. $^1O_2$ generation, PS aggregation and membrane/solution partition were studied in the presence of liposomes. Liposomes were irradiated with PS to evaluate membrane damage, which was measured by leakage of a fluorescent probe, TBARS generation, and structural changes detected by SAXS. **RESULTS AND DISCUSSION**: DMMB and DO15 were significantly more efficient in terms of damaging membranes, DO15 being faster and the only one that changed membrane structure. This results were attributed to membrane binding (which followed the order MB ≈ TBO < DMMB < DO15), and its effects on PS photochemistry. Though $^1O_2$ generation was similar for all compounds in ethanol, DO15 generated twice more $^1O_2$ in the presence of membranes than the other PS. PS aggregation (which generally decreases $^1O_2$ generation) was smaller for DO15 and higher for DMMB. However, only DO15 aggregates could lead to free radicals generation. **CONCLUSION:** PS/membrane interaction increases PS concentration near lipids, modulates the generation of reactive species by regulating PS aggregation, and affects $^1O_2$ generation near its biological target.

Key words: photosensitizers, singlet oxygen, membranes
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