Chemical Synthesis and Ex-Vivo Evaluation of Berberine Analogs as DNA-Binding Singlet Oxygen Photosensitizers

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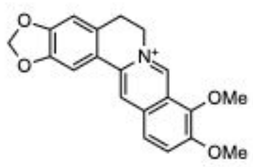
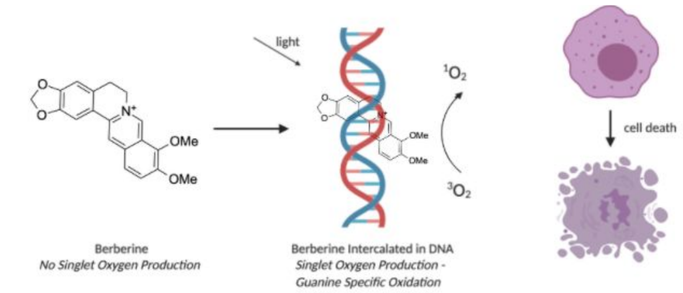
Background

Berberine is a natural product isoquinoline alkaloid that has previously been reported in literature for its medicinal properties, including antimicrobial, anticancer, antidiabetic, cardiovascular, and antidepressant properties. Its putative mechanism of action is derived from its ability to bind to DNA and to generate reactive singlet oxygen species upon photoirradiation. This singlet oxygen oxidizes guanine residues in DNA, resulting in cell death. Through semisynthetic modifications of berberine, more specifically nucleophilic additions or reductions at C-8, we can change the electronic structure of berberine, allowing for access to a greater library of analogs and yielding greater photosynthetic efficacy. Here we present ex-vivo experiments to quantify the singlet oxygen photosensitizing capabilities of berberine such that we can deduce the effect of berberine's structure onto its properties as a DNA-binding photosensitizer, further providing insight into its mechanism of action towards its medicinal properties. We utilized quantitative benchtop proton NMR to track singlet oxygen production by trapping it in a 4+2 hetero Diels-Alder reaction with cyclic dienes to quantify each analog's efficacy as a singlet oxygen photosensitizer.

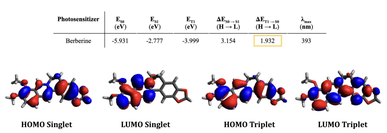


Putative Method of Action

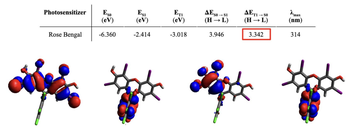


In-Silico Methods

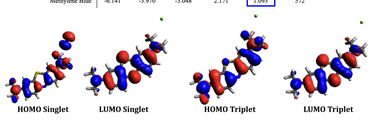
Photosensitizer	E_{S0} (eV)	E_{S1} (eV)	E_{T1} (eV)	$\Delta E_{S0\to S1}$ (H → L)	$\Delta E_{T1\to S0}$ (H → L)	λ_{max} (nm)
Berberine	-5.931	-2.777	-3.999	3.154	1.932	393



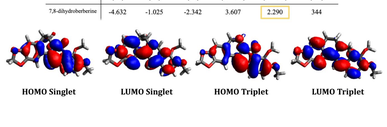
Photosensitizer	E_{S0} (eV)	E_{S1} (eV)	E_{T1} (eV)	$\Delta E_{S0\to S1}$ (H → L)	$\Delta E_{T1\to S0}$ (H → L)	λ_{max} (nm)
Rose Bengal	-6.360	-2.414	-3.018	3.946	3.342	314



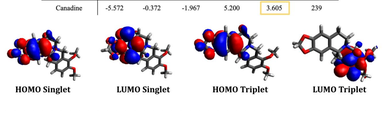
Photosensitizer	E_{S0} (eV)	E_{S1} (eV)	E_{T1} (eV)	$\Delta E_{S0\to S1}$ (H → L)	$\Delta E_{T1\to S0}$ (H → L)	λ_{max} (nm)
Methylene Blue	-6.141	-3.970	-5.048	2.171	1.093	572



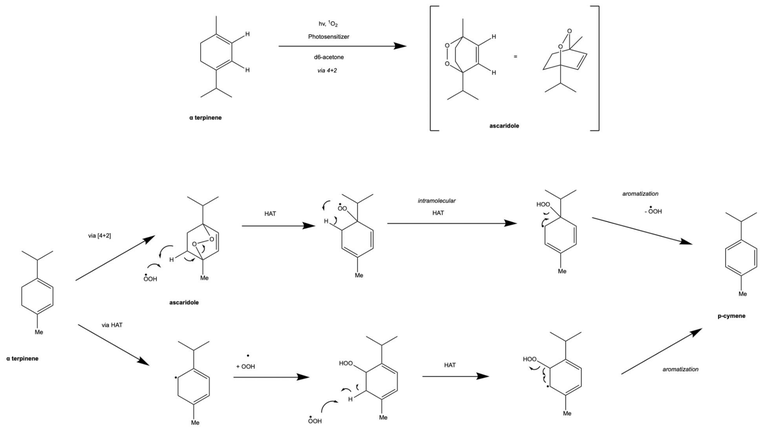
Photosensitizer	E_{S0} (eV)	E_{S1} (eV)	E_{T1} (eV)	$\Delta E_{S0\to S1}$ (H → L)	$\Delta E_{T1\to S0}$ (H → L)	λ_{max} (nm)
7,8-dihydroberberine	-4.632	-1.025	-2.342	3.607	2.290	344



Photosensitizer	E_{S0} (eV)	E_{S1} (eV)	E_{T1} (eV)	$\Delta E_{S0\to S1}$ (H → L)	$\Delta E_{T1\to S0}$ (H → L)	λ_{max} (nm)
Canadine	-5.572	-0.372	-1.967	5.200	3.605	239

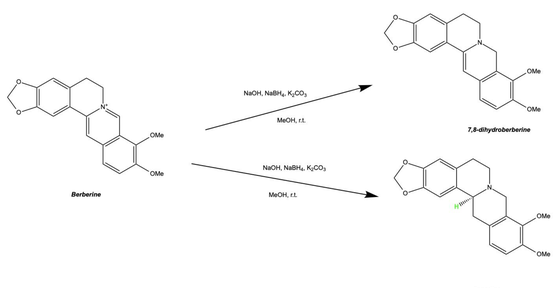


Synthesis



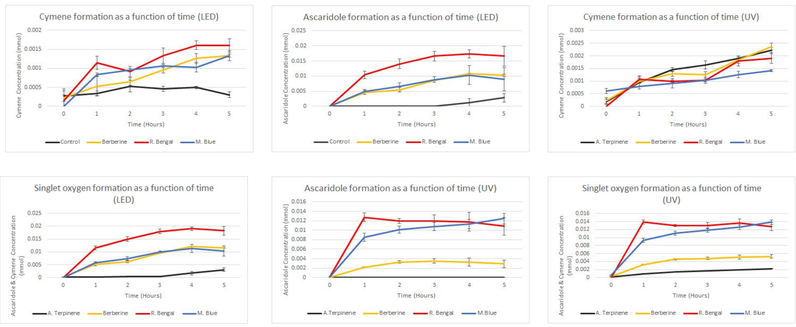
4+2 Hetero Diels-Alder Reaction between terpinene and singlet oxygen

Nucleophilic Additions to Berberine at C-8 via Grignard Reactions

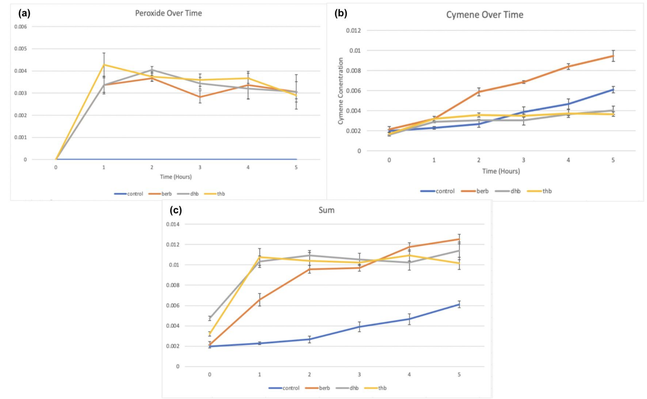


Reductions of Berberine at C-8

Singlet Oxygen Production



Singlet Oxygen Formation Over Time in Berberine Against Two Commercially Available Photosensitizers



Singlet Oxygen Formation Over Time in Berberine Against Two Reduced Berberine Analogs

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