Supporting Information: DNA Interstrand Cross-link Formation Initiated by Reaction Between Singlet Oxygen and a Modified Nucleotide.

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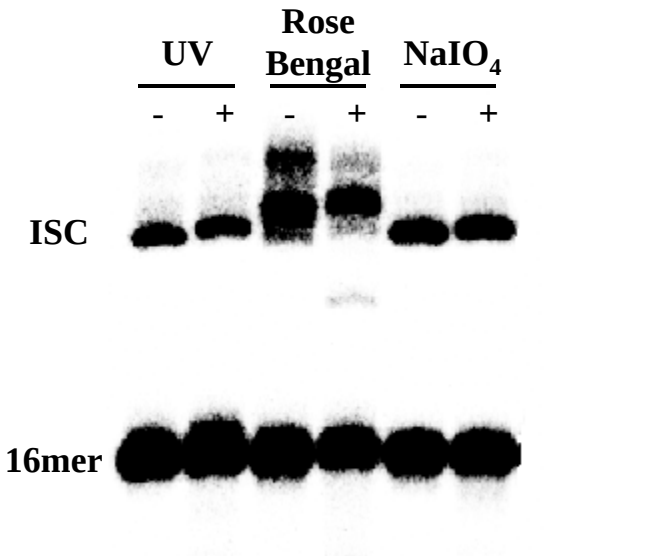
Contents:

General Methods. Oligonucleotides were synthesized as previously described.[1] Radiolabeling was carried out according to standard protocols.[2] U.V. photoreactions of oligonucleotides were carried out in Pyrex tubes in a Rayonet photoreactor fitted with 16 lamps having an output maximum at 350 nm. Rose Bengal sensitized white light photolyses were carried out in Pyrex tubes using tungsten lamp (250 W) with a 400 nm cut off filter. All photoreactions and thermoreactions were carried out in 10 mM potassium phosphate (pH 7.2) and 100 mM NaCl. After reaction, samples were precipitated from 0.3 N NaOAc pH 5.3 and calf thymus DNA (0.1 mM bp). Pellets were dried, and then resuspended in formamide loading buffer and subjected to 20% denaturing PAGE analysis. Percent ISC was determined by dividing the integrated volume for the cross-linked product band by the total integrated volume of DNA in the lane.

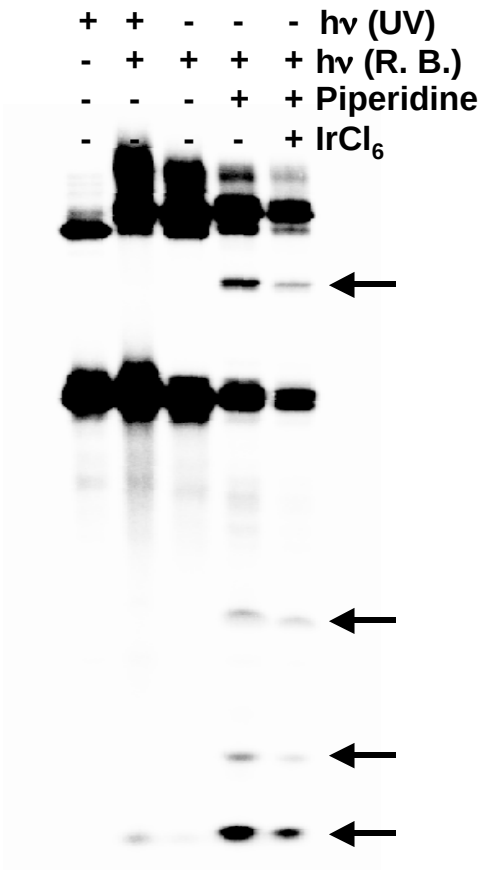
D_2O effect on the consumption of monomeric 2. White light photolyses of monomer **2** (50 μM) containing dA (5 μM) as an internal standard were carried out in H_2O, 50 % D_2O and D_2O as a solvent in the presence of Rose Bengal (10 μM). Sample was analyzed by reverse phase HPLC (C_{18}-μBondpak column (7.8 × 300 mm)) with UV detection at 260 nm. The peaks of interest (retention time: **2** (21.1 min), dA (8.7 min), Rose Bengal (14.3 min)) were quantified using the following gradient conditions: 5% to 35% MeCN in water, linearly for 25 min.

^{1}H NMR analysis of oxidation of monomeric 2 by $NaIO_4$ or Rose Bengal photosensitization. ^{1}H NMR experiment of the reaction of **2**(50 mM) with $NaIO_4$ (50 mM) was carried out in deuterated phosphate buffer (50 mM, pD 7.4) at room temperature. Reaction was initiated by adding periodate solution at room temperature. Data were collected prior to periodate addition, 10 min after addition and after 24 h at room temperature. Rose Bengal photosensitization was carried out in the same manner, except that the sensitizer was present at 50 μM, **2** (200 μM). The sample was irradiated for 5 min and the spectrum was immediately collected.

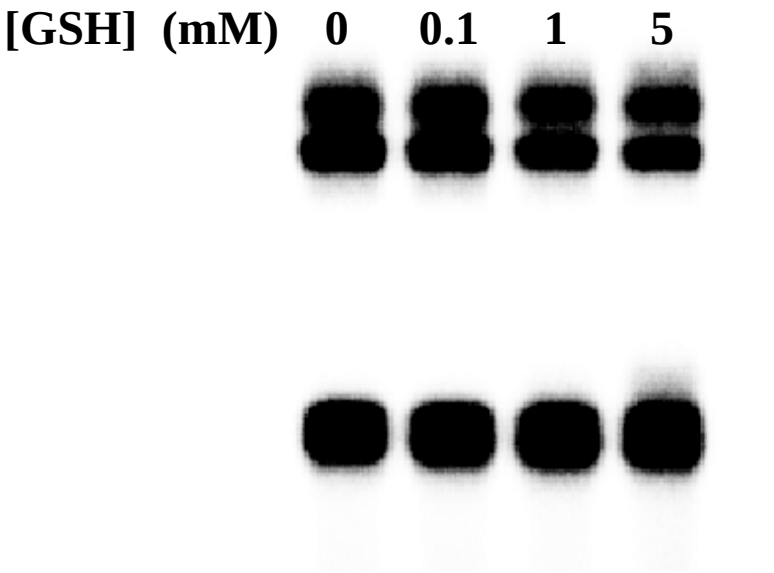
(1) Hong, I. S.; Greenberg, M. M. *J. Am. Chem. Soc.* **2005**, *127*, 3692-3693.
(2) Maniatis, T.; Fritsch, E. F.; Sambrook, J. *Molecular Cloning*; Cold Spring Harbor Laboratory, Cold Spring Harbor, NY., **1982**.



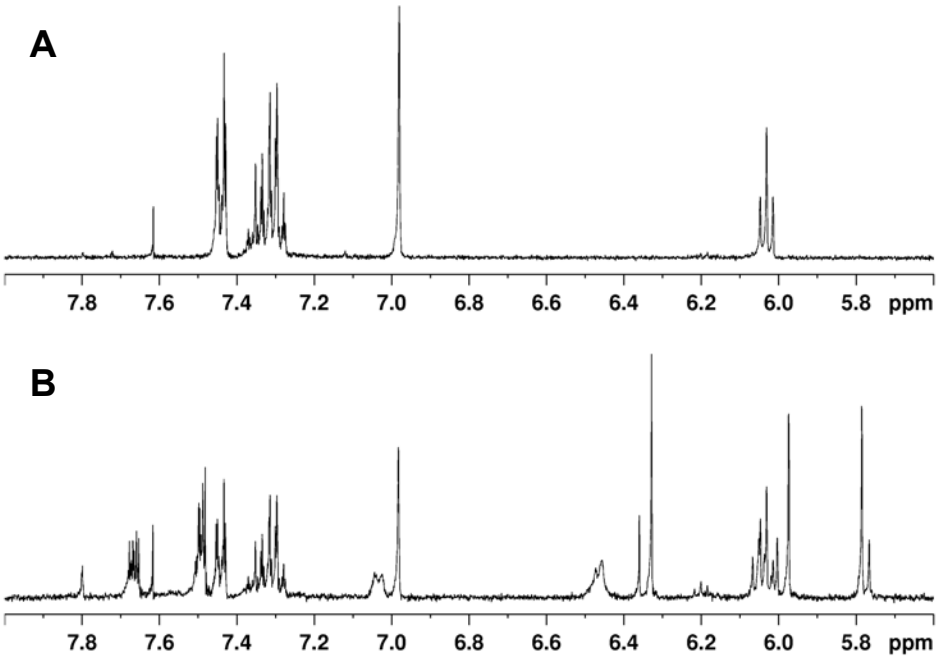
Supporting Information Figure 1. Phosphorimage autoradiogram of **3** (20 nM) decomposed by UV photolysis, Rose Bengal sensitized photolysis, reaction with $NaIO_4$.



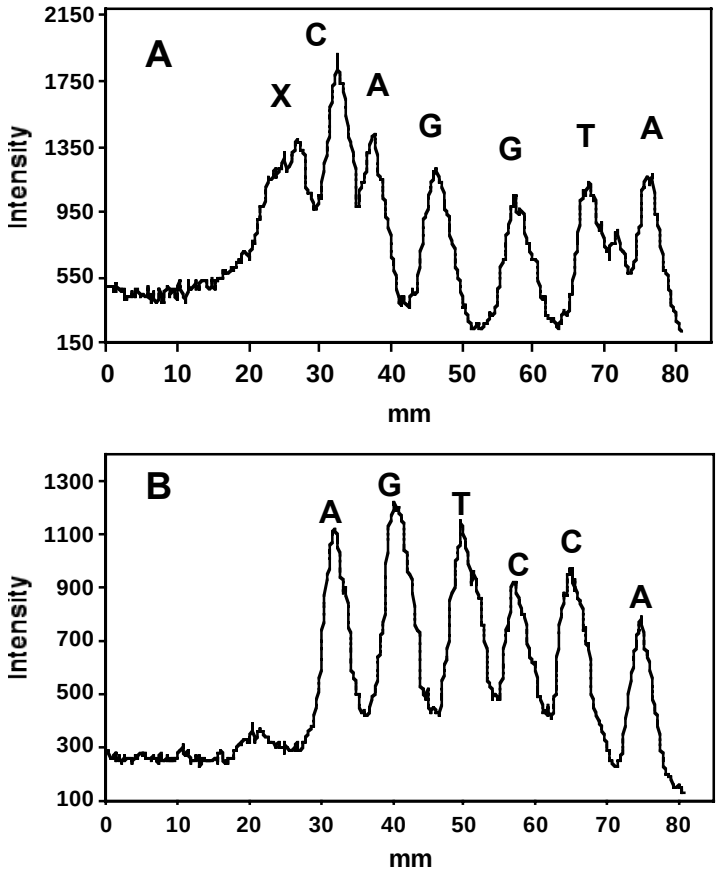
Supporting Information Figure 2. Phosphorimage autoradiogram of ISC products subjected to piperidine and $IrCl_6$ treatment. Arrows indicate cleavage bands resulting from alkali treatment.



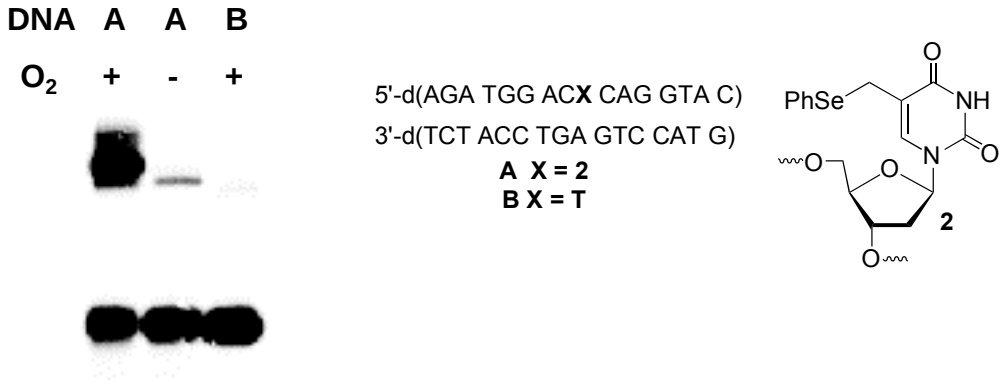
Supporting Information Figure 3. Phosphorimage autoradiogram of the effect of GSH on ISC formation in **3** exposed to Rose Bengal photosensitized photolysis. Photolysis time: 30 min, [Rose Bengal] = 100 μM.



Supporting Information Figure 4. ^{1}H NMR analysis of the photolysis of **2** (500 μM) in deuterated phosphate buffer (50 mM, pD 7.4) by Rose Bengal (50 μM) for 5 min. (A) Before photolysis (B) Immediately after photolysis.



Supporting Information Figure 5. Hydroxyl radical footprinting of ISC produced by reaction of **3** with $NaIO_4$. (A) Strand containing phenyl selenide **2** (B) complementary strand.



Supporting Information Figure 6. Sample phosphorimage autoradiogram of ISC formation in **A** and **B** (10 nM) exposed to Rose Bengal sensitized photolysis. Photolysis time: 30 min, [Rose Bengal] = 50 μM.

Supporting Information Table 1. Rose Bengal mediated ISC formation.

DNA duplex	O_2	ISC (%)
A	yes	55.2 ± 0.02
A	no	3.4 ± 0.6
B	yes	2.4 ± 0.1