Supporting Information

Direct and indirect photochemical reactions in viral RNA measured with RT-qPCR and mass spectrometry

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Rate Constant Analysis. For the UV experiments, first order reaction rate constants for each oligomer with each quantification method were calculated with linear regressions of ln (C) versus UV_{254} dose. Regressions on the MALDI decay data included all of the experimental data, as did the regressions on RT-qPCR data. For the $^1O_2$ experiments, pseudo first order reaction rate constants were determined by linear regression analyses of ln(C) versus time. Second order reaction rate constants were then calculated by dividing the pseudo first order rate constants by the steady-state singlet oxygen concentration (9.0×10^{-11} M).

Prediction of Oligomer Rate Constants with $^1O_2$. We compiled the limited data available from two previous publications to predict reaction rate constants of our oligomers. Wilkinson *et al.* reported that the second order rate constant for guanosine was $\leq 1 \times 10^6$ $M^{-1}s^{-1}$ in water and 6.2×10^6 $M^{-1}s^{-1}$ in D_2O.[1] Clagett and Galen reported the relative reaction rate constants of guanine, uridine, cytidine, and adenosine were 26:13:8:1.[2] Using a value for guanosine equal to 1×10^6 $M^{-1}s^{-1}$, we used the ratios suggested by Clagett and Galen to calculate the maximum rate constants for uridine, cytodine, and adenosine (5×10^5 $M^{-1}s^{-1}$, 3.1×10^5 $M^{-1}s^{-1}$, and 3.9×10^4 $M^{-1}s^{-1}$, respectively). There are several assumptions made here, including that the guanosine rate constant is equal to 1×10^6 $M^{-1}s^{-1}$, despite the fact that Wilkinson reported this as a maximum value, that nucleotides and nucleosides have the same reactivity with $^1O_2$, and that incorporation into an RNA oligomer does not impact the rate constants of the individual nucleotides. This prediction could be improved with more accurate rate constants for the reactions between nucleotides and $^1O_2$.

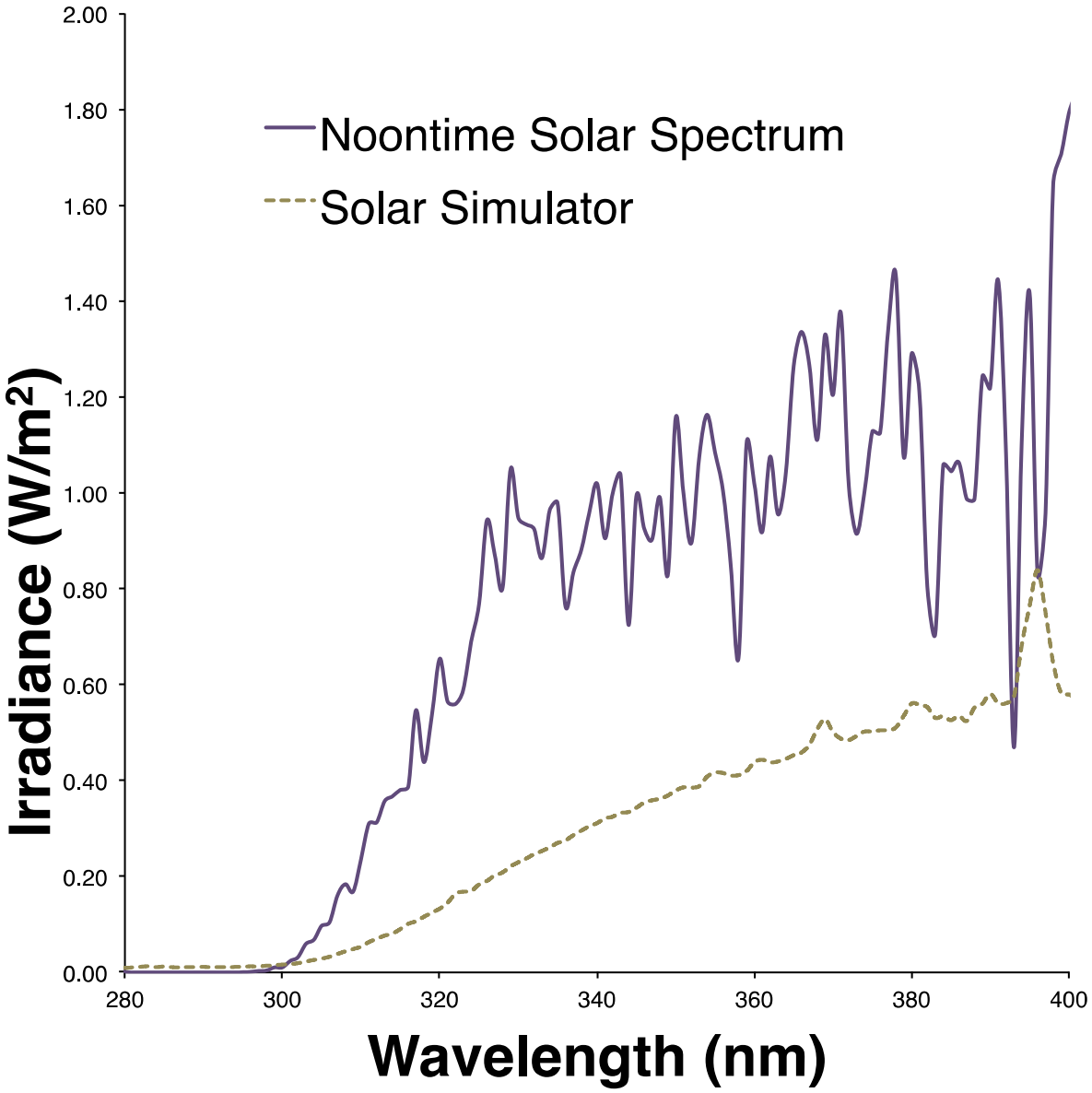

Figure S1. Comparison of solar simulator output spectrum and solar spectrum in Ann Arbor, MI (42.3º N, 83.7º W, 7/30/16, noontime, estimated with Quick TUV Caliculator, http://cprm.acom.ucar.edu/Models/TUV/Interactive_TUV/).

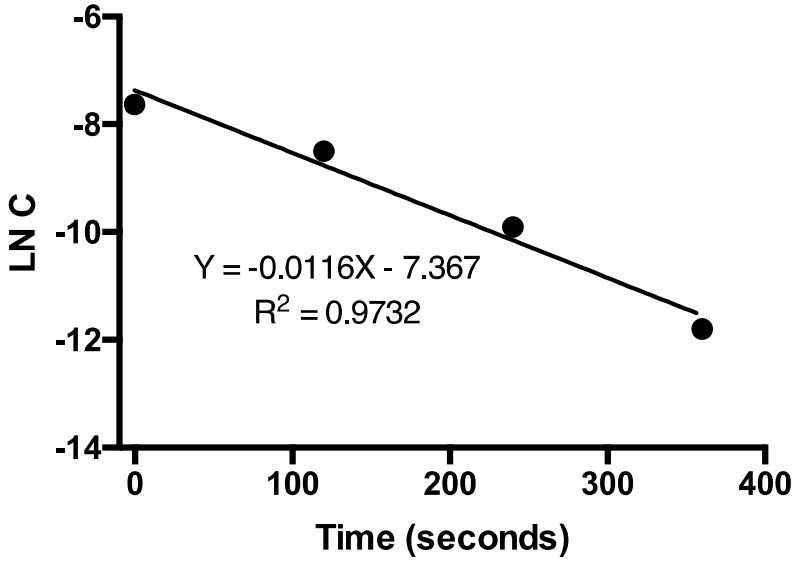

Figure S2. Decrease of furfuryl alcohol (FFA) concentration with simulated solar treatment. FFA serves as probe compound for measuring $^1O_2$ concentration.

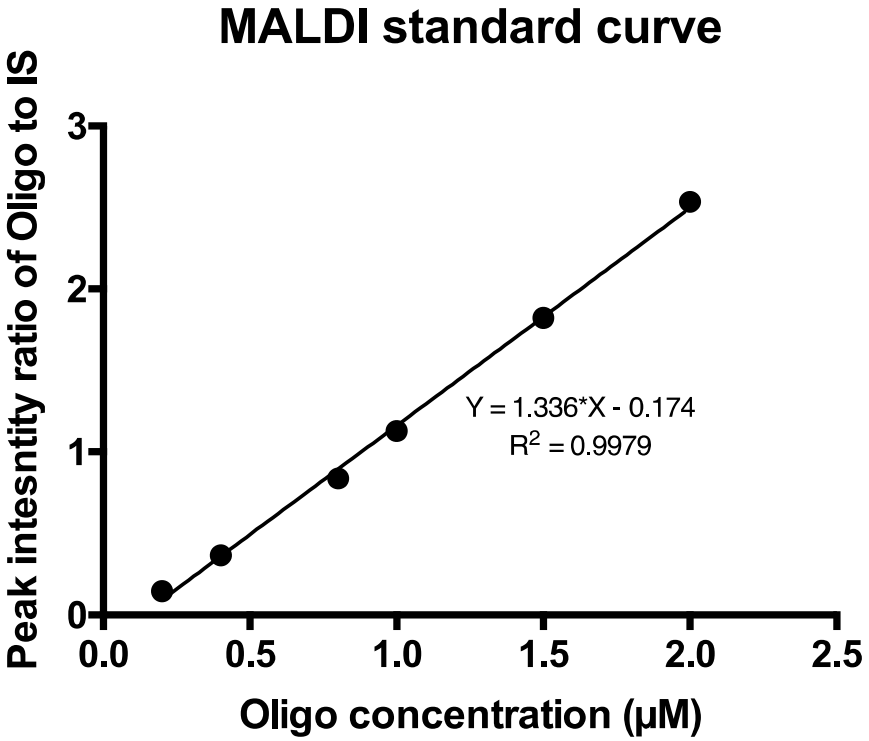

Figure S3. Quantitative MALDI-TOF-MS standard curve of Oligomer A. The concentration of 26-mer internal standard is 1 μM.

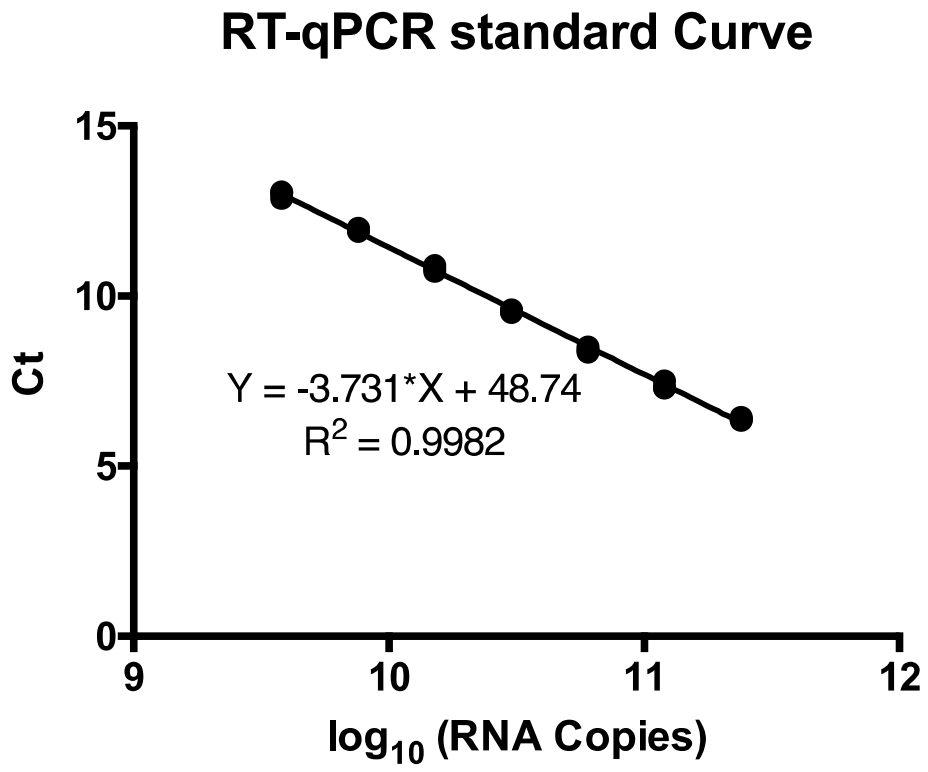

Figure S4. Step-loop RT primer based RT-qPCR standard curve of oligomer A.

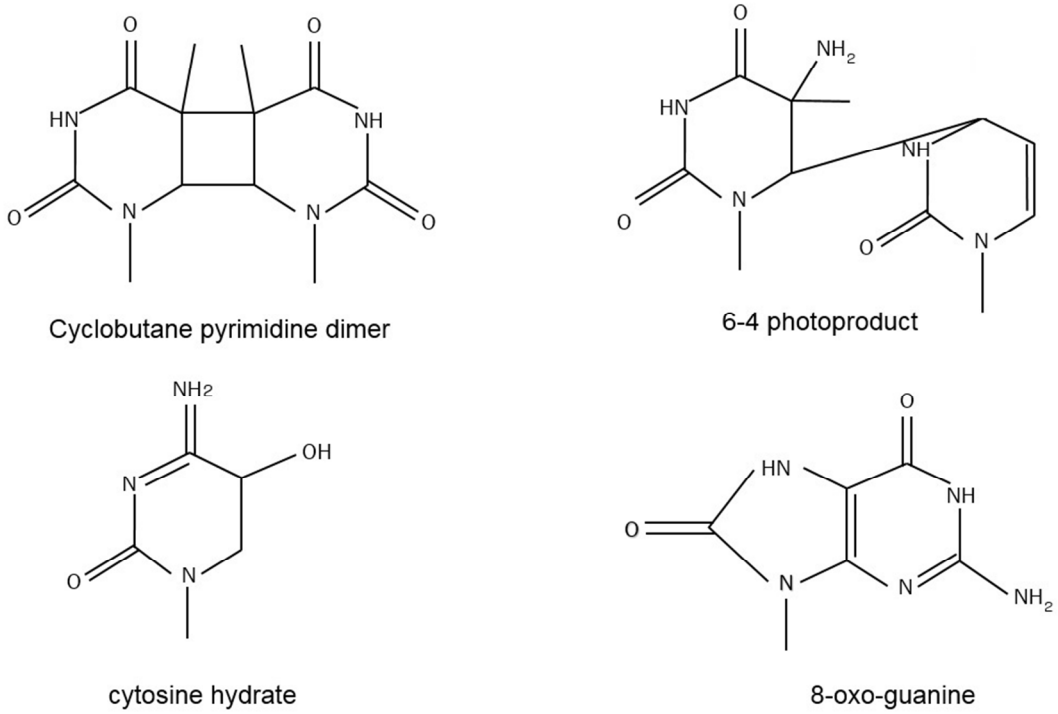

Figure S5. Chemical structure of three major DNA photoproducts and one major oxidation product reported in the literature.

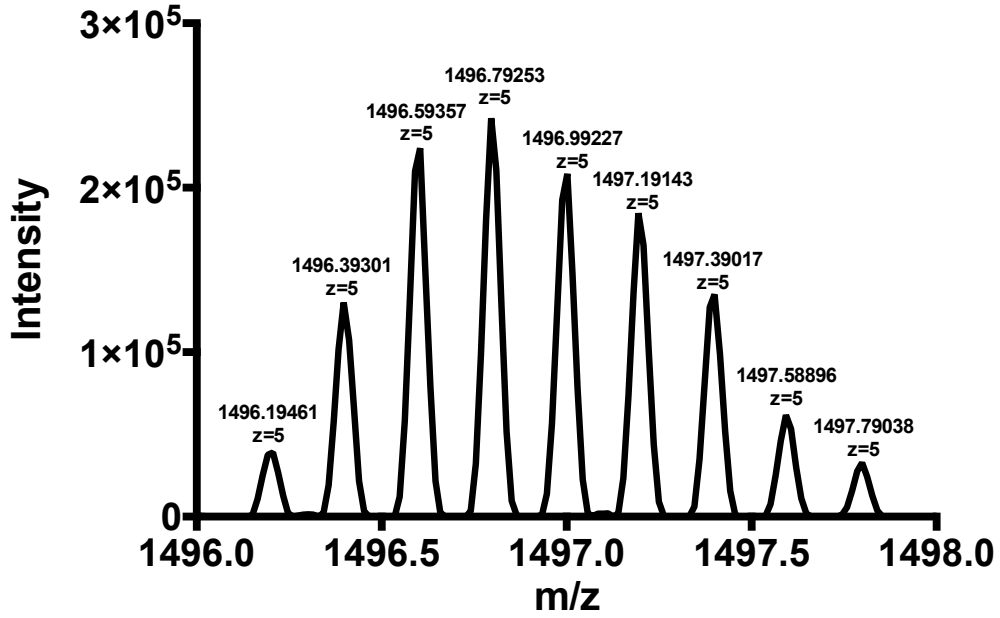

Figure S6. High-resolution mass spectra for double pyrimidine photohydrate after 20 minutes UVC reaction obtained by ESI-Orbitrap mass spectrometer.

Table S1 Time gradient of solvent A (2% HFIP + 0.4% TEA in Water) and solvent B (2% HFIP + 0.4% TEA in Methanol) for ESI-Orbitrap-MS analysis. The flow rate was 300 µL/min.

Time (min)	Solvent A (%)	Solvent B (%)
0.0	90	10
5.0	60	40
6.0	10	90
8.0	10	90
8.1	90	10

Table S2. Prediction of reaction rate constants for the two oligomers with $^1O_2$. The rate constants were predicted by summing up the products of the nucleoside rate constants (described above) and the number of each nucleoside in the oligomers. The sequences of Oligomer A and Oligomer B are provided in Table 1.

RNA segment	# A	# C	# G	# U	Rate constant ($M^{-1}s^{-1}$)
Oligomer A	6	10	1	7	7.8×10^6
Oligomer B	7	5	10	2	1.3×10^7

REFERENCES

(1) Wilkinson, F.; Helman, W. P.; Ross, A. B. Rate constants for the decay and reactions of the lowest electronically excited singlet state of molecular oxygen in solution. An expanded and revised compilation. *J. Phys. Chem. Ref. Data* **1995**, *24*, 663–677.

(2) Clagett, D.; Galen, T. J. Ribonucleoside reactivities with singlet (1Δg) molecular oxygen. *Arch. Biochem. Biophys.* 1971, **146**, 196–201.