Direct Evidence of Singlet Molecular Oxygen Generation from Peroxynitrate, a Decomposition Product of Peroxynitrite

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Supporting Information

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Figure S1. Quantification of O$_2$NOOH by spectro-iodometry. The quantification is based on the rapid reaction of O$_2$NOOH with iodide (eq. 1), which has an intense absorption at 352 nm ($\varepsilon = 26400$ M$^{-1}$ cm$^{-1}$). The reaction of H$_2$O$_2$ with iodide (eq. 2) undergoes at an appreciable rate only after the addition of ammonium molybdate as a catalyst. For the experiment, 2 ml of 8 mM KI and 20 µl of 2.4 M HNO$_3$ were pipetted into a cuvette. After recording the baseline, 20 µl of the diluted (1000 times) O$_2$NOOH solution in 2.4 M HNO$_3$ was added and the absorbance was measured for about 1 min. To determine the H$_2$O$_2$ concentration, 20 µl of 2% ammonium molybdate solution was added and the absorbance was recorded until a plateau was reached. I$_3^-$ has a strong absorption at 352 nm and its concentration was determined by using its absorption coefficient of 26400 M$^{-1}$ cm$^{-1}$.
**Figure S2.** Kinetics of the decay of monomolecular light emission at 1270 nm due to O₂ (¹Δg) generated during decomposition of 10 mM DHPNO₂ incubated in 0.1 M phosphate buffer pD 7.8: A) decay curve of data collected for 3810 s (63.5 min), B) expanded view of the Intensity – time curve in the first 500 s, which show the region selected for integration, and C), the area integrated from 300 – 400 s. Thermolysis of DHPNO₂ follows first-order kinetics [30]: based on the half-life of decomposition of DHPNO₂ at 37°C \( t_{1/2} = \ln 2/k = 23 \text{ min} \), the calculated value for the first-order rate constant \( k \) is \( 5.02 \times 10^{-4} \text{ s}^{-1} \). Taking into account that thermolysis of DHPNO₂ yields 59% O₂ (¹Δg) [30], the estimated rate of O₂ (¹Δg) production from 10 mM DHPNO₂ at 37°C is 2.96 μM.s⁻¹. The area obtained by integrating the light emission intensity over a
period of 100 s yielded a value of 58140 (arbitrary units), which corresponds to 296 μM of O₂ ("Δg"). This value was used to convert integrated area to [O₂ ("Δg")].
Figure S3. Time course for the emission of light from of O$_2$ ($^1\Delta_g$) generated during injection of 0.9 M phosphate buffer at pH 7.6 into 1.5 ml of 1 mM ONOO$^-$ in the absence of a) and in the presence of 10 mM b) NaNO$_2$ or c)NaNO$_3$. 
Figure S4. Influence of pD on the amount of EAS consumed during incubation with ONOO⁻. Reaction conditions are the same as those described in Figure S3.