Rate of ON−OO− Bond Homolysis and the Gibbs Energy of Formation of Peroxynitrite

2 AUTHORS, INCLUDING:

Gregory Poskrebyshev
California Institute of Technology

42 PUBLICATIONS   **455** CITATIONS

Rate of ON−OO⁻ Bond Homolysis and the Gibbs Energy of Formation of Peroxynitrite

Sergei V. Lymar* and Gregory A. Poskrebyshev†

Chemistry Department, Brookhaven National Laboratory, Upton, New York 11973

*Received: May 22, 2003; In Final Form: July 21, 2003*

Oxidation of the methyl viologen radical, $MV^{\bullet+}$, by the peroxynitrite anion, $ONOO^-$, occurs through an indirect pathway, which includes reversible homolytic dissociation of $ONOO^-$ into a pair of the $NO^\bullet$ and $O_2^{\bullet-}$ radicals (rate-determining step, $k_h = 0.017 \pm 0.001$ s⁻¹) followed by rapid oxidation of $MV^{\bullet+}$ by $O_2^{\bullet-}$ with $k_{so} = (3.5 \pm 0.2) \times 10^9$ M⁻¹ s⁻¹ at zero ionic strength. Direct oxidation of $MV^{\bullet+}$ radical through a bimolecular reaction with $ONOO^-$ is immeasurably slow ($k < 1 \times 10^3$ M⁻¹ s⁻¹). The $MV^{\bullet+}$ radical does not rapidly reduce $NO^\bullet$, suggesting that the one-electron reduction potential of the latter is significantly below −0.45 V (NHE), consistent with the recently reported reevaluations of $NO^\bullet$ reduction energetics. The equilibrium constant $K_h = (3.6 \pm 0.3) \times 10^{-12}$ M for the dissociation of $ONOO^-$ into $NO^\bullet$ and $O_2^{\bullet-}$ is determined from the measurements of rate constants for the forward and reverse (recombination of $NO^\bullet$ and $O_2^{\bullet-}$, $k_{-h} = (4.8 \pm 0.3) \times 10^9$ M⁻¹ s⁻¹) reactions. The free energies of formation in aqueous solution $\Delta_f G°(ONOO^-) = 68.5 \pm 1.4$ kJ/mol and $\Delta_f G°(ONOOH) = 30.8 \pm 1.5$ kJ/mol are derived from these data. The last value is fully consistent with the major role of the ONOOH homolytic dissociation into a pair of the $NO_2^\bullet$ and $OH^\bullet$ radicals in oxidations by peroxynitrite. Collectively, these results help to resolve disputes in the literature over the values for k_h, k_{-h}, K_h, $\Delta_f G°(ONOO^-)$, and $\Delta_f G°(ONOOH)$; measurements of all these quantities in this work disagree with their recent reevaluations, but largely agree with earlier reports.

Introduction

Many inorganic and practically all organic compounds are oxidized to some extent in the presence of peroxynitrite anion, $ONOO^-$, and/or its conjugate peroxynitrous acid, ONOOH. For this reason, peroxynitrite is commonly referred to as being a strong oxidant. Despite a long history of investigations, the mechanisms by which the oxidations are administered remain a subject of abiding debate. The contention concerns mainly the rates and the yields of homolytic dissociations involving the ON−OO⁻ and the ONO−OH bonds, as shown in Scheme 1. Efficient O−O bond homolysis in ONOOH would distinguish it from all other known hydroperoxides, which do not decompose in such a manner with significant rates under normal conditions. This homolysis generates strongly oxidizing $NO_2^\bullet$ and $OH^\bullet$ radicals and, as has been maintained by several research groups,[1−6] is the main mechanism for oxidations by ONOOH, the so-called "indirect mechanism". In opposition, it has been argued that such a homolysis does not occur to a significant extent and, thus, plays little or no role in the decomposition and oxidative chemistry of peroxynitrite.[7−9]

The Gibbs energy of formation for aqueous peroxynitrite, $\Delta_f G°(ONOO^-)$, is germane for understanding its redox properties and reactivity. It is also at the core of the ongoing debate on the role of homolysis pathways, which is not always explicit but can be understood with the help of Scheme 1. The recombination of $NO_2^\bullet$ and $OH^\bullet$ has been shown to partition between the formation of nitrate and peroxynitrous acid.[3,10] By virtue of microscopic reversibility, O−O bond homolysis must occur and its contribution to the decay of ONOOH depends on

SCHEME 1
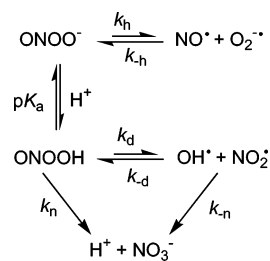
the magnitude of equilibrium constant $K_d = k_d/k_{-d}$, whose value is related to $\Delta_f G°(ONOO^-)$ through the $pK_a \approx 6.6$ of ONOOH. The first experimental value of $\Delta_f G°(ONOO^-)$ was obtained by Merényi and Lind[2] from evaluation of the equilibrium constant $K_h = k_h/k_{-h}$ for reversible homolytic dissociation of $ONOO^-$ (Scheme 1). Tetranitromethane, TNM, was used to scavenge the radical reaction products and the readily observable nitroform anion, NF⁻, was generated in the process. Subsequently, the validity of these measurements was challenged[11] and the challenge was rebutted.[12] Apparently, the measurements of NF⁻ accumulation kinetics in the presence of $ONOO^-$ are prone to complications that include, but are not limited to, concurrent NF⁻ formation through the peroxynitrite-independent alkaline hydrolysis of TNM and generation of the N_2O_3 reactive intermediate during radical scavenging; the latter may trigger a chain decomposition of $ONOO^-$ under certain conditions.[13] These complications may be at least partially responsible for the different outcomes of the k_{-h} determinations in the hands of different researchers using the TNM system.

The widely accepted earlier measurements[14−16] of the second parameter required for evaluating K_h, namely k_{-h}, have also recently been revised upward by about a factor of 3.[17] In the hope of resolving these issues, we have used pulse radiolysis

* Corresponding author. Tel: 631-344-4333. Fax: 631-344-5815. E-mail: lymar@bnl.gov.

† On leave from the Institute of Energy Problems of Chemical Physics, Russian Academy of Sciences, Moscow 117829, Russia.
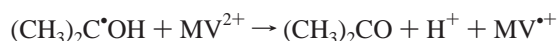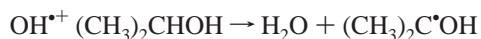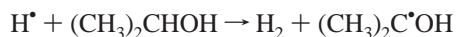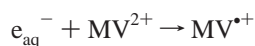
7992 *J. Phys. Chem. A, Vol. 107, No. 39, 2003*

Lymar and Poskrebyshev

to redetermine k_{-h} and to obtain data that allow determination of k_h and $\Delta_f G°(\text{ONOO}^-)$ without the use of TNM. The method involves measurements of the methyl viologen radical, $\text{MV}^{\bullet+}$, decay in the presence of ONOO^- and is free of complications. Our results are fully consistent with the major role of the ONOOH homolytic dissociation into a pair of the $\text{NO}_2^\bullet$ and $\text{OH}^\bullet$ radicals in oxidations by peroxynitrite. We also show that peroxynitrite anion is very inefficient as an outer-sphere one-electron oxidant.

Experimental Section

Analytical grade chemicals and Milli-Q purified (ASTM type I) water were used throughout. Peroxynitrite was prepared[5] in a tandem quench-flow mixing apparatus from NaNO_2 (taken in 10% stoichiometric excess) and H_2O_2 acidified with HClO_4; concentrated NaOH was used for quenching. Compressed nitric oxide mixtures in argon were prepared by filling an evacuated cylinder with individual compressed gases to the desired partial pressures. The gas mixtures were purified from N_2O_3 by passing the gas through a scrubbing column with 40% KOH and then through water in an all-glass-and-metal home-built apparatus. The apparatus was attached directly to the pulse radiolysis cell and all solutions in the cell and the apparatus itself were thoroughly purged with helium prior to introducing nitric oxide. The solubility of NO in water was taken as 1.9 mM/atm. The pulse radiolysis was carried out with 2 MeV electrons from a van de Graaff accelerator; pulse widths were in the range 0.06–0.8 μs. Dosimetry was performed with N_2O-saturated 10 mM KSCN solution using $G\epsilon = 4.87 \times 10^4$ ions (100 eV)$^{-1}$ M^{-1} cm^{-1} at 472 nm. All solutions were subjected to pulse radiolysis only once and then replaced with a fresh aliquot, except when the effect of repetitive irradiation of the same solution was investigated. The oxidation of $\text{MV}^{\bullet+}$ was followed at 600 nm, where the radical exhibits strong characteristic absorption with molar absorptivity of 1.4×10^4 M^{-1} cm^{-1}.[18] All experiments were done with temperature stabilization at 25 $\pm$ 0.1 °C. The uncertainties for all rate constants are based on their standard deviations obtained from either linear regressions or multiple determinations; the conventional method for propagation of independent errors[19] was used to calculate the standard deviations of derived quantities.

Results

All three primary radicals from water radiolysis (e_{aq}^-, $\text{OH}^\bullet$, and $\text{H}^\bullet$) can be converted into the $\text{MV}^{\bullet+}$ radical in an oxygen-free solution containing large excess of isopropyl alcohol, i.e., the reactions

$$\text{e}_{aq}^- + \text{MV}^{2+} \rightarrow \text{MV}^{\bullet+}$$

$$\text{H}^\bullet + (\text{CH}_3)_2\text{CHOH} \rightarrow \text{H}_2 + (\text{CH}_3)_2\text{C}^\bullet\text{OH}$$

$$\text{OH}^{\bullet+} (\text{CH}_3)_2\text{CHOH} \rightarrow \text{H}_2\text{O} + (\text{CH}_3)_2\text{C}^\bullet\text{OH}$$

$$(\text{CH}_3)_2\text{C}^\bullet\text{OH} + \text{MV}^{2+} \rightarrow (\text{CH}_3)_2\text{CO} + \text{H}^+ + \text{MV}^{\bullet+}$$

Under conditions employed in this study, all these reactions are sufficiently rapid to be complete within a few tens of microseconds after the radiation pulse.[20,21] The $\text{MV}^{\bullet+}$ radical is stable; accordingly, we have not detected its decay on time scales of seconds.[22] Importantly, no measurable loss of $\text{MV}^{\bullet+}$ was observed even in the presence of significant amount of the $\text{NO}^\bullet$ radical in solution (Figure 1, upper trace). In contrast, relatively rapid disappearance $\text{MV}^{\bullet+}$ occurred in the presence of perox-
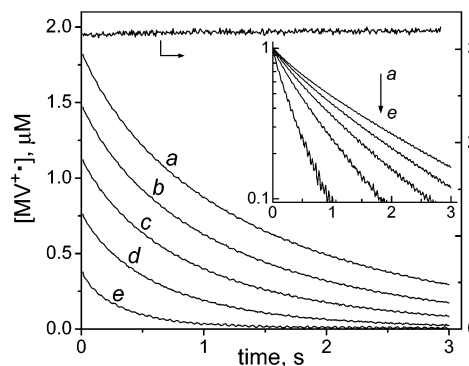

Figure 1. Decay of $\text{MV}^{\bullet+}$ in the presence of ONOO^- (traces a–e corresponding to different radiation doses, left axis). Conditions: 10 μM MV^{2+}, 98 μM ONOO^-, 1.3 M $(\text{CH}_3)_2\text{CHOH}$, 20 mM NaOH, Ar-saturated solution. The upper flat trace (right axis) shows the absence of $\text{MV}^{\bullet+}$ decay in the presence of $\text{NO}^\bullet$ and the absence of ONOO^-. Conditions: 1.7 mM MV^{2+}, 0.65 M $(\text{CH}_3)_2\text{CHOH}$, 32 mM NaOH, solution saturated with 1% $\text{NO}^\bullet$ in Ar (approximately 20 μM in $\text{NO}^\bullet$). The inset shows a semilogarithmic representation of the normalized traces a–e (log($[\text{MV}^{\bullet+}]_t/[\text{MV}^{\bullet+}]_0$) vs t).
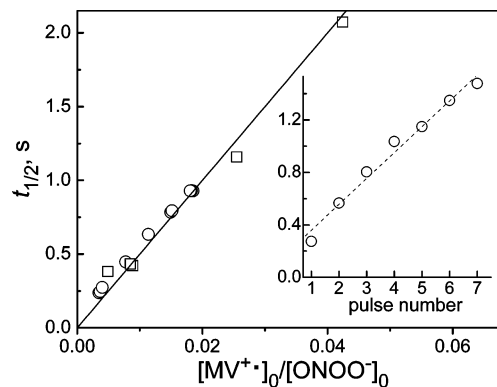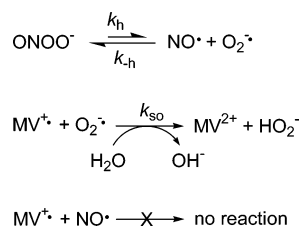

Figure 2. Half-life of the $\text{MV}^{\bullet+}$ radical in the presence of ONOO^- as a function of $[\text{MV}^{\bullet+}]_0/[\text{ONOO}^-]_0$. All solutions are 20 mM in NaOH and Ar-saturated. Key: (○) 10 μM MV^{2+}, 98 μM ONOO^-, 1.3 M $(\text{CH}_3)_2\text{CHOH}$; (□) 100 μM MV^{2+}, 100 μM ONOO^-, 0.65 M $(\text{CH}_3)_2$-CHOH. The solid line gives a linear fit through the origin with a slope of 50 s. The inset shows the half-life increase upon repetitive low-dose irradiation of the same solution; the time intervals between consecutive pulses allowed for a complete $\text{MV}^{\bullet+}$ decay.

ynitrite (Figure 1, traces a through e), which is attributable to the apparent net oxidation of $\text{MV}^{\bullet+}$ by ONOO^-.

Despite the large excess of ONOO^- over $\text{MV}^{\bullet+}$, the kinetics deviate significantly from exponential decay, so that the slopes in the semilogarithmic plot in Figure 1 decrease with time. Furthermore, the initial slopes depend on the initial concentrations of the viologen radical, $[\text{MV}^{\bullet+}]_0$. At essentially constant initial concentration $[\text{ONOO}^-]_0$, the decay half-time increases linearly with $[\text{MV}^{\bullet+}]_0$, as shown in Figure 2 for the two sets of conditions differing mainly in the concentration of added MV^{2+}. Although the same $t_{1/2}$ vs $[\text{MV}^{\bullet+}]_0/[\text{ONOO}^-]_0$ dependence is observed, the $\text{MV}^{\bullet+}$ radiation yield is about 20% lower at 10 μM than at 100 μM $[\text{MV}^{2+}]$ ($G \approx 5.9$), which may indicate that a minor fraction of reducing species (e_{aq}^- and/or $(\text{CH}_3)_2\text{C}^\bullet\text{-OH}$) have reacted with ONOO^-.

The observed kinetic peculiarities rule out direct oxidation of $\text{MV}^{\bullet+}$ by ONOO^- as the main pathway for the $\text{MV}^{\bullet+}$ decay. Indeed, if this were the case, all the slopes in Figure 1 (inset) would be equal and the half-lives in Figure 2 would scatter around a horizontal line. The observed kinetics are, however, completely consistent with the reaction mechanism shown in Scheme 2. Its salient features include reversible, rate-determin-

ON−OO⁻ Bond Homolysis

J. Phys. Chem. A, Vol. 107, No. 39, 2003 **7993**

SCHEME 2

$$ONOO^- \xrightleftharpoons[k_{-h}]{k_h} NO^\bullet + O_2^{\bullet -}$$

$$MV^{\bullet +} + O_2^{\bullet -} \xrightarrow{k_{so}} MV^{2+} + HO_2^-$$
$$\qquad\quad H_2O \qquad OH^-$$

$$MV^{\bullet +} + NO^\bullet \xrightarrow{\quad\times\quad} \text{no reaction}$$

ing, homolytic dissociation of $ONOO^-$, the rapid scavenging of $O_2^{\bullet -}$ by $MV^{\bullet +}$, and the absence of $MV^{\bullet +}$ decay due to concomitant accumulation of $NO^\bullet$.

The latter has been experimentally verified in Figure 1 and the very rapid reduction of $O_2^{\bullet -}$ by $MV^{\bullet +}$ has been reported.[23,24] The product of this reaction, hydrogen peroxide, also reacts with $MV^{\bullet +}$, but the rate constants for both H_2O_2 and HO_2^- are under $10\ M^{-1}\ s^{-1}$,[24,25] so that this reaction should be completely inconsequential under our conditions. As there is one report of the rapid $MV^{\bullet +} + H_2O_2$ reaction,[26] we have conducted a control experiment. No decay of $MV^{\bullet +}$ due to hydrogen peroxide was observed on the time scale of interest even at $[H_2O_2]$ many times exceeding the maximal $[MV^{\bullet +}]_0$ used in this study (Supporting Information, Figure S1). We thus suggest that oxidation by the superoxide anion is the dominant channel for the viologen radical decay. As this reaction proceeds, the $NO^\bullet$ radical accumulates and the nearly diffusion-controlled recombination of $NO^\bullet$ with $O_2^{\bullet -}$ begins to compete with the $MV^{\bullet +} + O_2^{\bullet -}$ reaction, slowing it down. These considerations lead to a readily verifiable prediction that the half-life of $MV^{\bullet +}$ should monotonically increase upon repetitive pulse irradiation of the same $ONOO^-$ solution. Such an increase has been indeed observed, as shown by the inset in Figure 2.
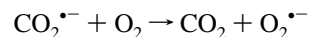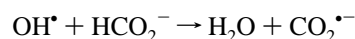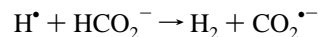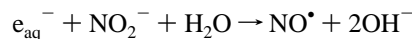
As long as $k_h \ll 10\ s^{-1}$, which is the case here, the concentration of $O_2^{\bullet -}$ can be treated as being in a steady-state and an analytical solution can be obtained for the kinetics of $MV^{\bullet +}$ decay described by Scheme 2 (Supporting Information, note 1). Although the resulting transcendental equation is unwieldy, a convenient expression for the half-life of viologen radical can be derived

$$t_{1/2} = \left\{ \frac{1}{2} + (\ln 2 - \frac{1}{2})\frac{k_{-h}}{k_{so}} \right\} \frac{1}{k_h} \frac{[MV^{\bullet +}]_0}{[ONOO^-]_0} \approx$$
$$\left(0.5 + 0.193\frac{k_{-h}}{k_{so}} \right)\frac{1}{k_h} \frac{[MV^{\bullet +}]_0}{[ONOO^-]_0}$$

As shown in Figure 2, a linear relationship between $t_{1/2}$ and the $[MV^{\bullet +}]_0/[ONOO^-]_0$ ratio was indeed observed with the slope 50 ± 1 s. The value of k_h can be obtained from this slope, if the ratio k_{-h}/k_{so} is known; notably, the small coefficient at this ratio significantly attenuates its influence on the slope. The two reported direct measurements of k_{so} (0.9×10^9 [23] and $2.8 \times 10^9\ M^{-1}\ s^{-1}$ [24]) disagree significantly. A similar situation exists with the value of k_{-h}, which is more critical for the purposes of the present study, for it will be used also to evaluate K_h. Relatively consistent early k_{-h} measurements ranging from 3.8×10^9 to $6.7 \times 10^9\ M^{-1}\ s^{-1}$ from three research groups were more recently questioned by Koppenol and co-workers, who reported about a 3 times larger k_{-h} value of $(16-19) \times 10^9\ M^{-1}\ s^{-1}$ (Table 1).

These discrepancies have prompted us to undertake re-determinations of k_{so} and k_{-h}. The value $k_{so} = (2.6 \pm 0.2) \times 10^9\ M^{-1}\ s^{-1}$ at 10 mM ionic strength, which is essentially

identical to the result of Levey and Ebbesen,[24] was obtained by measuring $MV^{\bullet +}$ decay with variable excess of $O_2^{\bullet -}$ in alkali (for details and comparison with previous reports, see Supporting Information, note 2). A typical result of k_{-h} determination is shown in Figure 3. In this experiment, both $NO^\bullet$ and $O_2^{\bullet -}$ were rapidly generated in nearly equal amounts from the primary radicals of water radiolysis in an air-saturated alkaline solution of formate and nitrite through the major reactions

$$e_{aq}^- + NO_2^- + H_2O \rightarrow NO^\bullet + 2OH^-$$

$$H^\bullet + HCO_2^- \rightarrow H_2 + CO_2^{\bullet -}$$

$$OH^\bullet + HCO_2^- \rightarrow H_2O + CO_2^{\bullet -}$$

$$CO_2^{\bullet -} + O_2 \rightarrow CO_2 + O_2^{\bullet -}$$

after which the much slower $NO^\bullet + O_2^{\bullet -} \rightarrow ONOO^-$ reaction was cleanly observed by monitoring the growth of characteristic $ONOO^-$ absorption around 300 nm.

Under our conditions, less than 10% of e_{aq}^- is predicted from known rate constants[20] to react with O_2 generating $O_2^{\bullet -}$; this small complication is partially offset by about the same fraction of $H^\bullet$ reacting with NO_2^- to yield $NO^\bullet$.[31] In a separate experiment without oxygen and nitrite, radiation yields of $G(e_{aq}^-) = 3.1$ and $G(OH^\bullet + H^\bullet) = 3.4$ radicals per 100 eV were determined in 1 M formate from the characteristic absorptions due to e_{aq}^- and $CO_2^{\bullet -}$.[32] With these data, a simple kinetic simulation using k_{-h} as the only adjustable parameter gives a good fit to the data with $k_{-h} = 4.8 \times 10^9\ M^{-1}\ s^{-1}$ (for simulation details, see Supporting Information, note 3). The standard deviation of k_{-h} from 18 individual fits is $0.14 \times 10^9\ M^{-1}\ s^{-1}$; this number is conservatively doubled in Table 1 to allow for slight influence on the fits of other fixed rate constants used in simulations. Our k_{-h} value agrees well with the $k_{-h} = (4.3 \pm 0.5) \times 10^9\ M^{-1}\ s^{-1}$ determination by Goldstein and Czapski[16] and is comfortably close to other early k_{-h} measurements (Table 1). In contrast, the latest k_{-h} value of $16 \times 10^9\ M^{-1}\ s^{-1}$ reported by Nauser and Koppenol[17] is clearly inconsistent with our data (Figure 3).

With k_{so} and k_{-h} measured in this work, the value of $k_h = 0.017 \pm 0.001\ s^{-1}$ is obtained from the slope of the solid line in Figure 2. This value is identical to k_h originally determined by Merényi and Lind with an entirely different experimental approach.[2] This excellent agreement lends a strong support to the mechanism of $MV^{\bullet +}$ oxidation presented in Scheme 2. Our data also clearly show that the homolytic dissociation of $ONOO^-$ is significantly suppressed by the presence of even micromolar concentrations of $NO^\bullet$, contrary to the results of Koppenol and co-workers.[11]

Discussion

From our measurements, the equilibrium constant for the homolysis of $ONOO^-$ is $K_h = k_h/k_{-h} = 3.6 \times 10^{-12}$ M, with about 10% uncertainty (Table 1). Thus, the free energy change in the homolysis reaction is $\Delta_h G° = 65.3 \pm 0.2$ kJ/mol, which, together with tabulated $\Delta_f G°$ values for $NO^\bullet$ and $O_2^{\bullet -}$,[30] gives the free energy of formation $\Delta_f G° (ONOO^-) = 68.5 \pm 1.4$ kJ/mol. This free energy practically coincides with that derived earlier by Merényi and Lind[2] and is significantly higher than the value of 59 kJ/mol recently proposed[11] as "preferred" by Koppenol and co-workers (Table 1). Considering the unusually generous uncertainty of 13 kJ/mol given to the latter number and that this number is not entirely experimental, unlike those reported previously and here, the reasons for its preferred

TABLE 1: Kinetic and Thermodynamic Parameters Pertaining to the Homolytic Decompositions of ONOO⁻ and ONOOH

parameter	value
k_h, 10^{-2} s^{-1}	1.7;[2] neg[a];[11] 2.3;[12] 1.7 ± 0.1 (this work)
k_{-h}, 10^9 M^{-1} s^{-1}	6.7 ± 0.9;[14] 3.8;[15] 4.3 ± 0.5;[16] 19 ± 2;[27] 16 ± 3;[17] 4.8 ± 0.3 (this work)
K_h, 10^{-12} M	3.1;[2] neg[a];[11] 3.6 ± 0.3 (this work)
$\Delta_f G°$(ONOO⁻), kJ/mol	42 ± 8;[7] 69.5;[2] 59 ± 13;[9,11] 67.4 ± 1.3;[28] 68.5 ± 1.4 (this work)[b]
$\Delta_f G°$(ONOOH), kJ/mol	4 ± 8;[7] 32.2;[2,29] 21 ± 13;[9] 29.7 ± 0.8;[28] 30.8 ± 1.5 (this work)[c]

[a] Negligibly small and inconsequential for reactivity. [b] A 1 kJ/mol uncertainty was assigned to the tabulated[30] values of $\Delta_f G°$(NO•) and $\Delta_f G°$(O$_2$•⁻) used for this estimate. [c] A 0.1 unit uncertainty in pK_a(ONOOH) was applied.
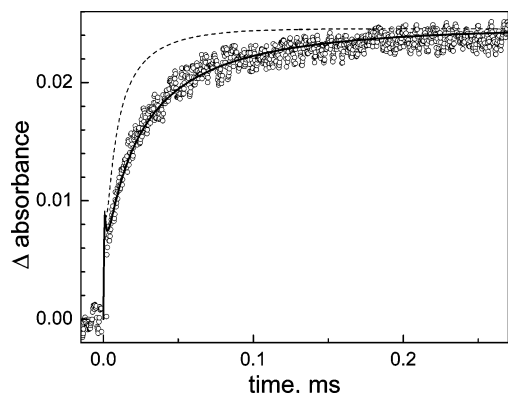

Figure 3. Kinetic trace recorded at 300 nm and showing accumulation of ONOO⁻ in air-saturated solution containing 1 M NaHCO$_2$, 10 mM NaNO$_2$, and 1 mM NaOH. Initial concentrations of radicals are 7.8 μM for [e⁻$_{aq}$] and 8.6 μM for [OH•] + [H•]. Solid and dashed lines give the simulated absorbance profiles with $k_{-h} = 4.8 \times 10^9$ M^{-1} s^{-1} and $k_{-h} = 16 \times 10^9$ M^{-1} s^{-1}, respectively. A prompt rise of initial absorbance is mainly due to the CO$_2$•⁻ radical absorption at this wavelength and is reproduced by the simulations.

designation are not clear. Accepting 6.6 ± 0.1 as the pK_a of ONOOH,[10,16,27,33] we obtain $\Delta_f G°$(ONOOH) = 30.8 ± 1.5 kJ/mol. Lately, the value of $\Delta_f G°$(ONOOH) = 29.7 ± 0.8 kJ/mol has been measured by yet another direct method.[28] It should be emphasized that the accurate measurement of $\Delta_f G°$(ONOOH) is much more than just an idle pursuit of precision because this free energy defines the equilibrium constant, K_d, for homolytic dissociation of ONOOH into NO$_2$• and OH• (Scheme 1) and thus controls the radical yield; the latter is important for understanding peroxynitrite chemistry and biology. Equally important, the reduction potentials of ONOO⁻ and ONOOH can only be obtained by means of their Gibbs energies of formation. The midpoint one-electron and two-electron reduction potentials for peroxynitrite at various pH are presented in Figure 4; the corresponding standard potentials are $E°$(ONOOH,H⁺/NO$_2$•,H$_2$O) = 2.12 V and $E°$(ONOOH,2H⁺/HNO$_2$,H$_2$O) = 1.68 V vs NHE.

With known values for $\Delta_f G°$(NO$_2$•) and $\Delta_f G°$(OH•),[30,36] we obtain the free energy change in the ONOOH homolysis reaction $\Delta_d G° = 57.3$ kJ/mol and its equilibrium constant $K_d = 9.2 \times 10^{-11}$ M. As pointed out in the Introduction section, the issue of NO$_2$• and OH• production from ONOOH is strictly quantitative, for denying the existence of this pathway would violate the microscopic reversibility principle. Both the decomposition of ONOOH and the recombination of NO$_2$• and OH• undoubtedly proceed through the intermediacy of a solvent cage containing these radicals, as has been previously pointed out on several occasions.[5,6,29] However, these details have no bearing on the considerations presented here and have been omitted from Scheme 1 for clarity. With the help of Scheme 1, the yield of radicals from ONOOH decomposition, Y_d, can be expressed through the experimentally determinable parameters
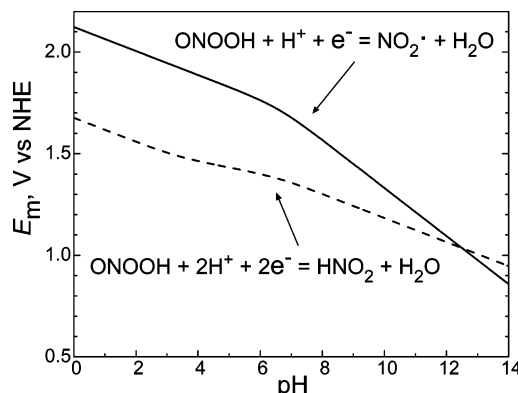

Figure 4. Variations with pH of the midpoint potentials, E_m, for one-electron (solid line) and two-electron (dashed line) reductions of peroxynitrite. The dependencies are based on the thermodynamic values for ONOO⁻ and ONOOH obtained in this work and the literature values for NO$_2$•,[30] water species,[34] and nitrite/nitrous acid.[34,35] At E_m, [ONOOH] + [ONOO⁻] = [NO$_2$•] for the one-electron and [ONOOH] + [ONOO⁻] = [HNO$_2$] + [NO$_2$⁻] for the two-electron process.

$$Y_d = \frac{k_d}{k_d + k_n} = \tau\, k_{-d} K_d$$

where $\tau = 1/(k_d + k_n)$ is the lifetime of ONOOH; at 25 °C, $\tau = 0.83$ s.[27] The two independent determinations of k_{-d} agree on $k_{-d} = (4.5 ± 1.0) \times 10^9$ M^{-1} s^{-1}.[3,10] These values and K_d obtained here predict the radical yield of about 35%, which is in a very satisfying agreement with 30−40% yields obtained through the measurements of various substrate oxidations by ONOOH.[1,5,6,37] In contrast, if the value $\Delta_f G°$(ONOOH) = 21 kJ/mol suggested by Koppenol and Kissner[9] were used for evaluating K_d, the predicted radical yield would have been Y_d < 1%, that is, unimportant for ONOOH reactivity.

The absence of MV•⁺ decay even in the presence of a significant amount of the NO• radical in solution (Figure 1) both plays a prominent role in devising Scheme 2 and is important in its own right. At first glance, the absence of MV•⁺ + NO• reaction may appear surprising, because two radicals are involved. However, there appears to be no intuitively reasonable low-energy pathway for formation of stable product. It has been recently argued that the one-electron reduction of NO• produces the nitroxyl anion in its triplet ground state, ³NO⁻, and that this reaction requires a very strong reductant, $E°$(NO•/³NO⁻) ≈ −0.8 V vs NHE.[38] The reduction potential $E°$(MV²⁺/MV•⁺) is only −0.45 V,[39] which is insufficient for rapid NO• reduction by MV•⁺. The only other available reaction mode, addition of MV•⁺ to NO•, does not generate an obvious product either, because the free valence in MV•⁺ is delocalized over the conjugated π-system of the two pyridinium rings.

The data in Figure 2 show that direct oxidation of MV•⁺ by ONOO⁻ is slow, if it occurs at all; its rate constant can be conservatively estimated as less than 1×10^3 M^{-1} s^{-1}. Even at this low rate, the direct reaction would substantially curve downward the dependence in Figure 2; for example, the longest

ON−OO⁻ Bond Homolysis

J. Phys. Chem. A, Vol. 107, No. 39, 2003 **7995**

half-life would decrease to about 1.6 s,[40] which is way outside the experimental uncertainty. The slowness of the direct reaction may appear surprising, as $MV^{\bullet+}$ is a strong reductant and $ONOO^-$ is a strong one-electron oxidant even in alkali (Figure 4), so that the overall two-step reaction

$$MV^{\bullet+} + ONOO^- \rightarrow MV^{2+} + ONOO^{\bullet 2-}$$

$$ONOO^{\bullet 2-} + H_2O \rightarrow NO_2^{\bullet} + 2OH^-$$
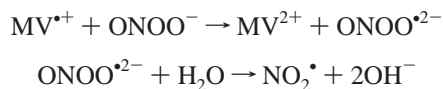
is exoergonic by 126 kJ/mol. The most likely explanation for the slowness of the overall process is that the first step is either thermodynamically uphill or strongly activated. Alternatively, the reduction of $ONOO^-$ could be water-assisted and concerted, i.e., proceed in one step without the intermediacy of $ONOO^{\bullet 2-}$. However, this reaction mode would involve simultaneous formation and breaking of several bonds and is unlikely to be rapid. Whatever the case, the $ONOO^-$ anion does not kinetically perform as a strong one-electron oxidant, even though it formally is one. Instead, the one-electron oxidative chemistry of the $ONOO^-$ anion is attributable to its Lewis basicity and the oxidation reactions occur through coupling of $ONOO^-$ with various Lewis acids, L, followed by peroxo bond breaking and attendant transfer of O^- to the acid with release of $NO_2^{\bullet}$,[41] e.g.,

$$L + ONOO^- \rightarrow LOONO^- \rightarrow LO^{\bullet-} + NO_2^{\bullet}$$

Examples of such a pathway include reactions of $ONOO^-$ with CO_2,[42] carbonyls,[6,43] and metal porphyrins.[44]

This is not to suggest, however, that the one-electron reduction of $ONOO^-$ cannot occur at all; as with HO_2^-, it may simply require an extremely strong reductant. The hydrated electron ($E^{\circ} = -2.87$ V[45]), for example, can be scavenged by $ONOO^-$.[46] Quite possibly, the $(CH_3)_2C^{\bullet}OH$ radical ($E^{\circ} = -2.1$ V in alkali[47]) is also capable of reducing $ONOO^-$ and the vestiges of these two reactions are responsible for the observed slightly decreased radiation yield of $MV^{\bullet+}$ at low $[MV^{2+}]$. However, this yield decrease should be the only effect of these reactions, because their $NO_2^{\bullet}$ product will rapidly oxidize a small fraction of the $MV^{\bullet+}$ radical and should not influence its further slow decay on the time scales of Figures 1 and 2.

Conclusions

1. Oxidation of the $MV^{\bullet+}$ radical by $ONOO^-$ occurs through an indirect pathway, which includes reversible homolytic decomposition of $ONOO^-$ into a pair of $NO^{\bullet}$ and $O_2^{\bullet-}$ radicals (rate-determining step, $k_h = 0.017 \pm 0.001$ s^{-1}) followed by rapid oxidation of $MV^{\bullet+}$ by $O_2^{\bullet-}$ (Scheme 2). Direct oxidation of the $MV^{\bullet+}$ radical through a bimolecular reaction with $ONOO^-$ is slow ($k < 1 \times 10^3$ M^{-1} s^{-1}).

2. The $MV^{\bullet+}$ radical does not rapidly reduce $NO^{\bullet}$, suggesting that the one-electron reduction potential of the latter is much below -0.45 V (NHE). This result is in agreement with recent reevaluations of the energetics of $NO^{\bullet}$ reduction to various forms of nitroxyl (HNO/NO$^-$).[38]

3. The equilibrium constant $K_h = (3.6 \pm 0.3) \times 10^{-12}$ M for the homolytic decomposition of $ONOO^-$ into $NO^{\bullet}$ and $O_2^{\bullet-}$ is determined from the measured rate constants for the forward and reverse (peroxynitrite-forming recombination of $NO^{\bullet}$ and $O_2^{\bullet-}$, $k_{-h} = (4.8 \pm 0.3) \times 10^9$ M^{-1} s^{-1}) reactions. From this K_h and the pK_a of ONOOH, the free energies of formation in aqueous solution $\Delta_f G^{\circ}(ONOO^-) = 68.5 \pm 1.4$ kJ/mol and $\Delta_f G^{\circ}-(ONOOH) = 30.8 \pm 1.5$ kJ/mol are obtained. The last value is fully consistent with the major role of the ONOOH homolytic decomposition into $NO_2^{\bullet}$ and $OH^{\bullet}$ radicals in oxidations by peroxynitrite.

4. The values for all k_h, k_{-h}, K_h, $\Delta_f G^{\circ}(ONOO^-)$, and $\Delta_f G^{\circ}(ONOOH)$ measured in this work are in good agreement with the previous determinations of these parameters,[2,14−16] but not with more recent estimates of these quantities by Koppenol and co-workers.[9,11,17]

Supporting Information Available: Kinetic trace showing the absence of $MV^{\bullet+}$ oxidation by H_2O_2; derivation of the integral rate law for Scheme 2; details concerning the measurement of k_{so}; details of numerical simulations for Figure 3. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Mahoney, L. R. *J. Am. Chem. Soc.* **1970**, *92*, 5262.

(2) Merényi, G.; Lind, J. *Chem. Res. Toxicol.* **1998**, *11*, 243.

(3) Merényi, G.; Lind, J.; Goldstein, S.; Czapski, G. *J. Phys. Chem. A* **1999**, *103*, 5685.

(4) (a) Coddington, J. W.; Hurst, J. K.; Lymar, S. V. *J. Am. Chem. Soc.* **1999**, *121*, 2438. (b) Lymar, S. V.; Khairutdinov, R. F.; Hurst, J. K. *Inorg. Chem.* **2003**, ASAP Article.

(5) Gerasimov, O. V.; Lymar, S. V. *Inorg. Chem.* **1999**, *38*, 4317.

(6) Hodges, G. R.; Ingold, K. U. *J. Am. Chem. Soc.* **1999**, *121*, 10695.

(7) Koppenol, W. H.; Moreno, J. J.; Pryor, W. A.; Ischiropoulos, H.; Beckman, J. S. *Chem. Res. Toxicol.* **1992**, *5*, 834.

(8) (a) Padmaja, S.; Kissner, R.; Bounds, P. L.; Koppenol, W. H. *Helv. Chim. Acta* **1998**, *81*, 1201 (b) Kissner, R.; Koppenol, W. H. *J. Am. Chem. Soc.* **2002**, *124*, 234. (c) Maurer, P.; Thomas, C. F.; Kissner, R.; Ruegger, H.; Greter, O.; Rothlisberger, U.; Koppenol, W. H. *J. Phys. Chem. A* **2003**, *107*, 1763.

(9) Koppenol, W. H.; Kissner, R. *Chem. Res. Toxicol.* **1998**, *11*, 87.

(10) Logager, T.; Sehested, K. *J. Phys. Chem.* **1993**, *97*, 6664.

(11) Nauser, T.; Merkofer, M.; Kissner, R.; Koppenol, W. H. *Chem. Res. Toxicol.* **2001**, *14*, 348.

(12) Goldstein, S.; Czapski, G.; Lind, J.; Merényi, G. *Chem. Res. Toxicol.* **2001**, *14*, 657.

(13) Goldstein, S.; Czapski, G.; Lind, J.; Merényi, G. *Chem. Res. Toxicol.* **1999**, *12*, 132.

(14) Huie, R. E.; Padmaja, S. *Free Rad. Res. Commun.* **1993**, *18*, 195.

(15) Kobayashi, K.; Miki, M.; Tagawa, S. *J. Chem. Soc., Dalton Trans.* **1995**, 2885.

(16) Goldstein, S.; Czapski, G. *Free Radic. Biol. Med.* **1995**, *19*, 505.

(17) Nauser, T.; Koppenol, W. H. *J. Phys. Chem. A* **2002**, *106*, 4084.

(18) (a) Watanabe, T.; Honda, K. *J. Phys. Chem.* **1982**, *86*, 2617. (b) Solar, S.; Solar, W.; Getoff, N.; Holcman, J.; Sehested, K. *J. Chem. Soc., Faraday Trans. 1* **1982**, *78*, 2467.

(19) Young, H. D. *Statistical Treatment of Experimental Data*; McGraw-Hill: New York, 1962.

(20) Ross, A. B.; Mallard, W. G.; Helman, W. P.; Buxton, G. V.; Huie, R. E.; Neta, P. *NDRL/NIST Solution Kinetics Database, Ver. 2.0*; National Institute of Standards and Technology: Gaithersburg, MD, 1994.

(21) In 20 mM NaOH used here, OH$^{\bullet}$ and $(CH_3)_2C^{\bullet}OH$ are about 80% and 65%, respectively, dissociated to their conjugate anions (Hickel, B.; Corfitzen, H.; Sehested, K. *J. Phys. Chem.* **1996**, *100*, 17186. Poskrebyshev, G. A.; Neta, P.; Huie, R. E. *J. Phys. Chem. A* **2002**, *106*, 11488. Laroff, G. P.; Fessenden, R. W. *J. Phys. Chem.* **1973**, *77*, 1283).

(22) The previous investigation of the alkaline methyl viologen−2-propanol system found about 10% $MV^{\bullet+}$ radical decay occurring within 0.5−1.5 ms; the time being inversely proportional to the radiation dose (Mulazzani, Q. G.; D'Angelantonio, M.; Venturi, M.; Hoffman, M. Z.; Rodgers, M. A. J. *J. Phys. Chem.* **1986**, *90*, 5347). The decay was attributed to a reaction between $MV^{\bullet+}$ and the nonreducing β-carbon radical, $^{\bullet}CH_2$-(CH$_3$)CHOH, occurring at some 7×10^8 M^{-1} s^{-1}. The OH$^{\bullet}$ radical is known to generate about 13% of β-carbon radical along with about 86% of strongly reducing α-carbon radical, $(CH_3)_2C^{\bullet}OH$ (Asmus, K.-D.; Möckel, H.; Henglein, A. *J. Phys. Chem.* **1973**, *77*, 1218). We have confirmed the presence of this short-lived dose-dependent decay; accordingly, all the

Lymar and Poskrebyshev

kinetics in the methyl viologen−2-propanol system were analyzed starting from 15 ms past the radiation pulse where the fast decay was complete.

(23) Farrington, J. A.; Ebert, M.; Land, E. J. *J. Chem. Soc., Faraday Trans. 1* **1978**, *74*, 665.

(24) Levey, G.; Ebbesen, T. W. *J. Phys. Chem.* **1983**, *87*, 829.

(25) Levey, G.; Rieger, A. L.; Edwards, J. O. *J. Org. Chem.* **1981**, *46*, 1255.

(26) Winterbourn, C. C. *FEBS Lett.* **1981**, *128*, 339.

(27) Kissner, R.; Nauser, T.; Bugnon, P.; Lye, P. G.; Koppenol, W. H. *Chem. Res. Toxicol.* **1997**, *10*, 1285.

(28) Merényi, G.; Lind, J.; Czapski, G.; Goldstein, S. *Inorg. Chem.* **2003**, *42*, 3796.

(29) Merényi, G.; Lind, J. *Chem. Res. Toxicol.* **1997**, *10*, 1216.

(30) Stanbury, D. M. *Adv. Inorg. Chem.* **1989**, *33*, 69.

(31) Lymar, S. V.; Schwarz, H. A.; Czapski, G. *J. Phys. Chem. A* **2002**, *106*, 7245.

(32) Hug, G. Optical Spectra of Nonmetallic Inorganic Transients in Aqueous Solution. U.S. Department of Commerce/National Bureau of Standards: Gaithersburg, MD, 1981.

(33) Lymar, S. V.; Hurst, J. K. *J. Am. Chem. Soc.* **1995**, *117*, 8867.

(34) Wagman, D. D.; Evans, W. H.; Parker, V. B.; Schumm, R. H.; Halow, I.; Bailey, S. M.; Churney, K. L.; Nuttall, R. L. *J. Phys. Chem. Ref. Data* **1982**, *11, Suppl. 2*.

(35) Park, J.-Y.; Lee, Y.-N. *J. Phys. Chem. A* **1988**, *92*, 6294.

(36) Schwarz, H. A.; Dodson, R. W. *J. Phys. Chem.* **1984**, *88*, 3643.

(37) Goldstein, S.; Czapski, G. *Inorg. Chem.* **1995**, *34*, 4041.

(38) (a) Shafirovich, V.; Lymar, S. V. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 7340. (b) Bartberger, M. D.; Liu, W.; Ford, E.; Miranda, K. M.; Switzer, C.; Fukuto, J. M.; Farmer, P. J.; Wink, D. A.; Houk, K. N. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 10958.

(39) Wardman, P. *J. Phys. Chem. Ref. Data* **1989**, *18*, 1637.

(40) Calculated by numerical simulation of the mechanism comprising reactions in Scheme 2 plus the direct MV$^{•+}$ + ONOO$^-$ reaction.

(41) Lymar, S. V. Peroxynitrite Chemistry. In *McGraw-Hill Yearbook of Science and Technology 2002*; McGraw-Hill: New York, 2001; p 263.

(42) (a) Goldstein, S.; Czapski, G. *Inorg. Chem.* **1997**, *36*, 5113. (b) Lymar, S. V.; Hurst, J. K. *Inorg. Chem.* **1998**, *37*, 294. (c) Pryor, W. A.; Lemercier, N. N.; Zhang, H.; Uppu, R. M.; Squadrito, G. I. *Free Radic. Biol. Med.* **1998**, *24*, 508.

(43) (a) Uppu, R. M.; Winston, G. W.; Pryor, W. A. *Chem. Res. Toxicol.* **1997**, *10*, 1331. (b) Merényi, G.; Lind, J.; Goldstein, S. *J. Am. Chem. Soc.* **2002**, *124*, 40.

(44) (a) Lee, J.; Hunt, J. A.; Groves, J. T. *J. Am. Chem. Soc.* **1998**, *120*, 7493. (b) Lee, J.; Hunt, J. A.; Groves, J. T. *J. Am. Chem. Soc.* **1998**, *120*, 6053. (c) Jensen, M. P.; Riley, D. P. *Inorg. Chem.* **2002**, *41*, 4788.

(45) Schwarz, H. A. *J. Phys. Chem.* **1991**, *95*, 6697.

(46) Merényi, G. Personal communication to S. V. Lymar.

(47) Schwarz, H. A.; Dodson, R. W. *J. Phys. Chem.* **1989**, *93*, 409.