Reaction of a Superoxochromium(III) Ion with Nitrogen Monoxide: Kinetics and Mechanism

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Abstract: The kinetics of the rapid reaction between $Cr_{aq}OO^{2+}$ and NO were determined by laser flash photolysis of $Cr_{aq}NO^{2+}$ in O_2-saturated acidic aqueous solutions, $k = 7 \times 10^8$ M^{-1} s^{-1} at 25° C. The reaction produces an intermediate, believed to be NO_2, which was scavenged with $([14]aneN_4)Ni^{2+}$. With limiting NO, the $Cr_{aq}OO^{2+}/NO$ reaction has a 1:1 stoichiometry and produces both free NO_3^- and a chromium nitrato complex, $Cr_{aq}ONO_2^{2+}$. In the presence of excess NO, the stoichiometry changes to $[NO]/[Cr_{aq}OO^{2+}] = 3:1$, and the reaction produces close to 3 mol of nitrite/mol of $Cr_{aq}OO^{2+}$. An intermediate, identified as a nitritochromium(III) ion, $Cr_{aq}ONO^{2+}$, is a precursor to a portion of free NO_2^-. In the proposed mechanism, the initially produced peroxynitrito complex, $Cr_{aq}OONO^{2+}$, undergoes O–O bond homolysis followed by some known and some novel chemistry of $Cr_{aq}O^{2+}$ and NO_2. The reaction between $Cr_{aq}O^{2+}$ and NO generates $Cr_{aq}ONO^{2+}$, $k > 10^4$ M^{-1} s^{-1}. $Cr_{aq}OO^{2+}$ reacts with NO_2 with $k = 2.3 \times 10^8$ M^{-1} s^{-1}.

Introduction

The reaction between nitrogen monoxide and oxyhemoglobin[1–3] is believed to be a major route for NO depletion in vivo.[1,3,4] This and analogous reactions of other superoxometal complexes are fast ($k > 10^6$ M^{-1} s^{-1}) and ultimately produce nitrate and nitrite ions via an initial peroxynitrito complex,[5–8] as shown in Scheme 1.

Metal peroxynitrito species also have been proposed as intermediates in metal-catalyzed isomerizations of peroxynitrite ions, $OONO^-$, which are believed to be involved in a number of damaging reactions in living organisms[3,9,10] and associated with diseases such as cancer, heart disease, inflammatory processes, and others.[11–13] Several metal complexes, most of them porphyrin-based, have been shown to catalyze the conversion of the potentially damaging peroxynitrite ions to the innocuous nitrate ions.[6,11,13–15] Often, these catalysts only

Scheme 1
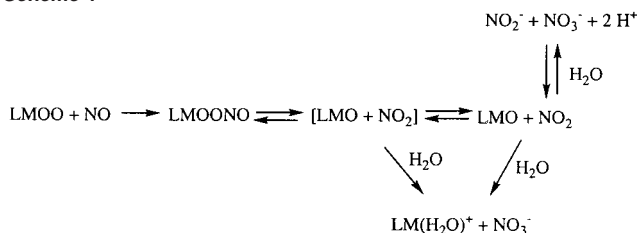
accelerate the natural reaction and still produce NO_2 (see Scheme 1), a strong oxidant and nitrating agent for biological tissues. This mechanism is closely related to that proposed for isomerization of the parent HOONO.[16–18] Some other, pharmacologically more promising candidates seem to operate by a different mechanism and involve much smaller amounts of NO_2, so that the catalysts provide some protection against nitration of biological tissues by $OONO^-$.[6,13,15,19,20] Current research is geared toward the identification of key mechanistic steps in these complex reactions to establish a basis for the development of pharmacologically useful compounds.

Recently, we observed a nitratorhodium complex as an intermediate in a reaction between a macrocyclic superoxorhodium(III) ion and NO.[7] All the evidence pointed to a mechanism whereby the rhodium nitrato species was formed by in-cage and/or out-of-cage recombination of NO_2 and L^2RhO^{2+}, which were in turn generated by homolytic O–O bond cleavage in

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the initially formed $L^2RhOONO^{2+}$ ($L^2 = Me_6$-[14]aneN_4). This, and the simultaneous observation of a nitrato complex of pentacyanocobalt(III) by another group,[21] are the first cases documenting nitratometal intermediates in the NO/superoxometal chemistry, although a possibility that such species might be involved had been considered earlier.[6,11,22]

We have now extended our studies to another superoxometal complex, $Cr_{aq}OO^{2+}$. There are several issues that we believe could be addressed uniquely by this particular complex: (1) The nitrato and nitrito complexes of chromium(III), $Cr_{aq}ONO_2^{2+}$ [23] and $Cr_{aq}ONO^{2+}$,[24] are sufficiently long-lived to be detected under the reaction conditions, an essential requirement for the detailed assignment of some mechanistic steps. (2) The nitrosyl complex, $Cr_{aq}NO^{2+}$, is stable and unreactive toward molecular oxygen, which provided a basis for the development of a new method for the determination of the kinetics of the $Cr_{aq}OO^{2+}$/NO reaction. (3) Another potential intermediate, $Cr_{aq}O^{2+}$,[25] is also known, allowing us to check independently some of the proposed steps and thus the validity of the overall scheme.

Experimental Section

Acidic aqueous solutions of $Cr_{aq}OO^{2+}$ (containing 0.1 M methanol as a stabilizer)[25] and $Cr_{aq}O^{2+}$ were prepared from Cr_{aq}^{2+} and O_2 as previously described.[26] Most of the work with both of these species was carried out in an atmosphere of O_2 or air, but for some experiments, solutions of $Cr_{aq}OO^{2+}$ were degassed by a stream of argon for 10 min. Such solutions still contained some ($<10^{-5}$ M) O_2, which is necessary to protect $Cr_{aq}OO^{2+}$ from rapid decomposition.

Solutions of $Cr_{aq}NO^{2+}$ were prepared[27] from Cr_{aq}^{2+} and NO and purified by ion exchange on a column of Sephadex C-25. From the UV spectral data and the reported[27] molar absorptivities at 449 nm ($\epsilon = 121$ M^{-1} cm^{-1}) and 238 nm ($\epsilon = 1170$ M^{-1} cm^{-1}), we obtained the molar absorptivity at the irradiation wavelength, $\epsilon_{266} = 700$ M^{-1} cm^{-1}. Gaseous NO (Matheson) was purified by passage through Ascarite, sodium hydroxide and water.[7] Stock solutions of NO were prepared by bubbling the purified gas through 20 mL of argon-saturated water for 25 min. Such solutions typically contained 1.7 mM NO and 0.2 mM nitrite ions.[7]

Solid [L^1Ni](ClO$_4$)$_2$ ($L^1 = $ [14]aneN_4) was prepared by a literature procedure.[28] Solid [(NH$_3$)$_5$CoNO$_2$](OTf)$_2$ (OTf = trifluoromethanesulfonate) was prepared from the chloride salt[29] and LiOTf (Aldrich). The possible contamination by the isomeric nitrito form was judged negligible on the basis of the UV−visible spectrum, which is quite sensitive to even small amounts of (NH$_3$)$_5$CoONO^{2+}.[29] Sodium nitrite (Aldrich) was used as received.

Laser flash photolysis (LFP) experiments utilized the third (355 nm) and fourth (266 nm) harmonics of the Nd:YAG laser, which is part of an Applied Photophysics laser flash photolysis system described earlier.[30] The diameter of the laser beam, and therefore the effective path length in all the measurements, was 0.5 cm. Other kinetic and spectral measurements utilized a Shimadzu 3101 PC spectrophotometer.

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The kinetics of the reaction between $Cr_{aq}OO^{2+}$ and NO were determined by LFP. In a typical experiment, a 0.15−0.90 mM solution of $Cr_{aq}NO^{2+}$ in O_2-saturated ([O$_2$] = 1.26 mM) aqueous 0.1 M HClO$_4$ was flashed (λ_{irr} 266 nm), and the absorbance changes were monitored at 293 nm, where $Cr_{aq}OO^{2+}$ exhibits an absorption maximum with $\epsilon = 3000$ M^{-1} cm^{-1} [31,32] and $Cr_{aq}NO^{2+}$ absorbs negligibly ($\epsilon \sim 100$ M^{-1} cm^{-1}). The signal-to-noise ratio was low, which necessitated the collection and averaging of 10−12 traces for each run.

The kinetics of the $Cr_{aq}OO^{2+}$/NO$_2$ reaction were determined by flash-photolyzing (λ_{irr} 355 nm) an acidic solution of (NH$_3$)$_5$CoNO$_2^{2+}$ (100 μM) containing $Cr_{aq}OO^{2+}$ (46−170 μM) and monitoring the absorbance decrease at 293 nm.

LFP of $Cr_{aq}NO^{2+}$ was also used to generate Cr_{aq}^{2+} for the reaction with NO. The reaction was carried out under pseudo-first-order conditions with a large excess of NO over Cr_{aq}^{2+} and monitored at 260 nm, where $\epsilon(Cr_{aq}NO^{2+}) = 735$ M^{-1} cm^{-1}.

The analyses of nitrite[33] and nitrate[34] were carried out in two types of experiments. In one, a solution of 1 mM $Cr_{aq}NO^{2+}$ in O_2-saturated 0.1 M HClO$_4$ was flashed 100 times. After each set of 5 flashes, the cell was removed from the cell compartment and the solution was shaken. The total concentration of $Cr_{aq}OO^{2+}$ produced was calculated from the absorbance change in each shot and the irradiated volume. The concentration of NO$_3^-$ produced was determined[34] after the metal ions had been removed by ion-exchange chromatography on ice-cold Sephadex C-25 resin, as described earlier.[7]

Another set of experiments utilized manual mixing of known concentrations of NO and $Cr_{aq}OO^{2+}$, followed by analysis of NO$_2^-$ and NO$_3^-$. As in our previous work,[7] the analysis of NO$_3^-$ was carried out at different times after the completion of the reaction to determine whether some of the NO$_3^-$ was complexed to Cr_{aq}^{3+}. Immediately after the mixing of $Cr_{aq}OO^{2+}$ and NO, 5 mL of the reaction solution was passed through a short column (2 cm) of ice-cold Sephadex C-25 ion-exchange resin to remove all the cationic chromium species. The resin was then rinsed with several milliliters of 0.01 M HClO$_4$ and the combined eluent was analyzed for free NO$_3^-$.

The remaining solution was analyzed without prior ion-exchange to determine total nitrate. The harsh conditions in the analytical procedure used[34] are believed to cause a complete dissociation of NO$_3^-$ from $Cr_{aq}ONO_2^{2+}$. This was confirmed in a third type of experiments, whereby the spent reaction solution was made alkaline by addition of dilute NaOH and allowed to stand for 2 h, until the aquation of $Cr_{aq}ONO_2^{2+}$ was complete. The solution was then reacidified, ion-exchanged, and analyzed for NO$_3^-$. The results were the same as those obtained for a fresh solution that had not been ion-exchanged.

The in-house distilled and deionized water was purified by passage through a Millipore water purification system. All the kinetic data are reported at room temperature (25 ± 1 °C).

Results

Kinetics of $Cr_{aq}OO^{2+}$/NO Reaction. Both reagents were generated in situ according to eqs 1 and 2, followed by the desired chemistry of eq 3. Arguments for the formation of $Cr_{aq}OONO^{2+}$ in eq 3 will be presented later. According to eqs 1 and 2, $Cr_{aq}OO^{2+}$ and NO are produced in a 1:1 ratio. The potential interfering reactions, autoxidation of NO[35] and the reverse of eq 1, were both unimportant at the low concentrations of NO used.

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$$Cr_{aq}NO^{2+} \underset{}{\overset{h\nu}{\rightleftharpoons}} Cr_{aq}^{2+} + NO \qquad (1)$$

$$Cr_{aq}^{2+} + O_2 \underset{k_{off}}{\overset{k_{on}}{\rightleftharpoons}} Cr_{aq}OO^{2+} \qquad (2)$$

$$Cr_{aq}OO^{2+} + NO \rightarrow Cr_{aq}OONO^{2+} \qquad (3)$$

Kinetic traces at 293 nm (Figure 1) exhibited an absorbance increase associated with reaction 2 followed by a much slower decrease, corresponding to reaction 3. The rising portion obeyed first-order kinetics with $k = 2.8 \times 10^5$ s^{-1}, in good agreement with the expectations based on the O$_2$ concentrations used and the known rate constant for reaction 2, $k_{on} = 1.6 \times 10^8$ M^{-1} s^{-1}.[31,32] The same kinetic behavior was observed throughout the wavelength range 270–450 nm, but the absorbance change was the greatest at 293 nm, as expected for the formation of $Cr_{aq}OO^2$ from $Cr_{aq}NO^{2+}$.

In the second step, reaction 3, the absorbance decreased to the same value it had prior to reaction 2, indicating that all $Cr_{aq}OO^{2+}$ had been consumed and no absorbing product was formed. The kinetic analysis utilized an equal-concentration second-order treatment, eq 4, where Abs$_t$, Abs$_{inf}$, and Abs$_0$ represent absorbances at time t, at the end of the reaction, and at zero time, respectively. The concentration of $Cr_{aq}OO^{2+}$ at zero time was calculated from the absorbance increase in the first, fast stage of the reaction (Figure 1).

$$Abs_t = Abs_{inf} + \frac{Abs_0 - Abs_{inf}}{1 + k_3[Cr_{aq}OO^{2+}]_0 t} \qquad (4)$$

Within the error, the same rate constant, $k_3 = (7.0 \pm 0.7) \times 10^8$ M^{-1} s^{-1}, was obtained under all the concentration conditions employed, which includes the variation in $[Cr_{aq}OO^{2+}]_0$ and $[NO]_0$ in the 4–23 μM range.

Several experiments were carried out in the presence of 0.1–1.0 mM L^{1}Ni^{2+}, a good scavenger for NO$_2$.[36] The results are shown in Figure 2. After the initial, rapid step (not shown) of eq 2, a further increase in absorbance (Figure 2B) takes place on a time scale comparable to that for the absorbance decrease in the absence of L^{1}Ni^{2+} (Figure 2A). Additional L^{1}Ni^{3+} is formed over a period of several seconds in the reaction of L^{1}Ni^{2+} with $Cr_{aq}O^{2+}$, $k = 10^3$ M^{-1} s^{-1}; see below.[37,38] The generation of strongly absorbing L^{1}Ni^{3+} during and after each shot reduces the amount of laser light available to $Cr_{aq}NO^{2+}$. This decreases both the amount of $Cr_{aq}OO^{2+}$ in each subsequent shot and the signal-to-noise ratio. Both of these factors made it impossible to collect precise kinetic data or determine the precise yield of L^{1}Ni^{3+} relative to $[Cr_{aq}OO^{2+}]$. Qualitatively, however, the rate of formation of L^{1}Ni^{3+} and the amount formed showed little or no dependence on $[L^1Ni^{2+}]$ in the range examined. Thus the formation of an oxidizing intermediate(s) in reaction 3 determines the rate of oxidation of L^{1}Ni^{2+}.

A cursory study of the $Cr_{aq}O^{2+}$/L^{1}Ni^{2+} reaction yielded a rate constant of 1×10^3 M^{-1} s^{-1} at 0.10 M H$^+$.

$Cr_{aq}OO^{2+}$/NO$_2$ Reaction. LFP of (NH$_3$)$_5$CoNO$_2^{2+}$ at 355 nm generates NO$_2$ (eq 5).[29] The cobalt(II) fragment rapidly aquates, which makes the photochemical reaction irreversible.

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(37) Another, slower reaction (time scale of minutes) that generated more L^{1}Ni^{3+} was traced to the generation of additional amounts of NO and NO$_2$ from HNO$_2$, produced initially in the L^{1}Ni^{2+}/NO$_2$ reaction.
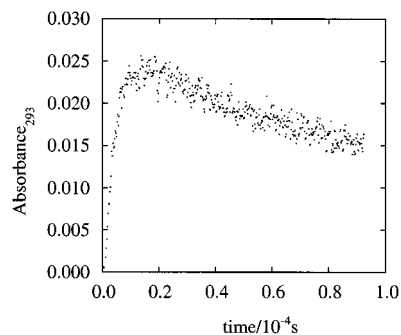(38) Stedman, G. *Adv. Inorg. Chem. Radiochem.* **1979**, *22*, 113–170.

Figure 1. Kinetic trace at 293 nm obtained upon flashing an O$_2$-saturated solution of 0.7 mM $Cr_{aq}NO^{2+}$ in 0.10 M HClO$_4$. The absorbance increase corresponds to reaction 2, and the decrease to reaction 3. Path length: 0.5 cm.

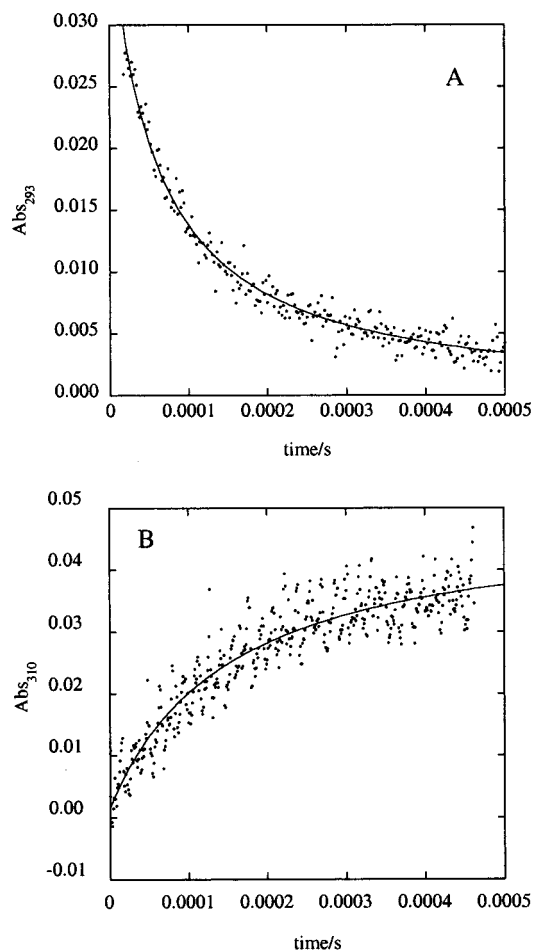**Figure 2.** Kinetic traces for the slow stage in $Cr_{aq}OO^{2+}$/NO reaction in the absence (A) and presence (B) of 1 mM L^{1}Ni^{2+} in 0.1 M HClO$_4$. In panel A, the loss of $Cr_{aq}OO^{2+}$ was monitored at 293 nm, and in panel B, the build-up of L^{1}Ni^{3+} was monitored at 310 nm. $Cr_{aq}OO^{2+}$ was produced by LFP of $Cr_{aq}NO^{2+}$ (A, 0.9 mM; B, 0.6 mM) in the presence of O$_2$ (eq 1–2). Path length: 0.5 cm. L^1 = [14]aneN$_4$.

The concurrent isomerization to (NH$_3$)$_5$CoONO^{2+} [29] caused no observable absorbance changes at 290 nm.

$$(NH_3)_5CoNO_2^{2+} \xrightarrow{h\nu, H^+} NO_2 + Co_{aq}^{2+} + 5\,NH_4^+ \qquad (5)$$
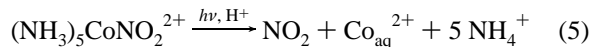
In the presence of $Cr_{aq}OO^{2+}$, a small bleach during the laser pulse was followed by an absorbance decrease at 290 nm, which we interpret as reaction 6, although the products have not been
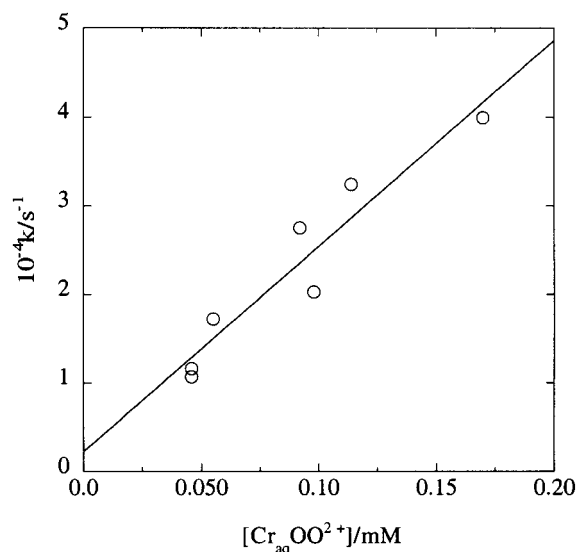
Figure 3. Plot of k_{obs} vs $[Cr_{aq}OO^{2+}]$ for the reaction between $Cr_{aq}OO^{2+}$ and NO_2, eq 6.

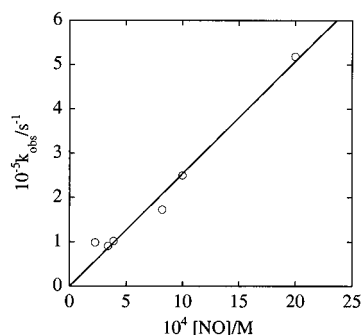**Figure 4.** Plot of k_{obs} vs $[NO]$ for the reaction between Cr_{aq}^{2+} and NO, reverse of reaction 1. Initial $[Cr_{aq}NO^{2+}] = 0.8$ mM.

determined. A plot of k_{obs} against the concentration of $Cr_{aq}OO^{2+}$ in Figure 3 gives $k_6 = 2.3\ (\pm\ 0.3) \times 10^8\ M^{-1}\ s^{-1}$.

$$Cr_{aq}OO^{2+} + NO_2 \rightarrow Cr_{aq}OONO_2^{2+} \qquad (6)$$

Cr_{aq}^{2+}/NO Reaction. The experiments utilized anaerobic conditions and excess NO (see Experimental Section) and yielded a rate constant for the reverse of reaction 1 of $k_{-1} = (2.5 \pm 0.1) \times 10^8\ M^{-1}\ s^{-1}$ at 0.10 M HClO$_4$ (Figure 4). This value is close to those for the capture of other radicals,[39] including O$_2$,[31,32] by Cr_{aq}^{2+} but much larger than our previously reported value for k_{-1} of $2.2 \times 10^6\ M^{-1}\ s^{-1}$.[40] The rate constant obtained in this work is a result of a direct measurement and as such more reliable than the earlier value, which was obtained in a competition study. It now appears that the outcome of the rapid competing reactions in the previous work[40] may have been distorted by mixing effects.

Stoichiometry, Products, and Intermediates. Three milliliters of an O$_2$-saturated solution of 1 mM $Cr_{aq}NO^{2+}$ that had received 100 laser shots generated a total of 0.31 μmol of $Cr_{aq}OO^{2+}$. The yield of nitrate produced was 0.30 μmol, which gives a stoichiometric factor $[NO_3^-]_\infty/[Cr_{aq}OO^{2+}]_0 = 1$. We

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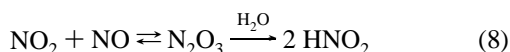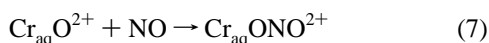(40) Melton, J. D.; Bakac, A.; Espenson, J. H. *Inorg. Chem.* **1986**, *25*, 3360−3367.

Table 1. Products and Observed Intermediates in $Cr_{aq}OO^{2+}$/NO Reaction[a]

$[Cr_{aq}OO^{2+}]_0$	$[NO]_0$	% $(Cr_{aq}ONO_2)^{2+ b}$	% $(Cr_{aq}ONO^{2+})^c$	% $(NO_3^-)_t{}^d$	% $(NO_2^-)_t$
51[e]	53	26	0	75	25[f]
55[e]	59	24	0	67	33[f]
10[g,h]	10	i	0	97[g]	0
48[h]	180	i	80	16	260

[a] In 0.1 M HClO$_4$. All the concentrations are given as micromolar, and the percentages are based on $[Cr_{aq}OO^{2+}]_0$. [b] The yield was calculated as a difference between the total and free nitrate. [c] Estimated from ΔAbs$_{240}$ in the kinetic traces corresponding to reaction 9. [d] Includes free NO$_3^-$ and $Cr_{aq}ONO_2^{2+}$. [e] Air-free. [f] Calculated under the assumption that no other products were formed. [g] The value is for free NO$_3^-$ in LFP experiments. Standard error is $\pm 30\%$. The products formed in 100 laser shots were accumulated, ion-exchanged, and analyzed for NO$_3^-$. [h] O$_2$-saturated solutions. [i] Not determined.

associate an estimated error of $\pm 30\%$ with this value because the $[Cr_{aq}OO^{2+}]_0$ was calculated as a sum of 100 individual contributions, each subject to a standard error of several percent; see Experimental Section. The analysis was carried out immediately after the photolysis, which identifies free NO$_3^-$ as a major product. The amount of bound nitrate, $Cr_{aq}ONO_2^{2+}$, could not be determined in this type of experiments because the solutions contained large amounts of unreacted $Cr_{aq}NO^{2+}$, which decomposed and generated nitrate in the time required for hydrolysis of $Cr_{aq}ONO_2^{2+}$.

The direct mixing of NO and $Cr_{aq}OO^{2+}$ resulted in a stoichiometry and yields of NO$_3^-$ that varied with the concentrations of $Cr_{aq}OO^{2+}$ and NO stock solutions (Table 1). A spectrophotometric titration at $\lambda = 293$ nm gave a stoichiometric ratio $R = [NO]/[Cr_{aq}OO^{2+}]$ in the range 2.4−3.1 when solutions of $Cr_{aq}OO^{2+}$ (42−86 μM) were titrated with 1.7 mM NO. This result was obtained with both argon and O$_2$-saturated solutions of $Cr_{aq}OO^{2+}$. The major product was nitrite, formed in an amount that was close to 3 times that of $Cr_{aq}OO^{2+}$ consumed. The yield of nitrate was only 16% under these conditions.

The ratio $R = 1.6−1.8$ was obtained in the initial stages ($\leq 30\%$ $Cr_{aq}OO^{2+}$ consumed) of titrations of argon-saturated solutions of $Cr_{aq}OO^{2+}$ with 0.4 mM NO. When the concentration of NO stock was decreased even more, to 0.16 mM, R decreased to 1.0−1.1. These ratios were somewhat larger in the later stages of titration as $Cr_{aq}OO^{2+}$ was being depleted. The decrease in R was accompanied by an increase in the yield of NO$_3^-$, until it became the major product ($\geq 75\%$) at R ~ 1 (Table 1). Clearly, local concentrations of NO have a pronounced effect on the outcome of the reaction.

Free NO$_3^-$ was a major form of the nitrate produced. Immediately after the reaction, the yields were 49% and 43% in two separate experiments. The total yields, determined as described in the Experimental Section, were 75% and 67% in the same two experiments, Table 1. On the basis of our previous work with L^{2}RhOO^{2+}, the difference between the total and initial $[NO_3^-]$ is assigned as the amount of complexed nitrate, $Cr_{aq}ONO_2^{2+}$. The less than 100% yield of total nitrate is most likely caused by the stoichiometry, expressed as $[NO]/[Cr_{aq}OO^{2+}]$, being greater than 1:1 when a full equivalent of NO is mixed with $Cr_{aq}OO^{2+}$. Any deviation from 1:1 stoichiometry will cause a decrease in $[NO_3^-]$ and increase in $[NO_2^-]$; see above. Also, a portion of NO$_2$, a probable reaction intermediate, may disproportionate to NO$_3^-$ and NO$_2^-$ in competition with $Cr_{aq}O^{2+}$/NO$_2$ reaction, which would also decrease the yield of NO$_3^-$.
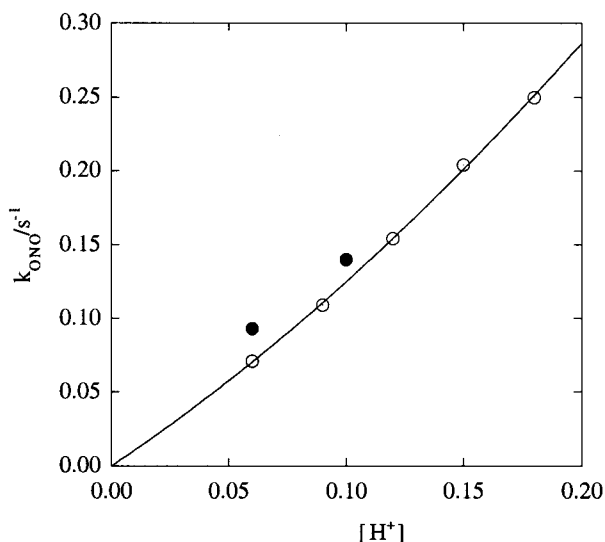
Figure 5. Plot of k_{ONO} vs [H$^+$] for the disappearance of Cr$_{aq}$ONO^{2+}, eq 9, which was generated as an intermediate in the Cr$_{aq}$OO^{2+}/NO reaction (○) or from the Cr$_{aq}$O^{2+}/NO reaction (●). Conditions: 17.4° C and 1.0 M ionic strength (HClO$_4$ + NaClO$_4$). Kinetics were monitored at 240 nm.

The increased consumption of NO under conditions of excess (stoichiometric or local) of NO suggests a mechanism whereby an intermediate(s) can be transformed to final products either by itself or in reaction with NO. A likely possibility involves the formation of a peroxynitrito complex, Cr$_{aq}$OONO^{2+}, eq 3, followed by O–O bond homolysis to yield Cr$_{aq}$O^{2+} and NO$_2$. The fraction of these two species that diffuses out of the solvent cage is expected to react with NO as in eqs 7 and 8. Reaction 8 is well documented,[38,43] and that in eq 7 was written in analogy with the recently reported reactions of NO with a macrocyclic chromium(IV) complex, *trans*-Cr(cyclam)(O)(ONO)$^+$ [41] and with MbFe^{IV}O[42] (Mb = myoglobin).

$$Cr_{aq}O^{2+} + NO \rightarrow Cr_{aq}ONO^{2+} \qquad (7)$$

$$NO_2 + NO \rightleftharpoons N_2O_3 \xrightarrow{H_2O} 2\,HNO_2 \qquad (8)$$

If the product of reaction 7 is indeed Cr$_{aq}$ONO^{2+}, then it should decay in a matter of seconds in an [H$^+$]-dependent process, eqs 9 and 10, accompanied by an absorbance decrease at 240 nm.[24] This is precisely what happened when a solution of Cr$_{aq}$O^{2+} (45 μM) was mixed with NO (74 μM) in dilute HClO$_4$. Figure 5 shows a plot of the observed first-order rate constants, measured at 240 nm and 17.4 °C, against [H$^+$]. The values of k_{ONO} at the two acid concentrations are close to those calculated from the literature data[24] for hydrolysis of Cr$_{aq}$ONO^{2+}. Obviously, the formation of Cr$_{aq}$ONO^{2+} in eq 7 is much faster than the observed decay, which places a limit on k_7 of $>10^4$ M^{-1} s^{-1}.

$$Cr_{aq}ONO^{2+} + H^+ \rightarrow Cr_{aq}^{3+} + HNO_2 \qquad (9)$$
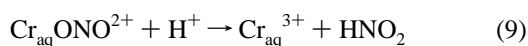
$$-d[Cr_{aq}ONO^{2+}]/dt = k_{ONO}[Cr_{aq}ONO^{2+}] = \\ (k_a[H^+] + k_b[H^+]^2)[Cr_{aq}ONO^{2+}] \quad (10)$$

(41) DeLeo, M. A.; Ford, P. C. *J. Am. Chem. Soc.* **1999**, *121*, 1980–1981.
(42) Herold, S. *Nitric Oxide: Biol. Chem.* **1999**, *3*, 36.
(43) Graetzel, M.; Henglein, A.; Taniguchi, S. *Ber. Bunsen-Ges. Phys. Chem.* **1970**, *74*, 292–298.

Scheme 2

Cr$_{aq}$OO^{2+} + NO
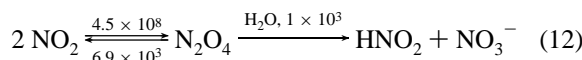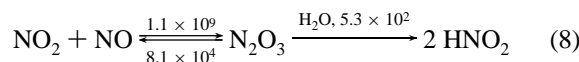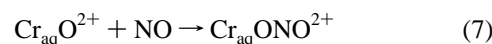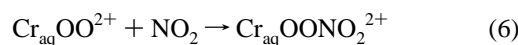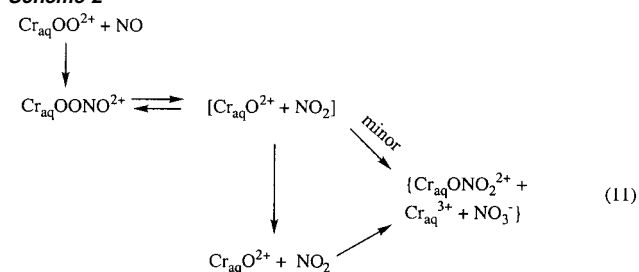
Cr$_{aq}$OONO^{2+} $\rightleftharpoons$ [Cr$_{aq}$O^{2+} + NO$_2$] $\xrightarrow{\text{minor}}$ {Cr$_{aq}$ONO$_2$$^{2+}$ + Cr$_{aq}$$^{3+}$ + NO$_3$$^-$} (11)

Cr$_{aq}$O^{2+} + NO$_2$

$$Cr_{aq}OO^{2+} + NO_2 \rightarrow Cr_{aq}OONO_2^{2+} \qquad (6)$$

$$Cr_{aq}O^{2+} + NO \rightarrow Cr_{aq}ONO^{2+} \qquad (7)$$

$$NO_2 + NO \underset{8.1\times10^4}{\overset{1.1\times10^9}{\rightleftharpoons}} N_2O_3 \xrightarrow{H_2O,\ 5.3\times10^2} 2\,HNO_2 \qquad (8)$$

$$2\,NO_2 \underset{6.9\times10^3}{\overset{4.5\times10^8}{\rightleftharpoons}} N_2O_4 \xrightarrow{H_2O,\ 1\times10^3} HNO_2 + NO_3^- \qquad (12)$$

Once the chemistry in eq 7 was confirmed, we searched for Cr$_{aq}$ONO^{2+} in the Cr$_{aq}$OO^{2+}/NO reaction. Upon addition of a 3-fold excess of NO to a solution of Cr$_{aq}$OO^{2+} (50 μM), the absorbance at 240 nm decreased and the kinetics exhibited the expected [H$^+$] dependence, as shown in Figure 5 along with the data for the decay of Cr$_{aq}$ONO^{2+} produced in the Cr$_{aq}$O^{2+}/NO reaction. Clearly, the same species was produced in the two kinds of experiments. The form of the rate law, eq 10, is the same as that reported[24] for genuine Cr$_{aq}$ONO^{2+}, and the values of k_a (1.06 ± 0.03 M^{-1} s^{-1}) and k_b (1.9 ± 0.2 M^{-2} s^{-1}) are close to the respective published values, 0.75 and 2.4, identifying Cr$_{aq}$ONO^{2+} as an intermediate in the reaction of Cr$_{aq}$OO^{2+} with excess NO. From the observed absorbance changes in the two types of experiments in Figure 5, we calculate the yield of Cr$_{aq}$O^{2+} in the Cr$_{aq}$OO^{2+}/NO reaction at >80%.

Discussion

The fast initial step in the Cr$_{aq}$OO^{2+}/NO reaction, $k_3 = 7 \times 10^8$ M^{-1} s^{-1}, is suggestive of radical coupling to form Cr$_{aq}$OONO^{2+}. All the other results presented strongly support reaction 3 as the initial step, as discussed below. Other possibilities, such as substitution at the chromium center, perhaps at the labile position trans to the coordinated superoxide,[25] or electron transfer seem highly unlikely in view of the large rate constant and the lack of driving force for either oxidation or reduction of Cr$_{aq}$OO^{2+} by NO.[25,44,45] This conclusion is reinforced by the observation that NO$_2$ (E_{ox} = 1.6 V, E_{red} = 1.04 V)[44,46] reacts with Cr$_{aq}$OO^{2+} with a rate constant of 2.3×10^8 M^{-1} s^{-1}, similar to that for NO despite the vastly different reduction potentials,[44,47–50] suggesting that both radicals attack at the terminal oxygen of Cr$_{aq}$OO^{2+}.

(44) Stanbury, D. M. *Adv. Inorg. Chem.* **1989**, *33*, 69–138.
(45) Kang, C.; Anson, F. C. *Inorg. Chem.* **1994**, *33*, 2624–2630.
(46) Koppenol, W. H.; Moreno, J. J.; Pryor, W. A.; Ischiropoulos, H.; Beckman, J. S. *Chem. Res. Toxicol.* **1992**, *5*, 834–842.
(47) Bartberger, M. D.; Fukuto, J. M.; Houk, K. N. *Proc. Natl. Acad. Sci. U.S.A.* **2001**, *98*, 2194–2198.
(48) Benderskii, V. A.; Krivenko, A. G.; Ponomarev, E. A. *Soviet Electrochem.* **1989**, *25*, 154–161.
(49) Lymar, S. V. Personal communication.
(50) For NO, E_{ox} = 1.21 V. The value for E_{red} was reported as 0.39 V, but it could be much lower if the ground state is a triplet, as recently suggested.[48] New experimental data[49] suggest that the potential is indeed much lower than the reported[44] 0.39 V. Another report gives E = −0.81 V.[48]

Table 2. Summary of the Rate Constants Determined in This Work[a]

reaction	$k/M^{-1}\,s^{-1}$
$Cr_{aq}OO^{2+} + NO$	$7.0\,(\pm0.7) \times 10^8$
$Cr_{aq}^{2+} + NO$	$2.5\,(\pm0.1) \times 10^8$
$Cr_{aq}OO^{2+} + NO_2$	$2.3\,(\pm0.3) \times 10^8$
$Cr_{aq}O^{2+} + NO$	$>10^4$
$Cr_{aq}O^{2+} + L1Ni^{2+}$	$\sim1 \times 10^3$

[a] At 25 °C, $[HClO_4] = 0.10$ M.

The change in stoichiometry from 3:1 (excess NO) to 1:1 (limiting NO), and the detection of close to stoichiometric amounts of $Cr_{aq}ONO^{2+}$ provide important mechanistic clues and, in combination with the kinetic and product data, suggest the mechanism in Scheme 2.

As the trapping of reaction intermediates by an external reagent can take place only outside of the solvent cage, the major portion of the two homolysis fragments, $Cr_{aq}O^{2+}$ and NO_2, must diffuse into the solution to be available for the reaction with NO and other potential scavengers, such as L^1Ni^{2+}. Most of the products under all the conditions are thus generated in bulk solution, with the in-cage product formation being a minor path at best.

Both $Cr_{aq}O^{2+}$ and NO_2[43] react rapidly with NO, which explains the 3:1 stoichiometry when excess NO is available. The detection of $Cr_{aq}ONO^{2+}$ by its decomposition kinetics provides strong support for this picture, and the limit, $k_7 > 10^4\,M^{-1}\,s^{-1}$, is consistent with the known, fast reactions of NO with $(cyclam)Cr(ONO)(O)^+$ $(3.1 \times 10^6\,M^{-1}\,s^{-1})$[41] and $MbFe^{IV}O$ $(9 \times 10^6\,M^{-1}\,s^{-1})$.[42]
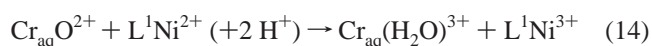
In the absence of extra NO, the $Cr_{aq}O^{2+}$ and NO_2 react with each other and yield either $Cr_{aq}ONO_2^{2+}$ or $\{Cr_{aq}^{3+} + NO_3^-\}$, and the stoichiometry is reduced to 1:1. Some NO_2 may also hydrolyze, as in eq 12. This chemistry is comparable to that observed in the L^2RhOO^{2+}/NO reaction, although the in-cage recombination played a much more prominent role in the rhodium case.[7] The rate constant for the $Cr_{aq}O^{2+}/NO_2$ reaction is not known, but it has to be large enough to compete with the rapid self-reaction of NO_2 in eq 12, $k_{NO2} = 4.5 \times 10^8\,M^{-1}\,s^{-1}$.[51] Given the reversibility of NO_2 dimerization,[51] the $Cr_{aq}O^{2+}/NO_2$ reaction will be competitive even if it has a rate constant that is 10–20 times smaller than k_{NO2}. The analogous reaction of NO_2 with $(TMPS)Fe^{IV}O$ [TMPS = *meso*-tetra(2,4,6-tri-methyl-3,5-disulfonato)porphine] has $k = 1.7 \times 10^7\,M^{-1}\,s^{-1}$,[22] and that with $MbFe^{IV}O$, $1 \times 10^7\,M^{-1}\,s^{-1}$.[6] $(TMPyP)Fe^{IV}O$ [TMPyP = 5,10,15,20-tetrakis(*N*-methyl-4′-pyridyl)porphyrin], on the other hand, reacts with NO_2 only slowly.[19]

When NO is present in excess, most of the NO_2 is removed in reaction 8, which makes reaction 6 unimportant. Even in laser flash photolysis experiments, which generate equimolar amounts of $Cr_{aq}OO^{2+}$ and NO, we estimate that no more than 25% of $Cr_{aq}OO^{2+}$ will react with NO_2 based on the rate constants k_3 and k_6; see Table 2. The exact amount will be smaller, but cannot be calculated precisely in the absence of kinetic data for one of the crucial competing reactions, that between $Cr_{aq}O^{2+}$ and NO_2.

The lifetime of $Cr_{aq}OONO^{2+}$ can be estimated from the results of scavenging experiments with L^1Ni^{2+} and NO. When

$Cr_{aq}OO^{2+}$ is mixed manually with excess NO, reactions 11, 7, and 8 are believed to be responsible for the 3:1 stoichiometry. Since the formation of $Cr_{aq}ONO^{2+}$ is complete in the mixing time (≤5 s), the lifetime of $Cr_{aq}OONO^{2+}$ must be also ≤5 s.

In the laser experiments in the presence of L^1Ni^{2+}, the formation of L^1Ni^{3+} is as fast as the disappearance of $Cr_{aq}OO^{2+}$. This observation can be rationalized by either direct reaction between L^1Ni^{2+} and $Cr_{aq}OONO^{2+}$, eqs 13 and 14, or rapid homolysis of $Cr_{aq}OONO^{2+}$ followed by L^1Ni^{2+}/NO_2 reaction.

$$Cr_{aq}OONO^{2+} + L^1Ni^{2+} \rightarrow Cr_{aq}O^{2+} + L^1Ni^{3+} + NO_2^- \quad (13)$$

$$Cr_{aq}O^{2+} + L^1Ni^{2+}\,(+2\,H^+) \rightarrow Cr_{aq}(H_2O)^{3+} + L^1Ni^{3+} \quad (14)$$

To account for the data quantitatively (the formation of L^1Ni^{3+} completed in ≤0.5 ms, independent of $[L^1Ni^{2+}]$ at concentrations greater than 0.2 mM), a rate constant for the scavenging step has to be $\geq10^8\,M^{-1}\,s^{-1}$. This value would seem too high for k_{13}, given that the related (but, presumably, less strongly oxidizing) hydroperoxo complex $Cr_{aq}OOH^{2+}$ oxidizes L^1Ni^{2+} more slowly by a factor of at least 10^7.[52] The L^1Ni^{2+}/NO_2 reaction $(k = 1.2 \times 10^8\,M^{-1}\,s^{-1})$, on the other hand, is sufficiently rapid to satisfy the above conditions but, of course, requires that the generation of NO_2 from $Cr_{aq}OONO^{2+}$ also take place in ≤0.5 ms. Without the kinetic data that would allow us to assess relative reactivities of coordinated hydroperoxide and peroxynitrite, the mechanism in eqs 13 and 14 cannot be ruled out completely, and the conservative estimate of ≤5 s must be placed on the lifetime of $Cr_{aq}OONO^{2+}$.

The short lifetimes of the two peroxynitrito ions in our work, $L^2RhOONO^{2+}$ and $Cr_{aq}OONO^{2+}$, make them similar to the peroxynitrites of hemoglobin,[5,6] myoglobin,[6] and iron[19,22] and manganese[13,20] complexes but different from another, purely inorganic ion, $(CN)_5CoOOONO^{3-}$, which is exceptionally stable.[8]

Most of the observed and postulated short-lived peroxynitrito complexes are believed[7,13,19,20,22] to decay by O–O bond homolysis followed by the reactions of NO_2 with high-valent metal-oxo species, LMO in Scheme 1, and dimerization/hydrolysis of NO_2. If this mechanistic assignment is correct, then the level of tissue protection by $ONOO^-$ isomerization catalysts must be related to the amount of NO_2 produced and the rate of its hydrolysis and oxidation by LMO in competition with nitration reactions. The hydrolysis of NO_2 should be common to all the systems, although the rates for this second-order process (eq 12) will be $[NO_2]$-dependent.

Even if the LMO/NO_2 reaction has a large rate constant, the small and equal concentrations of the two reagents may make the reaction slow enough for at least some NO_2 to be available for other reactions. Only the in-cage reaction (to yield $\{M^+ + NO_3^-\}$ or MNO_3) will make NO_2 nonscavengeable. The rate of in-cage reaction will be influenced by both overall thermodynamics and the geometry and steric crowding at the reaction center. In the case of $L^2RhOONO^{2+}$, only 40% of NO_2 and L^2RhO^{2+} diffuses into the solution.[7] In the chromium case the amount is $\sim100\%$. The data are insufficient to determine which

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(52) The kinetics of the $L^1Ni^{2+}/Cr_{aq}OOH^{2+}$ reaction are complex and the details have not been worked out, but at 0.2–1 mM L^1Ni^{2+}, the reaction would require at least several minutes for completion.

factors are dominating in determining the success of cage reaction, but the clean 1:1 $[L^2RhOO^{2+}]/[NO]$ stoichiometry under all the conditions shows that L^2RhO^{2+} reacts with NO_2 faster than $Cr_{aq}O^{2+}$ does.

An ideal catalyst for $ONOO^-$ isomerization should have $\sim100\%$ of LMO and NO_2 react inside the solvent cage. This is a difficult requirement, but if achieved, such a reaction would be indistinguishable from a purely intramolecular isomerization.

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