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Nitrosylcobalt(II) Tetraphenylporphinate: Femtosecond and Longer Studies of the Dynamics of NO Loss

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In recent years nitric oxide has become recognized as a biologically important molecule, regulating blood pressure, acting as a neurotransmitter, and participating in the ability of the immune system to kill tumor cells and intracellular parasites.[1−4] The dimeric enzyme nitric oxide synthase, NOS, which contains a pair of ferric-heme prosthetic groups, has been implicated as being responsible for the biological production of nitric oxide.[1,5,6] While the purpose and production of nitric oxide in humans are becoming understood, the storage and transport of this highly reactive, diatomic molecule require more elucidation. Metalloporphyrins, molecules known to form relatively stable complexes with nitric oxide, are abundant in nature and cannot yet be ruled out as storage/transport systems for this ground state radical. Additionally, activation of certain enzymes is thought to occur via binding of NO to the iron of the enzymatic heme residue.[1]

Several metalloporphyrin−nitric oxide complexes show NO release in significant yields subsequent to absorption of light in the molecular π-system.[7−15] The factors influencing ligand release and the nature of the electronic states between the initially excited one and the dissociative one are still poorly understood, largely because of the ultrashort time scale on which these processes take place and because of the difficulty of resolving the intense porphyrin-centered transition bands from the weak metal−metal and ligand−metal transitions. Reported here are some early results of an investigation that has been initiated to elucidate the mechanism(s) by which ligand release takes place in metalloporphyrin−nitric oxide complexes. Laser flash photolysis studies on time scales varying from microseconds to femtoseconds have been employed to characterize the spectral and dynamic properties of the electronic states of $\mathrm{(TPP)Co^{II}NO}$ and to delimit the rate constant for fragmentation of the Co−NO.

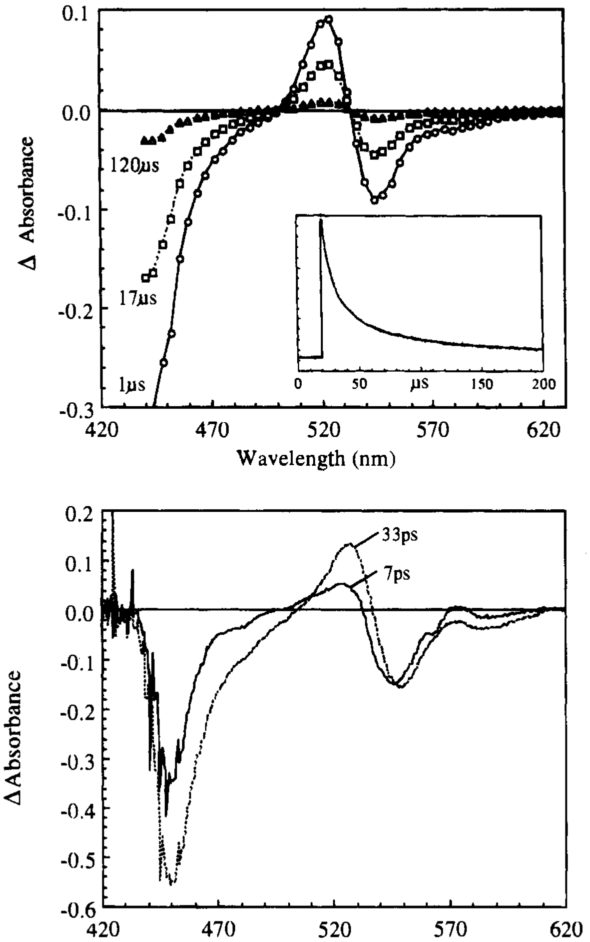

Figure 1. (a) Transient absorbance spectra at 0.5, 12, and 33 μs of $\mathrm{(TPP)Co^{II}NO}$ in Ar-saturated benzene excited at 425 nm with 7 ns pulses. Inset depicts the kinetic decay of the above sample monitored at 522 nm. (b) Transient absorbance spectra at 7 and 33 ps of $\mathrm{(TPP)Co^{II}NO}$ in Ar-saturated benzene excited at 532 nm with 30 ps pulses.

Solutions of $\mathrm{(TPP)Co^{II}NO}$[16] in Ar-saturated benzene at room temperature were excited with 7 ns pulses of 425 nm light from an OPOTEK optical paramagnetic oscillator (OPO) which was pumped by the third harmonic (355 nm) of a Q-switched Nd:YAG laser (Continuum Surelite I). Using the OPO, the excitation wavelength could be tuned to optimum wavelengths within the absorption spectra of the metalloporphyrins. Typical excitation energies employed were a few millijoules per pulse. Spectral and temporal resolution of the ensuing transients was recorded using a kinetic spectrometer system that has been described previously.[17] Transient difference spectra recorded postpulse are shown in Figure 1a. Clearly depicted are negative absorbance changes at 450 and 550 nm and a positive-going signal at 530 nm. This species decayed (under Ar-saturated conditions) to the original ground state spectrum by second-order kinetics (Figure 1a, inset) with a bimolecular rate constant of 7.9×10^9 M^{-1} s^{-1}. The spectral profile shown in Figure 1a is identical to that of the difference between the ground state spectrum of $\mathrm{(TPP)Co^{II}NO}$ and that of $\mathrm{(TPP)Co^{II}}$. This difference spectrum was present immediately after the 10 ns excitation pulse, indicating that the NO molecule is ejected from the metal

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(16) Nitrosylcobalt(II) *meso*-tetraphenylporphyrinate was prepared by introduction of NO gas, previously scrubbed of higher oxides by passage through a KOH column, into a nitrogen-saturated solution of $\mathrm{(TPP)Co^{II}}$ (Midcentury Chemicals) dissolved in benzene. After bubbling for $\sim$20 min, NO-saturated MeOH was added until precipitation of the adduct was complete. The reaction vessel was then evacuated and purged with N_2. The resulting precipitate was collected under N_2, dried, and stored under vacuum.

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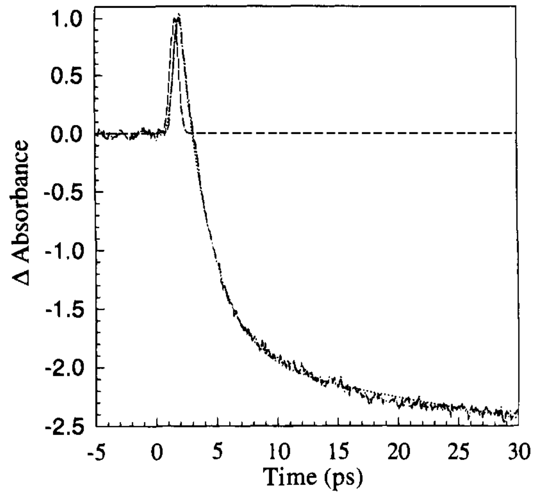

Figure 2. Transient absorption changes at 450 nm of (TPP)Co^{II}NO in Ar-saturated benzene excited at 400 nm with 500 fs pulses. The fitted (dotted) curve includes convolution with a Gaussian instrument response profile (dashed curve).

center in times shorter than this. Moreover, by employing an excitation pulse of 30 ps coupled with a diode array kinetic spectrography instrument as described earlier,[18] it was found that the same spectral feature was completely formed within the rise time of the detection system (Figure 1b). Thus NO release occurs instantaneously (within our time resolution) on the picosecond time scale. Close examination of Figure 1b shows that there is a spectral shift from 535 to 540 nm as the reaction proceeds from early in the pulse (7 ps) to near the end of the pulse (33 ps). In view of what follows, it is probable that this shift is real and is a consequence of the NO loss process occurring at the earlier times during the pulse.

In an attempt to directly observe the rate of loss of NO from the nitrosylcobalt^{II} porphyrin, sub-picosecond time resolution was employed. Ar-saturated solutions of (TPP)Co^{II}NO in benzene (A_{400} = 1.0 per 0.5 mm) were exposed to 500 fs pulses of 390 nm radiation from a frequency-doubled, amplified Ti: sapphire laser. The sample was flowed through the 0.5 mm excitation path length quartz cuvette to minimize photodegradation from prolonged exposure to the excitation radiation. A white continuum, generated by focusing the residual Ti:sapphire fundamental through a 1 cm path length cell of flowing ethylene glycol, was employed as probe beam. The probe wavelength of 450 nm was isolated from the continuum by an interference filter. Details of the femtosecond laser have been described.[19] The resulting time profile at 450 nm (Figure 2) shows initially a positive difference absorption, which subsequently converts to a negative difference absorption. This latter state is reached via two consecutive exponential processes, having rate constants of (4.7 ± 0.5) × 10^{11} and (9.0 ± 1.0) × 10^{10} s^{-1}, respectively. On the assumption that the species present at the end of the kinetic profile (at 30 ps) in Figure 2 is identical to that seen at 33 ps in the absorption spectrum in Figure 1b, i.e., the thermally-relaxed (TPP)Co^{II}, the time profile in Figure 2 reveals the evolution of the excitation and relaxation stages summarized in the following sequence:

$$S_0 \rightarrow S_2 \rightarrow S_1 \rightarrow CT \rightarrow {}^*(TPP)Co^{II} + NO \rightarrow (TPP)Co^{II}$$

where S_0, S_2, and S_1 refer to states localized on the porphyrin π-system, CT refers to a metal-centered electronic state, and the asterisk indicates vibrational excitation. The initial photoexcitation step is clearly seen in Figure 2 as the rapidly rising positive absorption. Subsequently two clearly distinguished kinetic processes (Figure 2) having rate constants of 4.7 × 10^{11} and 9.0 × 10^{10} s^{-1} lead to the overall negative difference absorption. These two processes together must account for the four schematic steps from the initially produced S_2 to relaxed (TPP)Co^{II}. The lower of the two distinguishable rate constants (9.0 × 10^{10} s^{-1}) probably arises from vibrational cooling of the freed porphyrin. This last process in hemes has been reported to have a lifetime in the region of 10 ps.[20] The fast process (k = 4.7 × 10^{11} s^{-1}) must therefore be the resultant of the three processes: $S_2 \rightarrow S_1$ internal conversion, $S_1 \rightarrow$ CT conversion, and NO liberation from the CT state. The $S_2 \rightarrow S_1$ process is expected to be very rapid and probably complete in less than 10^{-12} s. This premise, coupled with the assumption that the metal-centered CT state dissociates as soon as it is formed, leads to the conclusion that the process carrying the 4.7 × 10^{11} s^{-1} rate constant is the $S_1 \rightarrow$ CT conversion.

That the quantum yield for NO evolution in the directly excited case[8] is 1.0 ± 0.05 supports the conclusion that NO loss occurs extremely rapidly. The Co−NO bond is presumed to be very weak in these metalloporphyrin complexes,[9] and therefore it is not unreasonable to conclude that the $^1(\pi,d_z)$CT state possesses enough energy to cause Co−NO bond homolysis.[21] Furthermore, the CT state is the first state in the downward cascade of electronic excited states which involves the metal d_z orbital, and it is generally accepted that increasing the electron density in the d_z orbital induces ligand release from these types of compounds.[25] Moreover, a photoinduced process having a quantum yield of unity necessitates that all putative competing processes be significantly slower, a condition that is easier to adhere to if the NO-loss reaction occurs in sub-picoseconds.

While the above assignments are far from firm, the data clearly show that there is a richness of detail to be found at these ultrashort time scales. Experiments are currently underway to further characterize the dynamics of the electronic states preceding NO release in this and other metalloporphyrin−NO complexes and to understand which factors influence the efficiency by which ligand release occurs.

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