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Photochemical Dynamics of a Trimethyl-Phosphine Derivatized [FeFe]-Hydrogenase Model Compound

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Abstract

Though there have been many studies on photosensitizers coupled to model complexes of the [FeFe]-hydrogenases, few have looked at how the models react upon exposure to light. To extract photoreaction information, ultrafast time-resolved UV/visible pump, IR probe spectroscopy was performed on $\text{Fe}_2(\mu\text{-S}_2\text{C}_2\text{H}_4)(\text{CO})_4(\text{PMe}_3)_2$ (**2b**) dissolved in heptane and acetonitrile and the photochemical dynamics were determined. Excitation with 532 and 355 nm light produces bleaches and new absorptions that decay to half their original intensity with time constants of 300 ± 120 ps and 380 ± 210 ps in heptane and acetonitrile, respectively. These features persist to the microsecond timescale. The dynamics of **2b** are assigned to formation of an initial set of photoproducts, which were a mixture of excited-state tricarbonyl isomers. These isomers decay into another set of long-lived photoproducts in which approximately half the excited-state tricarbonyl isomers recombine with CO to form another complex mixture of tricarbonyl and tetracarbonyl isomers.

Keywords

Hydrogenase models; ultrafast; time-resolved vibrational spectroscopy; reaction dynamics; photochemical isomerization; molecular structure

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INTRODUCTION

Hydrogenases, a class of convergent-evolved metalloenzymes found in a variety of microorganisms, catalyze the reversible oxidation-reduction between H^+ and hydrogen gas [1–6]. These enzymes are generally utilized in metabolic processes, and they are unique in that they are the only known metalloenzyme with carbonyl and cyanide ligands occurring naturally in the active sites [6]. There are three main types of hydrogenases, which are classified based on the metal composition in the active site: the [Fe]-, [NiFe]-, and [FeFe]-hydrogenases. In general, [NiFe]-hydrogenases catalyze the oxidation reaction, while [Fe]- and [FeFe]-hydrogenases catalyze reduction though the [Fe]-hydrogenases are much rarer [6–10]. Furthermore, [FeFe]-hydrogenases commonly display a larger catalytic turnover rate than their [Fe]- and [NiFe]-hydrogenase counterparts [3]. Therefore, the [FeFe]-hydrogenases have the potential to be used as an alternative means of producing hydrogen gas that is not reliant on fossil fuels.

The general structure of the active site of [FeFe]-hydrogenases, shown in Figure 1, consists of a di-iron bond, a bidentate dithioate ligand where the sulfur atoms are connected by an amine and both chelate and bridge the di-iron center. In addition to a lone hydrogen ligand, a carbonyl ligand bridges the two irons with carbonyl and cyanide pendent ligands coordinated to each iron. However, this overall structure depends on the oxidation state of the enzyme [1, 11, 12]. The proximal iron of the active site is usually ligated to the sulfur of a cysteine, which has a $[Fe_4-S_4]$ cluster attached to it. The oxidation state of the di-iron center in its catalytically active form is $Fe^I Fe^II$ [13]. During catalysis, the di-iron center is reduced to $Fe^I Fe^I$, which causes the bridging carbonyl to move to the iron distal to the cysteine and $[Fe_4-S_4]$ cluster, leaving a vacancy across the di-iron bond for subsequent protons to bind [13].

The natural [FeFe]-hydrogenase enzymes and their active sites have been studied for their use as a catalyst. An [FeFe]-hydrogenase enzyme has been successfully complexed to TiO_2 electrodes [14] and CdS nanorods [15] with catalytic efficiencies of 70% and 20%, respectively. Furthermore, the native active site has been extracted and shown to retain catalytic activity in acidified water [16]. The enzyme has also been attached to Photosystem I through its distal $[Fe_4-S_4]$ in order to make the oxidation-reduction reaction light-driven [5].

Recent research has focused on synthesizing model compounds with similar structures to the [FeFe]-hydrogenase active site that can be used in hydrogen production. Many of these models are based on compounds that consist of two irons, two sulfur bridged connected by either an ethyl or propyl bridge, and six pendent ligands (carbonyl, phosphine, and/or cyanide) with three ligands on each iron [17, 18]. Model compounds with phosphine in place of cyanide ligands are of particular interest because they do not appear to have the same tendency as the cyanide models to become insoluble and inactive when acid is present [18, 19]. Figure 2 displays the structures of the lowest energy isomers of four of these models: $Fe_2(\mu-S_2C_3H_6)(CO)_6$ (**1a**), $Fe_2(\mu-S_2C_2H_6)(CO)_6$ (**1b**), $Fe_2(\mu-S_2C_3H_6)(CO)_4(PMe_3)_2$ (**2a**), and $Fe_2(\mu-S_2C_2H_4)(CO)_6(PMe_3)_2$ (**2b**). However, the catalytic turnover rates of all these model species are significantly lower than that of the native enzyme.

Many studies have also looked at attaching photosensitizers to [FeFe]-hydrogenase models in order to drive the reduction using light, with a mechanism much like Photosystem II [25–34]. Various types of photosensitizers have been used, including porphyrin and metalloporphyrin rings [27, 28, 31], ruthenium photosensitizers [25, 29, 30, 32, 33], and fullerenes [34]. These studies have mostly focused on the synthetic and structural characteristics of these model systems, but the photochemical dynamics of the active site models themselves have yet to be fully elucidated. Furthermore, the native active site of [FeFe]-hydrogenase enzymes have been shown to react upon exposure to UV-visible light [35, 36]. Multidimensional IR spectroscopy (2D-IR) has been performed on **1a** and $[\text{Fe}_2(\mu\text{-S}_2\text{C}_3\text{H}_6)(\text{CO})_4(\text{CN})_2]^{2-}$ in order to determine the vibrational mode energy and coupling dynamics and relaxation mechanisms [37, 38], which appear to be solvent-dependent and changes when cyanide ligands are present in the models. 2D-IR has also been coupled with 350 nm excitation for studies of **1a** [39]. Ultrafast time-resolved UV pump, IR probe spectroscopy exciting at a single frequency (350 nm) has also been performed on **1a** and **2a** to elucidate the photochemical dynamics of these models. Excitation at 350 nm of **1a** in heptane, heptyl cyanide, and acetonitrile produces photoproducts, which were assigned to pentacarbonyl species that form metastable adducts where the solvent binds to the CO vacancy, a behavior which is independent of solvent identity [40, 41]. Excitation at 350 nm of **2a** in heptane, methanol, and acetonitrile produces photoproducts that were assigned to a mixture of 4-CO and 3-CO photoproducts, with the 3-CO products recombining to produce a 4-CO product mixture with isomer ratios differing from the starting isomer ratios [42].

Ultrafast time-resolved, UV-visible pump, IR probe spectroscopy (TRIR) was previously performed by our group on **1a**, **1b**, and **2a** by exciting the sample at multiple wavelengths [43, 44]. These studies utilized 532 nm, 355 nm, and 266 nm excitation wavelengths which correspond to absorptions in the UV-visible spectra of these models. When **1a** in heptane and acetonitrile are excited at multiple UV-visible wavelengths, wavelength-dependent behavior was observed, with pentacarbonyl formation when using 266 nm excitation. With 355 nm excitation, an additional photoproduct consisting of electronic excitation accompanied by a longer Fe-Fe bond is observed. Using 532 nm excitation, both pathways are still observed, but the electronic excitation is significantly stronger [43]. Similar dynamics are seen in **1b** but with significantly more spectral overlap between the two photoproducts.

When **2a** is excited at multiple UV-visible wavelengths in heptane or acetonitrile, initial photoproducts were produced which were assigned to excited-state 3-CO photoproducts. About half the 3-CO photoproduct recombined with CO, producing a complex mixture of 3-CO and 4-CO photoproducts resulting in long-lived signals [44]. This behavior was distinctly different from that previously assigned to **2a**.

The photochemical dynamics of **2b** have not been previously studied. In this paper, the dynamic behavior of **2b** is measured and compared to the behavior of **2a** in order to determine the effects of changing the alkyl bridge length, as was done with **1a** and **1b** [43]. The UV-visible spectra of **2b** in hexane and acetonitrile is shown in Figure 3. Ultrafast UV and visible pulses at 532 nm, 355 nm, and 266 nm were used to excite the solution-phase molecules, and the time-dependent behavior was monitored with IR pulses tuned to the 5

micron carbonyl-stretching region. The results of these studies were compared with Density Functional Theory (DFT) calculations of the possible photoproducts to determine the potential relaxation and photoproduct pathways.

METHODS

Synthesis

All syntheses were conducted according to literature methods [9, 20] under an inert atmosphere using a glovebox and Schlenk line. All chemicals were obtained from Sigma Aldrich (Saint Louis, MO) and used as received unless otherwise noted [45]. The final purity of the products was ascertained by FTIR spectroscopy and compared to prior work [10,19]. Solids stored under inert atmosphere are generally stable but solutions were again examined by FTIR prior to each TRIR investigation.

Fe₂(μ-S₂C₂H₄)(CO)₆ (1b) [9]: To synthesize **1b**, 3.0 g of Fe₃(CO)₁₂ was dissolved in toluene, and 500 μL of ethanedithiol added. The mixture was stirred at 70 °C until the color changed from green to red. The resultant mixture was filtered, dried, and extracted with hexanes before filtering and drying a final time.

Fe₂(μ-S₂C₂H₄)(CO)₄(PMe₃)₂ (2b) [46]: To synthesize **2b**, 0.80 g of **1b** was dissolved in hexanes, and 0.88 mL of trimethylphosphine added. The solution was refluxed under argon at 70 °C overnight. The resulting solution was filtered and dried under vacuum. Quantitative FT-IR spectra in the CO region of **2b** dissolved in hexanes and acetonitrile are shown in the supplementary information (Figure S11).

TRIR

For TRIR measurements, approximately 30 mg of **2b** was dissolved in 50 mL of either n-heptane or anhydrous acetonitrile and constantly flowed through a sample cell with a 2 mm pathlength and CaF₂ windows. The sample concentrations used produced pump and probe static absorption features with an optical density (OD) less than one. To reduce degradation and preserve solutions, the flowing sample was purged with argon and light exposure was minimized. The excitation beam was shuttered at 10 Hz for infrared probe normalization and to permit complete sample exchange of the excitation volume.

For short-delay experiments (0 to 750 ps), a diode-pumped, Nd:Vanadate laser (10 ps, 2 W output at 532 nm, 80 MHz) was used to synchronously pump two tunable dye lasers, one tuned to 589 nm and the other to 656 nm, and to seed a 20 Hz Nd:YAG regenerative amplifier. The output from the Nd:YAG regenerative amplifier (532 nm, 25 mJ/pulse) was used to longitudinally pump three-stage dye cells to amplify the outputs from each of the dye lasers, and the residual Nd:YAG 1.06 micron output was used to generate optically delayed 70 ps to 50 ps excitation pulses at 532 nm, 355 nm and 266 nm (1 mJ, 150 μJ, and 50 μJ per pulse at the sample, respectively). The amplified dye lasers were combined in a 1 cm long LiIO₃ difference mixing crystal to produce 150 fs, tunable broadband IR probe pulses (ca. 80 cm⁻¹ full width at half-maximum FWHM), which passed through a timing delay line and split in two. One IR beam, which becomes the signal beam, was combined

with the pump beam before being focused into the sample cell. The other IR beam entered an unexcited location in the sample and was used as a reference. The IR beam was focused to about 100 μm in diameter through the sample cell, while the UV-visible excitation beams were focused to about 500 μm in diameter. The signal and reference beams then entered a monochromator and were detected by a 256×256 pixel InSb focal plane detector for spectral analysis (ca. 4 cm^{-1} FWHM spectral resolution). In order to cover the entire region of interest, two spectral regions were studied, one with the probe pulses centered at 1920 cm^{-1} , and the second with the probe centered at 1980 cm^{-1} . The spectra were combined by overlapping the roughly 60 cm^{-1} region the spectra had in common.

Long-delay experiments (10 ns to 15 ps) were conducted by using the femtosecond IR probe set-up as for the short-time experiments, except the pump beam was created using a separate diode-pumped, Nd:YAG Q-switched laser ($\sim 5 \text{ ns}$ pulse width) with pump-probe timing controlled electronically. We ensured that the same pump pulse beam diameters and energies were directed to the sample when using the Q-switched excitation source as for the picosecond experiments described above. All experiments were conducted at 298 K.

COMPUTATIONS & MODELING

Ground-State Compounds (FT-IR):

DFT calculations were conducted on all isomers of **2b** using Gaussian 03W (Gaussian, Inc.) [45, 47]. The ground-state isomers are shown in the first row of Figure 4 and numbered to be consistent with Ref. 44 [44]. Calculations were run using Becke and Perdew's BP86 functional [48–50], using the TZVP basis set [51, 52].

Ground-State Photoproducts via TRIR:

DFT, BP86 method [48–50], TZVP basis set [51, 52] calculations (Gaussian 03W, Gaussian Inc. [47]) were run on the various 3-CO loss products of **2b** (see Figure 4).

Modeling IR and TRIR Spectra:

Excel (Microsoft Office Professional Plus 2013) [45] was used to model the ground-state IR and TRIR spectra. To model the ground-state IR spectrum, the CO bands of each ground state, 4-CO isomer were modeled using the DFT-generated frequency and intensity data. Each band was represented by a Lorentzian distribution, with a full-width of 5 cm^{-1} for spectra in heptane and 9 cm^{-1} for spectra in acetonitrile. The isomers were manually combined in various ratios until the best match with the experimental spectrum was obtained.

The ground-state IR spectra (and the corresponding bleaches in TRIR modeling) contain components of two separate ground-state, 4-CO isomers (labeled **2b-1** and **2b-2** in Figure 4). In order to fit the FT-IR spectrum in hexanes, a ratio of 60% **2b-1** and 40% **2b-2** is used (in more-polar acetonitrile, the ratio shifts to 25% **2b-1** and 75% **2b-2**). The results of these fits are shown in Figure SI2 for hexanes and Figure SI3 for acetonitrile. The isomers present are similar to those found for **2a** [42, 44], however, **2b** seems to contain significantly more of the more-polar **2b-2** isomer in both solvents than the corresponding **2a** isomers.

The TRIR spectra were modeled using the IR spectrum as a negative signal, forming the bleaches. The remaining ground-state 4-CO and 3-CO isomers were used to model the potential new absorptions in the spectra as positive-going signals. These models were compared with the long-time TRIR signals to help determine which isomers might be present in the long-time data.

TRIR Time Constants:

The time dependent data of the major bands in the TRIR spectra of **2b** were modeled as a single exponential decay with a long-time offset. Data points 100 ps and later were initially fit using Microsoft Excel's Solver routine. The long-time offset generated was then subtracted off, allowing Microsoft Excel's LOGEST routine to be used to calculate a standard deviation for the time constant. Modifications of this procedure due to noisy data and poor fits are noted in the footnotes of the corresponding tables.

RESULTS

TRIR of **2b** in Heptane

2b was dissolved in n-heptane, and TRIR was performed by exciting the sample with 532 nm, 355 nm, and 266 nm pulses and collecting the transient IR spectra of the sample at various delay times before, during, and after excitation. Figure 5 displays the time-resolved IR difference spectra, which are the collected IR spectra subtracted by the unpumped (ground-state) IR spectra (which is the same as the FTIR spectrum shown d, red)) when the sample is excited at (a) 266 nm, (b) 355 nm and (c) 532 nm. Dark green represents early delay times, while dark blue represents later times. The bleach bands below the baseline represent reductions in the absorptions of the starting material, which line up nicely with the bands in the ground state FT-IR spectrum. The new absorption bands above the baseline, arise from photoproducts.

532 nm excitation: There are three distinct new absorption bands at 1892 cm^{-1} , 1920 cm^{-1} , and 1949 cm^{-1} which grow in with the pump-probe pulse convolution time of ca. 80 ps. The absorptions at 1892 cm^{-1} and 1949 cm^{-1} , along with the bleaches at 1908 cm^{-1} and 1958 cm^{-1} , all decay to about half the maximum AOD, with long-lived signals lasting out into the microsecond timescale (nanosecond and longer signals are essentially the same as the 742 ps spectra, see Figure S4). The 1920 cm^{-1} band starts out as bleach, which transitions to a new absorption. An apparent new absorption at around 2000 cm^{-1} was checked for reproducibility (data not shown), but the behavior appears to be an artifact in the IR spectra due to low light intensities and is not photochemical behavior.

The time-dependent behavior of the bleach and absorption bands presented in Figure 5 are shown in Figure 6, along with their associated best fits to an exponential decay plus a long-time offset (solid lines in Figure 6). The resulting time-constants and R^2 values are summarized in Table 1. Overall, both the absorptions and the bleaches decay with a time constant of $300 \pm 120\text{ ps}$ (the average is $450 \pm 85\text{ ps}$ if the fits requiring manipulations are removed from consideration).

355 nm excitation: As seen in the 532 nm heptane data, there are three distinct new absorption bands at 1891 cm^{-1} , 1921 cm^{-1} , and 1950 cm^{-1} . Both the bleaches and absorptions, except for the 1921 cm^{-1} absorption, decay to about half the original OD, with long-lived signals remaining into the microsecond timescale (See FigureS4). The 1921 cm^{-1} absorption appears to transition from a bleach to a new absorption.

The time-dependent behavior of the bleach and absorption bands are shown in Figure 7, along with their associated fits. The resulting time-constants and R^2 values for the linear fits are summarized in Table 2.

When averaged, both the absorptions and bleach bands decay with a time constant of $320 \pm 200\text{ ps}$. This time constant is consistent with the overall time constant found for 532 nm excitation in heptane.

266 nm excitation: The signal-to-noise ratio of the acquired 266 nm heptane data is too small to obtain reliable fits to extract the time-dependent photo-dynamics. However, the strongest bleaches observed in the 532 nm and 355 nm excitation data are also found in the 266 nm transient data and shown for comparison in Figure 5 a).

TRIR of 2b in Acetonitrile

2b was dissolved in acetonitrile, and TRIR was performed at 532 nm, 355 nm, and 266 nm. Figure 8 displays the resulting TRIR plots at 266 a), 355 b), and c) 532 nm where dark green is early time and dark blue is later delay time. The ground state FT-IR spectrum of **2b** in acetonitrile is also displayed in Figure 8 d) (red spectrum).

532 nm excitation: There are three distinct new absorption bands found at 1875 cm^{-1} , 1931 cm^{-1} , and 1967 cm^{-1} . Both the bleaches and new absorptions at 1875 cm^{-1} and 1931 cm^{-1} decay to about half the maximum OD, with long-lived signals lasting out into the microsecond regime (see Figure S4). The 1921 cm^{-1} absorption observed in the heptane data with 532 nm and 355 nm excitation is no longer present in the acetonitrile data. It has likely been lost due to the broadening from the more polar solvent, causing the new small absorption to be overwhelmed by the larger, broader bleach at 1904 cm^{-1} . Furthermore, there is a new absorption at 1967 cm^{-1} that does not correspond to any band in the heptane data. The absorption at 1967 cm^{-1} decays nearly to the baseline at long times. Again, the apparent new absorption in the region past the 1982 cm^{-1} bleach appears to be an artifact due to low light levels in that region.

The time-dependent behavior of the bleach and absorption bands and their associated fits are shown in Figure 9. The resulting decay time-constants and R^2 values for the linear fits are summarized in Table 3.

Overall, both the absorptions and bleaches decay with an averaged time constant of $480 \pm 220\text{ ps}$ (if only the unmodified fits are considered, the time constant is $390 \pm 140\text{ ps}$). This time constant is consistent with the those for the 355 nm and 532 nm excitation wavelengths for the heptane data. However, the uncertainties for all of the time constants are large.

355 nm excitation: There are three distinct new absorption bands at 1874 cm^{-1} , 1929 cm^{-1} , and 1960 cm^{-1} . The bleaches and absorptions, except the 1876 cm^{-1} absorption and the 1908 cm^{-1} bleach, decay to about half their maximum AOD, with long-lived signals persisting to the microsecond timescale (see Figure S4). Both the 1874 cm^{-1} absorption and 1908 cm^{-1} bleach appear to grow in and persist into long timescales with little decay, although this may be an experimental artifact. As in the 532 nm acetonitrile data, the 1921 cm^{-1} absorption observed in the heptane data with 532 nm and 355 nm excitation is no longer present in the acetonitrile data. The new absorption at 1960 cm^{-1} is very weak, if it is present at all.

The time-dependent behavior of the bleach and absorption bands are shown in Figure 10, with their associated fits. The resulting time-constants and R^2 values for the linear fits are summarized in Table 4.

Overall, the absorptions and bleaches decay with an average time constant of $230 \pm 33\text{ ps}$. Given the large uncertainties in the other time constants, this time constant is likely not significantly different from the time constants extracted from the other datasets.

266 nm excitation: Although the 266 nm acetonitrile data display stronger signals than the 355 nm acetonitrile data, the noise in the data was again too large to analyze the photochemical dynamics. However, the bleaches and adsorption features observed in the 355 nm excitation data appear to also occur to some degree in the 266 nm data and are shown in Figure 8 a) for comparison.

DISCUSSION

Consideration of all the data sets shows a pattern of signals that grow in with the excitation laser pulse, then decay with a time constant of a few hundred picoseconds with a long-lived persistent non-zero signal. Given the results presented above, two possibilities to explain these findings exist. For **1a** [43], two distinct sets of bands are clearly observed that grow in with the excitation laser pulse. One set of bands decays in about 150 ps, while the other set remains for microseconds. For **2a** [42, 44], two distinct sets of bands were seen, again with one growing in with the excitation pulse, then decaying away in with a time constant of about 150 ps. The second set of absorption features grows in with approximately this longer time constant.

Unfortunately, the data for **2b** is not as clear. The short-lived and long-lived absorption bands are almost entirely overlapping. While there is some evidence of bands that decay almost entirely to baseline, there are no clear bands that show a growth in intensity on the few-hundred picosecond timescale (with the possible exception of the 1920 cm^{-1} band in the heptane data, which may convert from an overlapping bleach to a new absorption on this timescale). This overlapping of bands is similar to what was observed for **1b** [43], where the exchange of a propyl bridge with an ethyl bridge also resulted in substantial overlap between the short-lived and long-lived bands. While it may be possible that slight perturbation of the Fe-Fe bond distance could affect the electronic structure and IR product bands, at present we

have no compelling explanation for these spectral observations arising from this nominally simple bridge substitution and further study is warranted.

In parallel to the mechanism determined for **2a** photochemistry [44], the short-lived photoproducts are assigned to CO-loss species likely formed within an dissociative electronic excited state. The signals from these short-lived photoproducts decay as the molecules relax from this excited state with a time-constant of a few hundred picoseconds.

Ground-State Photoproduct Identification

DFT calculations were employed to establish the identity of the ground-state photoproducts for **2b**. More complicated excited state DFT calculations would be required to determine the composition of the excited state photoproducts. However, ground-state DFT calculations have been shown to be sufficient for determining the composition and structures of the long-lived photoproducts for **1a-2a** reactions [43, 44].

Figure 11 a) displays potential ground-state spectra of possible product isomers, assuming all of the photoproducts are 4-CO isomers for **2b** in n-heptane. In these spectra, the negative-going bleaches are a 60%/40% mixture of isomers **2b-1** and **2b-2**, as in the heptane spectra (bandwidths are also appropriate for the heptane spectra). Note that none of the 4-CO isomers has any band lower in energy than the 1904 cm^{-1} band of isomer **2b-1**. This implies that the new absorptions observed below this band (centered around 1890 cm^{-1} in heptane and 1875 cm^{-1} in acetonitrile) cannot be explained based on 4-CO product isomers alone.

Figure 11 b) displays select 3-CO isomers of **2b** that could potentially model the 1875 cm^{-1} to 1890 cm^{-1} absorption bands found in all the TRIR data for **2b** compared to the bleaches present in the long-time data of **2b** in n-heptane. Adding 3-CO isomers produces better agreement with the TRIR data. While the inclusion of 3-CO isomers to describe the long-time signals is necessary to model the data, the data is not of sufficient quality to indicate which 3-CO isomers might be present. A total of nine 3-CO isomers are possible (not counting enantiomers, see Figure 4), and the long-time signal likely results from an admixture of these isomers. We also cannot rule out the presence of 4-CO isomers after reaction is completed. The long-time spectra are thus presumed to be generated by a complicated mixture of ground-state 3-CO isomers and a non-thermal population of 4-CO isomers.

While the above analysis used heptane bleaches and bandwidths, a similar analysis was conducted to model the acetonitrile data, and the conclusion is essentially the same. However, given the presence of a long-lived absorption at 1967 cm^{-1} in the acetonitrile data that is not present in the heptane data, the ensuing mixture of 3-CO and 4-CO product isomers is presumably different for the two solvents, although this may be due simply to the difference in the initial isomer ratio.

Table 5 summarizes the calculated energies ΔG values are the energy difference between a particular isomer and the lowest energy isomer) and the band frequencies and intensities for the ground-state 4-CO and 3-CO isomers. The structure of the 4-CO and 3-CO isomers are displayed in Figure 4.

The overall photo-dynamics for **2b** appear to be similar to the dynamics observed for **2a** [44]. For **2a** in both heptane and acetonitrile, two sets of photoproducts were identified when excited at 355 nm and 532 nm. The short-lived absorptions (135 ± 74 ps) were attributed to a mixture of electronically excited 3-CO photoproducts. These bands decayed and were replaced by a new set of bands that increased in intensity with a time constant of 214 ± 119 ps. The decay is attributed to vibrational relaxation and recombination with CO generating about half of the population as 4-CO ground state isomers in the original isomer ratio. The other half of the initially-excited molecules decayed back to the electronic ground state, either without recombining with CO (forming ground-state 3-CO photoproducts) or recombining with CO but with a non-thermal mixture of isomers, causing the new bands to grow in. This mechanism formed a complex mixture of ground state 3-CO and 4-CO isomers. The ratio of 4-CO isomers was different than the starting isomer ratio, resulting in signals from both 3-CO and 4-CO species. The particular ratios of product isomer populations appears to differ depending on the excitation wavelength and solvent used. This is likely due to differences in the UV-Vis absorption spectra of the different isomers and differences in the ratio of starting isomers [44]. Similar reaction dynamics appear to occur for **2b**, but the time constant for decay back to the ground state and recombination with CO seems to be somewhat longer (ca. 230 vs 150 ps).

The other major difference between the behavior of **2b** and **2a** is that for **2a**, the electronically excited and ground state species have distinctly different spectra, so transiently decaying and increasing product absorption bands were readily separated and analyzed. In **2b**, the spectra of the excited and ground state species overlap strongly, so the growth of new absorption bands is masked by decay of the already-existing bands. Similar spectral overlap effects were found when comparing product spectra for **1a** and **1b** [43]. The spectra of **1a** included two distinct sets of bands while the second set remained constant for microsecond time delays. The spectra for **1b** showed similar dynamics, but the sets of bands were entirely overlapping. These differences in behavior of the time-resolved spectra are somewhat surprising, since **1a** and **1b** differ only from the bridging disulfur ligand (propyl versus ethyl disulfide), and the steady-state IR spectra exhibit only a 2.5 cm^{-1} shift. **2a** and **2b** display more distinct differences in their steady-state IR spectra, but that difference is wholly attributed to a the steady-state isomer ratio with each isomer only showing a small shift in frequency. Further more detailed studies are required to fully understand the reason for observed differences in the time-dependent product spectra for propyl versus ethyl bridged species.

CONCLUSIONS

Excitation of **2b** with 532 nm and 355 nm light produces bleaches and new absorptions which decay to half their original AOD maxima. These remnant signals persist into the microsecond timescale. Initially, the bleaches and absorptions decay with average time constants of $ca\ 300 \pm 120$ ps and 380 ± 210 ps for **2b** in heptane and acetonitrile, respectively. The ratio of isomeric photoproducts generated from **2b** depend on both the excitation wavelength employed and the solvent. As was found earlier for **2a**, product bands for **2b** also are assigned to two sets of photoproducts [44]: 1) an initially generated set of

photoproducts is assigned to a mixture of 3- CO excited-state isomers and 2) the initial photoproduct population decays into another set of photoproducts which result from recombination of about half the excited-state 3-CO isomers with CO to form a complex mixture of 3-CO and 4-CO ground-state isomers. Further exploration of these assignments and comparison to similarly liganded analogs is warranted using more sophisticated measurement methods and spectral assignments.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Figure 1.
The structure of the [FeFe]- hydrogenase active site [1,11,12]

Figure 2.
The lowest energy isomers of: Fe₂(μ-S₂C₃H₆)(CO)₆ (**1a**), Fe₂(μ-S₂C₂H₄)(CO)₆ (**1b**),
Fe₂(μ-S₂C₃H₆)(CO)₄(PMe₃)₂ (**2a**), Fe₂(μ-S₂C₂H₄)(CO)₄(PMe₃)₂ (**2b**).

Figure 3.

The UV-visible spectra of **2b** dissolved in hexanes and acetonitrile. The excitation wavelengths (532, 355, and 266 nm) are marked.

Figure 4.

The structures of the ground-state 4-CO isomers (**2b-1**, **2b-2**, **2b-4**, **2b-5**) and corresponding 3-CO isomers. Isomers are labeled to be consistent with Ref. 44.

Figure 5.

The TRIR spectra of **2b** in n-heptane excited at a) 266 nm, b) 355 nm, and c) 532 nm. d) The FT-IR spectrum of **2b** in hexanes is also displayed for reference.

Figure 6. The time-dependent data and fits of a) new absorptions and b) bleaches for **2b** in heptane excited at 532 nm at 293 K.

Figure 7. Time dependent data and fits from the a) new absorptions and b) and bleaches for **2b** in heptane excited at 355 nm.

Figure 8.

TRIR spectra of **2b** in acetonitrile excited at: a) 266 nm, b) 355 nm, and c) 532 nm. d) The FT-IR spectrum of **2b** in acetonitrile is also displayed for reference.

Figure 9. The time dependent data and fits to the a) new absorptions and b) bleaches for **2b** in acetonitrile excited at 532 nm.

Figure 10.

The time dependent data and fits of the a) new absorptions and b) bleaches for **2b** in acetonitrile excited at 355 nm.

Figure 11.

Bleach bands for **2b** in n-heptane long-time data and the simulated spectra of: a) the 4-CO ground-state isomers of **2b** and b) select 3-CO ground-state isomers of **2b**.

Table 1.

Time decay constants (τ) and R^2 values for **2b** in heptane with 532 nm excitation.

	wavenumber(cm^{-1})	τ (Ps)	R^2
absorbance	1892	390 ± 31	0.94
bleach	1908	180 ± 43^a	0.68
absorbance	1920	270 ± 63^b	0.65
absorbance	1949	510 ± 31	0.96
bleach	1958	240 ± 84^c	0.45
bleach	1994	220 ± 41^d	0.79

^aSolver did not converge with last two points. Manually set offset to -0.06 and removed last two points for LOGEST.

^bSolver did not converge. Manually set offset to 0.0078 for LOGEST.

^cSolver did not converge. Manually set offset at -0.043 for LOGEST.

^dRemoved 633 and 674 ps data points for LOGEST.

Table 2.Time decay constants (τ) and R^2 values for **2b** in heptane with 355 nm excitation

	wavenumber(cm^{-1})	τ (ps)	R^2
absorbance	1891	250 ± 72^a	0.57
bleach	1908	270 ± 140^b	0.28
absorbance	1921	260 ± 47	0.76
absorbance	1950	210 ± 71^c	0.49
bleach	1956	720 ± 100^d	0.86
bleach	1994	190 ± 38^e	0.74

^a Solver did not converge. Manually set offset to 0.014 for LOGEST and removed 500 ps point.^b Solver did not converge. Removed 200 ps point and set offset to -0.024 for LOGEST.^c Solver did not converge. Manually set offset to 0.016 and deleted 100 ps point for LOGEST.^d Solver did not converge. Manually set offset at 0.015 and removed 600 and 674 ps data points for LOGEST.^e Removed 674 ns data point for LOGEST.

Table 3.

Time decay constants (τ) and R^2 values for **2b** in acetonitrile with 532 nm excitation.

	wavenumber(cm^{-1})	τ (ps)	R^2
absorbance	1875	550 ± 69	0.86
bleach	1904	250 ± 24^a	0.91
absorbance	1931	790 ± 110^b	0.85
bleach	1944	300 ± 22	0.95
absorbance	1967	640 ± 130^c	0.71
bleach	1983	330 ± 190	0.23

^a Solver did not converge. Manually set offset to -0.013 for LOGEST.

^b Had to eliminate first 100 and 133 ps data points for Solver to converge. All points included for LOGEST.

^c Deleted 100 ps data point for Solver to converge. All points included for LOGEST.

Table 4.Time decay constants (τ) and R^2 values for **2b** in acetonitrile with 355 nm excitation

	wavenumber(cm^{-1})	τ (Ps)	R^2
absorbance	1874	--- ^a	---
bleach	1908	--- ^b	---
absorbance	1929	240 ± 79	0.49
bleach	1941	270 ± 63	0.66
absorbance	1960	230 ± 52	0.66
bleach	1982	190 ± 59	0.52

^a Flat time dependence and last two data points make fits nonsensical.^b Flat time dependence and noise make fits nonsensical.

Table 5.

The G values, which are the energy difference compared to the lowest energy configuration, dipole moments, and the band frequencies and intensities for the 4-CO and 3-CO isomers of **2b**, calculated using DFT BP86/TZVP.

isomer	G (kcal/mol)	dipole moment (D)	bandfrequency (cm ⁻¹)	band intensity (arb. units)
2b-1	0.00	2.29	1904	415
			1911	885
			1949	1554
			1963	14
2b-1-1	54.42	1.84	1876	782
			1909	700
			1956	593
2b-1-2	52.61	3.90	1891	704
			1905	1127
			1956	663
2b-2	1.38	4.82	1913	427
			1923	509
			1952	1087
			1981	670
2b-2-1	56.62	5.37	1901	626
			1919	655
			1972	705
2b-2-2	53.31	3.01	1903	1172
			1906	525
			1949	545
2b-2-3	54.86	4.46	1893	419
			1906	1216
			1942	724
2b-2-4	54.49	6.49	1902	114
			1919	1328
			1961	944
2b-4	4.40	7.75	1925	2
			1946	1101
			1957	434
			1996	1089
2b-4-1	56.79	6.91	1912	494
			1926	671
			1972	1037
2b-5	5.32	7.42	1910	198
			1926	815

isomer	G (kcal/mol)	dipole moment (D)	bandfrequency (cm ⁻¹)	band intensity (arb. units)
			1948	1222
			1977	652
2b-5-1	56.56	6.14	1873	641
			1901	1258
			1928	590
2b-5-2	58.06	7.21	1898	333
			1923	1039
			1970	809