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Efficient, Light-Driven Reduction of CO_2 to CO by a Carbon Monoxide Dehydrogenase–CdSe/CdS Nanorod Photosystem

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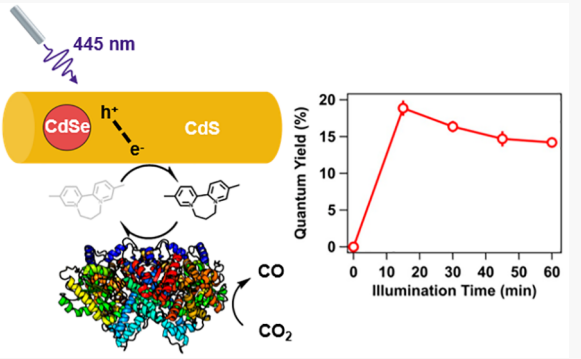
ABSTRACT: The solar conversion of CO_2 to low carbon fuels has been heralded as a potential solution to combat the rise in greenhouse gas emissions. Here we report the first light-driven activation of [NiFe] CODH II from *Carboxydothermus hydrogenoformans* for the reduction of CO_2 to CO. To accomplish this, a hybrid photosystem composed of CODH II and CdSe/CdS dot-in-rod nanocrystals was developed. By incorporating a low-potential redox mediator to assist electron transfer, quantum yields up to 19% and turnover frequencies of 9 s^{-1} were achieved. These results represent a new standard in efficient CO_2 reduction by an enzyme-based photocatalytic systems. Furthermore, successful photoactivation of CODH II allows for future exploration into the enzyme's not fully understood mechanism.



As the effects and scale of greenhouse gas emissions and climate change become increasingly apparent, significant effort has focused on developing new strategies for sustainable energy production.[1,2] A promising approach is the conversion of CO_2 into carbon neutral fuels using solar energy, but the reduction of CO_2 is challenging.[3−5] Current strategies employ photosensitizers coupled with transition metal complexes or heterogeneous semiconductor catalysts, but these systems suffer from poor efficiency, low photostability, and a lack of selectivity to prevent parasitic hydrogen evolution.[6,7] Metalloenzymes such as the [NiFe] carbon monoxide dehydrogenases (CODH) naturally reduce CO_2 with excellent efficiency and selectivity, both traits that are highly desirable for sustainable energy production.[8−10] Significant effort has focused on CODH I from *Carboxydothermus hydrogenoformans* (*ch*CODH I) as a catalyst, in combination with photosensitizers such as CdS nanorods or silver nanoclusters tethered to TiO_2 nanoparticles.[11−13] The overall quantum efficiency achieved with these photosystems was 1.5% at best, significantly lower than reported values for photoenzymatic hydrogen production which ranges from 10 to 80%.[14] In addition, while CODH I has been previously employed for photoconversion of CO_2, CODH II, also from *Carboxydothermus hydrogenoformans*, is the subject of extensive mechanistic studies in part from having a solved crystal structure.[15−17] The two CODH enzymes are related but are distinct in sequence (<60% identity) and native function.[18] Despite being a model CO_2 reducing enzyme, CODH II has yet to be incorporated in a photocatalytic system. Light induced activation of the enzyme will allow excellent control over the catalytic cycle and enable future studies into its cryptic mechanism.[19]

Herein, we report the first photoactivation of *ch*CODH II using mercaptopropionic acid (MPA) capped nanocrystalline seminconductor CdSe/CdS dot-in-rod (DIR) photosensitizers in conjunction with a diquat-based redox mediator for efficient reduction of CO_2 (Figure 1A). A core/shell DIR photosensitizer was selected due to its high efficiency in activating other redox enzymes such as hydrogenases.[14,20] While the conduction band edge of the bulk CdS is low enough (< −700 mV vs SHE at pH 7) for direct reduction of CODH II (−520 mV), a redox mediator was incorporated to improve electron transfer to the enzyme.[21,22]

Low electron availability was speculated to be the rate-limiting step in other CODH-based photosystems, as these enzymes are highly active.[12,13] The use of a redox mediator with a similar DIR photosensitizer successfully improved the quantum yield of photoenzymatic hydrogen production.[14] For the redox mediator, we selected DQ53 (Figure 1A) due to its low reduction potential (−635 mV), which should provide a sufficient driving force for the activation of CODH II.[23] Illumination of the photosystem reduces the mediator and lowers the solution potential, which can be calculated from the concentration of the mediator redox couple and the Nernst equation (Figure 1C). Continuous illumination of the photosystem in the presence of the enzyme that is actively

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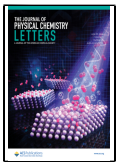

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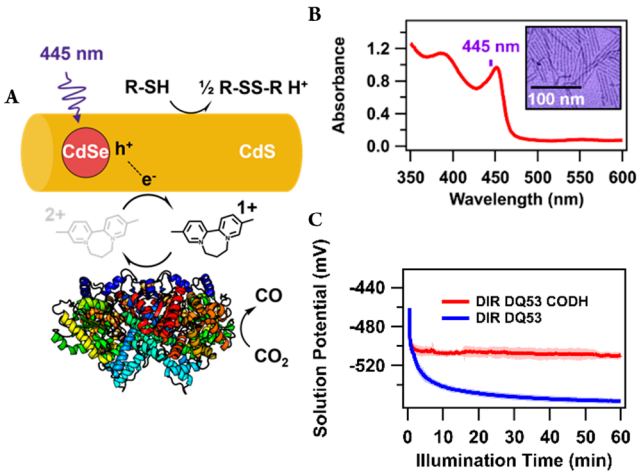

Figure 1. CODH-DIR photosystem. (A) Reaction scheme: illumination of CdSe/CdS dot-in-rod (DIR) generates the exciton that reduces mediator DQ53; electron transfer to CODH II (PDB: 1SUF) initiates enzyme turnover. (B) UV−vis spectrum of the DIR photosensitizer. Inset: TEM image of DIR. (C) Solution potentials of the photosystem generated by the mediator redox couple with or without enzyme, determined using the Nernst equation and mediator concentrations.

consuming the reduced mediator produces a steady state solution potential of approximately −510 mV, near the redox potential of the reversible reduction of CO_2.

CODH II reduces CO_2[24,25] but does not bind the molecule as tightly as CO.[26] CO_2 has been shown to be the substrate of the CODH from *Peptostreptococcus productus* rather than biocarbonate or carbonate.[24,25] Therefore, the headspace of the cuvette was purged with CO_2 to ensure maximum binding and turnover efficiency of the photocatalytic system. In addition, bicarbonate was added to maintain pH stability and to maintain a high concentration of CO_2 substrate. The solution was irradiated with a 445 nm diode laser while stirring continuously. CO production was measured via gas chromatography (GC) through interval (every 15 min) sampling of the headspace gas. A calibration curve of known volumes of CO was used to determine the amount of CO in the headspace. To monitor the reaction in real time, mediator reduction was observed through UV vis spectroscopy.

To confirm that CO production resulted from our photocatalytic system as described, control experiments were performed under identical conditions while removing one component (DIR, mediator, CODH II). No CO was detected in any of these control experiments, thus confirming the importance of incorporating each component in the photocatalytic system (Figure 2A). Direct reduction of CODH by the CdS nanorod is possible as electrostatic charges promote binding between the positively charged enzyme and negatively charged photosensitizer.[12,27] No CO was produced in control experiments without mediator present, however, suggesting that direct electron transfer from the DIR to CODH is inefficient. It is possible that the binding of CODH II to the DIR occurs in a geometry that does not facilitate electron transfer.

Over the course of 1 h, 1.77 μmol of CO was produced (Figure 2B) based on GC analysis. No H_2 or other CO_2 reduction products such as formate were detected in any assay, consistent with the high selectivity for CO production previously observed CODH II.[28,29] The turnover frequency (TOF) observed peaks at 8.6 s^{-1} for the initial time point (15 min) then steadily decreases to 6.4 s^{-1}, indicating that the rate of CO production decreases over time and after subsequent sampling (Figure 2C). This decrease is not likely due to the consumption of reagents (CO_2 and the sacrificial electron donor MPA) because they are present in large excess. Several explanations for this decrease are possible. CODH II is sensitive to minute quantities of oxygen, becoming deactivated in its presence. Reactivation of the enzyme is possible under reducing conditions, albeit with less activity.[17] It is likely that

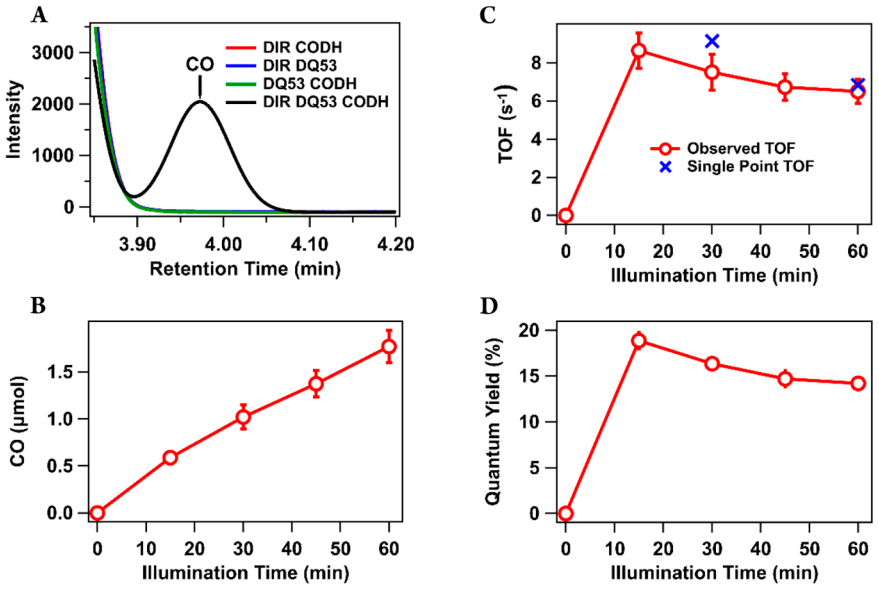

Figure 2. Photoreduction experiments. (A) GC traces of CO produced by the full CODH-DIR photosystem and control experiments each lacking a key component (DIR, mediator, enzyme). (B) Amount of CO produced over time using the CODH-DIR photosystem. (c) Turnover frequencies over time of the CODH-DIR photosystem. Blue × represents single time point TOFs under identical conditions without previous sampling. (d) Quantum yield of the CODH-DIR photosystem using 445 nm light. In all cases, error bars represent standard deviation of three trials.

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sampling of the headspace inadvertently introduces oxygen into the system, thereby lowering the catalytic effectiveness of CODH II. To investigate if the photocatalytic system is being hampered by accidental exposure to oxygen, trials with a single time point were performed. At 30 min, the TOF without previous sampling, or single point TOF, is significantly higher at 9.1 s^{-1} than the TOF of 7.5 s^{-1} after previous sampling (Figure 2c). Furthermore, the TOF of 8.6 s^{-1} at 15 min closely aligns to the 30 min single point TOF. These results demonstrate that headspace sampling lowers activity of the photocatalytic system, likely due to oxygen leakage.

In addition to accidental oxygen exposure, CO_2 reduction by CODH II has been shown to be inhibited by micromolar quantities of CO.[26] Thus, as more CO is produced over the course of an hour, the enzyme activity is likely being decreased by product inhibition. This becomes evident when comparing the TOF of the photocatalytic system without previous sampling vs after multiple sampling events. The single point TOF at 60 min is 6.8 s^{-1}, similar to the TOF of 6.51 s^{-1} after successive sampling. No prior oxygen leakage should occur in the single point experiment; therefore, it is likely the enzyme is being inhibited by the buildup of CO over time. The concentration of aqueous CO over time was calculated using Henry's law to compare against the known inhibition constant. After 60 min, the concentration of CO in solution is ∼30 μM, nearly 6-fold above the previously reported K_I of 5.4 μM.[26]

Apart from simply photoactivating CODH II, we sought to improve the quantum efficiency for enzyme-based, photocatalytic reduction of CO_2. The optimized photosystem gives a maximum quantum yield (QY) for CO production of ∼19% after 15 min of irradiation, which then levels off to ∼15% with subsequent sampling (Figure 2d). These results are an order of magnitude higher than those for other enzyme-based photosystems[13,30] and are comparable to the best molecular-based catalysts for the conversion of CO_2 to CO.[6,7,31−33]

In summary, we report the first photoactivation of CODH II for the reduction of CO_2. Utilizing nanocrystalline semiconductor CdS/CdSe DIR photosensitizers and the diquat based DQ53 redox mediator, micromole quantities of CO were detected over the course of an hour. Despite a steady decrease in CO production due to oxygen contamination and product inhibition, TOFs of ∼9 s^{-1} were observed. Furthermore, a quantum yield of ∼19% was achieved by the photocatalytic system, an increase of more than an order of magnitude when compared to previous reports. Our results demonstrate that CODH II can be photoactivated efficiently, enabling future study of its reaction mechanism by triggering the catalytic cycle with light and probing the catalytic cycle with time-resolved spectroscopic techniques. Overall, the photocatalytic system developed here sets the groundwork for further elucidation of the mechanism of CODH II and [NiFe] CODHs in general.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.jpclett.2c01412.

Materials and methods for synthesis and characterization of DQ53 and nanomaterial, GC method and calibration, photoreduction assay conditions, quantum efficiency calculations, and Supporting Information on photoreduction controls and assay (PDF)

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Notes

The authors declare no competing financial interest.

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