Supporting Information

Enhanced photocatalytic CO_2 reduction on biomineralized CdS via the electron conduit in bacteria

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Experimental Section

■ Chemicals and materials

S. oneidensis oneidensis MR-1 (SW) was purchased from American Type Culture Collection (Manassas, VA, USA). Luria-Bertani (LB) broth was purchased from Nanjing Reagent Company (Nanjing, China). Sodium thiosulfate pentahydrate, cadmium chloride, potassium bicarbonate ($KHCO_3$, ≥99.5%), sodium chloride, and magnesium sulfate were purchased from Sinopharm Chemical Reagent Co., Ltd. HEPES (4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid) and DMPO (5,5-dimethyl-1-pyrroline-N-oxide) was purchased from Aladdin (Shanghai, China). Ultrapure water (18.2 MΩ in resistivity, Mili-Q, Millipore) was used for all the experiments. All media and solutions were sterilized before use.

■ Instruments

The surface morphology and element composition of the samples were characterized by scanning electron microscopy with element mapping images (SEM, JEOL JSM-7800F, with an acceleration voltage of 5 kV), transmission electron microscopy (TEM, JEOL JEM 1400), and high-resolution TEM with element mapping images (HRTEM, JEM-2800). The XRD spectra were collected on Bruker D8 Advance X-Ray Powder Diffractometer. The UV–Vis diffused reflectance spectra (UV-vis DRS) of the samples were collected by a UV–Vis–near–IR spectrometer (UV3600-MPC3100) with 100% $BaSO_4$ as reference. The UV-Vis spectra were collected by Shimazu UV-3700 ultraviolet spectrophotometer. Fourier transforms infrared (FT-IR)

spectra were measured by a Nicolet 6700 spectrophotometer (Nicolet Co., USA). X-ray photoelectron spectroscopy (XPS) was analyzed by an ESCALAB250Xi spectrometer (Thermo Fisher Scientific Co., USA). Electron paramagnetic resonance (EPR) spectra were recorded by ESP-300E spectrometer (Bruker, Germany), and 5,5-dimethyl-1-pyrroline-N-oxide (DMPO) is employed as the spin trapping agent for $SO_3^{\bullet-}$. The 1H NMR spectroscopy was obtained with a 400 MHz liquid nuclear magnetic resonance spectrometer (NMR, Bruker advance III). The elemental content of the samples was measured by an inductively coupled plasma optical emission spectrometer (ICP-OES, ICPE-9810, PerkinElmer Co., U.S.A.). Confocal microscope images (CLSM) were taken through a TCS SP8 microscope (Leica, Germany).

■ *S. oneidensis* MR-1 cultivation and CdS nanoparticles biosynthesis

S. oneidensis MR-1 strains were grown in LB broth at 30 ℃ under 150 rpm shaking overnight to the late stationary phase. The bacterial cells were collected by centrifugation under 6000 rpm for 5 min and washed twice with M9 solution (85.5 mM NaCl and 10.0 mM $MgSO_4$). The cells were then incubated in 15 mL of an anaerobic mineral medium containing 1 mM $Na_2S_2O_3$ and 20 mM sodium lactate, with the initial OD_{600} of 0.5 (30 °C and 150 rpm rotation). After 12 h of incubation, 1 mM $CdCl_2$ was added, and cultured for a further 6 h to form CdS nanoparticles. The anaerobic mineral medium contains 50 mM HEPES, and 50 mM NaCl, which was purged with pure N_2 for 30 min to achieve an anaerobic condition. The obtained product was *S. oneidensis* MR-1@CdS (SW@CdS). And the amount of CdS was tested to be 1.50±0.05 mg

according to three ICP-OES analyses.

■ Preparation of CdS and SW@CdS

14 mg of $CdCl_2 \cdot 2.5H_2O$ was dissolved in 95 ml of deionized water with stirring (rotation rate of 500r/min). Then $Na_2S \cdot 9H_2O$ aqueous solution (12mg $Na_2S \cdot 9H_2O$ dissolved in 5 mL deionized water) was slowly dropped into the above solution. The mixture was centrifuged and washed several times with deionized water until the unreacted cadmium and sulfur ions were removed. Finally, CdS NPs were obtained after vacuum drying at 60℃. And SW-CdS was obtained by mixing 1.5 mg CdS NPs with SW solution.

■ SEM and TEM characterizations of SW/ SW-CdS/ SW@CdS

The samples were collected at 6000 rpm for 5 min, then fixed in 2.5 vol % glutaraldehyde at 4 ℃ for 4 h. The fixed cells were then dehydrated with different concentration gradients of ethanol solution, with the volume ratios of 25%, 50%, 75%, 90%, and 100% in sequence, each for 10 min. Finally, ITO and copper grids were dipped into the ethanol solution, and dried under ambient conditions for SEM and TEM testing, respectively. Before the SEM characterization, the samples were coated with Au.

■ ICP-OES analysis of the extracellular CdS amounts

15mL samples were collected (15000g, 10 min) and washed three times with

deionized water to remove the adsorbate. Then 200 μL samples were digested with 2 mL aqua regia (HNO_3/HCl =1:4) overnight. After dilution to 10 mL, the Cd contents in the digestion solutions were then quantified by ICP-OES following standard methods.

■ Viability test

Cell viability was studied by the LIVE/DEAD BacLight bacterial viability kit. Cells with intact or compromised membranes were differentiated based on the differential permeability of fluorescent dye. 800 μL concentrated aqueous solution of SW@CdS was pipetted into separated wells of a 96-well flat-bottom microplate, then 40 μL of the dye mixture (SYTO 9/propidium iodide, 1:1) was added to each well. After incubation in the dark for 20 min, the samples were imaged through a Carl Zeiss LSM 780 Axiovert inverted confocal microscope with an Achroplan oil immersion lens. SYTO 9 signals were detected using an excitation wavelength of 488 nm and an emission wavelength of 503 nm. The fluorescence agent (propidium iodide) was excited at 543 nm and the emission was collected through a long-band pass filter at 605 nm.

■ Photocatalytic activity tests

The washed SW@ CdS mixture was injected into the photocatalytic medium for CO_2 reduction. The medium contains 15 mL of M9 solution, 50 mM ~~K~~HCO_3, and 25 mM sodium sulfite in a photocatalytic reactor. The light source was a 300 W xenon lamp, the samples were irradiated at a distance of 18 cm from the lamp, and the optical power density was controlled at 100 mW m^{-2}. Before the Xe lamp irradiation, the

system needed to be purged with pure N_2 for 30 min and CO_2 for 15 min. During testing, the photocatalytic solution was stirred and CO_2 was purged (8 sccm). Online gas chromatography was used to determine the gaseous products (Aligent 7890B GC). The liquid products were monitored by ^1H NMR.

■ Photocurrent measurements and Mott-Schottky curves

Photocurrent measurements and Mott-Schottky curves of prepared catalysts were performed using a CHI760E electrochemical workstation (Chenhua, Shanghai) in conjunction with a three-electrode system. The counter electrode was a Pt foil, the reference electrode was an Ag/AgCl electrode, and the working electrode was an FTO conductive glass with a conductive side coated with a thin sample film. The prepared photocatalysts were mixed with 0.25% Nafion and ethanol to form a slurry, which was uniformly coated on the FTO conductive glass. The electrolyte was a 15 mL, 0.5 M Na_2SO_4 aqueous solution, and a 300 W Xe lamp was used to simulate sunlight.

■ Cyclic voltammetry curve

In a three-electrode system, cyclic voltammetry (CV) analyses were carried out using a CHI 760E electrochemical workstation (CH Instrument, China). The reference electrode was Ag/AgCl (+0.197 vs. SHE, according to the manufacturer), and the working electrode was carbon cloth (CC) with SW@CdS. CV curves from -0.8 to 0.2 V were performed at a scanning rate of 10 mV/s with sodium lactate as the carbon source. All voltages presented here were measured concerning Ag/AgCl.

■ Anaerobic environmental testing

We supplemented the SW or SW@ CdS groups with 20 mM sodium lactate as an electron donor and 0.1 % resazurin as an oxygen indicator. To compare, the group was also prepared but without SW. After a few seconds, only the SW or SW@ CdS groups showed a color change.

■ Heat-Treatment

To investigate whether the increased formate generation was linked to bacterial viability, the hybrid system was heat-treated following the method reported. Specifically, the SW@CdS hybrid was collected after CdS synthesis, washed, and heat-treated at 120 °C for 20 minutes to inactivate the *S. oneidensis* MR-1.

■ Quantification of ATP Level

The cells were centrifuged at 6000 rpm at 4ºC for 5 minutes and discarded the supernatant, and then lysed by the cell lysis buffer. After lysis, the cells were centrifuged at 12000 rpm at 4ºC for 5 minutes and took the supernatant for subsequent determination. The level of ATP was determined by the luciferase method through ATP Assay Kit (Beyotime Biotechnology), and the results of the ATP level were normalized by corresponding protein contents.

■ Quantification of $NADH/NAD^+$ Level

The cells were centrifuged at 6000 rpm for 5 minutes and then aspirated the

medium-added ice-cooled NADH/NAD^+ extract to lyse the cells. Then the cells were centrifuged at 12000 rpm at 4ºC for 5 minutes, and the supernatants were as the samples to be tested. The level of NADH/NAD^+ was determined through the WST-8 reaction by NADH/NAD^+ Assay Kit with WST-8 (Beyotime Biotechnology).

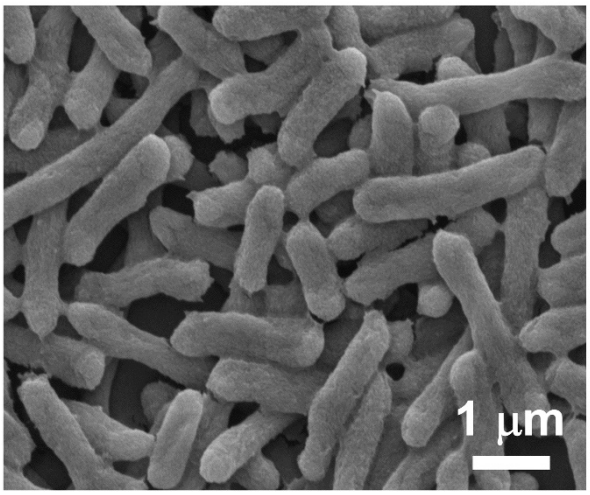


Fig. S1 SEM image of SW.

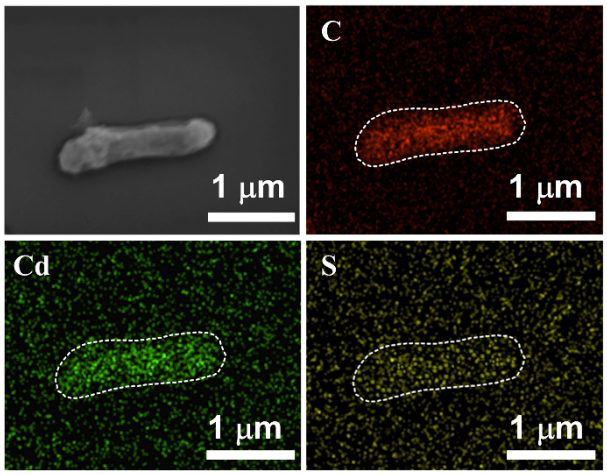


Fig. S2 SEM Energy-dispersive X-ray spectroscopy (EDS) elemental maps of SW@CdS.

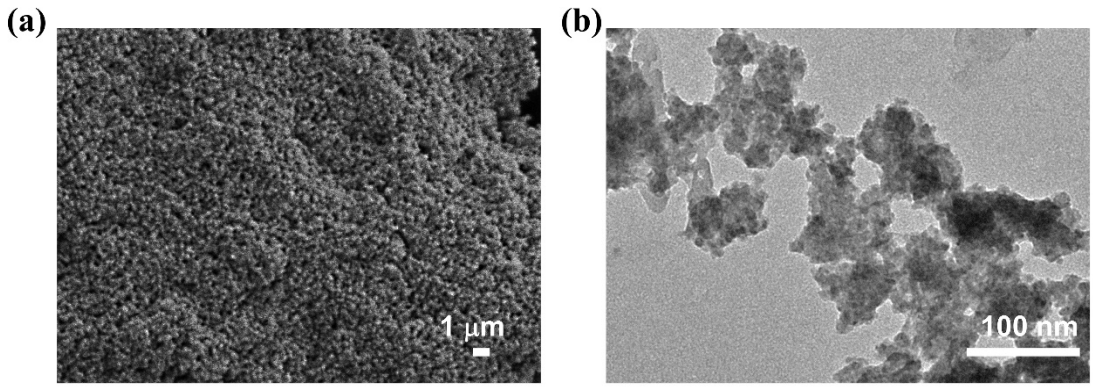


Fig. S3 SEM and TEM images of chemosynthetic CdS.

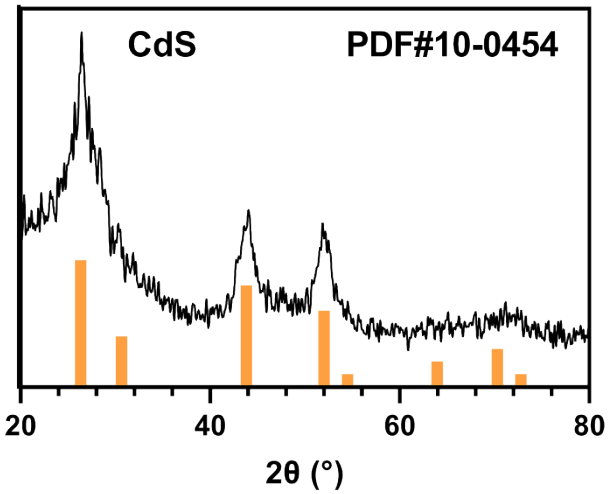


Fig. S4 XRD pattern of chemosynthetic CdS.

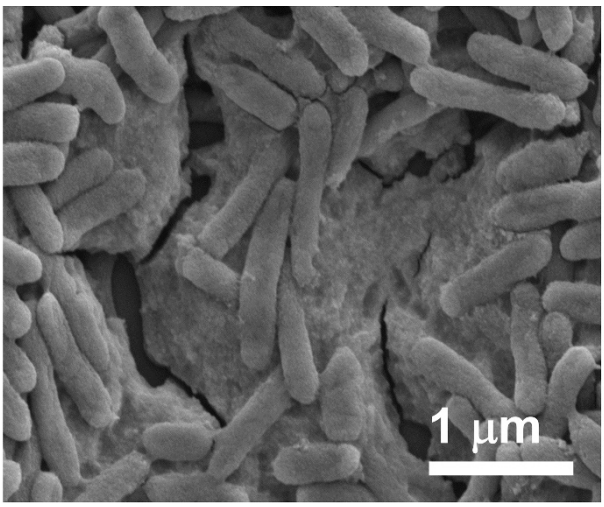


Fig. S5 SEM image of SW-CdS.

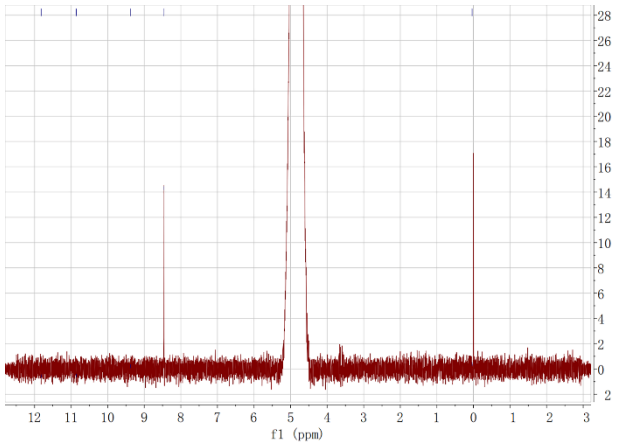


Fig. S6 A representative ^{1}H-NMR spectrum was obtained from the photoreaction solution for the SW@CdS/SO_3^{2-} system. TMSP was used as an internal standard for the quantification of liquid products.

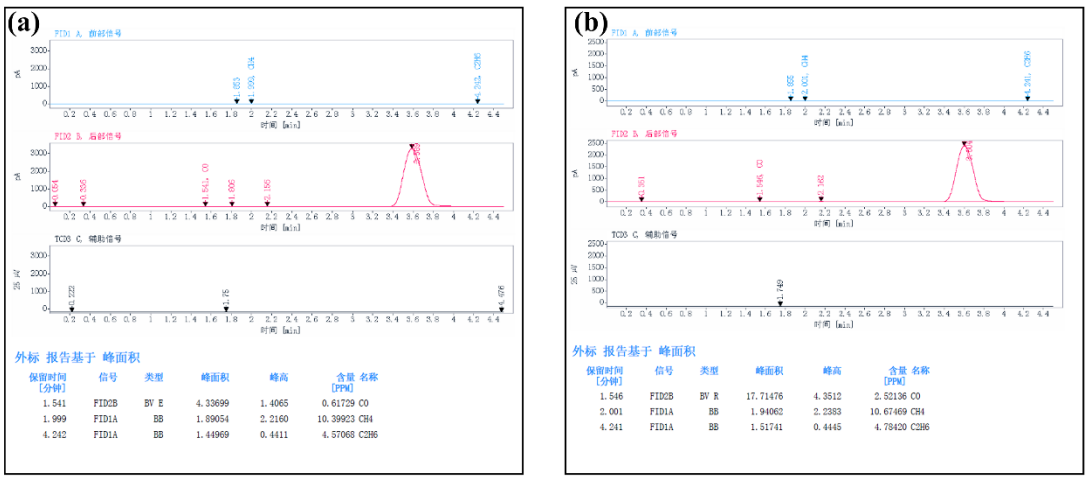


Fig. S7 The GC test data were collected before and during the CO_2 photoreduction for the SW@CdS/SO_3^{2-} system.

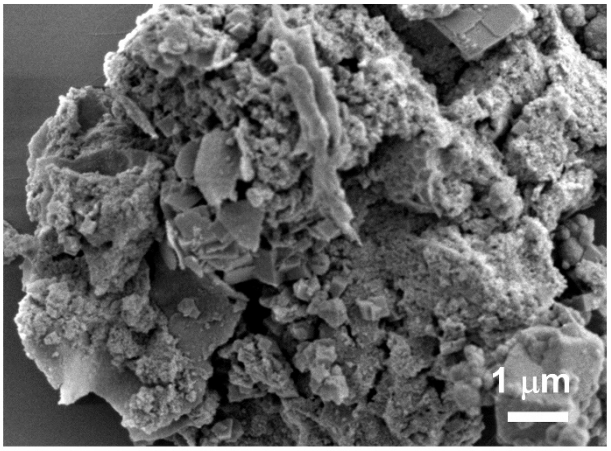


Fig. S8 SEM image of chemosynthetic CdS after 4-h illumination.

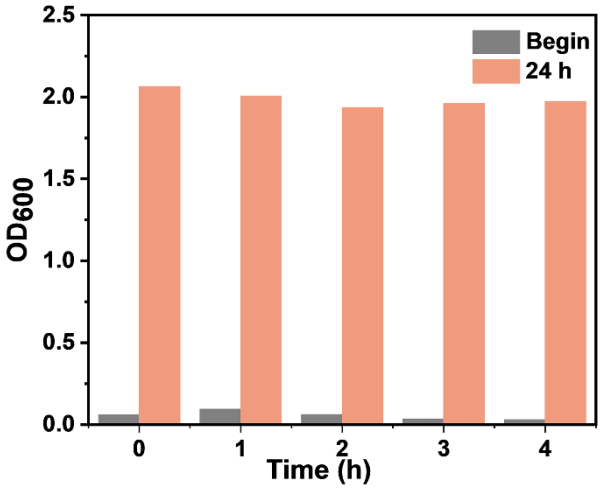


Fig. S9 The SW activity test during the 4-h illumination. For every hour, 500μL of solution from the photoreaction system was added to 15ml LB and cultured for 24 hours at 30 ℃ with 150 rpm shaking. OD_{600} was measured before and after culture.

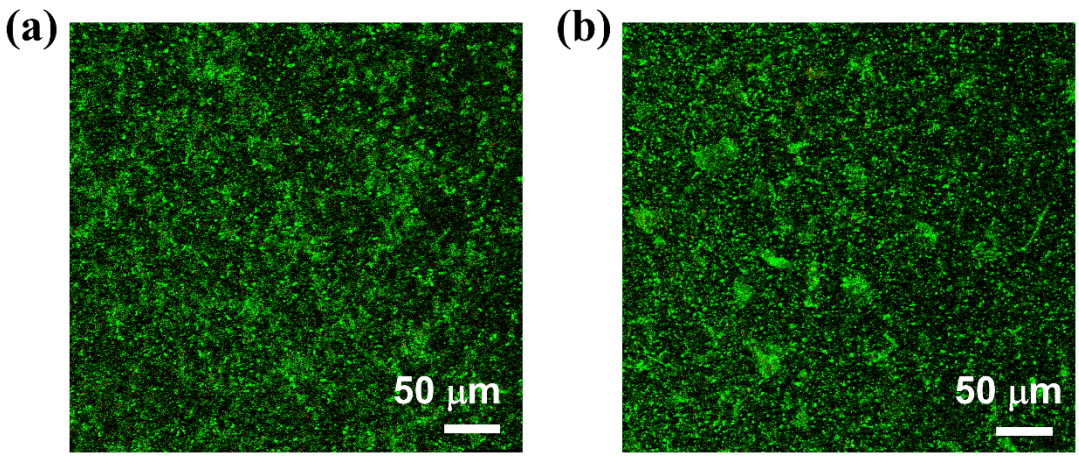


Fig. S10 CLSM images of *S. oneidensis* MR-1 stained with the Live/Dead BacLight kit were used to examine the viability of SW@CdS (a) before and (b) after a four-hour photoreaction.

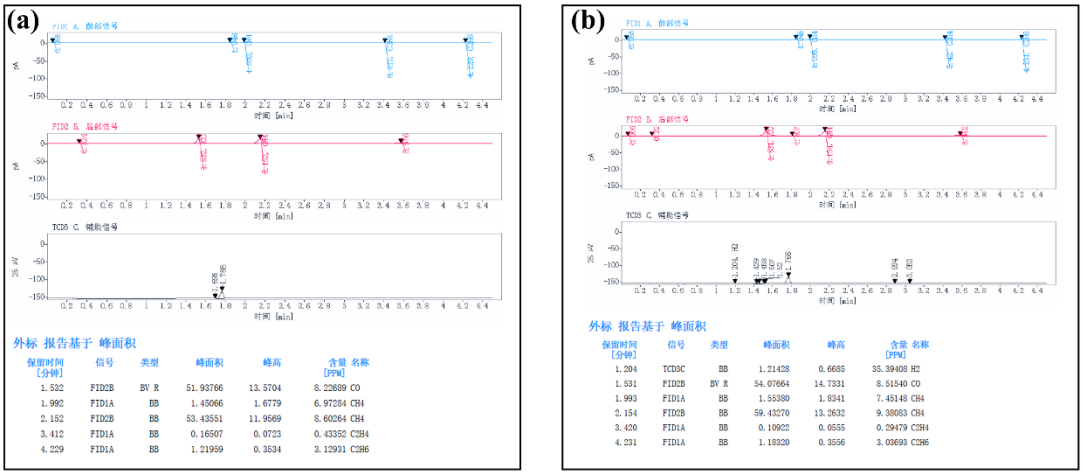


Fig. S11 The GC test data was collected before (a) and during (b) the photoreaction when using sodium lactate as an electron donor.

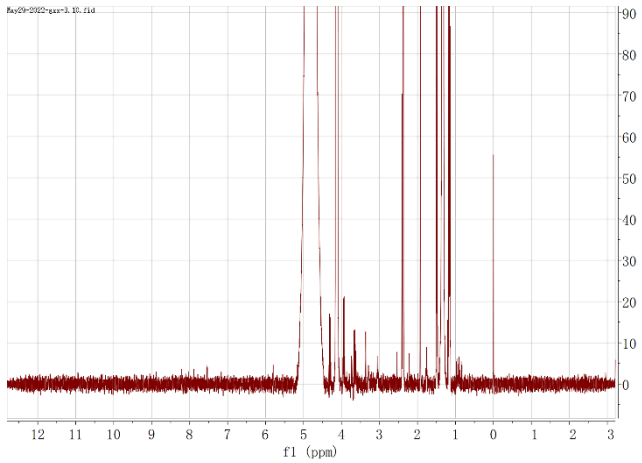


Fig. S12 The ^{1}H NMR spectrum of the SW@CdS/SO_3^{2-} system after the photoreaction when using sodium lactate as an electron donor.

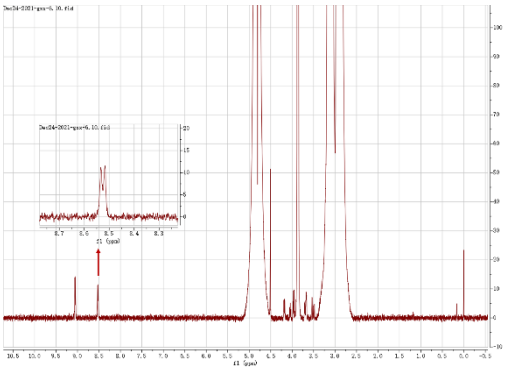


Fig. S13 ^{1}H NMR spectrum of the mechanism verification experiment after the reaction. The dithionite-driven MR-1-dependent reduction of CO_2 was carried out in a glass vial and it contained methyl viologen (0.5 mM) and dithionite (0.8 mg/mL; ca. 4.6 mM) with MR-1 (OD600≈0.5). There were two double peaks at 8.53 and 9.05, which belong to MV.

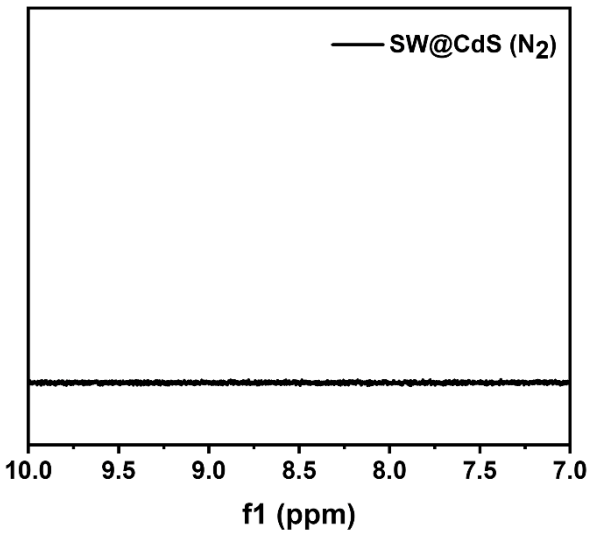


Fig. S14 The ^{1}H NMR spectrum of the SW@CdS/SO_3^{2-} system under a nitrogen atmosphere after four hours of illumination.

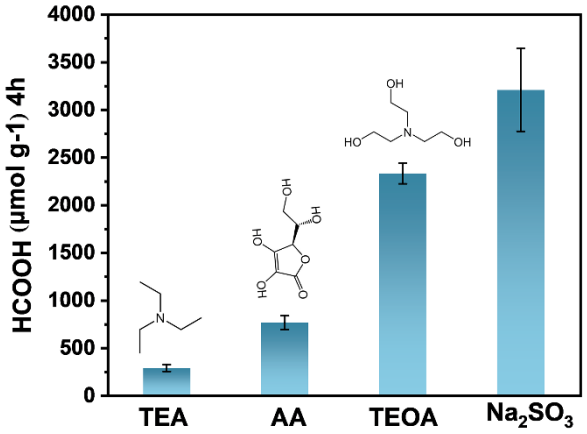


Fig. S15 For the SW@CdS photocatalyst, formate yield with triethylamine (TEA), ascorbic acid (AA), triethanolamine (TEOA), or Na_2SO_3 in solution.

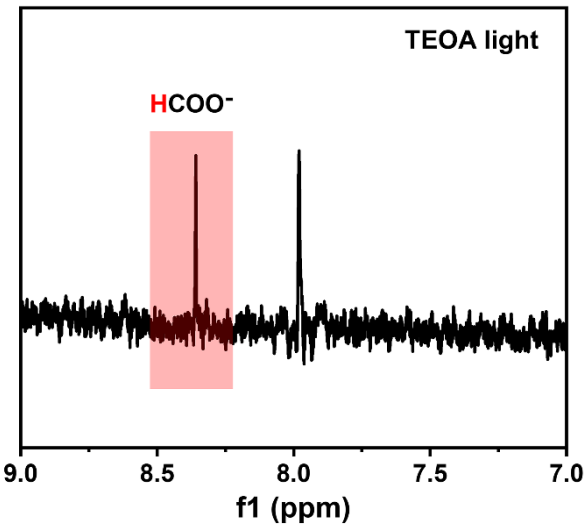


Fig. S16 The ^{1}H-NMR spectrum of the TEOA aqueous solution was obtained after four hours of illumination.

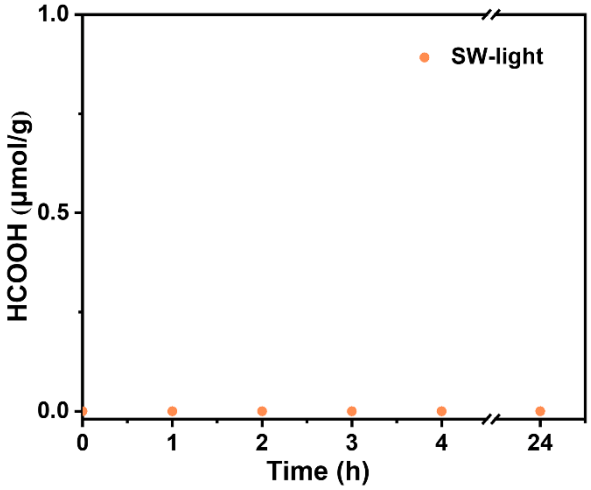


Fig. S17 The formate yield was measured over 24 hours when only SW was present in the photoreaction solution.

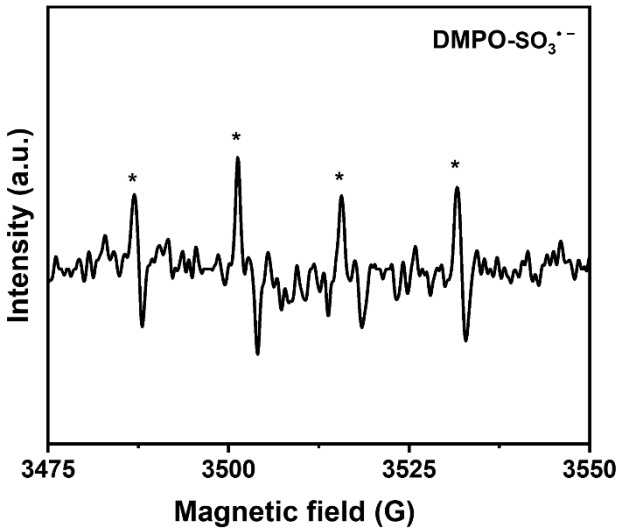


Fig. S18 EPR responses of the DMPO-$SO_3^{\bullet-}$ spin adduct by testing the suspension of SW@CdS/SO_3^{2-} system.

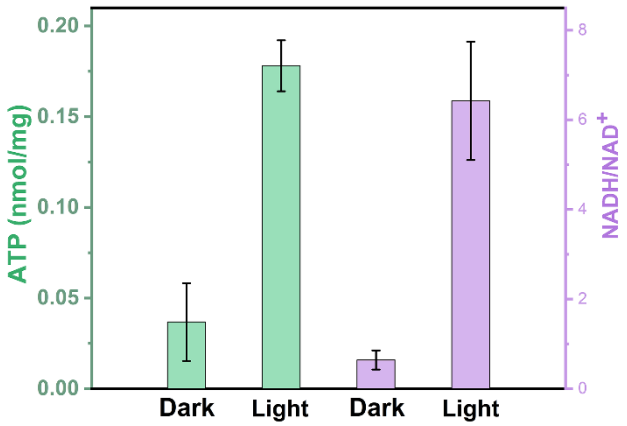


Fig. S19 The ATP level and NADH/NAD^+ ratio of the SW@CdS/SO_3^{2-} system were measured after four hours under dark or illumination.

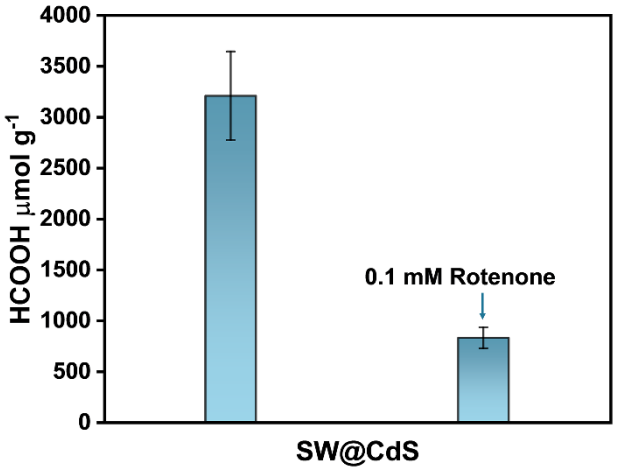


Fig. S20 HCOOH production by SW@CdS/SO_3^{2-} with or without 0.1 mM rotenone.

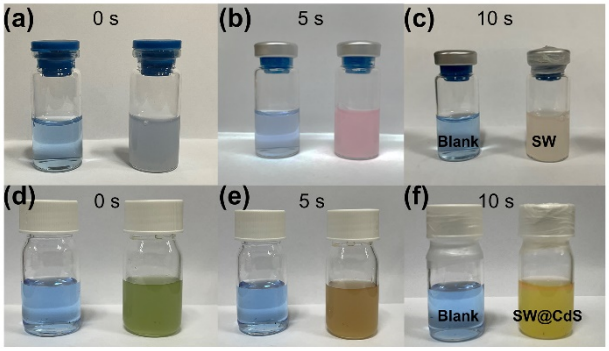


Fig. S21 Anaerobic environmental testing. When SW was added, the blue color (a) faded to pink (b) and then colorless (c) in seconds. As the attached video records, when SW@CdS was added, the cyan color (d) faded to brown (e) and then yellow (f) in seconds.

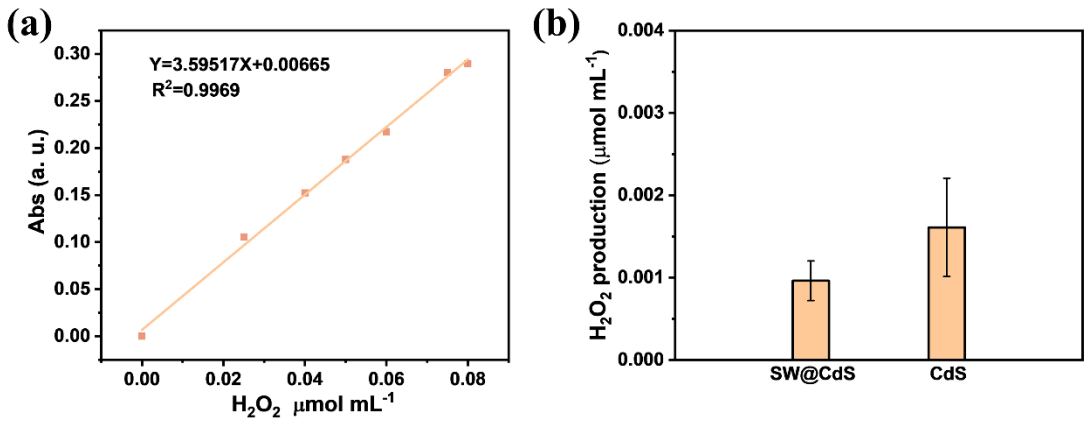
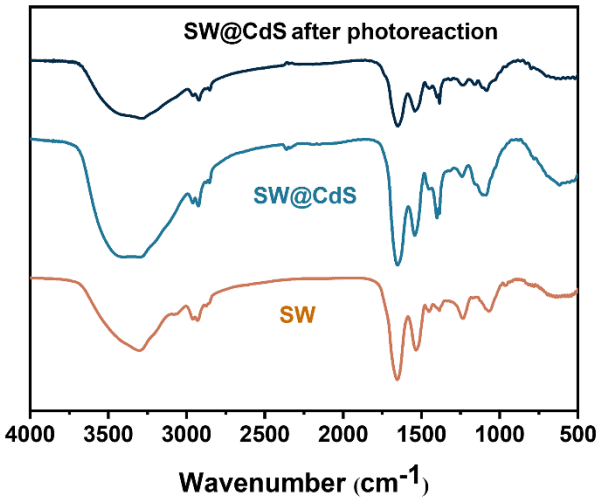


Fig. S22 (a) The linear fitting formula of standard H_2O_2 concentration. (b) The content of H_2O_2 production for SW@CdS and CdS under illumination. Irradiation time was 4 hours. (SO_3^{2-} was not added, considering the reaction between SO_3^{2-} and



H_2O_2)

Fig. S23 Fourier transforms infrared (FT-IR) spectra of SW, SW@CdS, and SW@CdS after 4-h photoreaction.

Table S1. Representative research in photocatalytic CO_2 reduction to HCOOH.

Photocatalyst	Light source	Reaction medium	Reaction condition	Products	HCOOH Selectivity	HCOOH Yield (μmol g^{-1} h^{-1})	Reference
CdS NPs	Hg arc lamps (250<λ<580 nm)	H_2O	0.1 MPa	HCOOH	100%	1.0	1
NH_2-Cu-MOF	300 W Xenon lamp	H_2O, Na_2SO_3	0.1 MPa	HCOOH, CO, CH_4	92.74%	138.7	2
Nb_2O_5-CuO	UVC lamp	$Na_2C_2O_4$ in H_2O	0.1 MPa	HCOOH, CH_4, CH_3COOH	25.9%	3	3
CuO/Ag/UiO-66	300 W Xe lamp	H_2O	0.3 MPa	HCOOH, CH_3OH	80.0%	60	4
Ce-MOF/ Bi_2MoO_6	300 W Xe lamp	H_2O	0.6 MPa	HCOOH, CH_4, CH_3OH	11.3%	74	5
PCN-222-Ni @UiO-67-NH_2	300 W Xe lamp (λ ≥ 420 nm)	H_2O	0.6 MPa	HCOOH, CO, CH_4	73.2%	146	6
V_{Zn}-rich ZnS	300 W Xe lamp	$KHCO_3$, K_2SO_3	0.1 MPa	HCOOH	85%	60	7
Ni-doped ZnS	300 W Xenon lamp (λ ≥ 420 nm)	$KHCO_3$, K_2SO_3	0.1 MPa	HCOOH, CO	94.69%	4389.1	8
Cd/ZnS: Cu	AM 1.5	$KHCO_3$, K_2SO_3	0.1 MPa	HCOOH	26.5%	3.2	9
Nb-doped TiO_2/g-C_3N_4	30 W white bulbs	H_2O	0.1 MPa	HCOOH, CO, CH_4	41.55%	698	10
TiO_2/CoPc	500 W tungsten–halogen lamp	H_2O, Na_2SO_3	0.1 MPa	HCOOH	82.35%	51.6	11
SW@MR-1	300 W Xe lamp (λ=350-780 nm)	H_2O, Na_2SO_3	0.1 MPa	HCOOH	100%	2650	This work
Photocatalyst	**Light source**	**Reaction medium**	**Reaction conditions**	**Products**	**HCOOH Selectivity**	**HCOOH Yield (μmol h^{-1})**	**Reference**

Engineered-*E coli*-CdS	Xenon lamp	VC in H_2O	0.1 MPa	HCOOH	100%	0.525	12
Eosin Y-MR-1	photosynthetic growth lamp	TEOA in H_2O	0.1 MPa	HCOOH, H_2	72.8%	0.039	13
SW@MR-1	300 W Xe lamp (λ=350-780 nm)	SO_3^{2-} in H_2O	0.1 MPa	HCOOH	100%	3.975	This work

References

1 J. T. S. Irvine, B. R. Eggins and J. Grimshaw, *Sol. Energy*, 1990, **45**, 27–33.

2 Q. Zhu, Y. Cao, Y. Tao, T. Li, Y. Zhang, H. Shang, J. Song and G. Li, *J. CO2 Util.,* 2021, **54**, 101781.

3 A. E. Nogueira, G. T. S. T. Silva, J. A. Oliveira, O. F. Lopes, J. A. Torres, M. Carmo and C. Ribeiro, *ACS Appl. Energy Mater.*, 2020, **3**, 7629–7636.

4 M. Zhang, X. Wang, X. Qi, H. Guo, L. Liu, Q. Zhao and W. Cui, *J. Catal.*, 2022, **413**, 31–47.

5 W. Cai, X. Yu, Y. Cao, C. Hu, Y. Wang, Y. Zhao and Y. Bu, *J. Environ. Chem. Eng.*2022, **10**, 107461.

6 H. Huang, Z. Fang, R. Wang, L. Li, M. Khanpour, T. Liu and R. Cao, *Small*, 2022, **18**, 2200407.

7 H. Pang, X. Meng, P. Li, K. Chang, W. Zhou, X. Wang, X. Zhang, W. Jevasuwan, N. Fukata, D. Wang and J. Ye, *ACS Energy Lett.*, 2019, **4**, 1387–1393.

8 H. Pang, X. Meng, H. Song, W. Zhou, G. Yang, H. Zhang, Y. Izumi, T. Takei, W. Jewasuwan, N. Fukata and J. Ye, *Appl. Catal., B,* 2019, **244**, 1013–1020.

9 X. Meng, G. Zuo, P. Zong, H. Pang, J. Ren, X. Zeng, S. Liu, Y. Shen, W. Zhou and J. Ye, *Appl. Catal., B,* 2018, **237**, 68–73.

10 N. T. Thanh Truc, L. Giang Bach, N. Thi Hanh, T.-D. Pham, N. Thi Phuong Le Chi, D. T. Tran, M. V. Nguyen and V. N. Nguyen, *J. Colloid Interface Sci.* 2019, **540**, 1–8.

11 S. Liu, Z. Zhao and Z. Wang, *Photochem. Photobiol. Sci.*, 2007, **6**, 695–700.

12 X. Wang, J. Zhang, K. Li, B. An, Y. Wang and C. Zhong, *Sci. Adv.*, 2022, **8**, eabm7665.

13 S. F. Rowe, G. Le Gall, E. V. Ainsworth, J. A. Davies, C. W. J. Lockwood, L. Shi, A. Elliston, I. N. Roberts, K. W. Waldron, D. J. Richardson, T. A. Clarke, L. J. C. Jeuken, E. Reisner and J. N. Butt, *ACS Catal.*, 2017, **7**, 7558–7566.