Nanowire–Bacteria Hybrids for Unassisted Solar Carbon Dioxide Fixation to Value-Added Chemicals

Chong Liu,†⊥ Joseph J. Gallagher,‡ Kelsey K. Sakimoto,† Eva M. Nichols,† Christopher J. Chang,*,†‡§# Michelle C. Y. Chang,*,†‡§# and Peidong Yang*†⊥§#

‡Department of Chemistry, †Department of Molecular and Cell Biology, ‡Howard Hughes Medical Institute, and §Department of Materials Science and Engineering, University of California, Berkeley, Berkeley, California 94720, United States

© Materials Sciences Division and §Chemical Sciences Division, Lawrence Berkeley National Laboratory, Berkeley, California 94720, United States

⊥Kavli Energy NanoSciences Institute, Berkeley, California 94720, United States

Supporting Information

ABSTRACT: Direct solar-powered production of value-added chemicals from CO2 and H2O, a process that mimics natural photosynthesis, is of fundamental and practical interest. In natural photosynthesis, CO2 is first reduced to common biochemical building blocks using solar energy, which are subsequently used for the synthesis of the complex mixture of molecular products that form biomass. Here we report an artificial photosynthetic scheme that functions via a similar two-step process by developing a biocompatible light-capturing nanowire array that enables a direct interface with microbial systems. As a proof of principle, we demonstrate that a hybrid semiconductor nanowire–bacteria system can reduce CO2 at neutral pH to a wide array of chemical targets, such as fuels, polymers, and complex pharmaceutical precursors, using only solar energy input. The high-surface-area silicon nanowire array harvests light energy to provide reducing equivalents to the anaerobic bacterium, Sporomusa ovata, for the photoelectrochemical production of acetic acid under aerobic conditions (21% O2) with low overpotential (η < 200 mV), high Faradaic efficiency (up to 90%), and long-term stability (up to 200 h). The resulting acetate (∼6 g/L) can be activated to acetyl coenzyme A (acetyl-CoA) by genetically engineered Escherichia coli and used as a building block for a variety of value-added chemicals, such as n-butanol, polyhydroxybutyrate (PHB) polymer, and three different isopenicol natural products. As such, interfacing biocompatible solid-state nanodevices with living systems provides a starting point for developing a programmable system of chemical synthesis entirely powered by sunlight.

KEYWORDS: Nanowires, artificial photosynthesis, bacteria, carbon dioxide fixation

N

Received: March 31, 2015
materials science and biology separates the demanding dual requirements for light-capture efficiency and catalytic activity, respectively, and provides a route to bridge efficient solar conversion in robust solid-state devices with the broad synthetic capabilities of living cells. This artificial photosynthesis strategy is distinct from the active area of microbial electrosynthesis, in that the nanomaterials carry out both light-harvesting and delivery of reducing equivalents. Here, as a first step, we demonstrate a stand-alone, solar-powered system composed of silicon (Si) and titanium dioxide (TiO₂) nanowire arrays as the light-capturing units and S. ovata as the cellular catalyst, which can effectively reduce CO₂ under mild conditions (e.g., aerobic atmosphere, neutral pH, and temperatures under 30 °C) and produce acetate for up to 200 h under simulated sunlight, with an energy-conversion efficiency of up to 0.38%. Such an system, where CO₂-reducing bacteria are directly interfaced with a photoactive semiconductor, to the best of our knowledge, represents the first example of microbial photoelectrosynthesis, which is different from conventional microbial electrosynthesis wherein microbes do not directly interact with light-absorbing devices. The nanowire—bacteria hybrids possess a high reaction rate of CO₂ reduction, and the presence of the nanowire array creates a local anaerobic environment that allows strict anaerobes to continue CO₂ reduction aerobically (21% O₂), which is important for practical application. Finally, the acetate intermediate represents a biosynthetic precursor to a wide variety of potential fine and
CO2 reduction to value-added fuels, chemicals, and materials, providing a versatile and amenable platform for solar-driven production of a variety of products with minimal modification.

The system starts by interfacing light-absorbing Si nanowire arrays with an acetogenic organism, S. ovata. Si nanowire arrays capture light for efficient solar energy conversion and provide high surface areas to interface with catalysts.22,26 The strictly anaerobic homoacetogen S. ovata metabolizes CO2 via the energy-efficient Wood-Ljungdahl pathway and has been reported to accept electrons from graphite electrodes to reduce CO2 to acetate under continuous sparging with 20% CO2/80% N2 with an overpotential η less than 200 mV at 0 V vs reversible hydrogen electrode (RHE) (see Methods), similar as reported in the literature.28 Additionally the Tafel slope of bacterial catalyzed CO2 reduction is distinctly different from that of abiotic proton reduction, implying different reaction mechanisms (n = 2). On average, each cell could produce (1.1 ± 0.3) × 106 molecules of acetate every second or ca. 1012 molecules of acetate over the course of about 5 days at ~0.2 V vs RHE (Supporting Information Figure 2e,d), comparable with its intrinsic rate of acetogenic metabolism (Supporting Information Note). Such nano-biohybrids, which operate at ambient temperature, possess a volumetric reaction rate of ca. 2 mol m−3 s−1, comparable to the rates in conventional gas phase catalysts (0.1−10 mol m−3 s−1) that require much higher temperatures (higher than 100 °C).32 It also corresponds to ca. 8 electrons s−1 nm−2 across the semiconductor/electrolyte interface (at ~0.2 V vs RHE), suitable to couple with efficient solar devices at 10 mA/cm2 when integrated into a high-surface-area electrode.1,2,24,36

The high reaction rate of the nanowire−bacteria hybrids was capable of reducing CO2 to acetate under continuous sparging with 20% CO2/80% N2, without apparent mass transport issues (Figure 2b,c). The cell loading of S. ovata within the nanowire array is 4.4 ± 1.0 times of that observed on a planar Si electrode (1.4 ± 0.1 vs 0.32 ± 0.07 cells per geometric μm2, n = 4) (Supporting Information Figure 2d), revealing increased contact interfaces between bacteria and electrodes in this high-surface-area platform. The proposed half-reaction of CO2 reduction is

\[
2\text{CO}_2 + 7\text{H}^+ + 8\text{e}^- \rightarrow \text{CH}_3\text{COO}^- + 2\text{H}_2\text{O}
\]

\[
E^0 = +0.143 \text{V vs RHE}
\]

From a classic electrochemical analysis without solar illumination (Figure 2d), the nanowire−bacteria hybrids were capable of reducing CO2 to acetate under continuous sparging with 20% CO2/80% N2 with an overpotential η less than 200 mV at 0 V vs reversible hydrogen electrode (RHE) (see Methods), similar as reported in the literature.28 Additionally the Tafel slope of bacterial catalyzed CO2 reduction is distinctly different from that of abiotic proton reduction, implying different reaction mechanisms (n = 2). On average, each cell could produce (1.1 ± 0.3) × 106 molecules of acetate every second or ca. 1012 molecules of acetate over the course of about 5 days at ~0.2 V vs RHE (Supporting Information Figure 2c,d), comparable with its intrinsic rate of acetogenic metabolism (Supporting Information Note). Such nano-biohybrids, which operate at ambient temperature, possess a volumetric reaction rate of ca. 2 mol m−3 s−1, comparable to the rates in conventional gas phase catalysts (0.1−10 mol m−3 s−1) that require much higher temperatures (higher than 100 °C).32 It also corresponds to ca. 8 electrons s−1 nm−2 across the semiconductor/electrolyte interface (at ~0.2 V vs RHE), suitable to couple with efficient solar devices at 10 mA/cm2 when integrated into a high-surface-area electrode.1,2,24,36
feedstock (Figure 1a). Analogous to the two photosystems found in nature,22,25,26 Si and TiO₂ nanowires were applied as two robust semiconductor light-absorbers to provide the thermodynamic driving force for CO₂ reduction.22 Specifically, an ion-conductive membrane was placed in between the two electrodes to separate reaction products, and the water-oxidizing TiO₂ nanowire electrode22 was placed in front of the nanowire–bacteria composite to absorb UV light and prevent possible bacterial photodamage (Figure 1a and Supporting Information Figure 1b). Without any additional energy input, nonzero photocurrent was observed (light chopping experiment in Figure 2e), and CO₂ reduction to acetate was confirmed (see Methods). The overall system produced about 0.3 mA/cm² photocurrent under simulated sunlight (AM 1.5G, 100 mW/cm²) and was stable for more than 120 h (Figure 2e, and Supporting Information Figure 3). Starting from an electrolyte free of organic compounds, acetic acid was steadily produced with a product selectivity (Faradaic efficiency) of 86 ± 9% (n = 6) (Figure 2e). The peak photocurrent reached 0.35 mA/cm², which corresponds to an energy conversion efficiency of 0.38% for acetic acid production (requires 1.08 V thermodynamically, see Methods). The acetic acid titers were ca. 1.2 g/L (20 mM) within 5 days and could reach over 6 g/L (ca. 100 mM) in M9-MOPS minimal medium (see Methods and Supporting Information Note). In separate control experiments, no acetic acid was detected without incorporation of S. ovata, and an isotope-labeling experiment proves that the acetate is produced from CO₂ (Supporting Information Figure 4). These results highlight our ability to upgrade CO₂ to chemicals beyond one-carbon targets. In next-generation designs, we are targeting even higher efficiencies through further improvement on peripheral limitations such as CO₂ mass transport in the electrolyte and the large band gap of the photoanode.26 Nevertheless, it represents a unique materials–biological hybrid for artificial photosynthesis, which demonstrates unassisted light-driven CO₂ fixation to acetic acid.

An interesting benefit of the nanowire array arises from its selective control of mass transport within the wire assem-

Figure 3. Enhanced oxygen tolerance for nanowire–bacteria hybrids. (a) A numerical simulation illustrates that integrating bacteria into a nanowire array allows for the survival of strict anaerobes in an aerobic environment. The oxygen concentration in the electrolyte decreases logarithmically from the nanowire array’s entrance, creating a local anaerobic environment. This is in contrast to the linear decrease of oxygen concentration for a planar electrode (Supporting Information Note and Figure 5). (b) Experimental demonstration of aerobic CO₂ reduction by S. ovata when Pt was additionally loaded onto the nanowire electrode, n = 3. Constant electrochemical bias (∼0.2 V vs RHE) was applied to the Si nanowire electrode, and the current was plotted against time. As highlighted in the plot, the sparging gas of the setup was switched from anaerobic (20% CO₂/80% N₂) to aerobic (21% O₂/10% CO₂/69% N₂) at t = 85 h.

Specifically, the design of nanowire–bacteria hybrids allows for the continuation of CO₂ reduction, a reaction catalyzed by strict anaerobe S. ovata, under a headspace containing 21% oxygen when an oxygen reduction reaction electrocatalyst (in the current case, Pt, see Methods) was loaded. The similar Tafel slopes of CO₂ reduction for planar and nanowire electrodes (Figure 2d) inform us that mass transport of protons and CO₂ was not a limiting factor within the nanowire array. However, with its limited solubility in water, oxygen can be depleted within the nanowire array logarhythmically, distinctly different from a planar counterpart (Supporting Information Note and Figure 5a,b). This arrangement effectively creates a local anaerobic environment at the bottom of the nanowire arrays, as supported by numerical simulation (Figure 3a and Supporting Information Figure 5c). Experimentally, after S. ovata had colonized the electrode anaerobically, we switched to an aerobic gas environment with 21% oxygen partial pressure (21% O₂/10% CO₂/69% N₂) (see Methods). Only the nanowire array loaded with Pt maintained its ability to reduce CO₂ and consistently produce acetic acid with a Faradaic efficiency of about 70% (t = 85 h, Figure 3b). Compared to the data obtained under anaerobic conditions (20% CO₂/80% N₂), the loss of Faradaic efficiency observed under aerobic conditions (∼15%) is related to the oxygen reduction reaction (Supporting Information Figure 6), which can be greatly minimized with improved design of the nanowire–bacteria hybrids (Supporting Information Note). In general, our observation implies that (1) in our system most, if not all, of the acetate is produced from bacteria interfacing directly with nanowires and (2) combining nanowire arrays with CO₂-reducing microorganisms can allow anaerobes to be used in a wider range of applications, such as CO₂ scrubbing from exhaust gas or even open air operation.

Taking advantage of the power of synthetic biology,18 a wide spectrum of complex organic molecules was synthesized by directly using the solar-derived acetate from oxygen-containing CO₂ feedstock. Under aerobic or microaerobic conditions, genetically engineered E. coli can activate acetate into the common biochemical intermediate acetyl-CoA, which then will
be used for the biosynthesis of a variety of complex molecules (Figure 4a). In principle, the acetate-consuming 
E. coli and the solar-powered CO₂-reducing nano-biohybrids can be positioned in a single aerobic reactor. However, for optimized yield these two processes are conducted in separate containers (Supporting Information Note). Here, as a proof of concept, the production of 
n-butanol,²⁹ PHB biopolymer,³¹ and three isoprenoid compounds³⁰ is demonstrated with H₂O and CO₂ as the starting materials and sunlight as the energy source (Figure 1b and Supporting Information Figures 7 and 8; see Methods). After the solar-powered acetate-production step, the accumulation of the target molecules is correlated with the consumption of acetate (n = 3, Supporting Information Figure 9a), implying the conversion of acetate into the desired products. The yield of target molecules was as high as 26% for 
n-butanol, 25% for one of the isoprenoid compounds (amorphadiene), and up to 52% for PHB biopolymer (Figure 4b and Supporting Information Figure 9b), comparable with literature values.²⁹−³¹ Taking into account the 0.38% efficiency from CO₂ to acetic acid, a solar energy-conversion efficiency of 0.20% is achieved from CO₂ to PHB biopolymer, a renewable and biodegradable plastic. Overall, the production of different organic products with vastly different synthetic pathways (Supporting Information Figure 7) proves the versatility of the integrated approach starting from one common biochemical building block, analogous to natural photosynthesis.

The results reported here outline a solar-energy conversion process that combines the strengths of semiconductor nanodevices and bacterium-based biocatalysts (Figure 1a). Key advantages of the nanowire-based device are the enhanced oxygen tolerance that allows exhaust gas to be directly fed into the system, thereby enabling use of strict anaerobes with aerobes, as well as the high measured CO₂ fixation activity of the nanowire–bacteria hybrid. Moreover, this modular platform simplifies the overall system design by allowing for the production of a variety of molecular targets, without any setup change in the components for light capture and CO₂ reduction into acetate, by varying only the downstream microorganisms.
**REFERENCES**