

# Electricity Production from Cellulose in a Microbial Fuel Cell Using a Defined Binary Culture

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Microbial fuel cells (MFCs) convert biodegradable materials into electricity, potentially contributing to an array of renewable energy production strategies tailored for specific applications. Since there are no known microorganisms that can both metabolize cellulose and transfer electrons to solid extracellular substrates, the conversion of cellulosic biomass to electricity requires a syntrophic microbial community that uses an insoluble electron donor (cellulose) and electron acceptor (anode). Electricity was generated from cellulose in an MFC using a defined coculture of the cellulolytic fermenter *Clostridium cellulolyticum* and the electrochemically active *Geobacter sulfurreducens*. In fed-batch tests using two-chamber MFCs with ferricyanide as the catholyte, the coculture achieved maximum power densities of 143 mW/m<sup>2</sup> (anode area) and 59.2 mW/m<sup>2</sup> from 1 g/L carboxymethyl cellulose (CMC) and MN301 cellulose, respectively. Neither pure culture alone produced electricity from these substrates. The coculture increased CMC degradation from 42% to 64% compared to a pure *C. cellulolyticum* culture. COD removal using CMC and MN301 was 38 and 27%, respectively, with corresponding Coulombic efficiencies of 47 and 39%. Hydrogen, acetate, and ethanol were the main residual metabolites of the binary culture. Cellulose conversion to electricity was also demonstrated using an uncharacterized mixed culture from activated sludge with an aerobic aqueous cathode.

## Introduction

The concerns of global fossil fuel depletion and environmental pollution from fossil fuel combustion are driving the search for carbon-neutral, renewable energy alternatives. As one of the most abundant renewable resources, cellulosic biomass, including waste products of agricultural and industrial activities, is particularly attractive in this context because of its relatively low cost and plentiful supply (1, 2). The U.S. Departments of Agriculture and Energy estimate the annual availability of 1.3 billion dry tons of biomass feedstock in the United States, which could displace 30% or more of the country's present petroleum consumption (3). Depending on the end-use application, this biomass could be converted to a variety of energy carriers such as ethanol (4), biodiesel (5), and hydrogen (6), as well as to electricity

indirectly derived from cellulose by coupling cellulolytic, fermentative hydrogen production with the catalytic oxidation of hydrogen at a fuel cell anode (7). This latter strategy is restricted by the theoretical fermentative hydrogen yield of 4 mol H<sub>2</sub>/mol hexose, with the balance of electrons appearing in soluble fermentation products and cells.

Using electrochemically active microorganisms as biocatalysts, microbial fuel cells (MFCs) are bioelectrochemical reactors that convert organic material directly into electricity (8). Unlike chemical or enzyme-based fuel cells, which are tailored to oxidize specific electron donors, MFCs have tremendous electron donor versatility. This includes simple substrates such as glucose, acetate, and lactate (9–11); complex substrates such as municipal and industrial wastewaters (12, 13); and even steam-exploded corn stover hydrolysate (14). MFCs can also be configured to produce hydrogen instead of electricity using an anaerobic cathode and a small applied voltage to reduce protons in the cathode chamber (15), thereby providing an approach for overcoming the 4 mol H<sub>2</sub>/mol hexose constraint of fermentation.

Although many types of bacteria have been found to be electrochemically active, none of them show cellulolytic activity, but rather require products of cellulose hydrolysis and fermentation as electron donors (9, 16–17). With the lack of an isolated microbe that can both hydrolyze cellulose and reduce solid extracellular electron acceptors, one strategy to directly produce electricity from cellulosic biomass, without the expense of chemical catalysts and overcoming the loss of electrons to soluble fermentation products, would involve a synergistic consortium of polymer-degrading, fermentative microorganisms and fermentation product-utilizing, electrochemically active microorganisms. *Clostridium cellulolyticum* degrades cellulose effectively, with the main fermentation products being acetate, ethanol, and hydrogen (18). This makes it a perfect match with *Geobacter sulfurreducens*, an electrochemically active bacterium that can oxidize all of these fermentation products (19) and use an electrode as an electron acceptor (9). In this study, a defined binary culture of *C. cellulolyticum* and *G. sulfurreducens* was tested in MFCs for *in situ* electricity production directly from cellulose. A mixed culture from activated sludge was also used to test the capability of natural inocula for cellulose–electricity conversion.

## Materials and Methods

**Cultures and Media.** *G. sulfurreducens* (ATCC 51573) and *C. cellulolyticum* (ATCC 35319) frozen stock cultures were cultured anaerobically in Balch tubes. The media used for the two strains had identical components except for the electron donor and electron acceptor. Both media contained (per liter) 1.05 g of NH<sub>4</sub>Cl, 1.5 g of KH<sub>2</sub>PO<sub>4</sub>, 2.9 g of K<sub>2</sub>HPO<sub>4</sub>·3H<sub>2</sub>O, 0.2 g of MgCl<sub>2</sub>·6H<sub>2</sub>O, 0.075 g of CaCl<sub>2</sub>·2H<sub>2</sub>O, 10 mL of trace mineral mix, and 10 mL of vitamin mix (20). In addition, *Geobacter* medium contained (per liter) 1.64 g of sodium acetate as electron donor and 13.7 g of ferric citrate as electron acceptor. *Clostridium* medium contained (per liter) 2 g of sodium carboxymethyl cellulose (CMC, MP Biomedicals, Inc.) and 0.5 g of yeast extract. The media were adjusted to pH 6.8 and flushed with N<sub>2</sub>–CO<sub>2</sub> (80:20) for *Geobacter* and N<sub>2</sub> (100%) for *Clostridium* before distributing and autoclaving in sealed vials. Activated sludge was obtained from the Pennsylvania State University Wastewater Treatment Plant and was stored at 4 °C until inoculation into MFCs.

**MFC Construction and Operation.** Two-chamber MFCs (21) were used in this study, despite their characteristically high internal resistance, because they allow the maintenance

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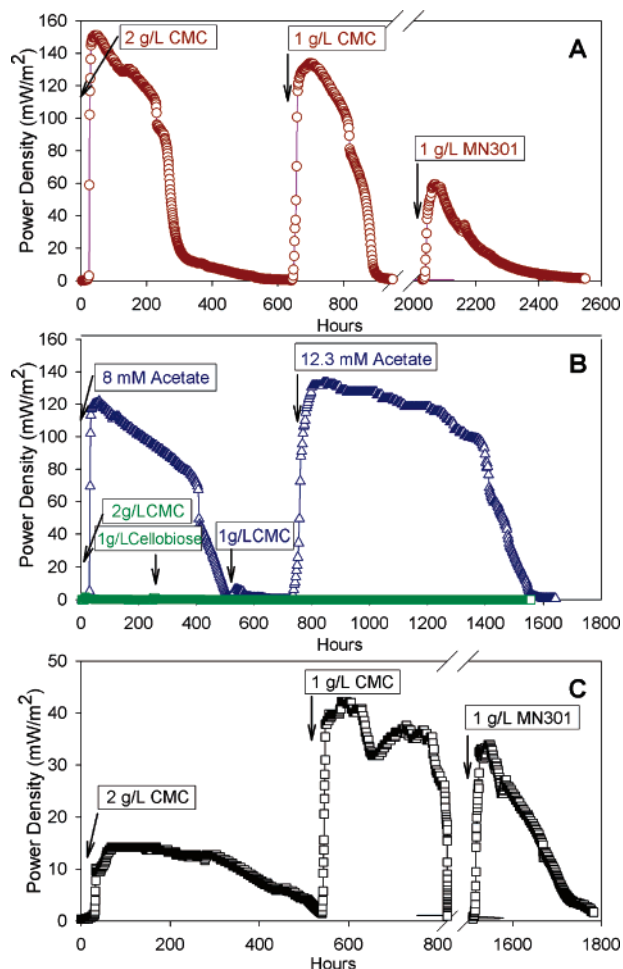
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of strictly anaerobic conditions in the anode chamber. Reactors were constructed from two medium bottles (310 mL capacity, VWR Inc.) joined by a glass bridge with a proton exchange membrane (Nafion 117, Dupont Co.) clamped between the two glass tubes. The anode chamber was sealed with a black rubber stopper in which a 10 mm diameter hole was drilled and then sealed by a butyl septum stopper to make a gastight seal. Anode chambers were filled anaerobically with the medium described above but with 0.1 g/L yeast extract and containing either acetate (without ferric citrate), CMC (soluble; substitution degree 0.65–0.90 carboxymethyl groups per 10 anhydroglucose units), or MN301 cellulose (insoluble; a combination of amorphous and microcrystalline cellulose; Macherey-Nagel, Duren, Germany) and mixed with magnetic stir bars. Cathode chambers were filled with 100 mM  $K_3Fe(CN)_6$  solution in 100 mM  $KH_2PO_4$  buffer (pH 7) for the pure- and binary-culture tests. For the mixed-culture MFC, the cathode chamber contained the 100 mM phosphate buffer into which air was continuously sparged to supply oxygen as the electron acceptor. Graphite plates (15.2-cm<sup>2</sup> surface area) were used as anodes. Plain carbon paper and carbon paper with Pt catalyst (0.35 mg/cm<sup>2</sup>; 10% Pt) on one side were used as cathodes in the defined culture systems and mixed-culture system, respectively. The electrodes were connected to a 1000  $\Omega$  external circuit using copper wires. A reference electrode (RE-5B, Bioanalytical Systems, Inc) was introduced into the anode chamber by embedding it through the stopper. All MFCs were operated in fed-batch mode at 30 °C, and sterile medium and  $K_3Fe(CN)_6$  were completely replaced when the voltage dropped below 40 mV. Each batch condition was repeated at least three times.

**Inoculation and Acclimation.** Sterilized anaerobic MFC anode chambers were inoculated with stationary-phase resuscitated pure cultures. *G. sulfurreducens* (5 mL) was transferred into MFC-1 containing 8 mM sodium acetate as the electron donor and the anode as the sole electron acceptor. Two other MFCs (MFC-2 and MFC-3) containing 2 g/L CMC as substrate were inoculated with 5 mL of *C. cellulolyticum*. When the voltage of MFC-1 stabilized at approximately 430 mV after 62 h, the anode of MFC-2 was replaced with the anode of MFC-1, which had been colonized by *G. sulfurreducens*. A new sterile graphite electrode was then inserted into MFC-1. Therefore, MFC-1 contained pure *Geobacter*, MFC-2 contained a coculture of *Geobacter* and *Clostridium*, and MFC-3 contained pure *Clostridium*. For the uncharacterized mixed culture MFC, 2 mL of activated sludge was transferred into an anode chamber containing either 2 g/L CMC or MN301 as substrate. All transfers were performed aseptically in an anaerobic chamber to prevent oxygen intrusion.

**Analyses.** Voltage (V) was continuously monitored using a data acquisition system (ADC22, Pico Technology, Ltd.) and periodically confirmed with a multimeter. Polarization data was collected by changing the external resistance using a variable resistor box during the stable power production stage of each batch experiment. The calculations of power density and Coulombic efficiency (i.e., the fraction of electrons removed from the electron donor that are recovered as current through the external circuit) were performed according to Cheng et al. (20). CMC concentration was determined using the phenol–sulfuric acid method for sugars after filtering samples through 0.2  $\mu$ m PES membrane filters (Pall Corp.) to remove cells. Biomass binding to MN301 was removed by centrifugation at 5900g for 5 min, washing with phosphate-buffered saline (130 mM NaCl, 10 mM  $Na_2HPO_4$ , pH 7.2), and resuspended in 0.2 M NaOH solution. After another equivalent centrifugation, the remaining MN301 cellulose pellets (not dissolved by NaOH as is the biomass) were analyzed using the phenol–sulfuric acid method for



**FIGURE 1.** Electricity generation by (A) binary culture, (B) *C. cellulolyticum* ( $\square$ ) or *G. sulfurreducens* ( $\Delta$ ), and (C) mixed culture.

sugars (22). COD was measured using a colorimetric assay (HACH Co.) after the removal of biomass (23), by filtration for CMC-fed reactors, and for MN301 fed reactors on both the filtrate and the solid after filtration and NaOH lysis and washing. For soluble metabolite analysis, samples were centrifuged and the supernatant was filtered through 0.2  $\mu$ m membranes and stored at –20 °C. The concentrations of organic acids and solvents were determined by gas chromatography (Varian Star 3400) with a flame ionization detector as previously reported (23). The anode headspace gas composition was analyzed for  $H_2$ ,  $CH_4$ ,  $CO_2$ , and  $N_2$  by gas chromatography (model 310; SRI Instruments) equipped with a thermal conductivity detector as previously reported (23). The internal resistance of the MFCs was measured by electrochemical impedance spectroscopy using a PC4/750 potentiostat (Gamry Ins.) with the anode as the working electrode and the cathode as the counter electrode (20). Conductivity was measured using a conductivity meter (Oaklon).

Scanning electron microscopy (SEM) was used to examine the anode biofilm and suspended microorganisms. Samples were prepared by 2% glutaraldehyde fixation. Biofilm and filtered solution samples were then prepared for SEM observation following the technique of Liu et al. (24), and the SEM (JSM 4500, JEOL) was operated at 20 keV.

## Results

**Electricity Generation by the Binary Culture.** Upon transferring the anode from MFC-1 into MFC-2, the power density rapidly increased to 151 mW/m<sup>2</sup> (Figure 1A) as *G. sulfurre-*

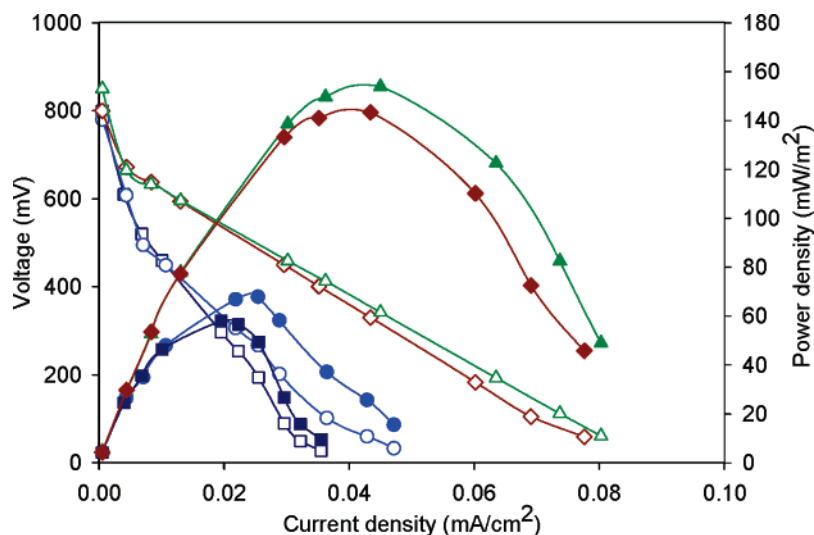


FIGURE 2. Voltage (hollow symbols) and power density (solid symbols) vs current density obtained by varying the external circuit resistance (50–50 000 $\Omega$ ). *G. sulfurreducens* in 12.3 mM acetate ( $\Delta$  and  $\blacktriangle$ ); binary culture in 1 g/L CMC ( $\diamond$  and  $\blacklozenge$ ); and binary culture in 1 g/L MN301 ( $\square$  and  $\blacksquare$ ) and after NaCl addition ( $\circ$  and  $\bullet$ ).

TABLE 1. Summary of Substrate Degradation and Electron Recovery

parameter	<i>Geobacter</i> (acetate)	<i>Clostridium</i> (CMC)	coculture (CMC)	coculture (MN301)	sludge (CMC)	sludge (MN301)
COD degradation <sup>a</sup> (%)	76 $\pm$ 3	16 $\pm$ 5	38 $\pm$ 3	27 $\pm$ 4	32 $\pm$ 4	34 $\pm$ 7
cellulose degradation (%)	NA	42 $\pm$ 5	64 $\pm$ 4	49 $\pm$ 6	41 $\pm$ 5	40 $\pm$ 7
Coulombic efficiency <sup>b</sup> (%)	51 $\pm$ 4	0	47 $\pm$ 4	39 $\pm$ 6	27 $\pm$ 4	22 $\pm$ 5

<sup>a</sup> All COD were measured after removal of biomass. <sup>b</sup> Based on COD removed.

*ducens* oxidized the CMC fermentation products of *C. cellulolyticum*. After attaining this level, the power density slowly declined, but electricity was generated for 24 days. When the anode solution was replaced with new medium containing 1 g/L CMC, power production resumed quickly even though no pre-fermentation by *C. cellulolyticum* occurred, and the maximum power density of 143 mW/m<sup>2</sup> was comparable to that of the initial batch. After five repeated cycles (complete data not shown), 1 g/L MN301 cellulose was used as the substrate instead of CMC. The maximum power density achieved from MN301 was 59.2 mW/m<sup>2</sup> after 3 days, and the power density dropped slowly to 0.76 mW/m<sup>2</sup> over the next 20 days (Figure 1A). Comparable results were obtained with three repeated MN301 additions.

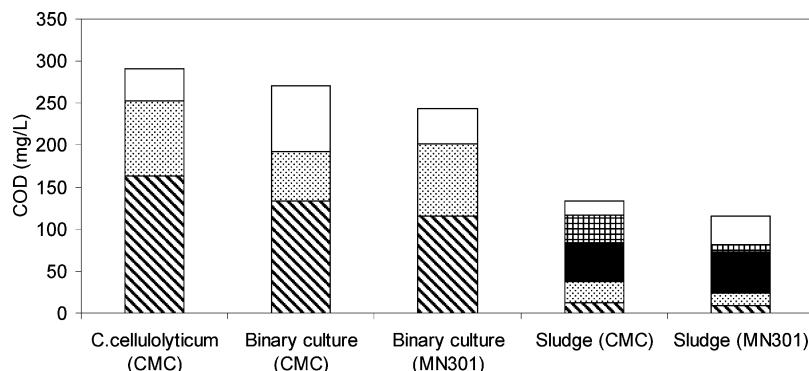
For the pure-culture MFCs, as expected, *G. sulfurreducens* produced power quickly when fed acetate in MFC-1 (Figure 1B) (9). However, when the substrate was changed to CMC, power was not produced. Moreover, no cellulose degradation occurred in that cycle, indicating CMC was not degraded by *G. sulfurreducens*. When acetate was added back into MFC-1 after a period of 193 h with only CMC, power production resumed very quickly, indicating that the cells were still alive. In MFC-3, *C. cellulolyticum* did not show power production from either CMC or cellobiose (Figure 1B), even though it did degrade these substrates. Very small amounts of power (max. 1.16 mW/m<sup>2</sup>) of short duration were detected immediately following each medium transfer, but this was presumably not due to microbial activity.

To compare electron recovery and power production between the *Geobacter* pure culture and the binary culture, new medium containing 12.3 mM acetate was fed to the *G. sulfurreducens* MFC (MFC-1) to provide the same electron equivalence (98.4 mM) as the cellulose-fed coculture system, assuming acetate was the only soluble fermentation metabolite produced from the cellulose (2 mol acetate/mol hexose). The differences in power production across a range

of current densities were obtained by varying the external resistance (50–50 000  $\Omega$ ) (Figure 2). The power density achieved by *G. sulfurreducens* with acetate was comparable to that from the CMC-fed binary culture, with maximum power densities of 154  $\pm$  4.1 and 143  $\pm$  7.2 mW/m<sup>2</sup>, respectively. The power density achieved from the MN301-fed binary culture was lower, with a maximum value of 59.2  $\pm$  3.5 mW/m<sup>2</sup>. When MN301 was used instead of CMC, the internal resistance of the system increased from 770 to 890  $\Omega$  and the conductivity of the anode solution decreased from 7.43 to 5.81 ms/cm. After adding 3 g/L NaCl to the MN301 medium, the solution conductivity increased from 6.08 to 11.34 ms/cm, but the maximum power density only increased to 67.9 mW/m<sup>2</sup> (Figure 2), indicating that decreased solution conductivity did not explain the significant drop in power density with MN301.

When fresh cultures of *G. sulfurreducens* and *C. cellulolyticum* were inoculated together into new medium with a sterile anode, a longer lag time (42 h) was observed and gradual power generation (max. 91.6 mW/m<sup>2</sup>) was detected, but when the anode solution was replaced with fresh CMC medium, power generation resumed quickly (max. 134 mW/m<sup>2</sup>) (data not shown), which indicated that the pre-fermentation and anode pre-colonization (MFC-2) were helpful for startup but not required in a cellulose-fed MFC.

**Electricity Generation by a Mixed Culture.** With 2 g/L CMC as the substrate, the uncharacterized mixed culture generated electricity after a lag time of 51 h following inoculation, but the maximum power density was only 14.9 mW/m<sup>2</sup>. The power density increased after subsequent medium changes, presumably indicating the enrichment of functionally relevant community members (Figure 1C). The maximum power density achieved by the mixed culture after three medium changes was 42.2  $\pm$  6.1 mW/m<sup>2</sup> on CMC and 33.7  $\pm$  4.9 mW/m<sup>2</sup> on MN301. However, a lower cathode potential due to the use of aqueous oxygen instead of



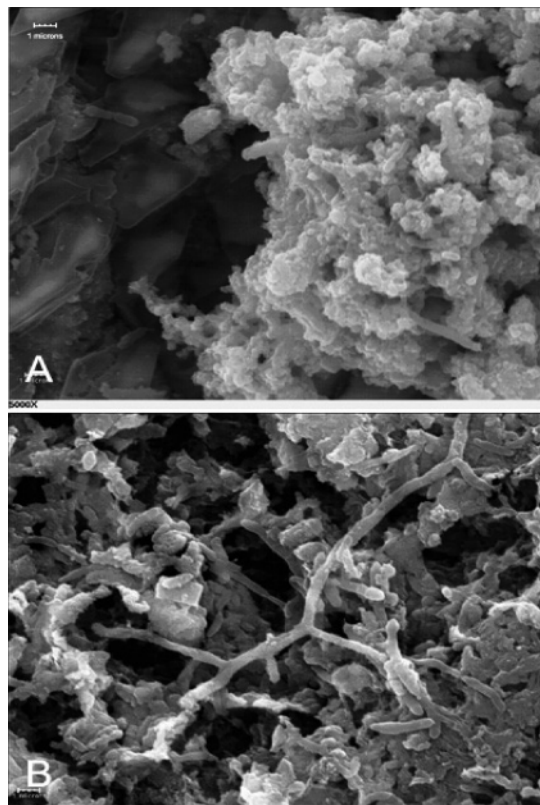
**FIGURE 3. Soluble metabolite concentrations (expressed as mg/L COD) for MFCs at the end of fed-batch cycles: uncharacterized COD (□), propionate (hatched), butyrate (■), ethanol (dotted), and acetate (diagonal striped).**

ferricyanide precludes a direct comparison between the mixed- and pure-culture systems.

**Cellulose Degradation and Coulombic Efficiency.** Cellulose degradation, COD removal, and Coulombic efficiency for the various MFC conditions are compared in Table 1, based on analyses of samples from the end of batch cycles. *G. sulfurreducens* consumed  $76 \pm 3\%$  of the COD when using 12.3 mM acetate as the electron donor, and directed approximately half of these electrons to anode reduction. For the 1 g/L CMC tests,  $42 \pm 5\%$  of the sugars present in CMC were metabolized in the pure *C. cellulolyticum* system, and most of the COD of those sugars remained in solution as other metabolites, resulting in a COD removal of only  $16 \pm 5\%$ . With no current produced in this system, the COD loss was presumably attributable to biomass generation, hydrogen production, and neutral metabolites diffusing to the cathode chamber. The coculture degraded  $64 \pm 4\%$  of the CMC in 16 days, which was 52% more than the pure culture, showing relief of product inhibition through their consumption by *Geobacter*. The corresponding COD removal by the coculture using CMC was  $38 \pm 3\%$ , with electricity production accounting for nearly half of these electrons and most of the other degraded CMC recovered as residual soluble fermentation products as described below. The coculture degraded  $49 \pm 6\%$  of the MN301, which was less than that observed with CMC, but still higher than that of the CMC-fed *C. cellulolyticum* pure culture. This MN301-fed system had a similar degree of COD loss relative to the extent of cellulose degradation (55%) as the CMC reactor, but a slightly lower Coulombic efficiency.

The sludge inoculum showed a similar amount of degradation with both CMC and MN301 ( $41 \pm 5\%$  and  $40 \pm 7\%$ , respectively). Only a quarter of the electrons removed from solution were transferred to the anode. The Coulombic efficiencies with the mixed culture were lower than with the binary culture presumably due to oxygen diffusion from the cathode and nonproductive metabolisms such as methanogenic activity. The final pH ranged from 5.2 to 5.9 for the various conditions, including binary- and mixed-culture systems.

**Metabolite Variation.** *C. cellulolyticum* fermented cellulose primarily into acetate, ethanol, and hydrogen as expected (18), with acetate plus ethanol accounting for nearly 90% of the soluble COD products (Figure 3). *Geobacter* consumed some of these as electron donors in conjunction with anode reduction. Hydrogen comprised approximately 50% of the headspace in both CMC-fed reactors on day one and declined slowly until it reached 15% in the headspace of the coculture reactor and 29% in the *Clostridium* reactor. By comparison, the metabolites in the mixed-culture MFC were more diverse, with acetate, butyrate, propionate, and ethanol as the main components (Figure 3). Their total residual concentrations were less than those in the coculture



**FIGURE 4. SEM images of (A) coculture anode biofilm (5000×), and (B) mixed-culture anode biofilm (5000×).**

system. There was no hydrogen detected in the headspace of the mixed-culture system, and the methane proportion was greater than 70%.

**Anode Biofilms.** Scanning electron micrographs taken of the anode biofilms showed various biofilm morphologies for different communities (Figure 4). The binary culture formed tight microcolony structures, while the mixed culture from activated sludge showed heterogeneous cell morphologies and a looser biofilm structure. Lower magnification images showed that the coculture biofilm had patchy coverage of the anode, while the mixed culture formed a more uniform biofilm over the surface of the graphite plate.

## Discussion

The binary culture of *C. cellulolyticum* and *G. sulfurreducens* converted cellulose into electricity, without enzymatic pretreatment or an exogenous catalyst. In this defined system, *C. cellulolyticum* fermented cellulose mainly into acetate, ethanol, hydrogen, and carbon dioxide, and *G. sulfurreducens* transferred electrons from some of these fermentation

products to the anode via anaerobic respiration. Since no known microbial strain performs this complete conversion by itself, this complementary pairing of cellulolytic and exoelectrogenic activities is currently necessary for electricity production from cellulose. It has been proposed that this coupling of fermentation with anaerobic respiration has a thermodynamic advantage over complete anaerobic oxidation of a fermentable substrate by a single microbe, based on the energy yield per electron transferred in the respective reactions (17, 25). This is consistent with the finding that the majority of glucose was fermented in Fe(III)-reducing sediments instead of being directly oxidized to CO<sub>2</sub> with Fe(III) oxide as the electron acceptor (26), even though a glucose-oxidizing iron reducer has subsequently been isolated from this environment (16). Therefore, a consortium of cellulolytic fermenters and electrochemically active bacteria might have a competitive advantage over a hypothetical cellulolytic exoelectrogen.

The maximum power density achieved by the coculture with CMC (143 mW/m<sup>2</sup>) was comparable to the power output using acetate as the electron donor for *G. sulfurreducens* (154 mW/m<sup>2</sup>). These results were higher than those of a similar two-chamber system (75 mW/m<sup>2</sup>) that used 20 mM acetate, a mixed-culture inoculum, and ferricyanide as a catholyte (27). However, the MN301-fed coculture system produced less than half this power density, for reasons other than MN301 effects on solution conductivity. The chemical functionalization of CMC makes it soluble and more accessible for degradation than native cellulose. Amorphous and microcrystalline MN301 is more similar to native cellulose, and the lower rate of MN301 hydrolysis and fermentation by *C. cellulolyticum* presumably contributed to the lower power density with this substrate. With both cellulosic substrates, COD removal was less than cellulose degradation because *G. sulfurreducens* did not use all of the available fermentation products by the time power production decreased in each batch cycle, as confirmed by the residual acetate and ethanol concentrations. A possible explanation for this is that the final pH usually dropped to approximately 5.2 due to the acidic products of fermentation, which may have inhibited the activity of *G. sulfurreducens*. This was supported by a control experiment, showing that power production resumed quickly from a nonproductive pH 5.2 reactor after an injection of sterilized NaOH, which increased the pH to 7.0. Since the buffer capacity of the medium was designed based on common *Clostridium* media, *Geobacter* probably could not tolerate such a range of pH variation.

The activated sludge inoculum also generated electricity from cellulose. However, the different experimentally measured cathode open-circuit potentials (+427 mV vs normal hydrogen electrode (NHE) for aqueous oxygen and +577 mV vs NHE for ferricyanide) and different overpotential losses associated with aerobic and ferricyanide cathodes preclude a direct comparison of power density between these experiments. Ferricyanide was used as the cathodic electron acceptor in the defined culture experiments (27, 28), which enabled oxygen-free operation of the cathode compartment. This was critical because both species used in these tests are strict anaerobes. Based on previous experience, *G. sulfurreducens* can tolerate a slight amount of oxygen once acclimated to the anode, but it was difficult to initiate the growth of either *G. sulfurreducens* or *C. cellulolyticum* in MFCs using aqueous oxygen as the cathodic electron acceptor, unless an oxygen scavenger such as cysteine is added to the anode chamber. However, cysteine can also be used as an electron donor for electricity production (21). In contrast, aqueous air cathodes, which are more practical for eventual MFC applications, were used in the mixed-culture fuel cell because microaerophilic and/or facultative bacteria would be present and could grow on the PE membrane, scavenge oxygen, and

reduce its diffusion into the anode chamber. The Coulombic efficiency was also lower in the mixed culture than in the binary culture, presumably due to competing metabolisms and oxygen diffusion through the membrane. The presence of methane in the headspace showed that methanogenesis was a significant competitive process to extracellular electron transfer. However, the results of these tests showed that natural consortia can also achieve cellulose–electricity conversion, but more efficient cultures would improve this process.

SEM pictures showed different biofilm architectures between the defined binary culture and the undefined mixed culture. While the performance data indicate that *C. cellulolyticum* remained in the binary-culture system at sufficient density to rapidly produce metabolites for *Geobacter* growth, the SEM pictures do not reveal the specific localization of the two functional species. More specific identification tools, such as fluorescent *in situ* hybridization, may be needed to ascertain the detail of the interaction between the two strains.

This research demonstrates the use of whole cells as biocatalysts for cellulose-derived electricity production, and it shows promise as a new method to accomplish both waste biomass treatment and electricity generation. Optimizing the reactor configuration, especially for particulate substrates, is necessary for the further development of cellulose-derived electricity production. In addition, running reactors in continuous mode and increasing the buffer capacity should increase power density, cellulose degradation, and electron recovery (29). Higher power is anticipated if physical and chemical constraints of the reactors are addressed (30). Furthermore, this MFC system employs a unique microbial ecology in which both electron donor and acceptor are insoluble. It should be very helpful for studying the microbial communities and improving our understanding of how bacteria degrade solid substrates and how they transfer electrons to solid electron acceptors.

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