Bioelectrochemical Systems for Anaerobic Digester Biogas Upgrading

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ABSTRACT
Anaerobic digester biogas, which contains a mixture of CO_2, CH_4 and trace gases (e.g., H_2S, N_2, H_2, NH_3, siloxanes) may be enriched in energy content (i.e., "upgraded") using bioelectrochemical systems (BESs). However, to-date, BES biogas upgrading studies have used bicarbonate or commercial gas mixtures instead of anaerobic digester biogas. Therefore, the objective of this study was to: i) compare the performance of a methanogenic BES between CO_2-fed and biogas-fed cycles; ii) assess the biocathode performance when fed with a varying biogas composition (i.e., variable CO_2 and CH_4 content); and iii) evaluate the effect of applied cathode potential on biocathode CH_4 production when fed with anaerobic digester biogas. Two BESs were developed: BES1, which was used to compare performance between a CO_2-fed cycle and a biogas-fed cycle when the gases were fed at atmospheric pressure (1 atm, absolute pressure); and BES2, which was used to compare 5 typical CO_2-fed cycles with 19 cycles fed by pressurizing the cathode headspace (1.4 atm) with anaerobic digester biogas. Low BES1 biocathode CH_4 production was observed during the biogas feeding, likely due to CO_2 substrate limitations. In BES2, headspace pressurization eased substrate limitations and the maximum CH_4 production rate occurred during a biogas-fed cycle (1.85 mmol/d), which was 350% higher than the maximum CH_4 production rate during a CO_2-fed cycle (0.41 mmol/d). CH_4 and H_2S in the biogas were theoretically capable of producing 4% of the total charge transfer from the anode to the cathode at -0.8 V (vs. SHE) applied cathode potential, and a maximum of 35% of the total charge transfer from the anode to cathode was observed at -0.55 V. This study indicates that anaerobic digester biogas is a promising biocathode feedstock for BES biogas upgrading. Further research is needed to determine how each individual component of anaerobic digester biogas affects system performance.

KEYWORDS: Biocathode, carbon dioxide, methane, methanogenesis, sulfide, trace gases

1. INTRODUCTION
Anaerobic digester biogas contains a mixture of carbon dioxide (CO_2), methane (CH_4) and trace gases (e.g., H_2S, N_2, H_2, NH_3, siloxanes). In order to expand the use of biogas to the vehicle fuel and natural gas markets, biogas must be processed such that its CH_4 content is higher than 80-96% (v/v), CO_2 is below 2-3% and H_2S is below 5 mg/m^3, along with requirements for O_2, NH_3 and siloxanes [5]. The processing of biogas to increase its energy content and remove impurities is referred to as "biogas upgrading". Physical/chemical biogas upgrading methods include water scrubbing, absorption, pressure swing adsorption, membrane and cryogenic separation. These technologies often require expensive consumables (e.g., membranes, sorbents), energy-intensive processes (e.g., pressurization, sorbent regeneration) or produce a CO_2 waste stream (e.g., membrane separation) [5,8]. Biological methods, such as algal CO_2 removal, often require large areas or produce biomass

that must be harvested and further processed [9]. Instead, bioelectrochemical systems (BESs) are able to directly convert biogas CO_2 to CH_4 without these disadvantages.

Previous studies have demonstrated the effectiveness of BES biocathodes to utilize bicarbonate [10], gaseous CO_2 [1,11,2,12], or a CO_2/nitrogen (N_2) mixture [3,4], for CH_4 production. However, the use of anaerobic digester biogas as a BES biocathode feedstock has not been explored. Unlike bicarbonate or commercial gases, biogas contains trace gases that could affect BES performance by altering the microbial community at the biocathode and/or bioanode, if gas is transported across the BES proton exchange membrane. Indeed, a prior study showed that the Nafion 117 membrane, commonly used in BESs, is permeable to CO_2, N_2, CH_4 and hydrogen (H_2) [7]. As a potential electron donor, H_2 could be used by methanogens for CO_2 conversion to CH_4, or stimulate other types of microbial processes (e.g., acetogenesis). Other gases, such as H_2S and NH_3, could inhibit microbial activity [13,14]. Alternately, gases may be transported across the membrane and become oxidized at the anode, donating electrons to the circuit, as shown in Equations 1-4, below. Indeed, bioanodes have been reported to perform, and produce current from the reactions listed in Equations 1-4 [15-17].

$CO_2 + 8H^+ + 8e^- = CH_4 + 2H_2O$ $E^{0\prime} = -0.24$ V (Equation 1)

$N_{2(g)} + 6H^+ + 6e^- = 2NH_{3(g)}$ $E^{0\prime} = +0.09$ V (Equation 2)

$SO_4^{2-} + 10H^+ + 8e^- = H_2S(g) + 4H_2O$ $E^{0\prime} = +0.31$ V (Equation 3)

$S^0_{(s)} + 2H^+ + 2e^- = H_2S_{(g)}$ $E^{0\prime} = +0.14$ V (Equation 4)

To date, BES biogas upgrading studies have typically used bicarbonate or commercial gaseous mixtures to study biocathodic methanogenic BES performance; thus, the effect of feeding with anaerobic digester biogas is unknown. Therefore, the objective of this study was to: i) compare the performance of a methanogenic BES between CO_2-fed and biogas-fed cycles; ii) assess the biocathode performance when fed with a varying biogas composition (i.e., variable CO_2 and CH_4 content); and iii) evaluate the effect of applied cathode potential on biocathode CH_4 production when fed with anaerobic digester biogas.

2. MATERIALS AND METHODS

2.1. Stock Anaerobic Digester. A stock anaerobic digester was developed with inoculum from a mesophilic, municipal anaerobic digester, fed with a mixture of dextrin/peptone and pre-reduced medium [18], maintained at 22ºC under continuous mixing. The digester had a retention time of 21 d and was batch-fed every 3 and 4 days weekly with a mixture of dextrin/peptone stock solution, to an initial dextrin/peptone concentration of 1.4 g COD/L and 1.9 g COD/L upon feeding, respectively; the mean organic loading rate was 480 mg COD/L-d. The COD removal was approximately 60% over the course of a feeding cycle and the pH ranged between 7.0 and 7.3. Biogas was continuously collected in an acid-brine displacement system to maintain a low pressure in the digester headspace. The mean biogas production was 300 mL/d and consisted of 53-66% CH_4 and 34-47% CO_2.

2.2. BES Setup. A batch-fed BES (BES1) was developed as previously described [2,7], with an acetate-fed bioanode and a CO_2-fed methanogenic biocathode. A second BES (BES2) was developed using biofilm-attached carbon felt from the BES1 anode and cathode electrodes as inoculum for the BES2 anode and cathode, respectively. Each BES bioanode was filled with 250 mL anolyte medium [7], flushed with N_2 and fed with sodium acetate to an initial concentration of 1,000 mg COD/L at the start of each feeding cycle. Cathodes were filled with 250 mL catholyte medium [2], and were maintained at -0.8 V (all potentials are vs. SHE, unless otherwise noted) using a Gamry Interface 1000 potentiostat (Warminster, PA). Anodes and cathodes were magnetically mixed and the two BESs were maintained at 22±2ºC. Initially, each biocathode headspace was flushed with 100% CO_2 and then pressurized to 1.6 atm (absolute pressure). Following the development of stable BES operation (e.g., current,

acetate removal, CH_4 production), BES1 was used to compare system performance when the biocathode was fed CO_2 or biogas at 1 atm, absolute pressure. Similarly, BES2 was used to compare BES performance when the CO_2 or biogas was fed to the biocathode at higher pressures (1.4 atm, absolute pressure). After development of consistent performance under CO_2-fed conditions, the BES2 biocathode was then fed daily with anaerobic digester biogas.

2.3. BES Operation. The performance of BES1 during a 3-d CO_2-fed cycle prior to biogas exposure was compared with its performance during a subsequent 3-d cycle when fed with anaerobic digester biogas. Gas production and composition, anode acetate removal, anolyte and catholyte pH, cell voltage, electrode voltages, and current were monitored, as described below (Section 2.4). Next, BES2, which was developed as a CO_2-fed biocathode, was fed with biogas at a higher pressure (1.4 atm) than in BES1 (1 atm) to overcome substrate (i.e., CO_2) limitation. BES2 performance before and after biogas introduction was compared. Next, in order to assess the BES2 operational stability under varying biogas feed composition, system performance was monitored over 24 1-d feeding cycles, during which the cathode was flushed and pressurized daily with anaerobic digester biogas with varying CO_2 and CH_4 content. Finally, the effect of applied voltage on BES2 performance was assessed when the biocathode was fed with anaerobic digester biogas by incrementally decreasing the applied cathode potential from -0.80 V to -0.50 V. To ensure a consistent biogas composition for the voltage tests, anaerobic digester biogas was collected in a locking, Tedlar gas bag, from which gas was used for flushing and pressurizing the cathode. For each of the biogas-fed cycles (-0.80 V to -0.50 V), the maximum possible contribution of CH_4, NH_3 and H_2S to the current was calculated. CH_4 was measured in the headspace of the anode at the end of the 1-d cycle and Henry's law was used to estimate the dissolved CH_4 available for possible oxidation at the anode (K_H = 776.4 atm/M) [7]. Due to the difficulty of measuring trace amounts of H_2S, only the initial biogas H_2S concentration could be measured. Thus, the H_2S available in the anode for oxidation was estimated assuming complete equilibrium of H_2S between anode and cathode, and between liquid and gas phases (K_H = 9.37 atm/M) [19,20]. Trace NH_3 is also difficult to measure and, therefore, the biogas NH_3 content was estimated by measuring the ammonia in the digester mixed liquor and assuming equilibrium with the gas phase ($K_H = 1.61 \times 10^{-2}$ atm/M) [19,20]. The biogas NH_3 in the BES was then assumed to achieve complete equilibrium between anode and cathode, and between liquid and gas phases. The total amount of charge transferred during a feeding cycle was calculated by integrating the area underneath the plot of current vs. time to obtain the Coulombs of charge transferred. To estimate the maximum amount of charge transferred from the oxidation of CH_4, H_2S and NH_3, the moles of each component were converted to moles of electron equivalents, using Equations 1, 2 and 3, respectively. Next, the moles of electron equivalents were converted to total number of electrons using Avogadro's number, followed by conversion to Coulombs of charge (-6.242×10^{18} electrons/Coulomb).

2.4. Analytical Methods. pH and ammonia was measured as described in *Standard Methods* [21]. Total gas production was measured using a pressure transducer (resolution ±1.974 atm, accuracy to 0.002 atm; Sper Scientific, Scottsdale, AZ). Gas composition (CO_2, CH_4, N_2, H_2) and acetate were measured by gas chromatography (GC) with thermal conductivity (TCD) and high-performance liquid chromatography (HPLC), respectively, as previously reported [22]. Anode and cathode potential were measured against adjacent Ag/AgCl electrodes using a handheld multimeter. Current was measured semi-continuously using Gamry Interface 1000 potentiostat (Warminster, PA). Current density is reported with respect to the proton exchange membrane (PEM) surface area. Periodic cyclic voltammetry was conducted using the potentiostat at a scan rate of 100 mV/s and a range of -1.0 V to -0.2 V.

3. RESULTS AND DISCUSSION

3.1. BES1 Performance. Following development and stable operation, BES1 was monitored over a 3-d CO_2-fed cycle and a subsequent 3-d biogas-fed cycle (Figure 1), without cathode headspace pressurization. The pH of the anolyte and catholyte remained between 6.8 and 7.3 during both the CO_2-fed and biogas-fed cycles. As in the prior CO_2-fed cycle, the current density increased immediately upon feeding with biogas (Figure 1a), and then declined over the remainder of the 3-d feeding cycle. However, during the biogas-fed cycle, the initial current density peak was slightly (13%) higher than in the CO_2-fed cycle, and the total charge transferred increased incrementally (5%) between the CO_2-fed cycle and the biogas-fed cycle. Although H_2S and NH_3 are minor components of anaerobic digester biogas, they could be transported to the anode through the PEM and donate electrons to the anode electrode [17,16,15]. Indeed, the anaerobic digester biogas contained approximately 0.3% H_2S (v/v) and 0.00012 - 0.00013% NH_3 (v/v). While the current increase may be due, in part, to the transport of H_2S, CH_4 or NH_3 from the cathode to the anode and their subsequent oxidation, other cause(s) may be likely, as further discussed in Section 3.3, below.

Because of greater availability, five-fold more CO_2 was removed from the headspace during the CO_2-fed cycle than during the biogas-fed cycle. However, the BES1 biocathode was able to remove CO_2 to a lower concentration in the biogas-fed cycle than in the CO_2-fed cycle (Figure 1b). In both cycles, CO_2 removal primarily occurred within the first day due to dissolution into the catholyte and microbial uptake/utilization. CH_4 production was slower in the biogas-fed cycle (0.06±0.03 mmol/d) than in the CO_2-fed cycle (0.19±0.01 mmol/d) (Figure 1c), which was likely due to CO_2 substrate limitations in the biocathode. CO_2 limitations were also indicated by the final gas composition; at the end of the CO_2-fed cycle, the biogas contained 10.5% CO_2, while at the end of the biogas-fed cycle, the biogas contained only 1.7% CO_2. Although a small amount of N_2 (transported from the anode) was present in the cathode headspace at the end of the biogas-fed cycle, the CH_4 content was 97.8%, which is well within the requirement for biogas use as vehicle fuel and in natural gas pipelines [5]. In contrast, the final gas of the CO_2-fed cycle contained 89.1% CH_4, which would require additional processing. During the CO_2-fed cycle, the CO_2 to CH_4 conversion efficiency was 29%, whereas during the biogas-fed cycle, the conversion efficiency was 63%. Within one day of biogas feeding, the biogas in the cathode headspace contained 96% CH_4 due to the rapid removal of CO_2. Thus, a shorter incubation period (< 2 d) could be used for batch biogas upgrading.

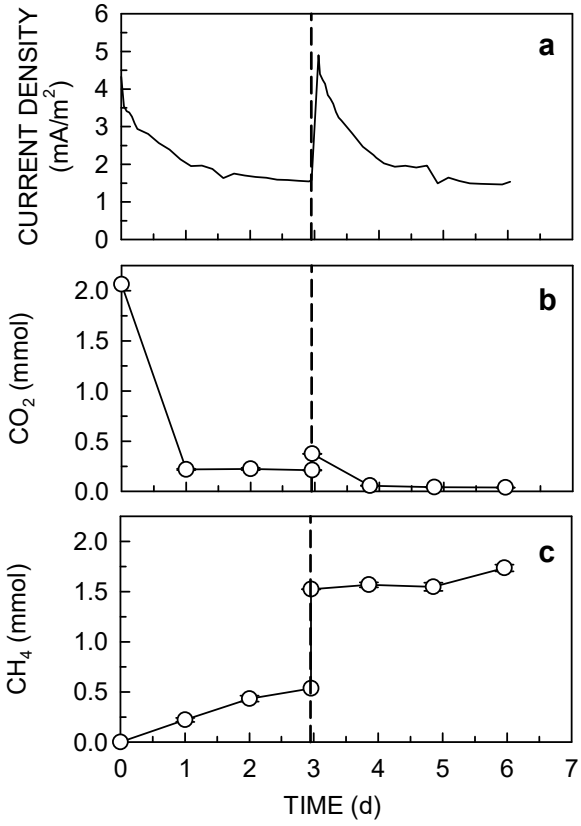

Figure 1. BES1 time course of current density (a), cathode headspace CO_2 (b), and cathode headspace CH_4 (c) when fed with 100% CO_2 (days 1-3) and anaerobic digester biogas (days 3-6) at 1 atm absolute pressure. The dashed line indicates the switch from CO_2 to biogas cathode headspace feed. Error bars represent mean ± standard deviation; $n = 3$.

3.2. BES2 Performance. To assess the performance of a biogas-fed biocathode operation at a shorter (1 d) incubation period, BES2 was developed and tested. Initially, the BES2 biocathode was fed with CO_2 until steady operation was achieved and then switched to biogas, using a pressurized headspace to help alleviate substrate (i.e., CO_2) limitations. The pH of the anolyte remained between 6.5 and 7.0 during all cycles. The catholyte pH ranged between 6.5 and 7.1 over the course of a 7-d incubation during CO_2-fed cycles. However, when fed with biogas, the catholyte pH reached 7.3-7.4 over the course of a 7-d incubation. It is likely that the larger amount of CO_2 available in the CO_2-fed cycles lowered the catholyte pH and led to a greater buffering capacity (i.e., HCO_3^-, CO_3^{2-}) of the catholyte.

The BES2 biocathode removed CO_2 to a lower concentration when fed with biogas than when fed with CO_2 (Figure 2b). Despite a variable initial CO_2 level during the biogas-fed cycles, the final CO_2 in the cathode headspace was 0.25 ± 0.07 mmol (n=19), indicating removal of CO_2 to a consistent level. During CO_2-fed cycles, a consistent biocathode CH_4 production rate was achieved (0.38 ± 0.02 mmol/d; n = 4; Figure 2a). When the biocathode was first fed with biogas (day 14, Figure 2a), CH_4 production was low and, on the second biogas feeding, CH_4 was even lost from the cathode headspace (Table 1). This loss could be due to the transport of CH_4 across the membrane to the anode compartment and possible oxidation. Remarkably, the CH_4 production improved dramatically on the third feeding with biogas (day 16), producing CH_4 at more than twice the rate than in the CO_2-fed cycles (Table 1). The elevated CH_4 production rate continued upon the fourth biogas feeding (day 17) and remained elevated above the CO_2-fed CH_4 production rates throughout the remainder of the 24 cycles. Based on CV tests conducted at the beginning of cycle 5 (CO_2-fed) and cycle 20 (biogas-fed), no significant redox peaks were observed, with the exception of one CO_2/CH_4 oxidation and reduction peak, which was smaller in the biogas-fed cycle.

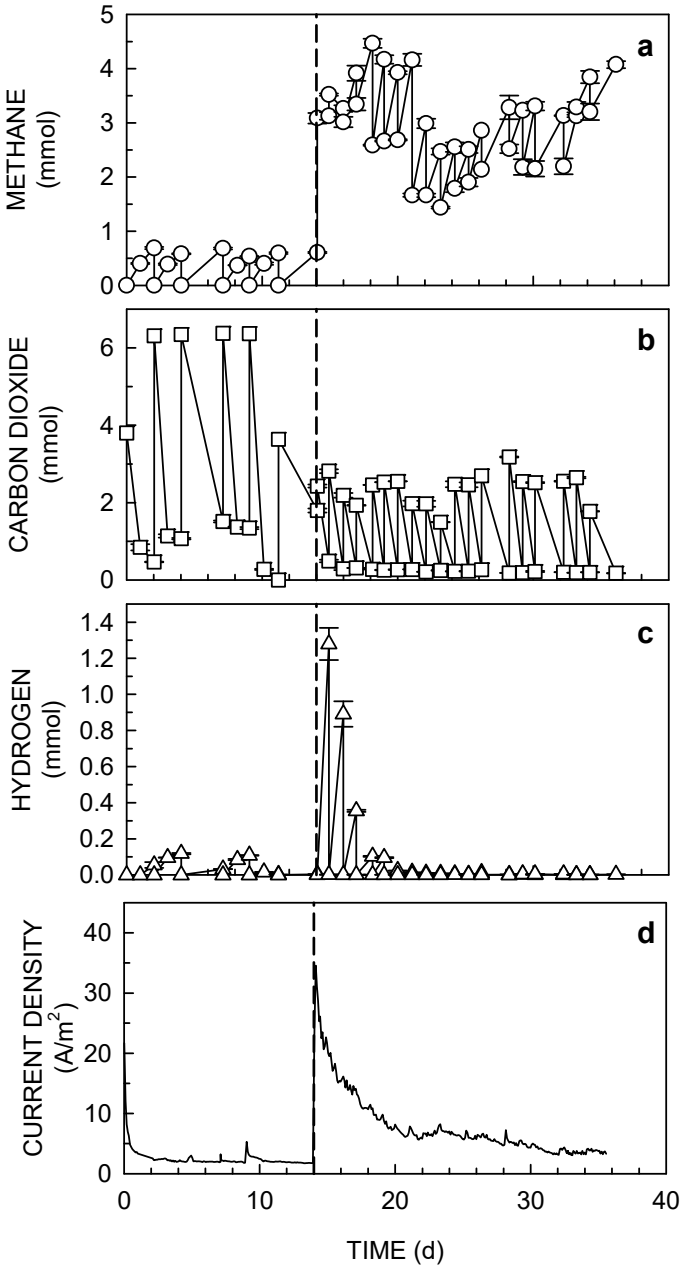

Figure 2. BES2 cathode headspace CH_4 (a), CO_2 (b) and H_2 (c), and system current density (d).

Table 1. BES2 performance over 24 1-d cycles, during which the biocathode was fed with CO_2 (cycles 1-5) and a variable composition biogas from an anaerobic digester (cycles 6-24).

Cycle Number	Cycle Days (d)	Initial CO_2 Partial Pressure (atm)	Initial CH_4 Partial Pressure (atm)	Anode Potential (V vs. Ag/AgCl)	Cell Potential (V)	Fraction of Removed CO_2 Converted to CH_4	Mean CH_4 Production Rate[a] (mmol/d)
1	0-2	0.92	0.00	0.93-1.28	2.13-2.85	0.21	0.41 ± 0.02
2	2-7	1.53	0.00	0.68-0.76	1.90-1.91	0.11	0.39 ± 0.01
3	7-9	1.54	0.00	1.07-1.16	2.23-2.34	0.14	0.35 ± 0.01
4	9-11	1.54	0.00	1.00-1.07	2.14-2.20	0.11	0.37 ± 0.02
5	11-14	1.54	0.00	1.00-1.09	2.14-2.23	0.13	0.21 ± 0.01
6	14-15	0.59	0.75	1.21-1.29	2.69-2.93	0.02	0.05 ± 0.02
7	15-16	0.68	0.85	1.19-1.21	2.57-2.69	-0.10	-0.24 ± 0.06
8	16-17	0.53	0.73	1.20-1.21	2.56-2.58	0.48	0.94 ± 0.17
9	17-18	0.47	0.81	1.20-1.21	2.51-2.55	0.68	0.93 ± 0.14
10	18-19	0.60	0.63	1.21	2.48-2.51	0.72	1.85 ± 0.08
11	19-20	0.61	0.65	1.21	2.46-2.49	0.56	1.26 ± 0.03
12	20-21	0.62	0.65	1.21	2.43-2.46	0.65	1.39 ± 0.11
13	21-22	0.48	0.40	1.18-1.19	2.38-2.41	0.75	1.30 ± 0.09
14	22-23	0.48	0.40	1.17-1.18	2.39-2.41	0.47	0.75 ± 0.07
15	23-24	0.36	0.47	1.19	2.42	0.36	0.60 ± 0.09
16	24-25	0.60	0.43	1.20	2.41-2.42	0.32	0.71 ± 0.09
17	25-26	0.60	0.46	1.19-1.20	2.41	0.44	0.99 ± 0.08
18	26-28	0.65	0.52	1.19-1.20	2.39-2.41	0.46	0.56 ± 0.22
19	28-29	0.57	0.61	1.15-1.20	2.34-2.35	0.24	0.70 ± 0.08
20	29-30	0.42	0.53	1.15-1.16	2.33-2.35	0.48	1.23 ± 0.17
21	30-32	0.41	0.52	1.15-1.16	2.31-2.34	0.42	0.47 ± 0.14
22	32-33	0.42	0.53	1.15-1.16	2.31-2.32	0.39	0.99 ± 0.15
23	33-34	0.44	0.80	1.15-1.17	2.30-2.31	0.23	0.55 ± 0.15
24	34-36	0.43	0.78	1.16	2.32	0.55	0.46 ± 0.17

[a] Mean ± standard deviation; $n = 3$.

To better understand whether a correlation exists between the initial CO_2 and/or CH_4 partial pressure in the biocathode headspace and the CH_4 production rate, the initial partial pressure values were plotted vs. the mean CH_4 production rate (Figure 3). During the first 5 cycles, only CO_2 was fed to the biocathode at a partial pressure of approximately 1.6 atm and the CH_4 production rate was relatively consistent. Throughout the remaining 19 cycles, the CO_2 and CH_4 initial partial pressures were varied between 0.40 atm and 0.80 atm. Despite the lower initial partial pressure of CO_2 and, thus, lower substrate availability, during the 19 biogas-fed cycles, the mean CH_4 production rate was higher than during the CO_2-fed cycles, with the exception of cycles 6 and 7, in which the biocathode was exposed to biogas for the first time. The biogas composition was varied over time, such that the CO_2 partial pressure was higher than the CH_4 partial pressure at times (cycles 13-14, and 16-18), and lower at other times (cycles 6-12, 15, and 19-24). The highest CH_4 production rates occurred during cycles in which the CO_2 and CH_4 partial pressures were nearly equal (cycles 10-13). However, an analysis of the CO_2 and CH_4 partial pressures relative to the CH_4 production rate showed that there was no statistical correlation between the partial pressures and CH_4 production rate ($p = 0.259$-0.518). The increase in CH_4 production rate during the biogas-fed cycles, in which CO_2 was less available, indicates that CO_2 was not limiting, even at lower initial partial pressures. Furthermore, the lack of correlation between CH_4 partial pressure and the CH_4 production rate indicates that CH_4 did not directly affect the CH_4 production rate at the biocathode. However, the presence of CH_4 in the biogas may indirectly affect CH_4 production if the CH_4 is transported across the proton exchange membrane to the anode,

where it may become oxidized and contribute to current in the system. Because the CH_4 partial pressure in the cathode sets the net driving pressure of CH_4 across the membrane, more CH_4 transport is expected at higher CH_4 partial pressures [7]. Thus, if CH_4 were the only biogas component to contribute to current at the anode through its oxidation, a correlation between the initial CH_4 partial pressure and the CH_4 production rate might be expected, if CH_4 did not affect the efficiency of CH_4 production at the biocathode. Because there is no direct correlation between initial CH_4 partial pressure and CH_4 production rate, it is likely that there are instead multiple other factors, in addition to CO_2 and CH_4 partial pressures, affecting the CH_4 production rate during biogas-fed cycles.

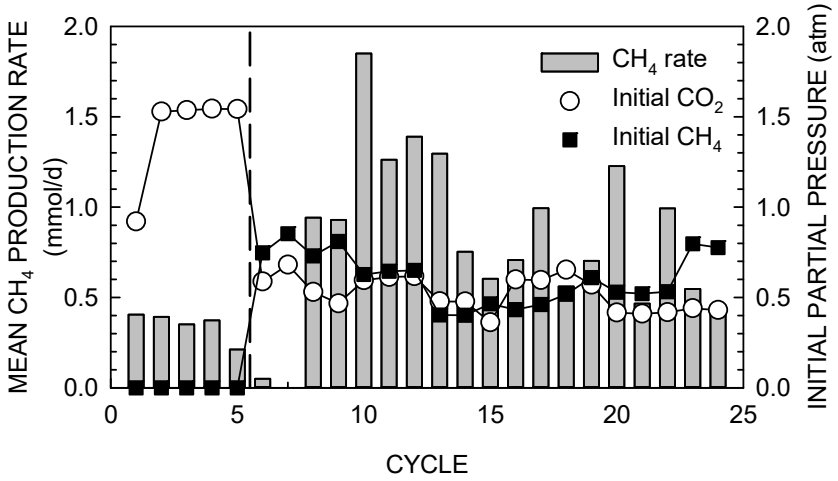

Figure 3. BES2 mean 1-d biocathode CH_4 production rate (vertical bars; left y-axis), initial CO_2 partial pressure in the cathode headspace (circles; right y-axis), and initial CH_4 partial pressure in the cathode headspace (squares, right y-axis), before and after changing the biocathode feed from CO_2 to anaerobic digester biogas (dotted vertical line).

3.3. BES2 Performance at a Range of Applied Cathode Potentials. To assess the effect of the applied cathode potential on the performance of the biogas-fed biocathode, the potential was decreased incrementally from -0.80 V to -0.50 V over seven 1-d feeding cycles. The BES performance at each applied potential is summarized in Table 2. The pH of the anolyte and catholyte remained between 6.8 and 7.2 for the duration of all feeding cycles. Anode acetate removal was lower at cathode potentials more positive than -0.80 V, although there was not a linear correlation between acetate removal and applied potential. At two potentials (-0.70 V and -0.55 V), no acetate removal was observed. Nevertheless, current was still produced at the anode during the -0.70 V and -0.55 V cycles. Furthermore, more total charge was transferred through the circuit during the -0.70 V and -0.55 V cycles than other cycles with higher acetate removal. One possible reason that the anode acetate removal did not correlate directly with the applied voltage is that acetate may been converted to carbon storage molecules in anodic biomass and utilized later for carbon oxidation, when electrons are donated to the anode [23,7]. Thus, the acetate removal rate is not necessarily equivalent to the anodic carbon oxidation rate and, therefore, correlations with the cathode applied potential may not be possible.

However, as expected, the mean CH_4 production rate generally declined as the applied potential became more positive, with the exception of slight increases between the -0.70 V and -0.65 V cycles, and between the -0.60 V and -0.55 V cycles (Table 2). At an applied potential of -0.5 V, the mean CH_4 production rate was less than half of that achieved

at -0.8 V. Because the cathode provides the reducing power to the methanogenic biofilm in the cathode, the applied potential is important for biocathode CH_4 production. In a separate study, a similar BES biocathode operated under open circuit conditions produced 99.5% less CH_4 in 1-d than when operated with an applied potential of -0.80 V [7]. The small amount of CH_4 produced under open circuit conditions was likely due to hydrogenotrophic methanogenesis using H_2 produced during the fermentation of microbial cells, or acetoclastic methanogenesis using acetate that diffused across the proton exchange membrane [7]. In the present study, the CH_4 production rate was normalized to the surface area of the proton exchange membrane to compare with other reported biocathode CH_4 production rates (Table 3). The normalized CH_4 production rate for the biocathode in this study when fed CO_2 (666 mmol/d-m^2) was comparable with other reported values for biocathodes fed with CO_2, bicarbonate or a CO_2/N_2 mixture. However, the maximum rate achieved with biogas (Table 1, cycle 10) and the biogas cycle during the -0.80 V applied potential test (Table 2) were 92% and 27% larger, respectively, than the next largest reported value. Thus, the influence of biogas feeding on biocathode CH_4 production was significant.

Table 2. Performance of BES2 when fed with biogas at various applied cathode potentials.

Cathode Potential (V vs. SHE)	Mean CH_4 Production Rate[a] (mmol/d)	Final Biocathode CH_4 [a] (%)	Final Biocathode CO_2 [a] (%)	Anode Acetate Removal[a] (%)	Anode Potential (V vs. Ag/AgCl)	Cell Potential (V)
-0.80	1.22 ± 0.07	79.9 ± 1.4	3.7 ± 0.1	21.2 ± 0.3	1.14-1.17	2.28-2.33
-0.75	0.98 ± 0.04	76.1 ± 0.9	4.1 ± 0.1	9.3 ± 1.1	1.09-1.10	2.14-2.17
-0.70	0.87 ± 0.12	78.3 ± 2.5	4.8 ± 0.1	NR[b]	1.04-1.07	2.05-2.08
-0.65	0.97 ± 0.05	78.4 ± 1.0	5.4 ± 0.1	9.1 ± 5.3	1.03-1.04	1.98-2.01
-0.60	0.74 ± 0.04	72.4 ± 0.9	6.2 ± 0.2	13.4 ± 0.4	1.02-1.04	1.93-1.94
-0.55	0.86 ± 0.14	70.7 ± 2.7	6.4 ± 0.1	NR	1.02-1.05	1.88-1.90
-0.50	0.53 ± 0.08	76.7 ± 1.7	8.3 ± 0.1	4.0 ± 0.1	0.16-0.36	1.15-1.34

[a] Mean ± standard deviation; $n = 3$.
[b] No removal.

Table 3. Comparison of reported CH_4 production rate values for methanogenic biocathodes, normalized to the surface area of the proton exchange membrane.

CH_4 Production Rate (mmol/m^2-d)	Reference
200	[1]
603	[2]
666	Present study, CO_2-fed (Table 1, Cycle 1)
1,067	[3]
1,103	[4]
1,519	[6]
1,562	[7]
1,980	Present study, biogas-fed (Table 2, -0.80 V)
3,003	Present study, biogas-fed (Table 1, Cycle 10)

In the present study, while the final CH_4 content of the cathode biogas varied, the highest CH_4 fraction (80%) was achieved at -0.80 V and the lowest fraction (71%) at -0.55 V. The final CO_2 content increased almost linearly ($R^2 = 0.934$) with increasing (i.e., more positive) applied potentials, ranging from 4% in the -0.80 V cycle to 8% in the -0.50 V cycle. The remainder of the gas in the cathode at the end of each cycle was N_2, which came with the

biogas or was transported from the anode headspace to the cathode. H_2 was not detected in the headspace of the cathode at the end of the cycles.

The maximum anode potential, as measured by an adjacent Ag/AgCl reference electrode, occurred at the beginning of each feeding cycle, and declined only slightly from 1.17 to 1.07 V (vs. Ag/AgCl) between the -0.80 V and -0.70 V cycles (Table 2). Then, the maximum anode potential remained relatively constant (1.04-1.05 V vs. Ag/AgCl) for the -0.65 V through -0.55 V cycles. When -0.50 V was applied to the cathode, the maximum anode potential declined by 66% from its value during the -0.55 V cycle (0.36 V vs. Ag/AgCl), indicating a threshold cathode potential between -0.50 V and -0.55 V, at which the relatively high anode potential could not be sustained.

The total charge transferred through the circuit, from anode to cathode, during the last CO_2-fed cycle of BES2 (at -0.8 V) was 148 C. However, during the -0.8 V cycle with the biogas, 200 C of total charge was transferred (Figure 4), which represents 35% more electron transfer than in the CO_2 cycle. One possible reason for the increased charge transfer is transport of trace gases from the cathode to the anode and their subsequent oxidation at the anode. For each of the biogas-fed cycles (-0.80 V to -0.50 V), the maximum possible contribution of CH_4, NH_3 and H_2S to the current was estimated (Figure 4). Although 1 mole of NH_3 theoretically releases 6 moles of electrons (Equation 2), the maximum estimated charge transfer from NH_3 oxidation at the anode was relatively insignificant (5.5×10^{-6} C) because NH_3 is a very small component of the biogas ($\leq 0.00013\%$, v/v), exerting a mean partial pressure of $1.26 \times 10^{-6} \pm 0.11 \times 10^{-6}$ atm in the biogas throughout this study. Thus, because NH_3 preferentially partitions to the aqueous phase, a very small amount of NH_3 is carried with biogas into the BES cathode. Therefore, the two most important reduced components of biogas that may donate electrons to the anode are CH_4 and H_2S (Figure 4). However, even CH_4 and H_2S are likely to be relatively minor components of the BES current generation at more negative applied cathode potentials (i.e., $\leq$ -0.70 V). At more positive potentials, CH_4 and H_2S may play a more prominent role in the current production and subsequent biocathode methane production. Previous studies have shown that bioanodes are capable of oxidizing H_2S into SO_4^{2-} (Equation 3), $S^0_{(s)}$ (Equation 4) and other oxidized sulfur species [15]. More recently, a study described CH_4 oxidation coupled with current generation at a bioanode [24]. Therefore, it is important to further examine the effect of these biogas components (particularly CH_4 and H_2S) on the performance of the entire BES (i.e., biocathode and bioanode).

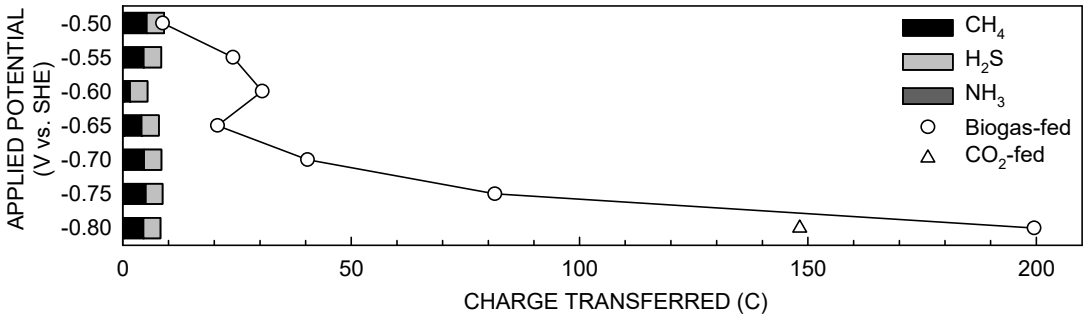

Figure 4. Total charge transferred in a 1-d cycle of BES2 when the biocathode was fed with CO_2 at -0.80 V (triangle), or biogas (circle) at various applied cathode potentials, overlaid with the estimated maximum total charge that the biogas components (CH_4, NH_3 and H_2S) could contribute by migrating to the anode and becoming oxidized.

At the most negative cathode potential (-0.80 V), the difference in charge transfer between the biogas-fed and CO_2-fed cycles cannot be completely explained by the presence of trace gases (Figure 4). Thus, other mechanism(s) may be involved in the observed increase in charge transfer following biogas feeding, which provides a lower cathode substrate (CO_2) concentration than during feeding with 100% CO_2.

The biocathode CH_4 production was significantly affected over the course of the first two feeding cycles with biogas (Figure 3), possibly indicating a shift in microbial metabolism or microbial community composition. However, the effect of biogas feeding on the microbial community is not currently known; analysis of the 16S rRNA gene sequencing of the anode and cathode biofilms before biogas exposure and after biogas feeding for approximately 40 cycles is currently underway. It is likely that a change in the microbial community of both the anode and cathode affected the biocathode CH_4 production. Although the biocathode archaeal community consisted primarily of a phenotype most closely related to *Methanobrevibacter arboriphilus*, the bacterial community composition has a significant effect on biocathode CH_4 production [2], which might be affected by the trace gases typically present in anaerobic digester biogas.

CONCLUSIONS

In the biogas-fed cycle at -0.80 V applied cathode potential, 35% more charge was transferred from the anode to the cathode than in the 100% CO_2-fed cycles, despite the lower biocathode substrate (i.e., CO_2) availability. However, only 16% of the increase in charge transfer could be explained by the oxidation of trace gases transported from the cathode to the anode. Therefore, other mechanism(s) must be responsible for the observed increased charge transfer and increased CH_4 production rates when the biocathode was fed with anaerobic digester biogas, as compared with 100% CO_2 feedings. It is likely that a change in microbial community and/or metabolism due to the presence of trace gases was a significant contributor to the increase in charge transfer and CH_4 production when the biocathode was fed with biogas instead of CO_2. Microbial community analysis of anode and cathode biofilm 16S rRNA genes before and after exposure to biogas is currently under investigation.

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