Supplemental Information

Enrichment of microbial electrolysis cell (MEC) biocathodes from sediment microbial fuel cells (sMFC) bioanodes

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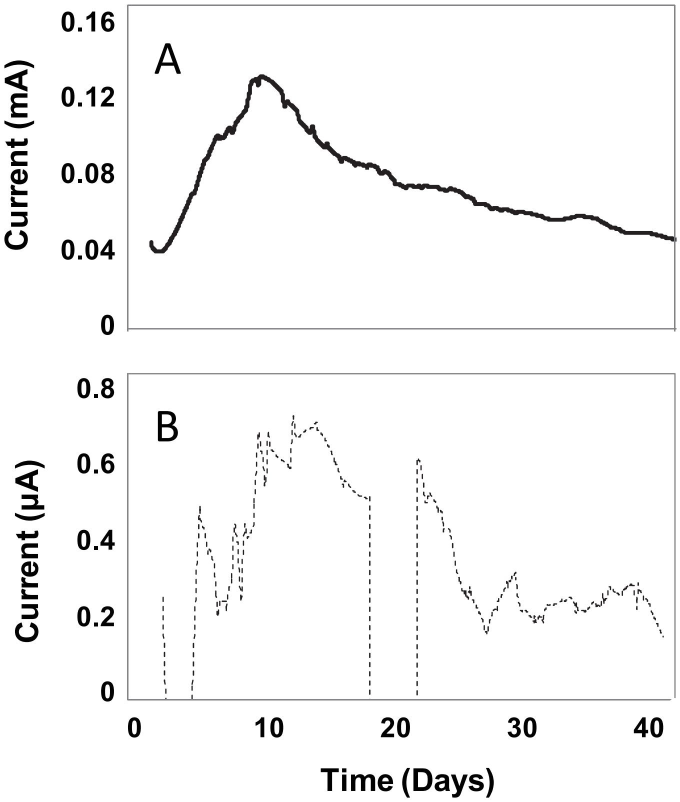

FIG. S1. Test tube sMFCs inoculated with (A) Harbor silt or (B) Bay sand developed exoelectrogenic activity as recorded by voltage drop across 1k Ohm external resistor. For these unpoised sMFCs exoelectrogenic activity peaks within 10 to 14 days and current amplitude is higher for Harbor than Bay, likely a result of the higher organic material or fine grain structure of this substrate. Moving average trend-line of 150 data points is shown to minimize fluctuation attributable to periodic addition of water lost to evaporation

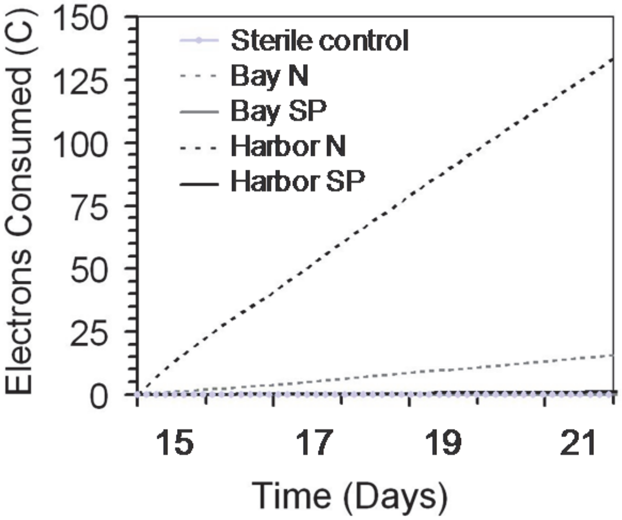

FIG. S2 Consumption of electrons in Coulombs at an applied WE potential of −539 mV in PBS media degassed with N_2/CO_2 ratio of 80%/20% [vol/vol] after 7 days

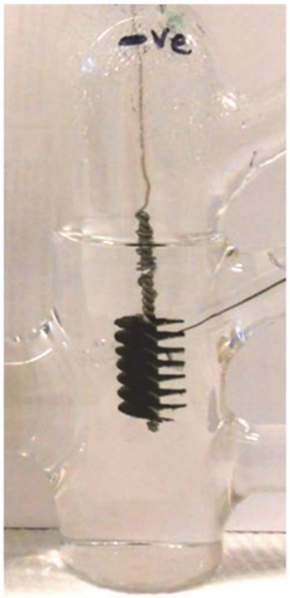

FIG. S3. Sterile control brush cathode developed no biofilm growth and cathode medium remained clear after week-long operation at applied potentials of −339 mV, −439 mV then −539 mV (shown)

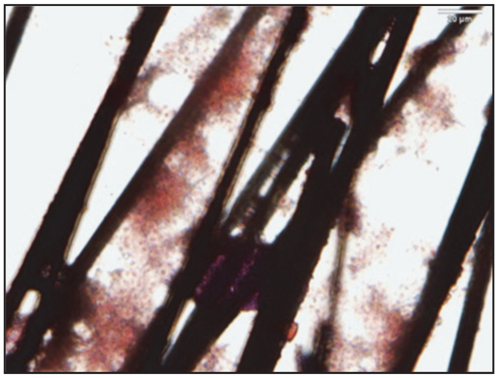

FIG. S4. Micrograph of Gram stained graphite fibers recovered from the functional biocathode produced from Harbor N sample prior to 16s Clone Library analysis on day 61. Graphite fibers appear to show closely associated film, including possible biofilm microorganisms, on this electron-accepting, biogas-producing cathode (Total Magnification, 400x).

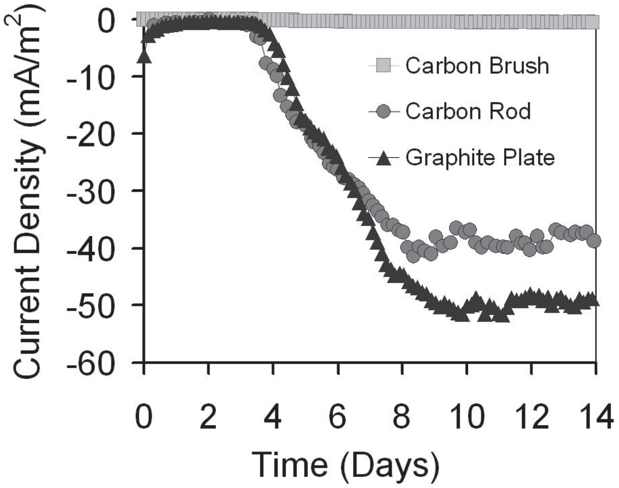

FIG. S5. Development of negative current (expressed as current density) after transfer of biocathode inoculum, obtained from Harbor N cathode chamber on day 45, into three new BES reactors each with different cathode material (carbon fiber brush, carbon rod, or graphite plate) at an applied WE potential of −539 mV in PBS minimal media degassed with N_2/CO_2 ratio of 80%/20% [vol/vol] for 14 days

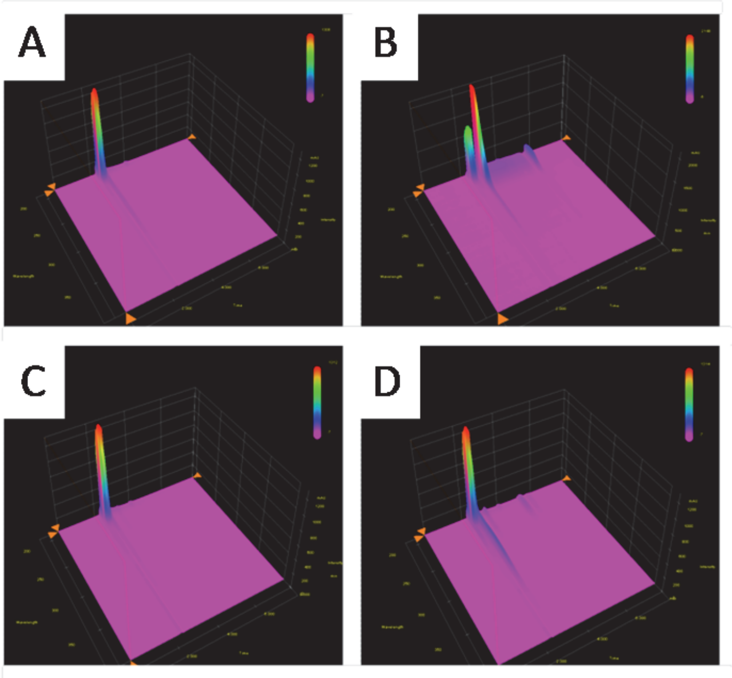

FIG. S6. HPLC analysis showing: sterile media (a), sterile media spiked with acetate (b), two week old cathode media from Bay SP reactor (c), and two week old cathode media from Bay N (d). Retention time is shown on X axis, absorbance is shown on Y axis and amplitude of absorbance depicted by peak height on Z axis

Table S-T1: Electrons transferred from cathode, biogas (CH_4 and H_2) released to headspace, and total inorganic carbon (T.I.C.) depleted from cathode compartment over 7 day operations at applied cathode potentials of –439 mV or –539 mV vs SHE. Media degassed with N_2/CO_2 ratio of 80%/20% [vol/vol] prior to start of each week

Species	**Bay SP**		**Harbor SP**		**Bay N**		**Harbor N**	
	–439	–539	–439	–539	–439	–539	–439	–539
Q (C)	0.11	0.08	0.62	0.93	0.68	15.49	84.00	135.00
e^- (mmol)	0.00	0.00	0.01	0.01	0.01	0.16	0.87	1.40
T.I.C. (mmol)	0.24	0.43	-0.18	0.49	1.07	0.83	1.72	2.71
CH_4 (mmol)	nd	nd	trace	nd	nd	nd	0.61	0.45
H_2(mmol)	nd	nd	trace	trace	nd	nd	nd	trace

(nd, not detected)

Calculation of total inorganic carbon consumed

We assume equilibrium at pH 7 for following calculations.

First, we adjust Henry's constant $k_{H,pc}(25\ °C) = 29.41\ L\ atm\ mol^{-1}$ [2, 3] for $T = 30\ °C = 303.15\ K$ according to van 't Hoff equation [1] using $C = 2400\ K$ for CO_2 [3]:

$$k_{H,pc}(T) = k_{H,pc}(T^{\Phi}) \exp\left[-C\left(\frac{1}{T} - \frac{1}{T^{\Phi}}\right)\right] \quad (1)$$

$$k_{H,pc}(30°C) = 29.41 \exp\left[-2400\left(\frac{1}{303.15} - \frac{1}{298.15}\right)\right] = 33.59\ \frac{L\ atm}{mol} \quad (2)$$

k_H: Henry's law constant; T: thermodynamic temperature; T^{Φ}: standard temperature (298.15 K); C: constant [3].

Using Henry's law [1],

$$k_{H,pc} = \frac{p_{CO2}}{c_{CO2(aq)}} \quad (3)$$

We can calculate concentration of dissolved CO_2 for $p_{CO2} = 0.2$ atm (20% CO_2) and $k_{H,pc} = 33.59\ L\ atm\ mol^{-1}$

$$c_{CO2(aq)} = \frac{p_{CO2}}{k_{H,pc}} = \frac{0.2}{33.59} = 0.00595\ \frac{mol}{L} \quad (4)$$

$c_{CO2(aq)}$: aqueous molar concentration of CO_2; p_{CO2}: partial pressure of CO_2

We obtain moles of $CO_2(aq)$

$$n_{CO2(aq)} = 0.00595 \cdot 120 \cdot 10^{-3} = 0.714\ mmol \quad (5)$$

Assuming pH 7, we can calculate each species of the carbonic acid system in equilibrium. Since about 99% of carbonic acid (H_2CO_3) is in the form of $CO_2(aq)$ [4], we expect according to equation (4)

$$[CO_{2_{(aq)}}] = [H_2CO_3^*] = 5.95\ mM \quad (6)$$

Moles of carbonic acid species: $n_{H2CO3*} = 0.714$ mmol

In order to calculate bicarbonate concentration [4], we use

$$\mathrm{K_{a,1}} = \frac{[\mathrm{H^+}][\mathrm{HCO_3^-}]}{[\mathrm{H_2CO_3^*}]} \qquad (7)$$

We obtain

$$[\mathrm{HCO_3^-}] = \frac{10^{-6.3} \cdot 5.95\ \mathrm{mM}}{10^{-7}} = 29.82\ \mathrm{mM} \qquad (8)$$

Moles of bicarbonate species: $n_{HCO3^-} = 3.578$ mmol

To calculate concentration of carbonate species [4], we use

$$\mathrm{K_{a,2}} = \frac{[\mathrm{H^+}][\mathrm{CO_3^{2-}}]}{[\mathrm{HCO_3^-}]} \qquad (9)$$

We obtain

$$[\mathrm{CO_3^{2-}}] = \frac{10^{-10.3} \cdot 29.82\ \mathrm{mM}}{10^{-7}} = 0.0149\ \mathrm{mM} \qquad (10)$$

Moles of carbonate species: $n_{CO3^{2-}} = 0.002$ mmol

Therefore, moles of dissolved total inorganic carbon $n_{i.c.(aq)}$ is

$$\mathrm{n_{i.c.(aq)}}(t_0) = \mathrm{n_{H_2CO_3^*}} + \mathrm{n_{HCO_3^-}} + \mathrm{n_{CO_3^{2-}}}$$

$$= 0.714 + 3.578 + 0.002 = 4.32\ \mathrm{mmol} \qquad (11)$$

Calculation of moles $n_{i.c.(aq)}$ after 7 days:

Assuming that ratio of $\frac{n_{CO2(g)}}{n_{i.c.(aq)}} = \mathrm{const.}$ [4]; we have

$$\frac{\mathrm{n_{CO2(g)}(t_0)}}{\mathrm{n_{i.c.(aq)}(t_0)}} = \frac{\mathrm{n_{CO2(g)}(t_7)}}{\mathrm{n_{i.c.(aq)}(t_7)}} \qquad (12)$$

$n_{CO2(g)}$ (t_n): moles of headspace CO_2 gas after n days;

$n_{i.c.(aq)}$ (t_n): moles of dissolved inorganic carbon after n days

For “Harbor N” at -439 mV (2nd week), we have

$$n_{\text{i.c.(aq)}}(t_7) = \frac{0.427 \cdot 4.32}{0.653} = 2.83 \text{ mmol} \qquad (13)$$

Consumed moles of dissolved inorganic carbon:

$$n_{\text{i.c.(aq)}}(\text{consumed}) = n_{\text{i.c.(aq)}}(t_0) - n_{\text{i.c.(aq)}}(t_7) = 4.32 \text{ mmol} - 2.83 \text{ mmol} = 1.49 \text{ mmol} \qquad (14)$$

Overall moles of total inorganic carbon, $n_{\text{t.i.c.}}$, consumption is:

$$n_{\text{t.i.c.}}(\text{consumed}) = n_{CO2(g)}(\text{consumed}) + n_{\text{i.c.(aq)}}(\text{consumed}) = 0.225 + 1.49 = 1.72 \text{ mmol} \qquad (15)$$

Calculation of electrons consumed

For “Harbor N” sample at -439 mV (2nd week), we have

$$\frac{Q}{96485\ C/F} \ \frac{1 \text{ mole e}^-}{1 \text{ F}} = \frac{84\ C}{96485\ C/F} \ \frac{1 \text{ mole e}^-}{1 \text{ F}} = 0.87 \text{ mmole e}^- \qquad (16)$$

Q: electric charge

References:

(1) Physical Chemistry, P.W. Atkins, Oxford University Press, 2006

(2) CRC Handbook of Chemistry and Physics, 76th Edition, D.R. Lide and H.P.R. Frederikse, ed(s), Inc., Boca Raton, FL, 1995

(3) The Solubility of Gases in Liquids (1966) R. Battino and H. Lawrence Clever, Chem. Rev., 66(4), 395-463

(4) Aquatic Chemistry, 3rd Edition, W. Stumm and J.J. Morgan, Wiley-Interscience, Inc. ,NY, 1996