## **Supporting Information**

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## **SI Materials and Methods**

**Cultivation.** Geobacter sulfurreducens strain DL1 (51573; ATCC) was maintained under anaerobic condition (80:20 N2:CO<sub>2</sub>) in freshwater media containing 40 mM fumarate and 10 mM sodium acetate as described previously (1–4). Cysteine (10  $\mu$ M) was added as a reductant.

Interdigitated Microelectrode Arrays Fabrication. Interdigitated microelectrode arrays (IDAs) (5) were fabricated in house photolithographically using a sacrificial resist (NR 71-1000PY; Futurex) by deposition of 100 Å titanium and 1,000 Å gold onto piranha etch-cleaned microscope slides using a Temescal e-beam metal evaporator. The photoresist (NR7-3000P) was used as a blocking layer to ensure that only the microelectrode bands of the array were exposed. Masks were designed in house using L-Edit software. Blanks of a layer of low reflective chrome covered with AZ photoresist were exposed in a Heidleberg DWL-66 laser pattern generator. The patterned gold electrode was connected to a copper-coated circuit board and shielded cable before packaging in water-insulating epoxy (Scotchcast; 3M). The IDAs were tested by cyclic voltammetry in (ferrocenylmethyl)trimethylammoniumhexafluorophosphate (6) in phosphate buffer with 100 mM potassium chloride to ensure that IDAs functioned electrochemically as expected. Before use in Geobacter culture, IDAs were autoclaved for sterility. The biofilm was grown using established methods (4) by poising both electrodes as anodes at +0.300 V vs. Ag/AgCl [approximately +0.500 V vs. standard hydrogen electrode (SHE)] in media containing excess acetate (10 mM) until a self-determined limiting catalytic current of 50  $\mu$ A (25  $\mu$ A for each electrode) was achieved, corresponding to a biofilm thickness of 18 µm (on average) that was sufficiently thick to span the gap between adjacent interdigitated microelectrode bands (Fig. S1).

**Electrochemical Measurements.** All electrochemical experiments were performed in a 250-mL water-jacketed single-chamber electrochemical cell (AFCELL3; Pine Research Instrumentation) maintained at 30 °C in a similar manner as pervious described

- 1. Bond DR, Lovley DR (2003) Electricity production by Geobacter sulfurreducens attached to electrodes. *Appl Environ Microbiol* 69:1548–1555.
- Caccavo F, Jr., et al. (1994) Geobacter sulfurreducens sp. nov., a hydrogen- and acetateoxidizing dissimilatory metal-reducing microorganism. *Appl Environ Microbiol* 60: 3752–3759.
- Strycharz-Glaven SM, Snider RM, Guiseppi-Elie A, Tender LM (2011) On the electrical conductivity of microbial nanowires and biofilms. *Energy Environ Sci* 4:4366–4379.
- Strycharz SM, et al. (2011) Application of cyclic voltammetry to investigate enhanced catalytic current generation by biofilm-modified anodes of Geobacter sulfurreducens strain DL1 vs. variant strain KN400. Energy Environ Sci 4:896–913.

(3, 4, 7) with the following exception. The two electrode experiments were performed using a biopotentiostat (AFCBP1; Pine Instruments), a counter electrode (graphite rod), and a reference electrode (Ag/AgCl, 3 M KCl; CH Instruments).

Microscopy. Representative biofilm-modified IDAs were prepared for SEM using the method in the work by Rollefson et al. (8) in which samples were fixed in 2% (vol/vol) gluteraldehyde in 0.15 M sodium cacodylate at pH 7.4 with 0.15% safranin O, post-fixed for 2 hr in 1.5% (vol/vol) osmium tetroxide, dehydrated using serial dehydration in ethanol, treated with hexamethyldisilazane (HMDS), affixed to standard SEM mounts, sputter coated with a thin layer of platinum, and imaged using a Carl Zeiss SMT Supra 55 scanning electron microscope at 5 kV. Representative biofilm-modified IDAs were imaged using confocal laser scanning microscopy. The IDAs were removed from electrochemical reactors after completion of electrochemical measurements and rinsed two times in 1× PBS, pH 7.4 (10× PBS solution, EN7859CX; Excelleron). Biofilms were stained according to the manufacturer's instructions with the LIVE/DEAD BacLight Bacterial Viability Kit (L7012; Invitrogen). Staining was carried out in 1× PBS, pH 7.4, for 10 min at room temperature in the dark. After incubation with the stain, IDAs were rinsed one time with 1× PBS, pH 7.4, and allowed to destain in 1× PBS, pH 7.4, for 10 min. IDAs were broken off from the electrode housing unit and mounted into a single-welled chambered coverglass slide (155361; Lab Tek) with several microliters of mounting oil (Prolong Gold Antifade, P36930; Invitrogen). Biofilm imaging was carried out using a Nikon TE-2000e inverted confocal microscope (Nikon) with a Nikon CFI Apo TIRF 100× (n.a. 1.49) oil objective. Two wavelengths, 488 and 514 nm, were used to excite the fluorescent stains. A mininimum of eight fields were imaged and processed with ImageJ software program (http:// imagej.nih.gov/ij/). Three random image stacks were used to determine the mean biofilm height by measuring the height at 18 random points for each stack using ImageJ.

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- Szentirmay MN, Martin CR (1984) Ion-exchange selectivity of Nafion films on electrode surfaces. Anal Chem 56:1898–1902.
- Strycharz-Glaven SM, Tender LM (2012) Study of the mechanism of catalytic activity of G. sulfurreducens biofilm anodes during biofilm growth. *ChemSusChem* 5: 1106–1118.
- Rollefson JB, Stephen CS, Tien M, Bond DR (2011) Identification of an extracellular polysaccharide network essential for cytochrome anchoring and biofilm formation in Geobacter sulfurreducens. J Bacteriol 193:1023–1033.



**Fig. S1.** Confocal laser scanning microscopy of a fully grown biofilm on an IDA. *Upper* is a representative slice of the biofilm perpendicular to the IDA surface. The mean biofilm height (n = 17) was 18.0  $\mu$ m. *Lower* is a representative slice of the biofilm parallel to the IDA surface. (Scale bar: 10  $\mu$ m.)

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