Experimental

Chemicals. Unless otherwise specified, all chemicals were purchased from Sigma-Aldrich GmbH (Schnelldorf, Germany). All solutions were prepared using water purified with PURELAB UHQ II system from ELGA Labwater (High Wycombe, UK) or with a Milli-Q system (Millipore, Milford, CT, USA).

Enzymes. Trametes hirsuta Lc was obtained from the basidiomycete *Trametes hirsuta* (Wulfen) Pilát, strain *T. hirsuta* 56, provided by the laboratory collection of the Moscow State University of Engineering Ecology (Russia). The basidiomycete was grown by submerged cultivation and the Lc was isolated from a culture medium as describe in.[19] The homogeneous preparation of the enzyme (protein concentration 10 mg/mL), as judged from SDS-PAGE, was stored in 100 mM phosphate buffer, pH 6.5 at –20 °C.

Dichomera saubinetii CDH was purified from the culture supernatant of the ascomycete *Dichomera saubinetii*, CBS 990.70, obtained from the Centralbureau voor Schimmelcultures (Baarn, The Netherlands). The procedures for cultivation and purification of the enzyme were similar to *Myriococcum thermophilum* CDH.[18] The homogeneous preparation of the enzyme (protein concentration 2.3 mg/mL) was stored in 50 mM acetate buffer, pH 5.5 at –80 °C.

Electrochemical measurements.

Electrode preparation. The enzyme-modified electrodes serving as working electrodes were made from rods of spectrographic graphite electrodes (SPGE, Ringsdorff Werke GmbH, Bonn, Germany, type RW001, 3.05 mm in diameter). The surface of the SPGE was prepared by first polishing with fine emery paper (Tufback Durite, P1200), then thoroughly rinsed with Millipore water and finally allowed to dry.

Electrode bio-modification. Biocathodes were prepared by placing an aliquot of 10 µl of enzyme solution (10 mg/mL of Lc) on the electrode surface, allowed the solution to dry at the surface of the electrode in air at room temperature for 20 min. After that the electrode was again rinsed with water. Bioanodes were prepared by placing an aliquot of 5 µL of enzyme solution (2.3 mg/ml of CDH) on the entire surface of the electrode. The electrode was dried in air at room temperature for 15 min and then stored overnight at 4 °C. Before use, the electrode was thoroughly rinsed with Milli-Q water to remove weakly adsorbed enzyme.

Voltammetry. Polarisation curves were recorded using linear scan voltammery with a scan rate of 1 mV/s. Bio-cathodes were studied in an one-compartment electrochemical cell containing 20 mL of solution. $Hg|Hg_2Cl_2|KCl_{sat}$ (242 mV *vs.* NHE) and a platinum mesh were used as the reference and the counter electrodes, respectively. A potentiostat/galvanostat 2059 combined with the function generator 7800 from Amel Instruments (Milano, Italy) was used to control the potential at the electrodes. Bio-anodes were studied in an electrochemical cell of 50 mL containing a $Ag|AgCl|KCl_{sat}$ (197 mV *vs.* NHE) reference electrode and a platinum foil counter electrode operated by electrochemical analyser (BAS CV 50W, Bioanalytical Systems, West Lafayette, IN, USA).

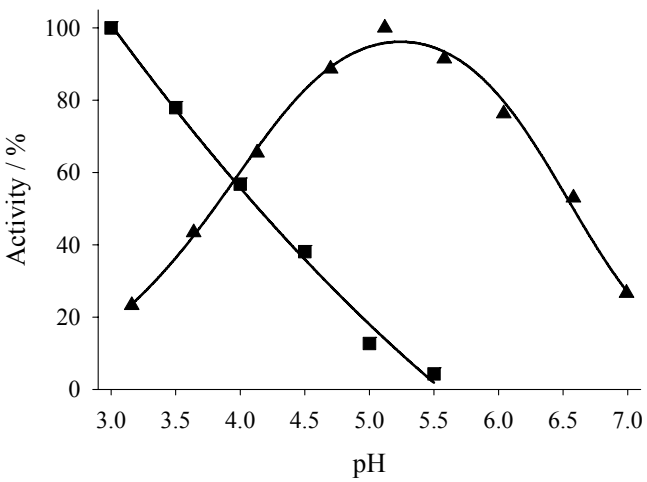
Steady-state potential measurements. For steady-state potential measurements an AUTOLAB PGSTAT 30 (EcoChemie, Utrecht, The Netherlands) equipped with GPES 4.9 software was used. The reference electrode was an $Ag|AgCl|KCl_{sat}$ electrode (Radiometer, Copenhagen, Denmark) and the enzyme-modified electrodes were used as the indicator electrodes placed in a one-compartment 50 mL electrochemical cell. The equilibrium potential values were registered under air saturated conditions.

Studies of the biofuel cell performance. The AUTOLAB PGSTAT 30 was used in potentiostat mode connecting the bioanode (CDH-modified SPGE) as the working electrode and the biocathode (Lc-modified SPGE) as a combined reference and counter electrode. For the time stability tests, an external load of 1 MΩ was connected between the two electrodes and the potential was measured with time both with and without stirring. The power output of the cells was calculated from the potential and resistance values according to Ohm's law. In this work all potentials are given *vs.* NHE.

Supplementary Table S1. Steady-state potentials (mV, *vs.* NHE) of spectrographic graphite electrodes modified with CDH or Lc (air saturated 0.1 M citrate-phosphate buffer, pH 4.5 with or without 5 mM sugars).

Conditions	Anode	Cathode
Air-saturated buffer	150	850
Air-saturated buffer + Glucose	110	840
Air-saturated buffer + Cellobiose	90	850
Air-saturated buffer + Lactose	85	855

Supplementary Fig. S1. DET activity versus pH for *D. saubinetii* CDH (triangles) and *T. hirsuta* Lc (squares).



Supplementary Fig. S2. Polarisation curve recorded with CDH-modified SPGE as the working electrode and Lc-modified SPGE as combined reference and counter electrode (scan rate: 1 mV s^{-1}, starting potential: -800 mV). As fuels, 5 mM lactose, cellobiose or glucose solutions (0.1 M citrate-phosphate air-saturated buffer, pH 4.5) were used. The measurements were performed in quiet (non stirred) conditions (filled symbols) and at 800 rpm (empty symbols).