

## ABSTRACT

Hydrogen production using fermentation leaves behind many soluble substrates like acetate. In this project, we use Microbial Electrolysis Cells to produce hydrogen from cellulose fermentation end products. We successfully produced hydrogen using a synthetic solution of cellulose fermentation end products with first an unacclimated inoculum, then a pre-acclimated inoculum taken from cultures acclimated to each single substrate present in the fermentation effluent which increased hydrogen yield from 799.9 ± 292.9 to 975.1 ± 114.8 mL H<sub>2</sub>/g COD. We also tested real cellobiose and corn stover effluents and obtained 1030.4 ± 301.5 and 749.4 ± 175.7 mL H<sub>2</sub>/g COD respectively.

## Introduction

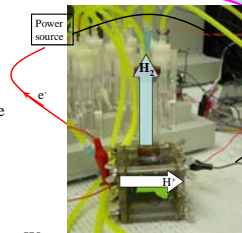
Hydrogen production is becoming increasingly important in view of using hydrogen in fuel cells. However, most of the production of hydrogen so far comes from the combustion of fossil fuels. Ecologically clean and renewable methods of producing hydrogen are microbial cellulose fermentation and Microbial Electrolysis Cells (MECs), also known as bioelectrochemically assisted microbial reactors or BEAMRs. The fermentation of biomass does not recover all the energy available as hydrogen and there are still many soluble products like acetate left in solution after fermentation has been completed.

In this project, we hypothesized that hydrogen could still be produced by the soluble fermentation products (acetate, lactate, formate, succinate and ethanol) using a MEC. In a MEC, soluble substrates are oxidized by the bacteria which release electrons to an electrical circuit. At the cathode these electrons combine with protons from the solution, with the addition of a small voltage, to produce hydrogen.

## Materials and Methods

### 1. Materials

- Reactors:
  - > 4 cm wide reactor
  - > brush anode, Pt on carbon cloth cathode
  - > 8 cm gas collection tube,
  - > Gas bags for gas collection
- Inoculum:
  - > Primary clarifier effluent from PSU Waste Water Treatment Plant
  - > Pre-acclimated cultures to the different single substrates (acetic acid, succinic acid, lactic acid, formic acid, ethanol)
- Substrates:
  - > **Synthetic effluent:** 26mM acetic acid, 5.6 mM succinic acid, 1.8 mM lactic acid, 0.6 mM formic acid, 14 mM ethanol + 50 mM PBS buffer + Minerals + Vitamins → COD=3.7g/L, pH=7
  - > **Cellobiose fermentation effluent:** pH=7.2, COD=4.2 g/L
  - > **Corn Stover fermentation effluent:** pH=7.3, COD=4.9 g/L



### 2. Methods

- Start-up:
  - > First operated in MFC mode for inoculation until 3 cycles obtained same maximum voltage then switched to MEC mode.
- MEC mode:
  - > Voltage applied: 0.5 V
  - > Sparged with N<sub>2</sub> before each batch
  - > Gas production and flow rate monitored with respirometer
  - > Gas analysis of reactor and gas bag using Gas Chromatograph

## RESULTS

### Gas analysis

#### Gas production (Table 1):

- > Acclimation increased and stabilized gas production
- > Acclimation increased proportion of hydrogen produced
- > Real fermentation effluents produce about the same amount of gas but with less hydrogen and less stable composition than synthetic effluent

#### Hydrogen maximum production rate (Table 1):

- > Acclimation improved production rate, close to single substrate rate
- > Real effluents lower production rates but still same order of magnitude

substrate	total gas production (mL)	%H <sub>2</sub>	%CH <sub>4</sub>	%CO <sub>2</sub>	Q (m <sup>3</sup> H <sub>2</sub> /m <sup>3</sup> /d)
synthetic (non-accli)	97.6 ± 31.5	76.0 ± 6.5	10.7 ± 8.8	13.3 ± 3.6	0.59 ± 0.21
synthetic (accli)	111.8 ± 10.2	79.1 ± 2.5	10.0 ± 2.3	10.7 ± 1.1	1.11 ± 0.13
cellobiose effluent	104.6 ± 16.99	69.2 ± 4.1	16.4 ± 3.7	14.4 ± 1.4	1.00 ± 0.19
corn stover effluent	96.8 ± 16.1	69.0 ± 6.3	12.4 ± 3.2	18.6 ± 3.4	0.96 ± 0.16
acetic acid	66.06 ± 3.3	88.6 ± 1.6	0.0 ± 0.0	11.4 ± 1.6	1.17 ± 0.07

Table 1: gas production and composition averaged over 2 reactors run in duplicates for 3 batches

#### Gas yield (Figure 1):

- > Acclimation increased and stabilized hydrogen yield
- > Cellobiose effluent increased hydrogen yield but also methane and carbon dioxide yields
- > Lower yields for all fermentation effluents than pure acetate substrate

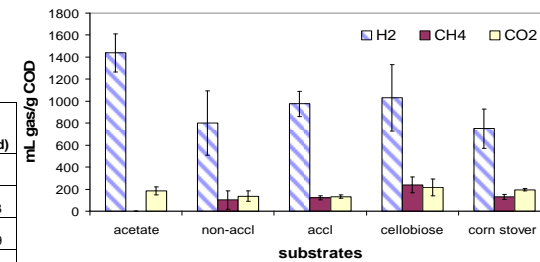


Figure 1: gas yield in mL gas produced/g COD consumed averaged over 2 reactors run in duplicates for 3 batches

### Energy efficiencies, H<sub>2</sub> recovery

#### Electrical energy efficiency

$$\eta_e = \frac{\text{energy in } H_2 \text{ produced}}{\text{energy input from power source}}$$

- > over 150%
- > much better with acclimation (100% higher efficiency)
- > same order of magnitude as single substrate

substrate	n <sub>e</sub>	n <sub>ess</sub>	RH <sub>2</sub> (%)
non-accli	152.2 ± 49.6	46.0 ± 16.0	51.5 ± 18.8
accli	269.4 ± 22.2	62.3 ± 6.8	62.7 ± 7.4
corn stover	225.4 ± 53.4	48.9 ± 11.5	48.2 ± 11.3
cellobiose	222.8 ± 27.1	61.3 ± 14.2	66.3 ± 19.4
acetic acid	278.3 ± 35.5	84.2 ± 8.8	92.6 ± 11.2

Table 2: electrical energy efficiency, overall energy efficiency, hydrogen recovery rate

#### Hydrogen recovery

- > Over 50% for synthetic substrate but less than single substrate
- > Acclimation improved recovery by more than 10%
- > Cellobiose effluent best hydrogen recovery, more than 65%

#### Overall energy efficiency

$$\eta_{ess} = \frac{\text{energy in } H_2 \text{ produced}}{\text{energy input from power source and substrate}}$$

- > over 45%
- > much better with acclimation (15% increase)
- > same order of magnitude as single substrate

## CONCLUSIONS

- ✓ Hydrogen production has been achieved in MEC reactors using a synthetic solution containing soluble fermentation products and real cellulose fermentation effluents.
- ✓ Acclimated inoculum improved hydrogen production, hydrogen maximum production rate, energy recoveries and hydrogen recovery
- ✓ More than 200% electrical energy recovered as hydrogen with real fermentation effluents and good hydrogen recovery compared to fermentation (only 20% with cellulose fermentation compared with up to 66% from the fermentation effluent in this study)
- ✓ Cellobiose fermentation effluent is the most promising fermentation effluent to use in MEC (higher hydrogen recovery and production rate)
- ✓ Still problematic methane production (10% of the gas produced with synthetic effluent and around 15% for real effluents)

## Future Work

Future experiments will investigate:

- > Continuous mode MEC system
- > Pure culture work to avoid methanogenesis

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