WP3 Biochemical Fuel Production

Truus de Vrije, Ana López Contreras

WFBR

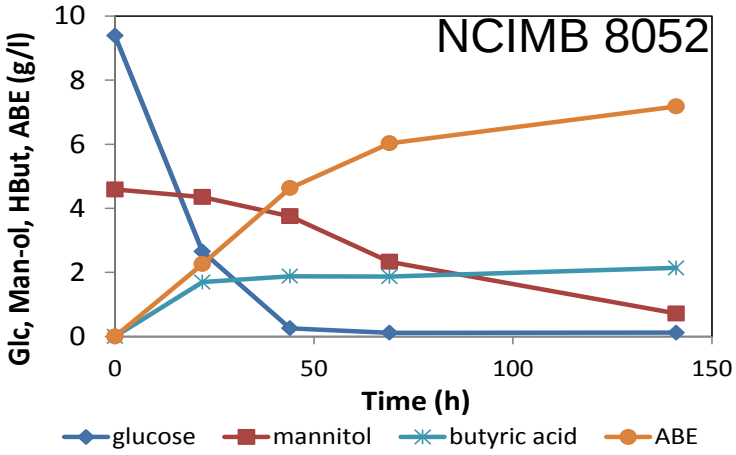
04-07-2017

Task 3.3 Efficient fermentation of seaweed to ABE (M1-40)

- ABE production from brown seaweed
- Improve substrate consumption, mannitol
- Consolidated bio-processing

ABE production from *Laminaria digitata*

- Hydrolysate prepared by DTI (MAB3 project)
- Very low salt content (conductivity 4.8 mS/cm)
- 24 g/l of soluble sugars
  - glucose 10.0; glucan 9.9; mannitol 3.3 g/l
- 90% substrate consumption
- 7.2 g/l of ABE by *C. beijerinckii*
- $Yield_{ABE}$ = 0.33 g ABE / g sugar



- Low rate of mannitol consumption

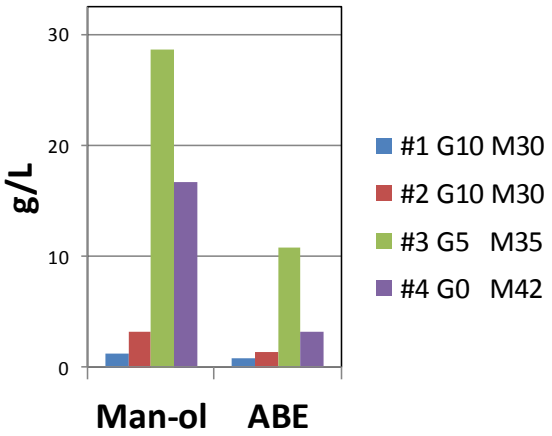
Adaptation of Clostridia to mannitol

- Aim: improve mannitol consumption: amount & rate
- Approach: adaptation by repeated cultivation in medium with mannitol – glucose mixture

(Preliminary results)

- *C. acetobutylicum*
  - Increase of mannitol consumption
  - On mannitol only (culture 4), less ABE pr

  subcultures
- *C. beijerinckii*
  - Mannitol consumption did not improve significantly



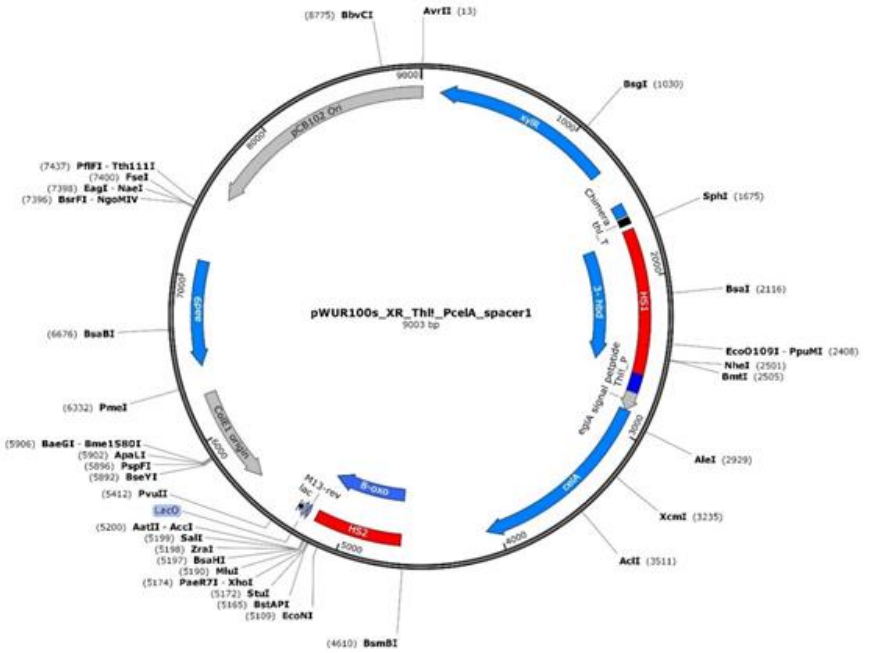
Consolidated Bio-Processing

- Introduction of genes in *Clostridium* for polymer degradation (laminarin, xylan, glucan)
- Tools for genome editing in ABE-producers developed: CRISPR-Cas9 ✓

Wasels et al, JMM, 2017

- New *celA*-construct

(cellulase, from *N. patriciarum*)



Consolidated Bio-Processing

- *C. beijerinckii* NCIMB 8052 harboring plasmid pMTL500E_celA (by classical transformation)
- Results:
  - ABE production from laminarin & lichenan
  - Low ABE production from soluble sugars in *Saccharina* (dried and milled)

- Laminarin in untreated *Saccharina* not easily available for degradation, pre-treatment necessary or additional enzymes