Genomic optimisation of hydrolysis in biogas production

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Introduction

Biogas is a promising renewable energy source due to its methane content. Methane itself seems to have high energy and heating potential. Its heating value was estimated as higher (55.5 MJ/kg) compared to diesel and wood (Fountoulakis and Manios 2009). Another advantage of biogas production is the possibility of the use of many different organic wastes as a substrate in the process (Amon *et al.* 2007; Li *et al.* 2011).

Biogas is produced in the anaerobic digestion process that consists of 4 stages: hydrolysis, acidogenesis, acetogenesis and methanogenesis (fig. 1) (Li et al. 2011). Each stage is conducted by specific microbial community able to interact with each other in syntrophic manner (Weiland 2010; Worm *et al.* 2010). The last step can be carried out by hydrogenotrophic (CH_4 production from H_2 and CO_2) or acetotrophic (CH_4 production from acetate) methanogens (Garcia *et al.* 2000). It has been already proved that there is syntrophy between some particular methanogens and some groups of bacteria (Stams *et al.* 1992).

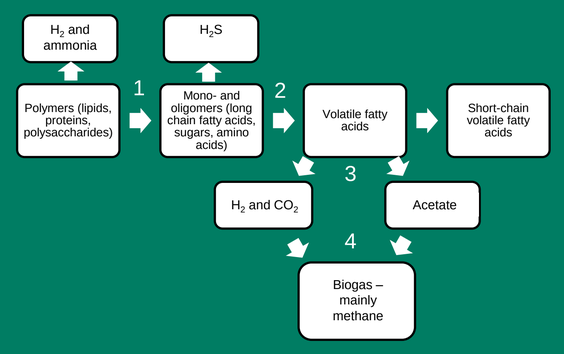

Fig. 1. Diagram showing the biogas production process. Designation: 1 – hydrolysis, 2 – acidogenesis, 3 – acetogenesis, 4 - methanogenesis

The project assumption is that there might be a syntrophic relationship (based on H_2 partial pressure) between some fermentative hydrogen-producing bacteria and hydrogenotrophic methanogens. In addition, hydrogenotrophic methanogens were proposed by Ziganshin *et al.* (2011) to give lower methane yields compared to acetoclastic methanogens. Thus, gene-monitoring specific for the particular hydrogen-producing bacteria (being in correlation with hydrogenotrophic *Archaea*) will bring the information about the methane yield (low specific gene abundancy – high methane yield). The project will bring a robust molecular biology technique of monitoring the methane production in anaerobic digesters.

Aims

The study on the possible shifts in microbial community as a result of various reactor performance – especially in terms of feedstock

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The investigation of possible syntrophy between candidate division hydrogen-producing bacteria and hydrogenotrophic methanogens and its influence on methane yields by using metagenomic tools

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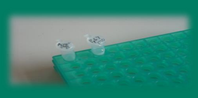
The development of robust technique (targeting the biomarker – gene of candidate division bacteria) to monitor methane yields in anaerobic digesters

Methods

Collection of a sample from plant- and lab-scale digesters

DNA extraction – Power Soil DNA Isolation Kit (Thermo Scientific)



454-pyrosequencing (bacterial and archaeal 16S investigation) and processing results in QIIME

Designing and testing lab-scale digesters

Results

The lab-scale reactors were designed, built and tested with inoculum and feedstock (slurry). The estimated biogas production – 0.5L/24 hours. HRT determined at 25 days.

Fig. 1. Lab-scale 1L reactor

Fig. 2. Lab-scale 1L reactors in water bath

Design:
- 1L glass vessel with 5 ports
- Manual stirrer (stirred once a day)
- HRT 25 days
- Mesophilic temperature
- Feedstock – slurry
- Inoculum – full-scale AD (using slurry and grass silage as feedstock)
- All reactors in triplicates – both control and treatment digesters
- Treatment digester – additional H_2
- Biogas volume measurement – displacement method (using acidified salt; own system)
- Biogas content measurement – portable gas analyzer

Conclusions and future plans

The designed reactors will be used to test the hypothesis and to find the correlation between specific microorganism presence and high (or low) biogas production. The treatment reactors will be run in triplicates with reference to control digesters. The biogas production will be monitored and samples will be taken for further analysis every day. Some interesting samples will be chosen and analysed by pyrosequencing which will reveal the microbial content of the samples. Moreover, samples from the other digesters (differing in feedstock and performance) will be investigated in the same manner to support the hypothesis.

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