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Application of complex anaerobic microbial community in the „Power-to-Biomethane” technology

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Summary of the Ph.D. thesis

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INTRODUCTION

The global energy consumption increases continuously and rapidly while the available fossil energy carriers become limited. This, together with the alarming climate change, put a special emphasis on the development of the renewables. The rapidly expanding renewable energy technologies, i.e. photovoltaics and wind power, generate fluctuating “green electricity”. The excess “green electricity” needs to be stored independently from the electricity grid. H_2 is a promising alternative energy carrier despite of the existing underdeveloped storage, transportation technologies. H_2 can be converted to CH_4 via hydrogenotrophic methanogenesis. This microbiological by-pass route offers an affluent solution of the energy storage and transportation problems. The “Power-to-Gas” technology directs the fluctuating “green electricity” to the more flexible natural gas grid by converting it to CH_4 . Bio CH_4 is chemically indistinguishable from the fossil “natural gas” and can replace “natural gas” in every applications the non-renewable and environmentally unfriendly energy carrier.

APPLIED MATERIALS AND METHODS

The total volume of the reactors was 160 mL (Wheaton glass serum bottle, Z114014 Aldrich). The H₂ and the CO₂ were manually injected into the gas phase with disposable plastic syringe. The gas composition changes were followed with gas chromatography (GC). The fermentation liquid was analysed by pH meter, and the microbial metabolisms catalysed volatile organic acid concentration changes were determined by high performance liquid chromatograph (HPLC) equipped with a refractive index detector.

The H₂/CO₂ rich gas phase induced microbial composition changes were investigated by metagenomic analysis. The samples were sequenced according to the Ion Torrent PGM™ protocols. The microbial communities were investigated by read-base and genome-centric analyses in mesophilic and thermophilic fermentations. Via the genome-centric metagenomic analyses enzymes of H₂ utilizing and methanogenic pathways were determined.

RESULTS

In this Thesis work, I investigated the application of a mixed microbial community developed from the fermentation effluents of biogas plants. The results corroborated the advantages of using mixed anaerobic communities in the Power-to-Gas conversion. The major findings were as follows:

1. In a batch-type fermentation system H_2 was daily injected into the reactor head space. Although H_2 is poorly soluble in the aqueous medium, it was converted by the hydrogenotrophic methanogens as indicated by the increased yield of CH_4 and concomitant reduction of CO_2 . The depletion of the residual organic substrate supply and dissolved CO_2 resulted in diminishing CH_4 production in time. This was also accompanied by H_2 accumulation in the headspace and elevation of pH due to the lack of the buffering capacity of CO_2/HCO_3^- . At mesophilic temperature these effects were apparent on days 11 and 14 in the case of 48.94 mL (40.78%) and 37.75 mL (31.46%) daily H_2 dosages, respectively. The reactors receiving 21.4 mL (17.83%) daily H_2 dosage converted the H_2 to CH_4 with 97.4% efficiency throughout the 29-days long experimental period. At

thermophilic temperature the conversion took place substantially faster.

2. The effects of a systematic organic substrate supply were also investigated. The regular organic substrate addition mimicked the industrial conditions of the in-situ P2G process on the one hand, and on the other hand, it was expected to balance the dangerous pH increase caused by the dissolved CO₂ depletion. The results indicated an intricate and sensitive equilibrium between H₂/CO₂ consumption and system stability at both mesophilic and thermophilic temperatures. The inhibition of the acetoclastic pathway resulted in the accumulation of volatile organic acids (VFAs) and acidification of the pH. Close to the optimal conditions stable microbial activity could be maintained. Surplus CH₄ evolution of 23.5% (mesophilic) and 10.15% (thermophilic) were measured, respectively.
3. The stoichiometric mixture of H₂+CO₂, i.e. 4H₂+CO₂, successfully modeled the ex-situ P2G industrial process conditions. Long term bioCH₄ production was achieved at balanced pH. The hydrogenotrophic methanogens converted the injected H₂ to CH₄ with 90%, 74% efficiency at mesophilic and thermophilic temperatures, respectively. The bioCH₄ content of the reactor's

headspace was over 97% in the mesophilic and over 95% in the thermophilic reactors. Both values are compatible with the quality requirements for injection into the natural gas grid.

4. The read based metagenomic analyses revealed a shift in the microbial community. In the mesophilic reactors the H₂/CO₂ rich headspace diminished the genera *Candidatus Cloacimonas*, *Methanosarcina*, *Sedimentobacter* and the *Clostridium* while the genera *Bacteroides*, *Romboutsia* and the H₂ utilizing *Methanobacter* and *Methanoculleus* became predominant. At thermophilic temperature the abundance of the genera *Ureibacillus*, *Lutispora*, *Herbinix*, *Clostridium*, *Bacillus* and the *Tepidanaerobacter* decreased and the growth of *Actinotalea*, *Methanobacterium*, *Cellulomonas* and the hydrogenotrophic *Methanothermobacter* was facilitated.
5. 34 and 43 bins were assembled from the mesophilic and thermophilic consortia, respectively. The genome-centric read-based analyses corroborated and validated each other. In the H₂/CO₂ rich environment hydrogenotrophic-, acetotrophic methanogenesis and reductive acidogenesis specific genes were observed in the assembled genomes. Methylophilic

methanogenesis or sulfate reduction specific genes were scarcely detected.

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MTMT identification number: 10053019

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