Genetic analyses of microbial communities in anaerobic digesters reveal enhanced methanogenesis during calcium nitrate dosage†

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† patented application

Aim of Study

Empirical measurements and observations

Calcium nitrate dosage to anaerobic digesters resulted in several improvements of the fermentation process, including:

- **Increased yield of methane**
- **Reduced hydrogen sulfide concentration**
- **Substrate savings**
- **Improved viscosity**
- **More robust plant operation**

In this study the biological effects of calcium nitrate dosage on the anaerobic microbial communities of several biogas plants were investigated: Microbial community structures and metabolic activities in control and treated fermenters of operating plants were determined.

Hypothesis

Calcium nitrate shifts the competitive balance between denitrifiers, sulfate reducing bacteria (SRB) and methanogens to the benefit of methane production.

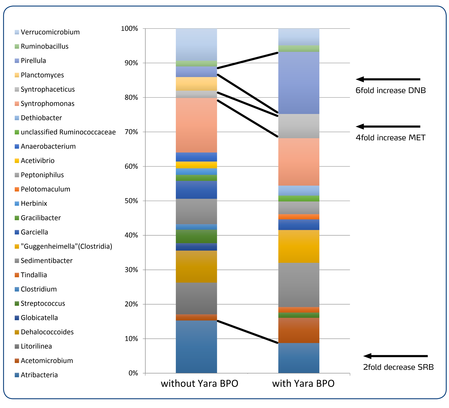
Methods

Nucleic acids from samples of a time series collected were extracted. High-throughput sequencing and analyses of taxonomic marker genes identified microbial community structures. Quantitative real time PCR of specific RNA transcripts revealed the activities of methanogens and denitrifiers (for SRBs not shown).

Results (examples)

Community structure

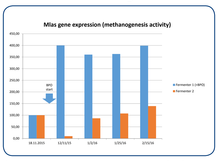
Genetic analyses of bacterial and archaeal community structures revealed remarkable differences between a BPO-treated and a non-treated control digester.



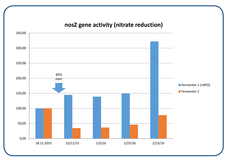
Bacterial community structures in digesters without and with Yara BPO. The figure illustrates a decrease of specific sulfate reducing bacteria (SRB) and an increase of specific denitrifying bacteria (DNB) and also methanogens (MET).

- Increase of denitrifiers (N-metabolism)
- Increase of methanogens (methane production)
- Decrease of species involved in S-metabolism (sulfate reduction)

Gene activity



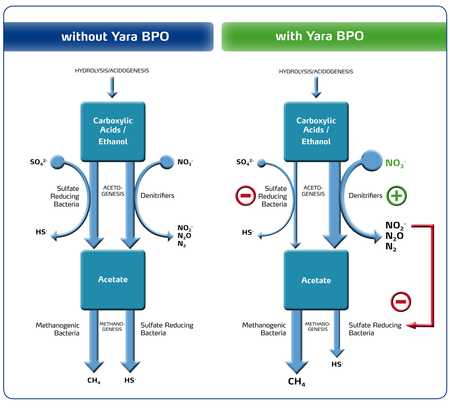
Increased gene activity involved in methanogenesis



Increased gene activity involved in nitrate reduction

Mechanistic model

Based on the data obtained from genetic analyses, we propose the following mechanistic model as one of the possible mode of actions of calcium nitrate in anaerobic digesters:



Conclusions

During dosage of calcium nitrate to anaerobic digesters, we found

- Enhanced methanogenesis
- Enhanced denitrification
- Decreased sulfide production (results not shown)

Denitrifying bacteria benefit from the additional supply of an energy source (calcium nitrate) for the breakdown of organic substrates (carboxylic acids and alcohols). Therefore, denitrifiers, are able to outcompete sulfate reducing bacteria, which utilize the same substrates. Furthermore, nitrite as a product of denitrification inhibits the ATP-Sulfurylase, an enzyme needed for the conversion of sulfate into sulfide (Carlson et al. 2015).

Further effects of calcium nitrate on fermentation process steps are currently under investigation.

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