Screening of methanogens in a Simultaneous Bioreactor System (SBRS) with multiple determination under high pressure conditions

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Introduction

Biomethanisation is a biotechnological process for the production of methane (CH_4), applying methanogenic microorganisms.

For the conversion CO_2-hydrogenotrophic methanogens, which belong to the domain of Archaea utilize molecular hydrogen (H_2) as a reductant together with CO_2 as a carbon and energy source to produce auto catalytically CH_4, water and biomass according to (1)[1-3].

$$CO_2 + 4\ H_2 \rightarrow CH_4 + 2\ H_2O \quad (1)$$

In the field of CO_2 based biological-methane-production (BMP) not many methanogenic strains were yet examined.

Scope of work

To perform reproducible CO_2-BMP experiments in quadruplicates at elevated pressure levels a Simultaneous Bioreactor System (SBRS) was developed.

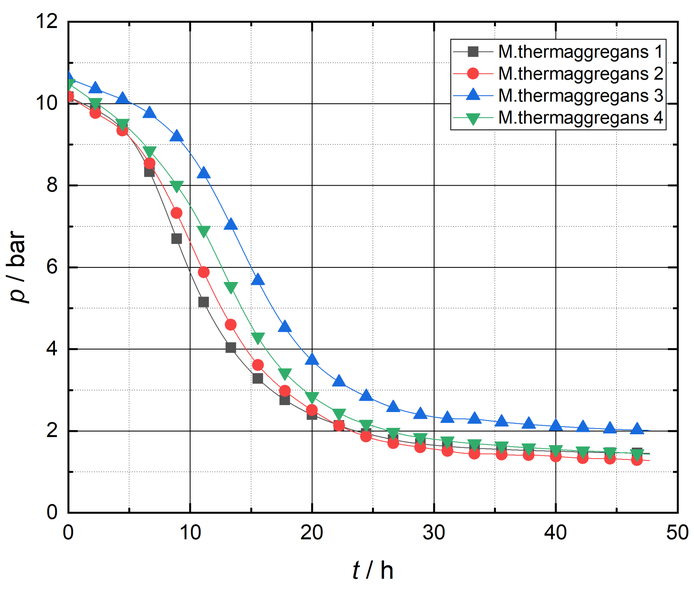
As a proof of principle, various CO_2-hydrogenotrophic methanogenic strains were cultivated in the SBRS and the reproducibility of the system was evaluated.

Experimental I - SBRS

The developed SBRS consist of four structurally identical bioreactors (R1-R4) made of stainless steel and can be used for screening methanogens in a closed batch cultivation mode up to 50 barg. Excepting the gassing line, all reactors can be operated independently, where each reactor has a total volume of 160 mL.

Each of the four vessels is equipped with an individual heating jacket as well as a digital pressure sensor on the top of each vessel to measure and control the pressure and temperature online. For better mixing of the cultures, the whole system is grounded on a lab shaker. A detailed flow sheet of the reactor system is shown in Figure 1.

Curves of the experiments



1) *M. thermaggregans*, 10 barg

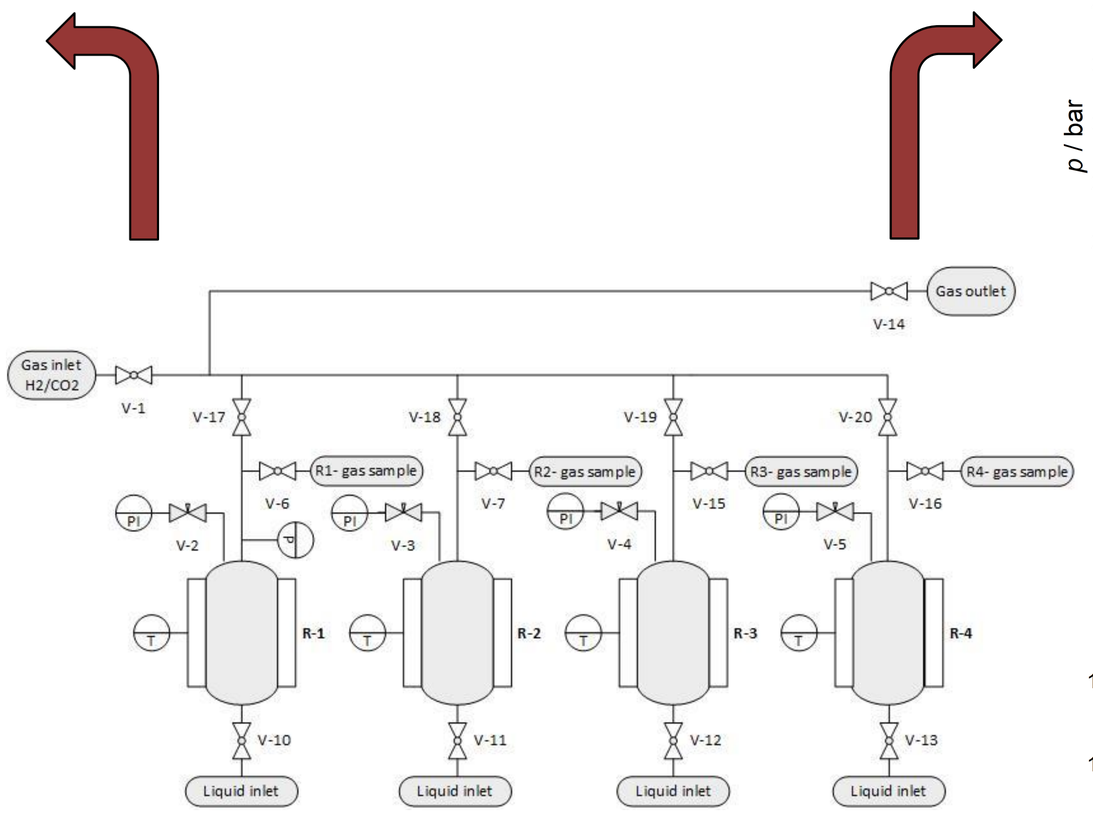
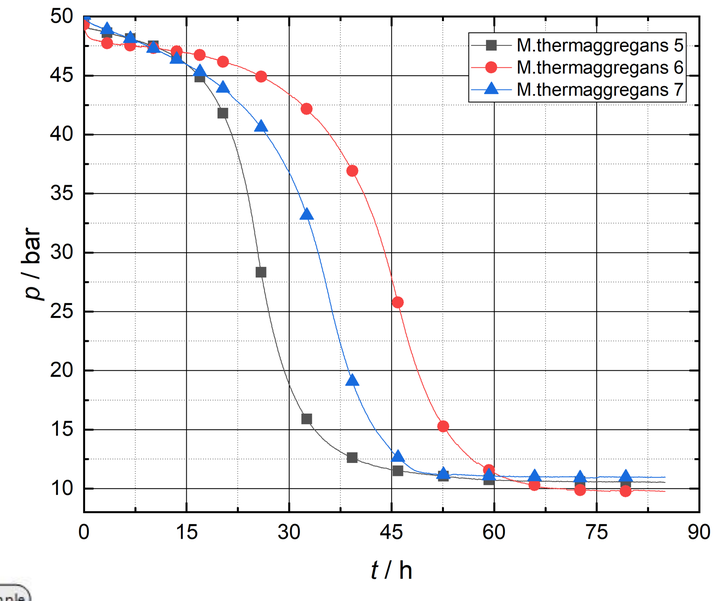
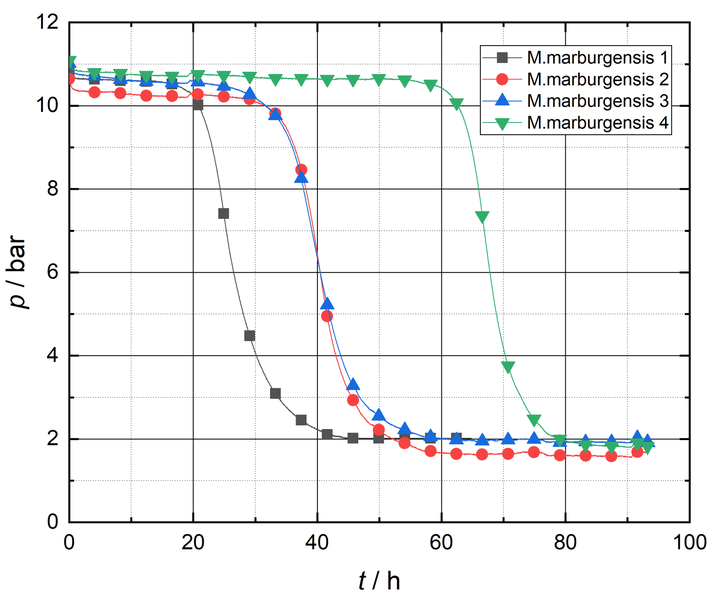


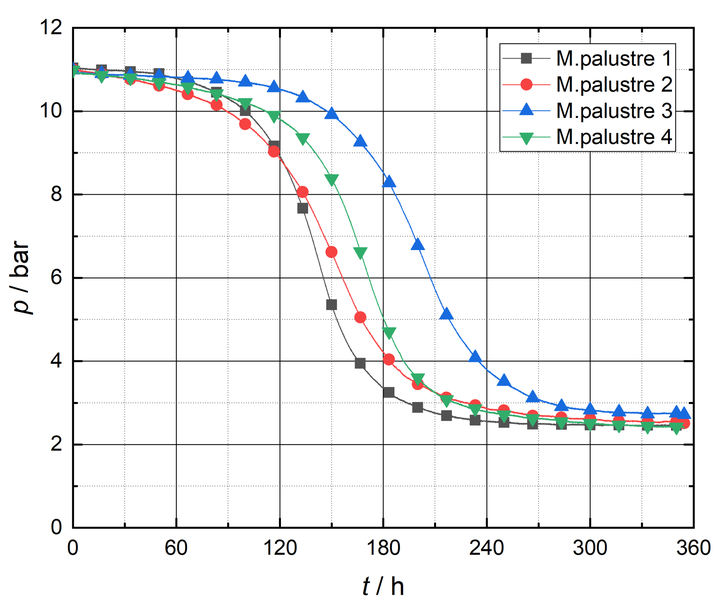
Figure 1: Flowsheet of the developed SBRS



2) *M. thermaggregans*, 50 barg



3) *M. marburgensis*, 10 barg



4) *M. palustre*, 10 barg

Experimental II - implementation

The SBRS was tested with three different methanogenic strains:

- *Methanothermobacter marburgensis* DSM 2133
- *Methanobacterium thermaggregans* DSM 3266
- *Methanobacterium palustre* DSM 3266

obtained from Deutsche Stammsammlung für Mikroorganismen und Zellkulturen.

The experiments were performed with a H_2:CO_2 mixture (80 Vol.-% H_2 in CO_2) and the strains were cultivated in *M. marburgensis* medium[4].

The monitoring of the experiments was carried out primarily by means of pressure measurements.

The production of CH_4 was checked by GC measurements for each experiment, whereby the gas samples were only taken at the end of the experiments.

Results and conclusion

To test the reproducibility between the four vessels of the new developed SBRS, three strains were cultivated at different pressure levels (10 and 50 barg). Following the reaction stoichiometry (Equation 1) methanogenic CH_4 production leads to a pressure drop in the reactor. This may indicate gas conversion and indirectly the growth of the methanogenic strain.

The expected pressure drop was seen in all experiments (Graph 1-4) and a conversion < 95 % could be achieved. The experiments show also a good comparability of the four reactors. Therefore we concluded, that the SBRS is a suitable high throughput bioreactor system for fast characterization and screening of methanogens and gas converting microorganisms.

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References

[1] Seifert et al., 2013

[2] Rittmann et al., 2015

[3] Rittmann et al., 2014

[4] Lecker et al., 2017