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RECYCLES WORKSHOP

Metagenomics and metabarcoding approaches to describe ecological systems and infer their development

5th, 6th & 7th of July 2022

Microbial diversity dynamics in a UASB reactor targeting sulfate reduction

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GA: 872053 – H2020 - MSCA - RISE-2019

European Commission

Outline

TABLE OF CONTENTS

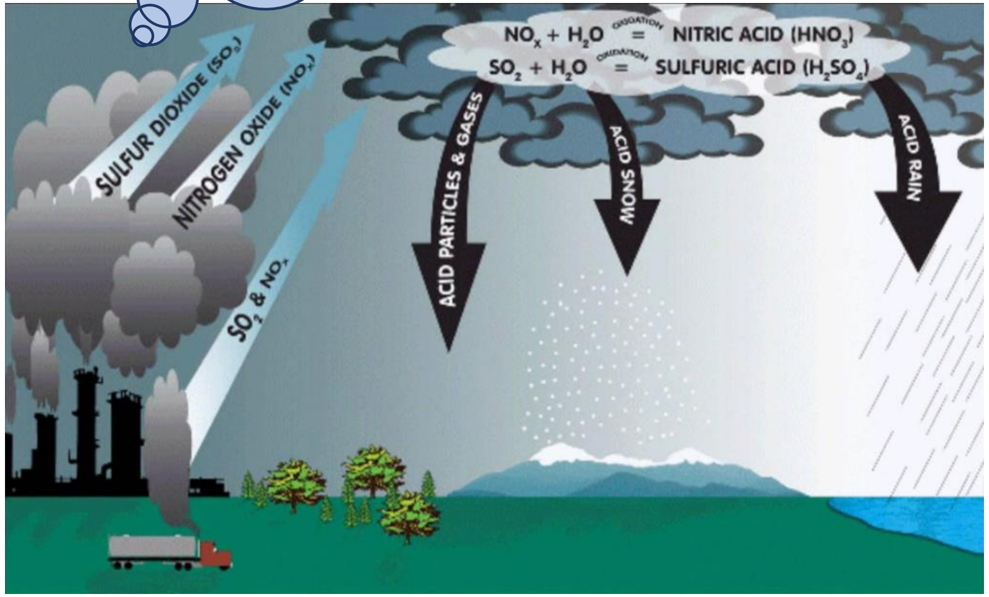
1. Introduction

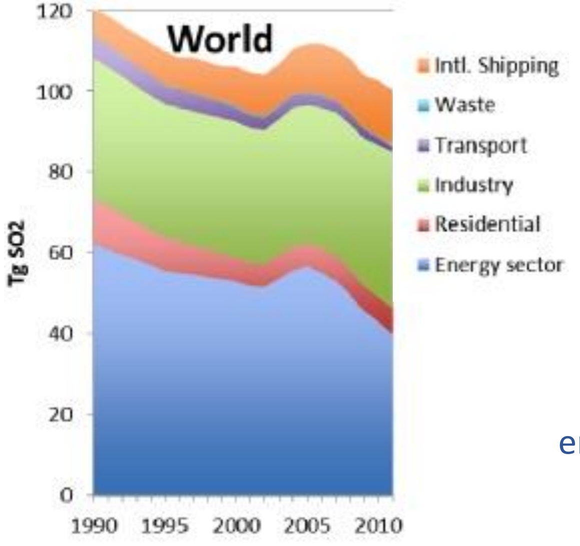
COMBUSTION PROCESS

Flue gases containing **SO_x**, **NO_x**, **CO_2** and other compounds

Consequences of SO_x release into the atmosphere

- Human health (respiratory illness)
- Environment (acid rain)





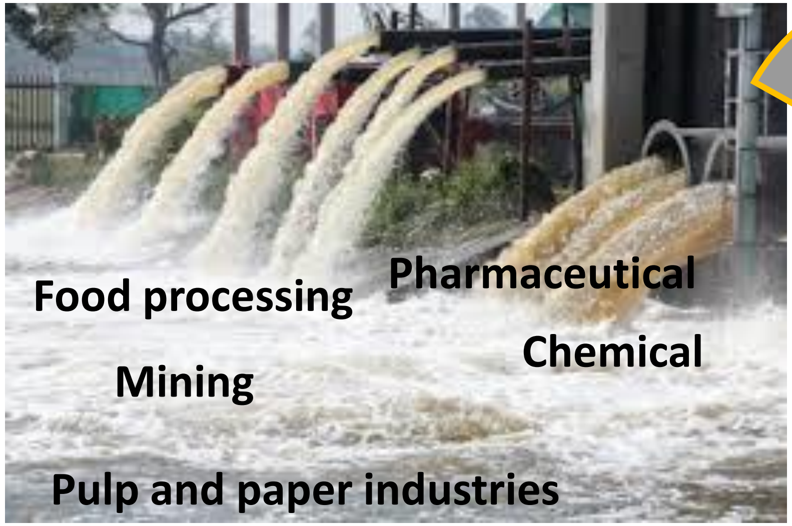
Z Klimont et al, 2013

INDUSTRIAL SECTOR

emitted 40 MT of SO_2 in 2011

1. Introduction

COMBUSTION PROCESS



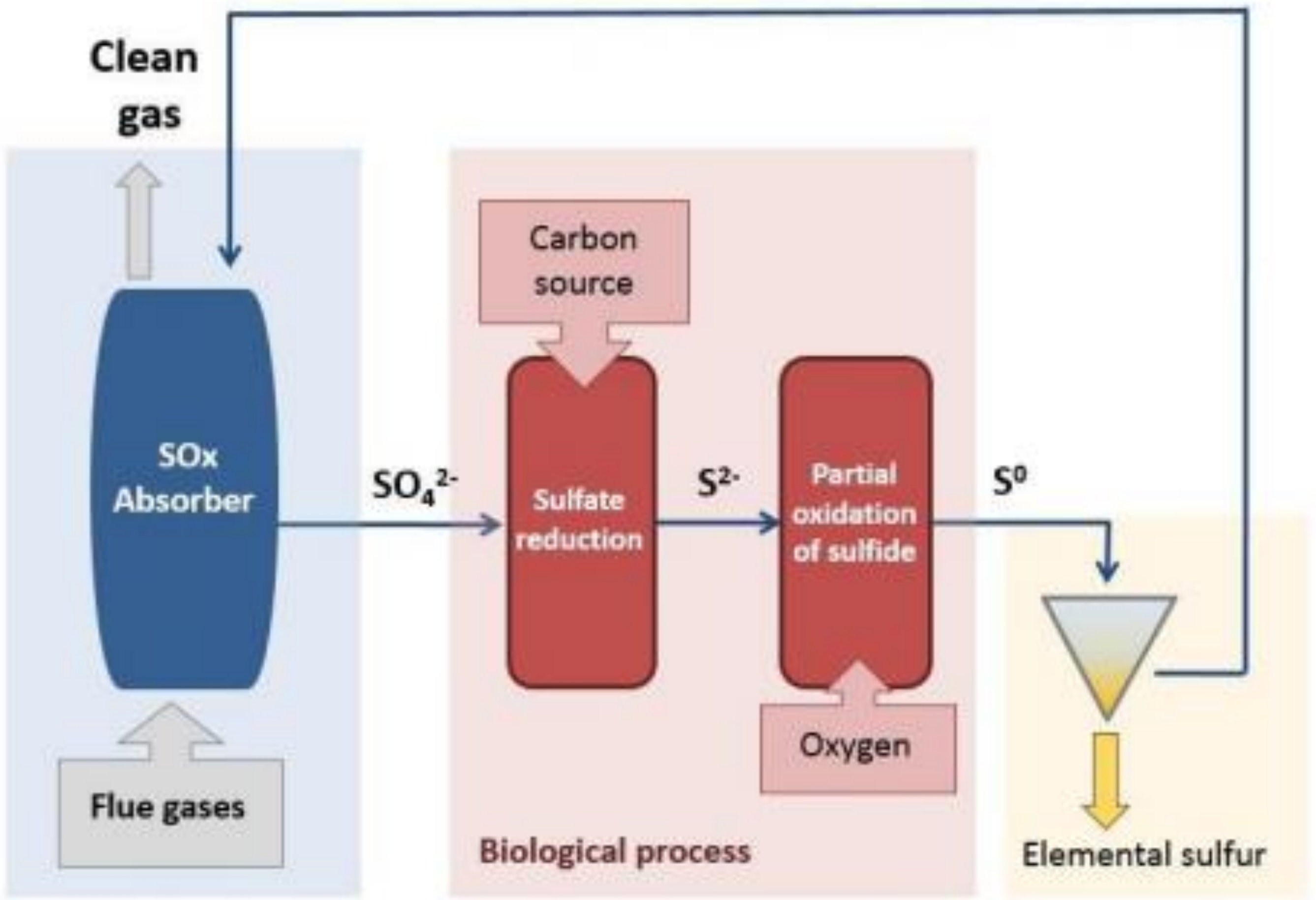
sulfite/sulfate$_{(aq)}$

Chemical

- The use of catalysts or chemical products generate effluents that must be further treated
- Costly and not environmentally friendly

✌ Biological treatment

SONOVA PROCESS: two-stage bioscrubber



Step 1: scrubbing

$SO_2 + OH^- \rightarrow HSO_3^-$

Reactor 1: reduction

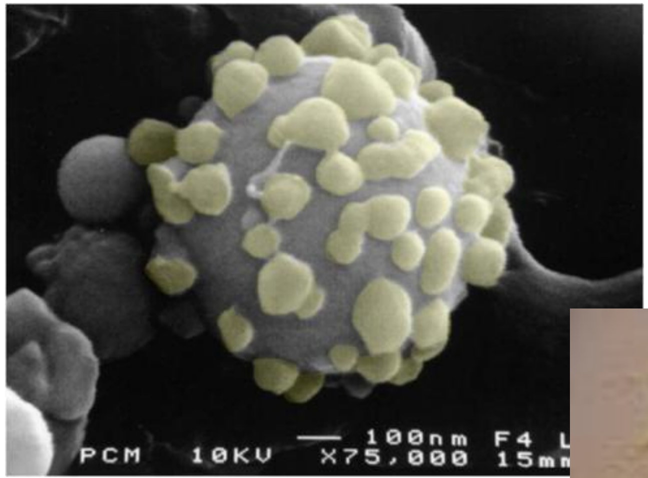
$HSO_3^- + 6\,[H] \rightarrow HS^- + 3\,H_2O$

$SO_4^{2-} + 8\,[H] \rightarrow HS^- + OH^- + 3\,H_2O$

Reactor 2: partial oxidation

$HS^- + 0.5\,O_2 \rightarrow S^0 + OH^-$

1. Introduction

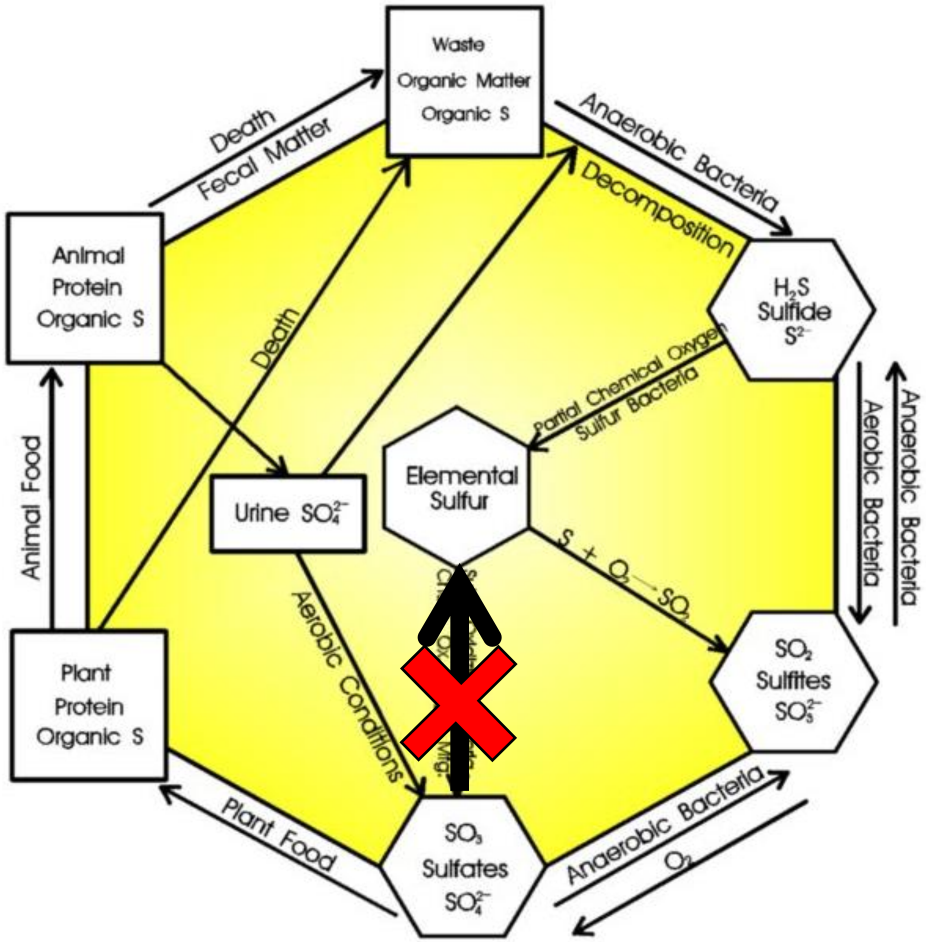


Biological desulfurisation
- ✓ Less use of chemicals
- ✓ Valuable product (sulfur)

Biosulfur has different properties than chemical sulfur!

1. Introduction

The Sulfur Cycle



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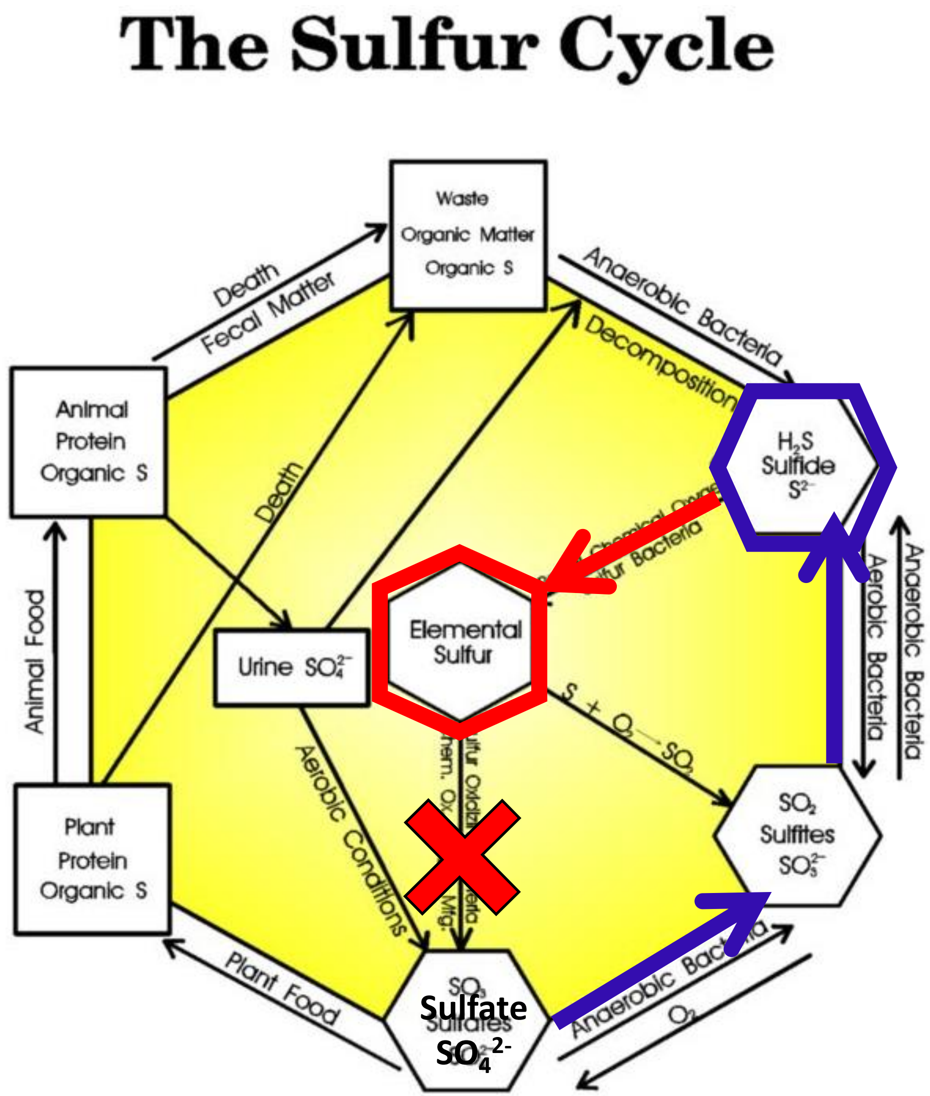


BIOLOGICAL SULFUR RECOVERY??

Two-step process for S recovery:

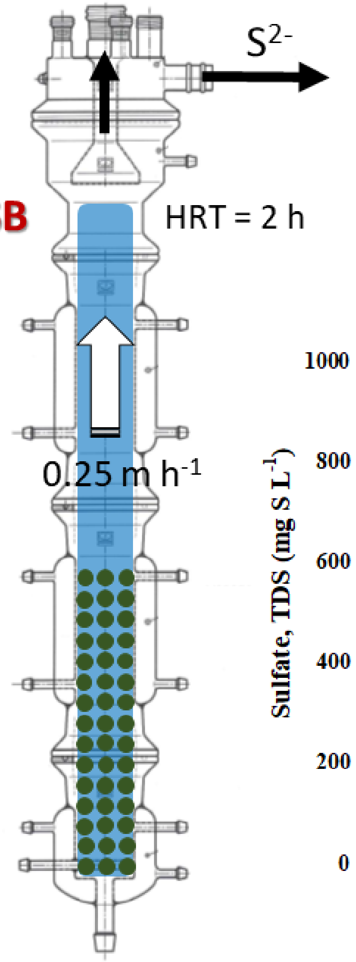
1. **Anaerobic sulfate reduction**

$$SO_4^{2-} + C_{organic} \rightarrow S^{2-}$$

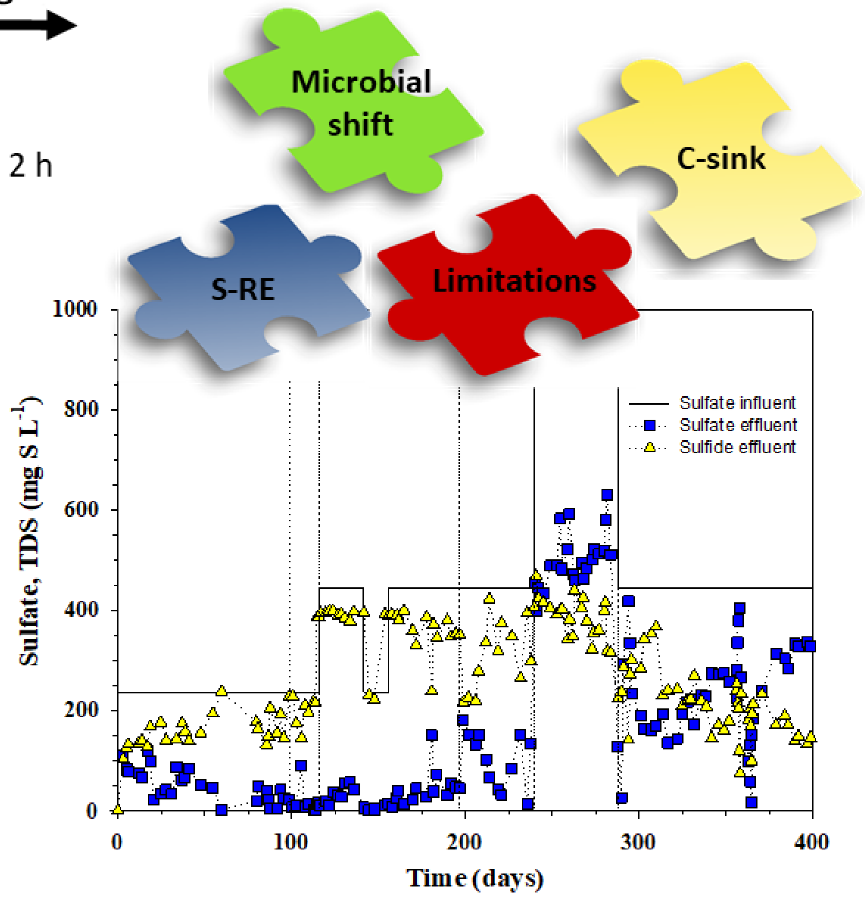
2. **Aerobic partial sulfide oxidation**

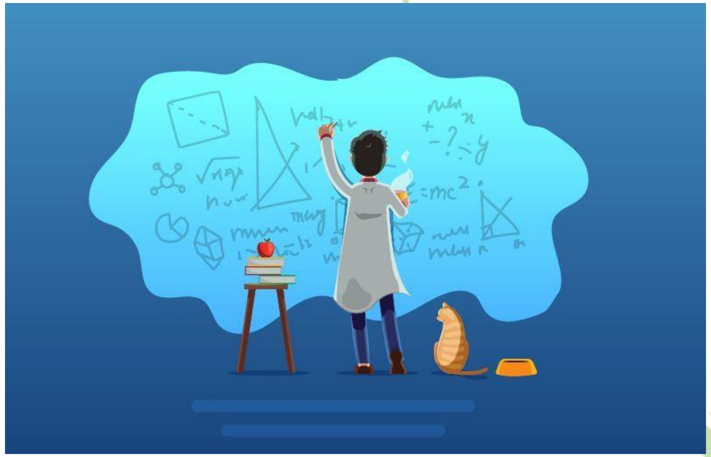
$$S^{2-} + ½\ O_2 + 2H^+ \rightarrow S^0 + H_20$$

2. Goals



Biological Sulfate reduction



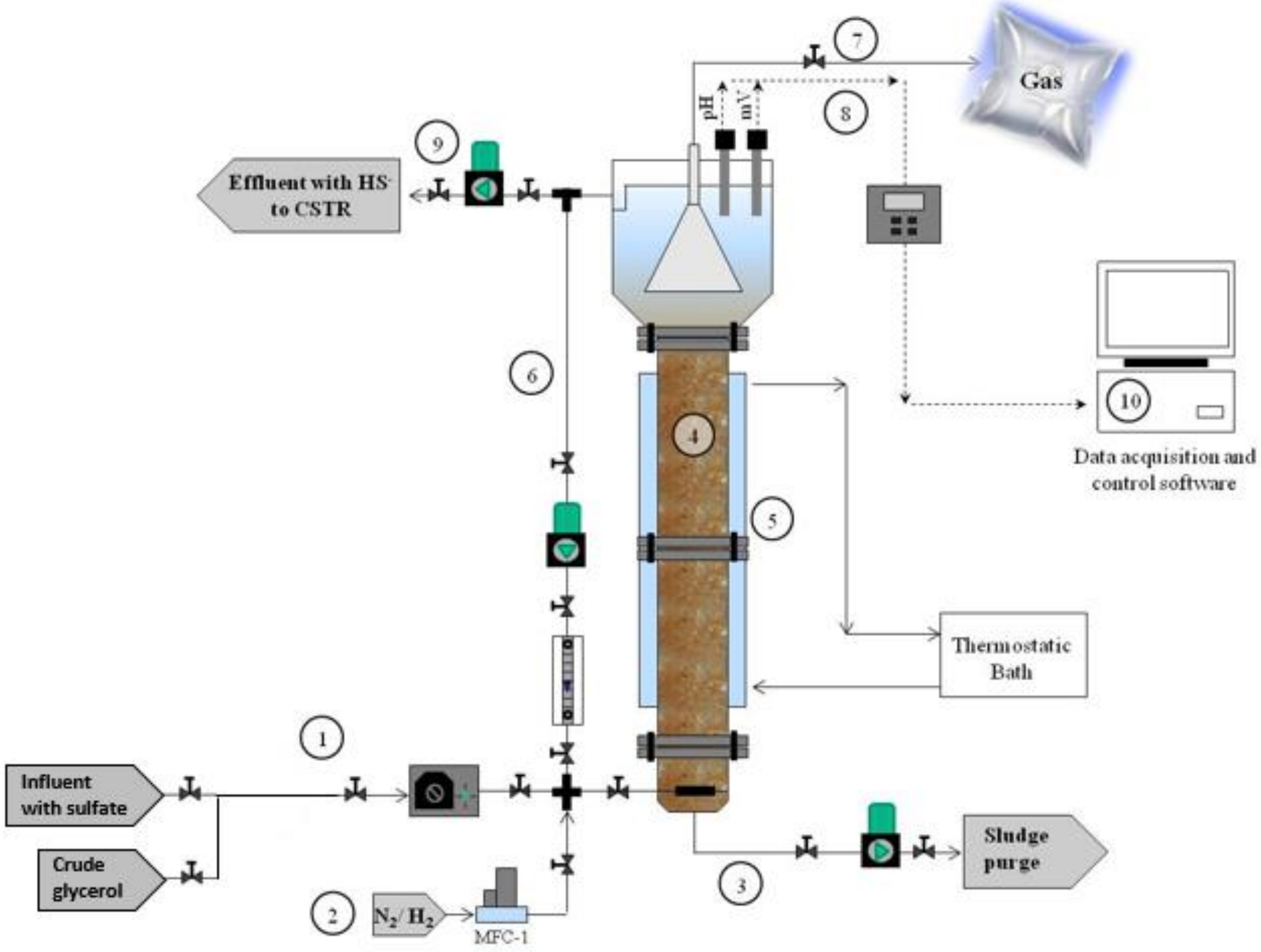


MM: Sulfate + **Crude glycerol**

3. Methodology

Reactor type: Upflow Anaerobic Sludge Blanket reactor (UASB)
Reactor volume = 2.5 L
Inoculum = granular sludge from a methanogenic UASB (paper recycling industry)
Granular sludge volume = 1-1.35 L

$\mathbf{v_{upflow}}$= 0.25 m h^{-1}
$\mathbf{pH_{inlet}}$ ≈ 8.4-8.8
T = 35 °C
HRT ≈ 2 h



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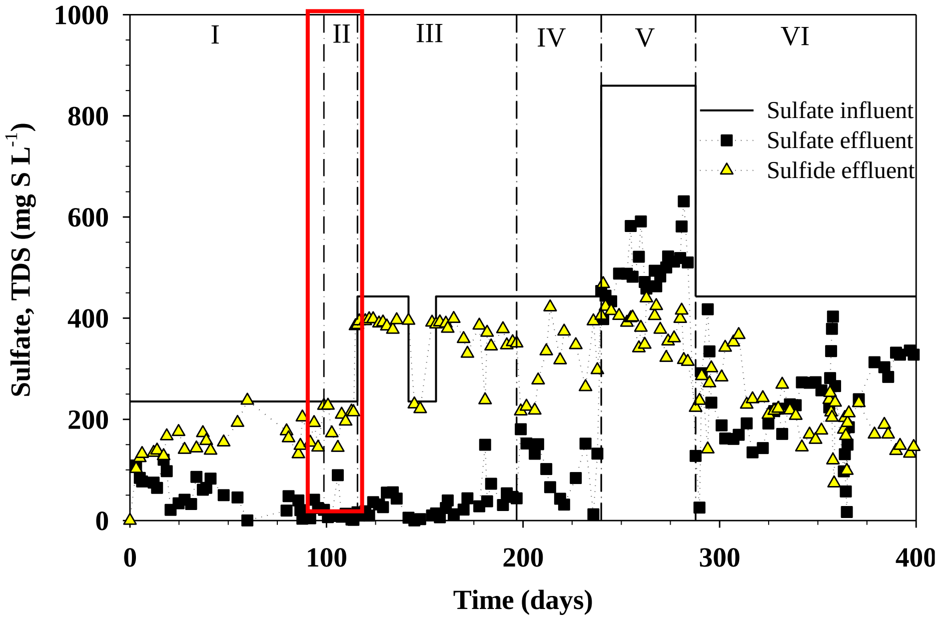
Fernández Palacios, 2020



Period	Days	Sulfate$_{inlet}$ (mg S L^{-1})	OLR (kg O_2 m^{-3}d^{-1})	COD/S$_{inlet}$ (g O_2 g^{-1} S)
I	0-99	235±17	12.0±2.1	3.8±0.8
II	99-115	235±17	15.8±4.6	5.3±0.6
III	115-197	442±47	24.4±6.9	5.4±1.0
IV	197-238	442±47	27.1±2.5	5.4±0.6
V	238-288	859±30	25.4±2.6	3.1±0.2
VI	288-400	442±47	25.7±6.9	4.5±1.4

3. Methodology

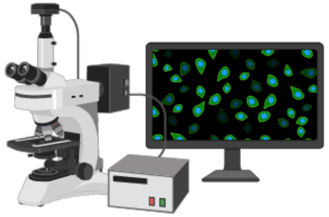
Physical-chemical data ⟷ **Microbial population**

Sulfate$_{inlet}$ (mg S L^{-1})	SLR (kg S-SO_4^{2-} m^{-3}d^{-1})	OLR (kg O_2 m^{-3}d^{-1})	TOC/S$_{inlet}$ (g C g^{-1} S)
252.8±20.8	5.1±0.7	7.6±1.6	1.5±0.3

16S rRNA gene amplicon sequencing by using Illumina platform

+

FISH

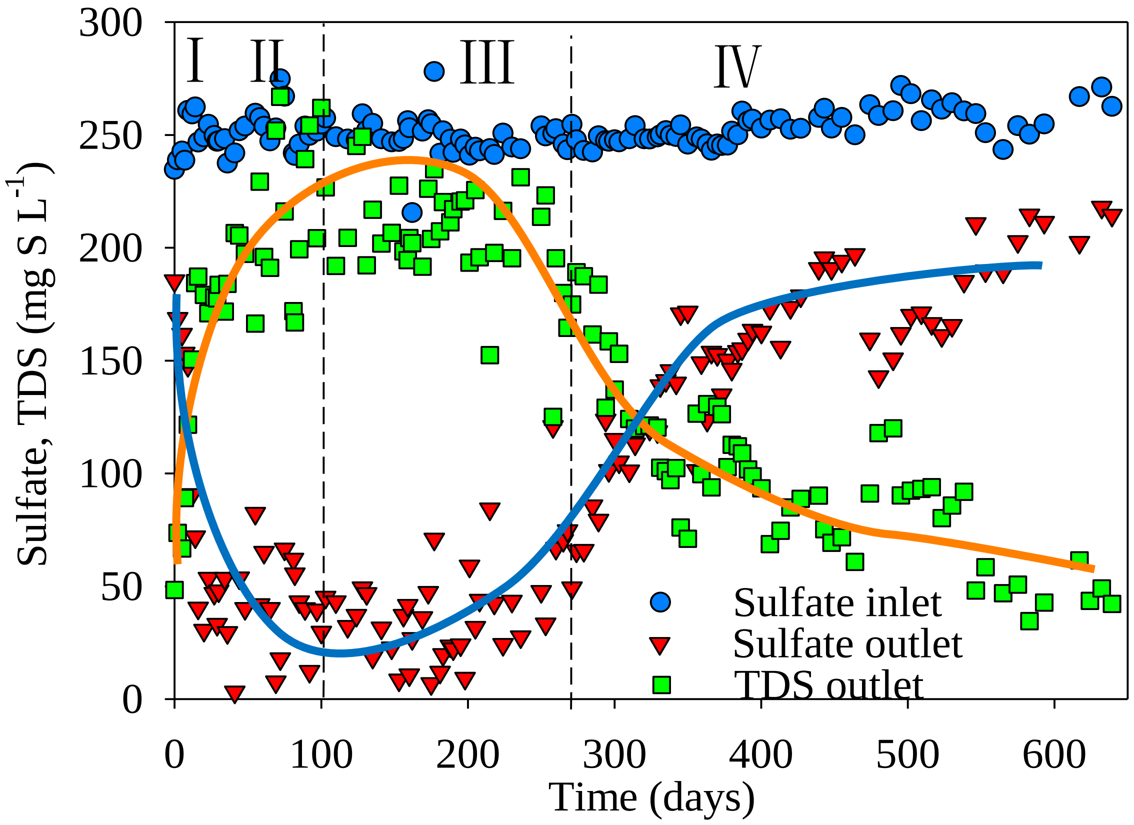


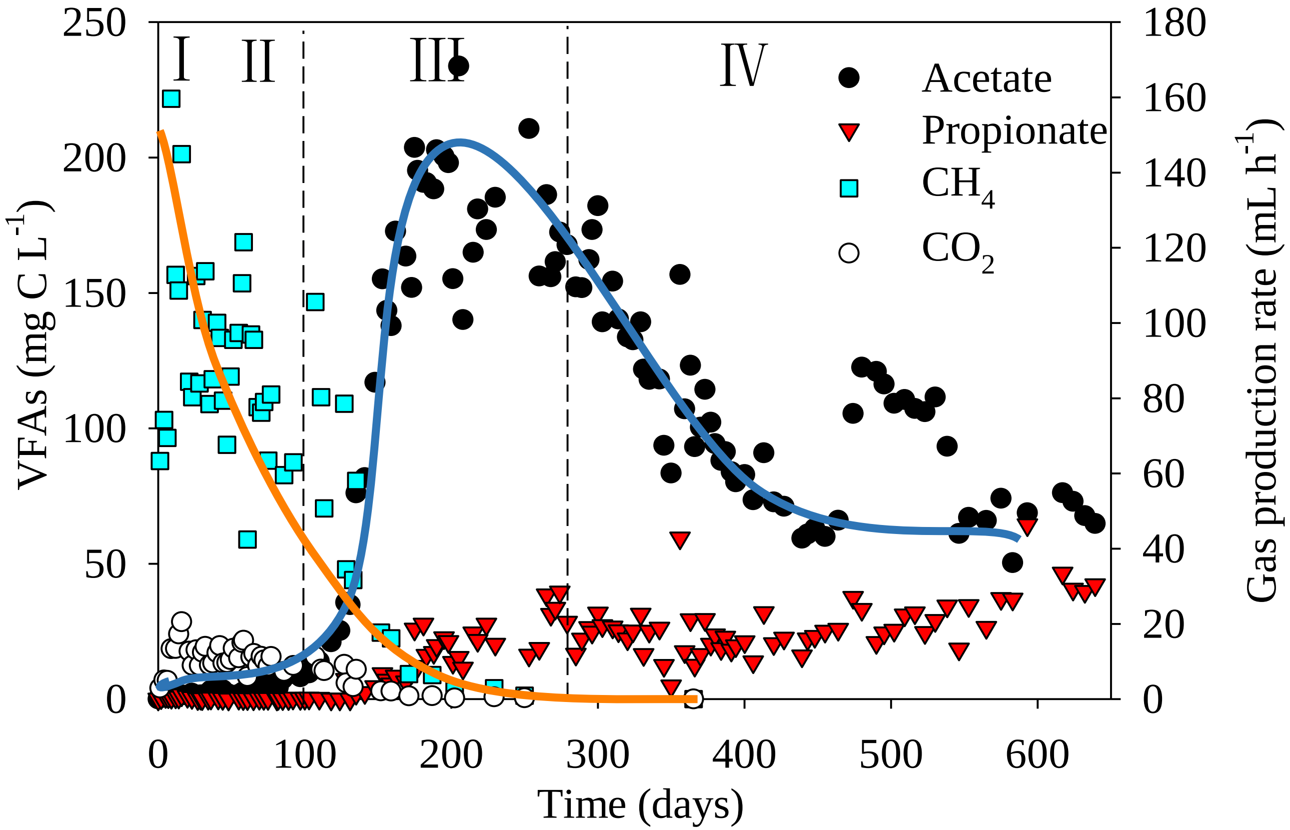
INOCULUM

+

Day 85, 149, 173, 230, 294, 538 of operation

4. Performance results

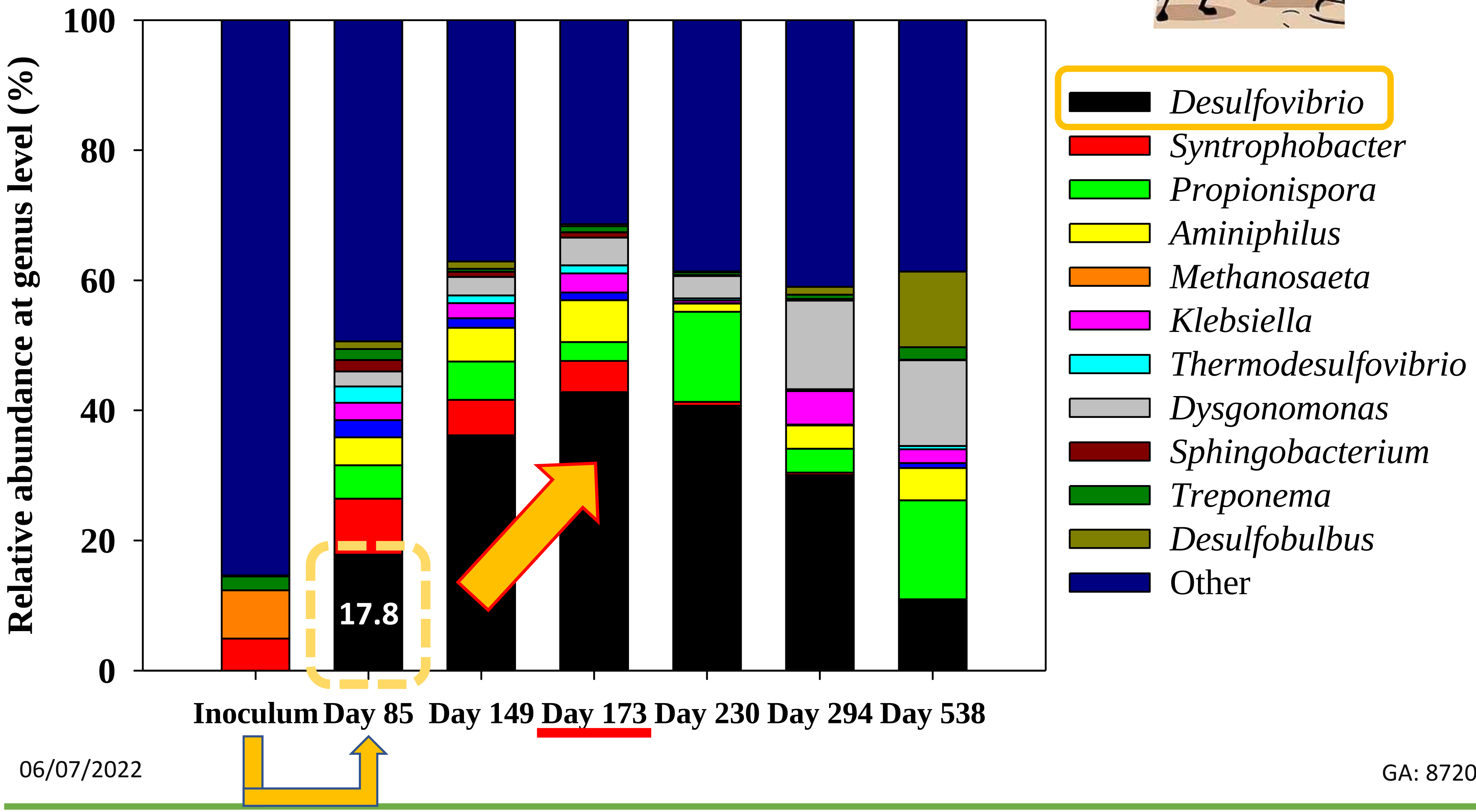


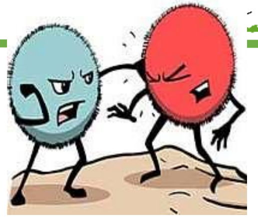


VFAs accumulation (mainly acetate) coincided with a decrease in CH_4 production

4. Performance results

Microbial community dynamics: 16S rRNA sequencing

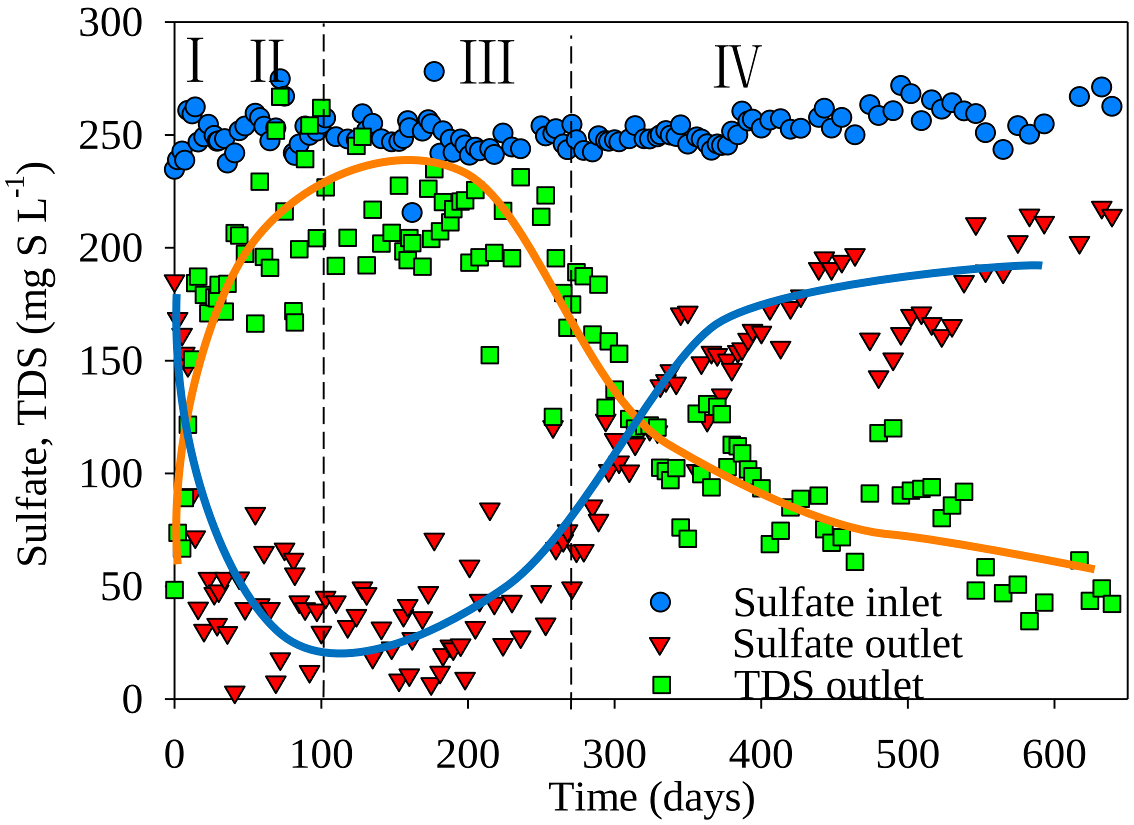


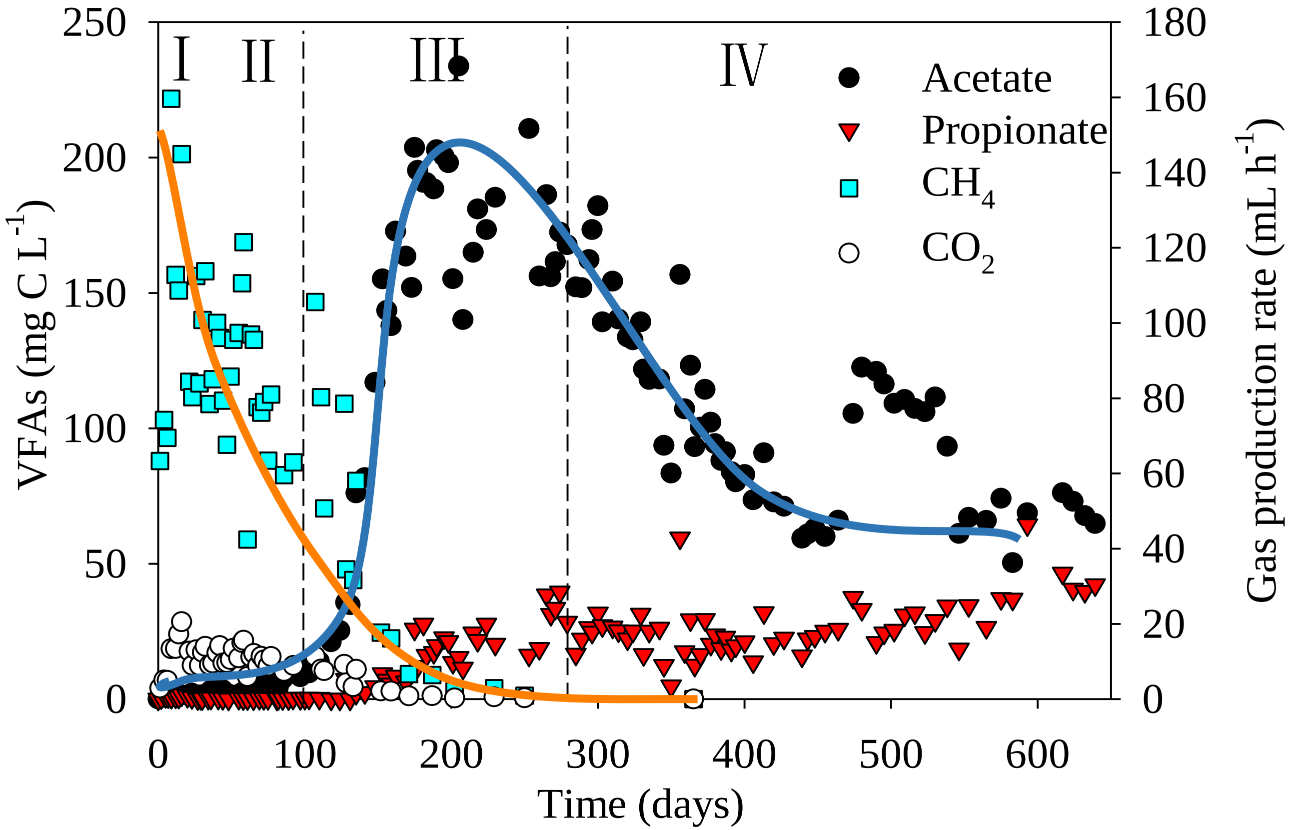


Day 173

Desulfovibrio
42.8 %

4. Performance results





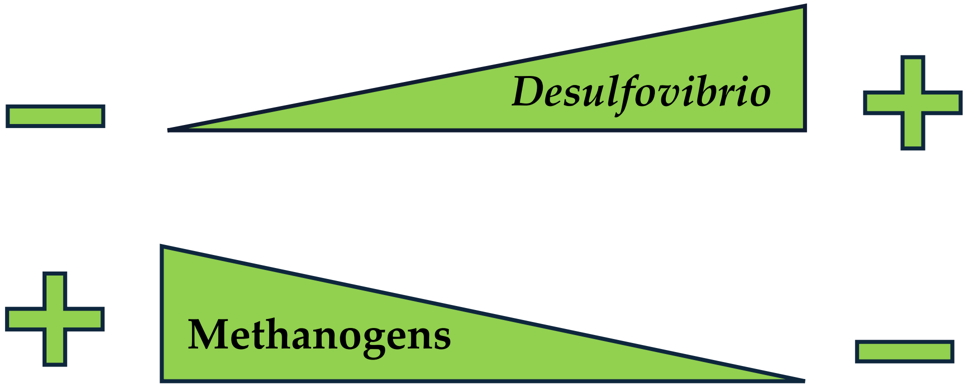
SR capacity increase correlated well with *Desulfovibrio* predominance

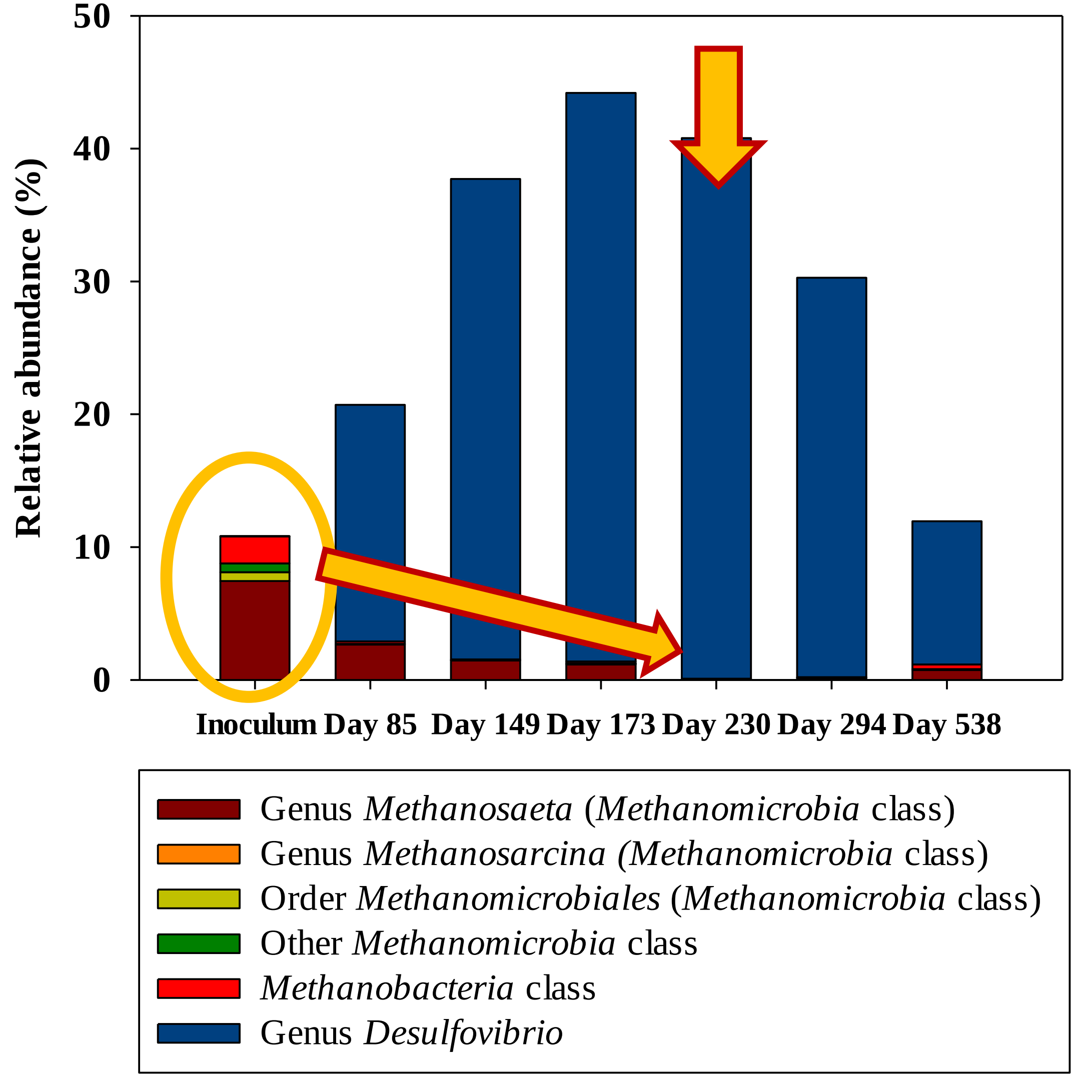
4. Performance results

Microbial community dynamics: 16S rRNA sequencing

	Methanogens (%)	SRB (Genus *Desulfovibrio*) (%)
Inoculum	10.8	[illegible]
Day 85	2.9	[illegible]
Day 149	1.5	[illegible]
Day 173	1.4	[illegible]
Day 230	0.0	[illegible]
Day 294	0.2	[illegible]
Day 538	1.2	[illegible]

Methanogens washout confirmed





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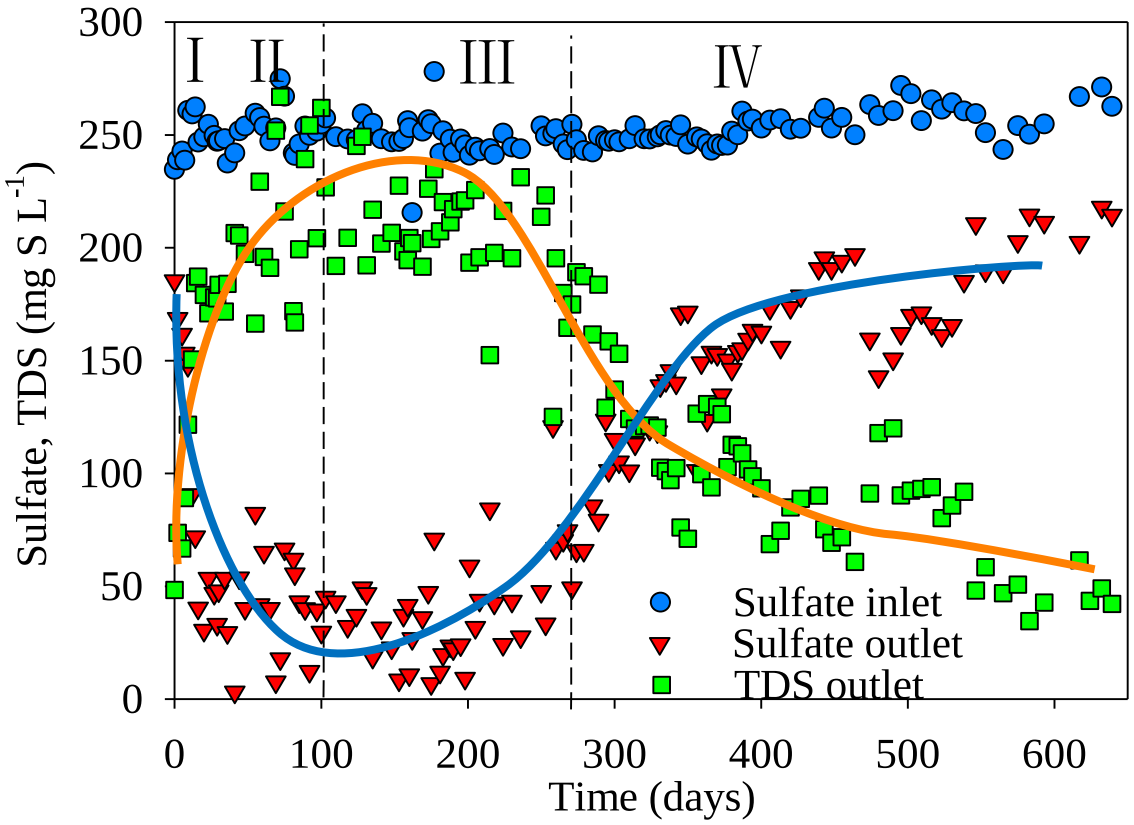
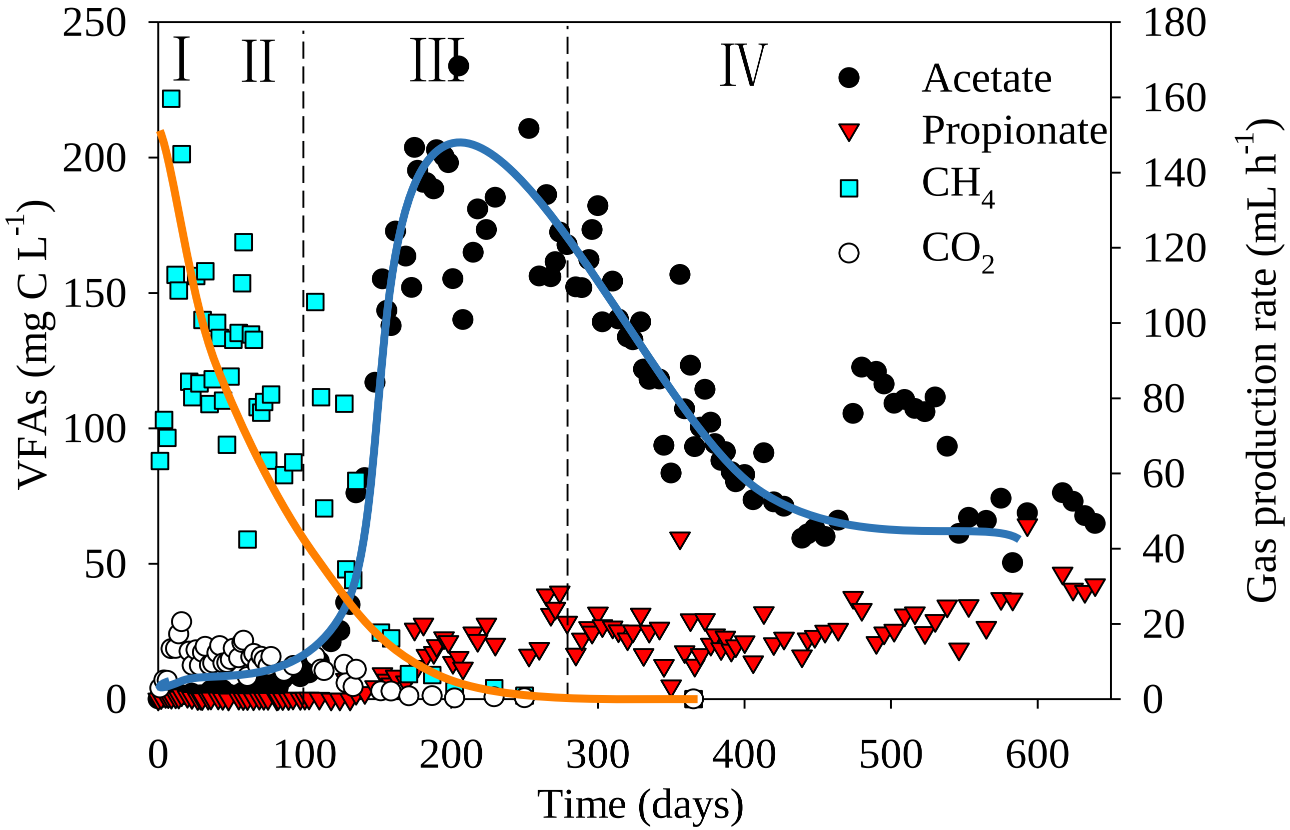
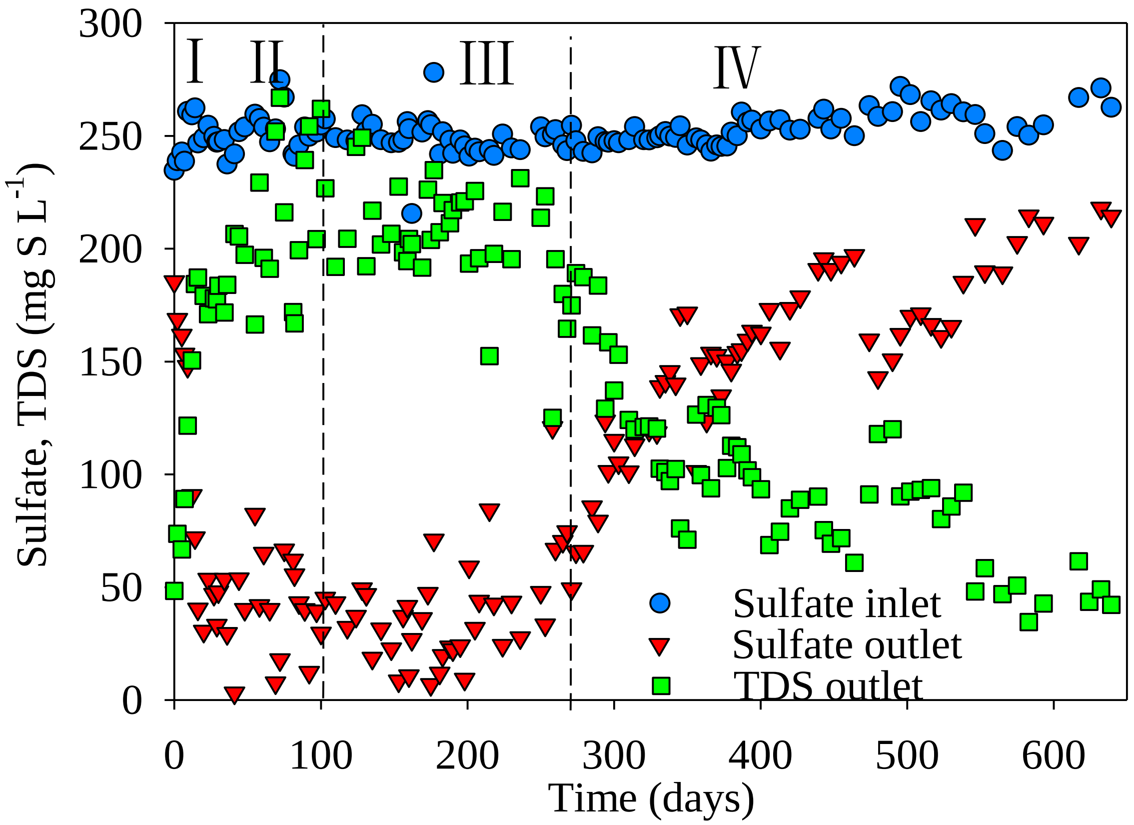
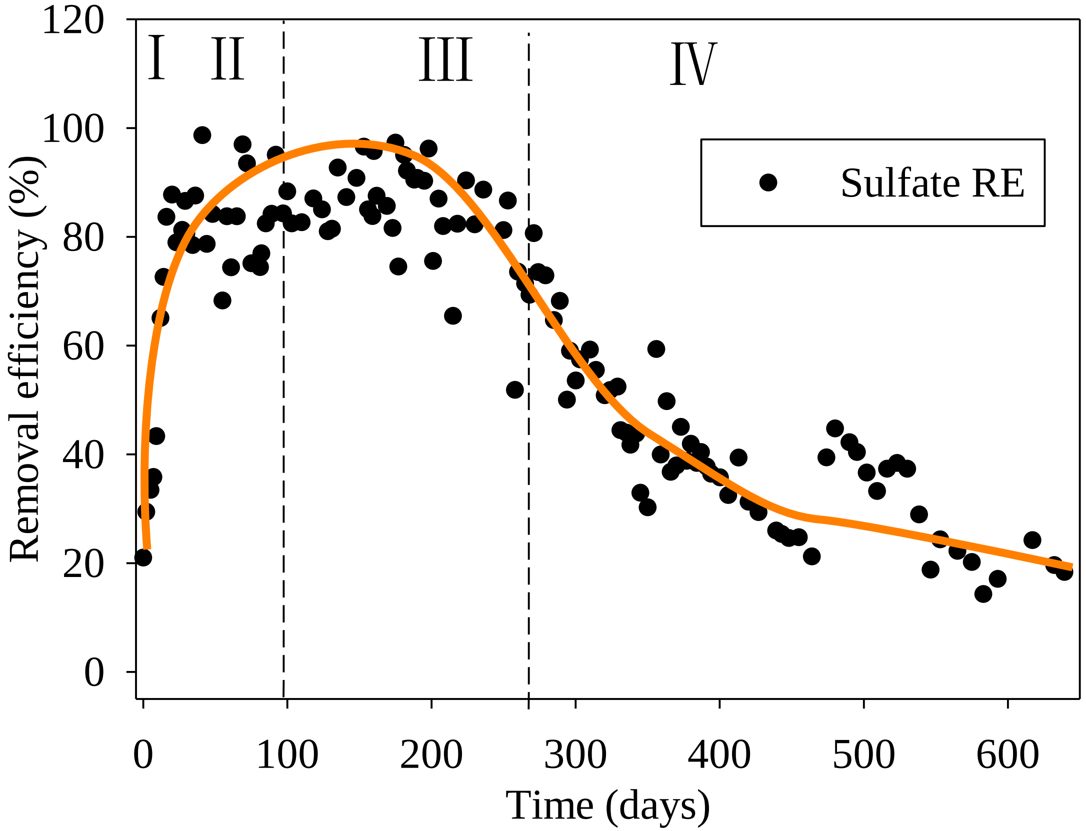




Methanogens wash out correlated well with loss of methanogenic capacity and VFA accumulation

4. Performance results

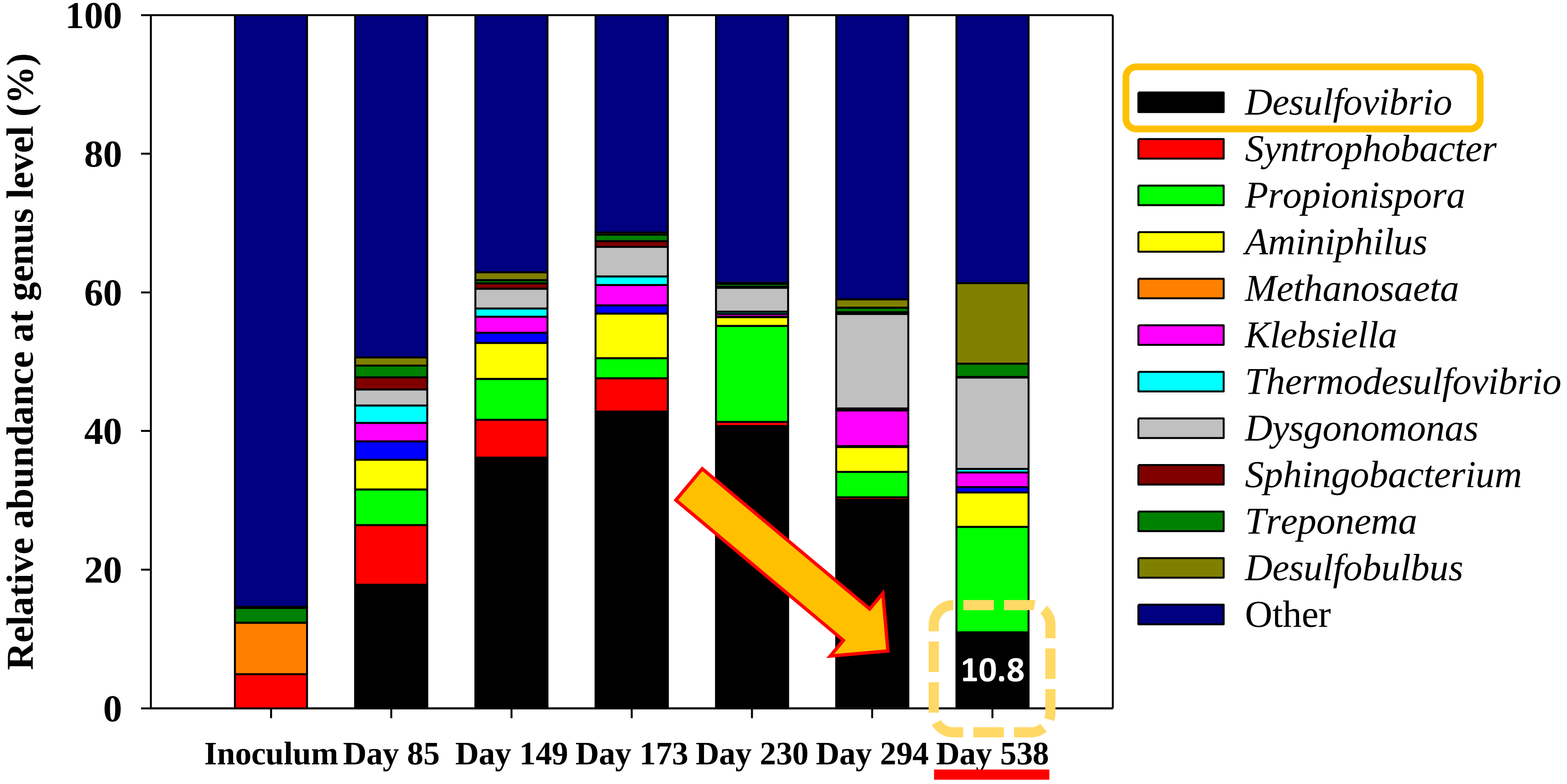


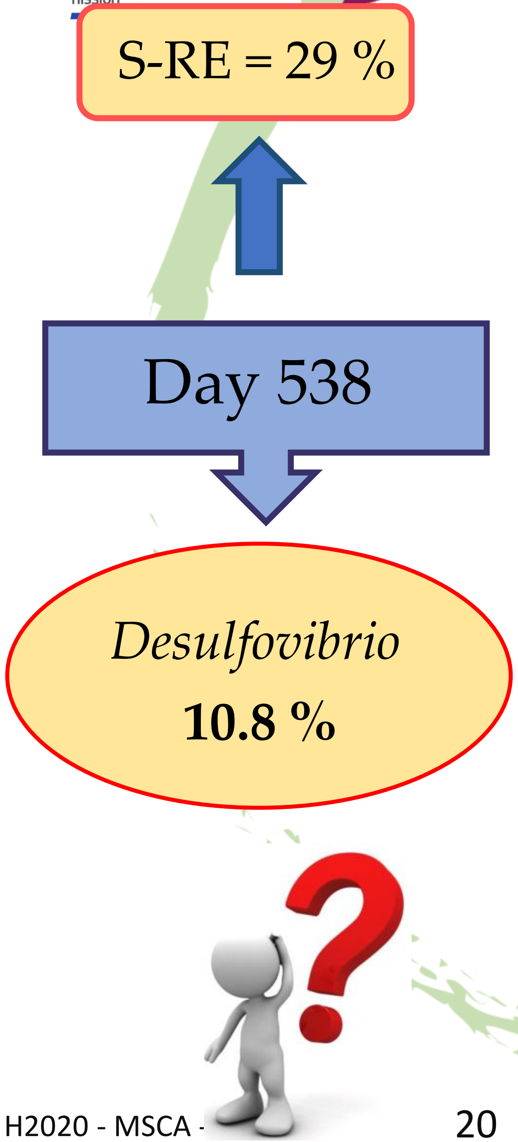


Long-term performance loss related to microbial dynamic changes?

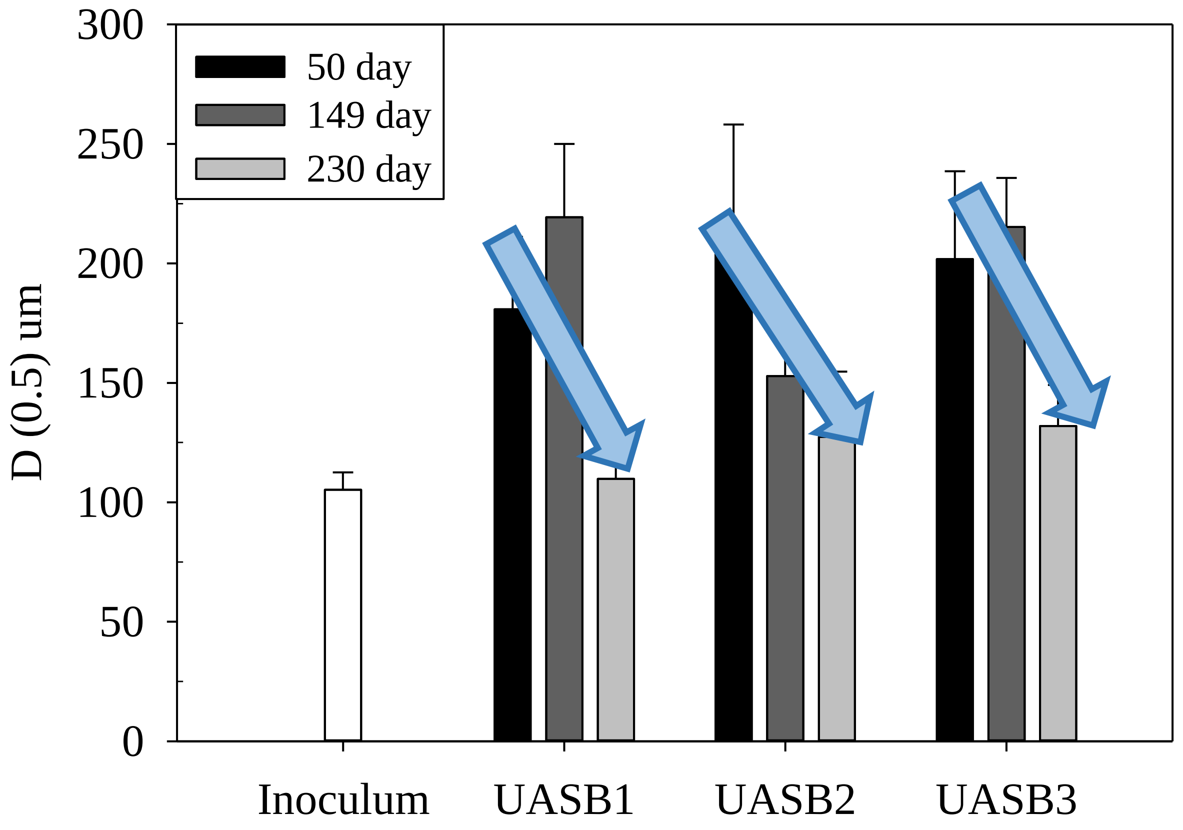
4. Performance results

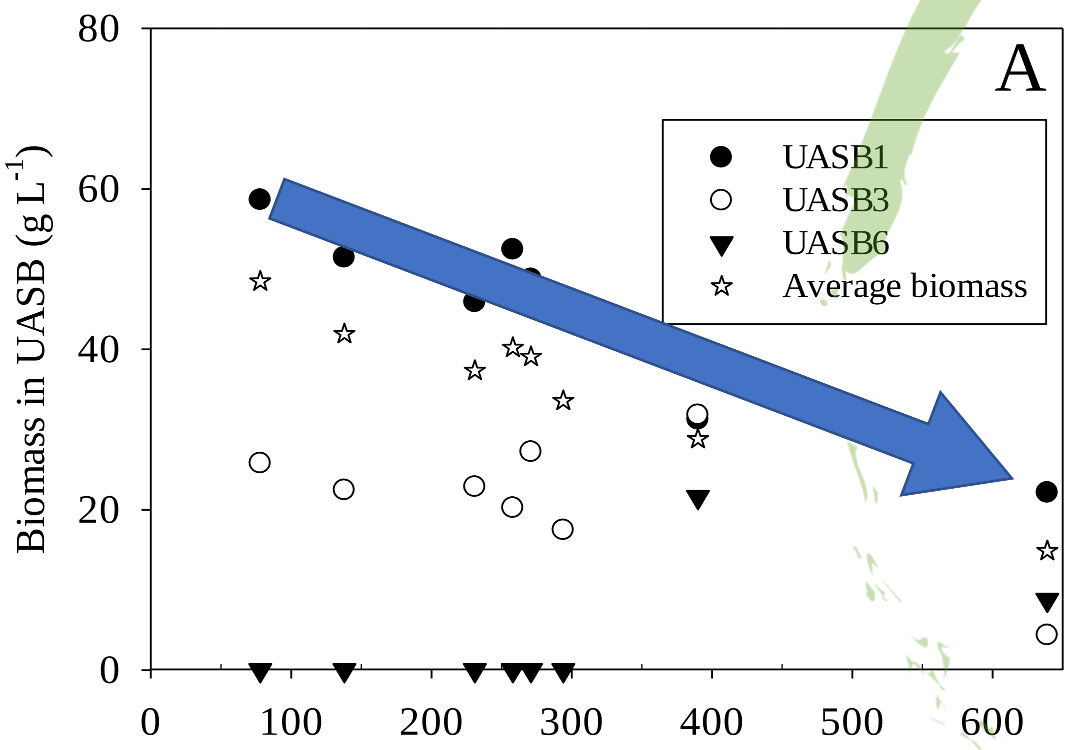
Microbial community dynamics: 16S rRNA sequencing





4. Performance results



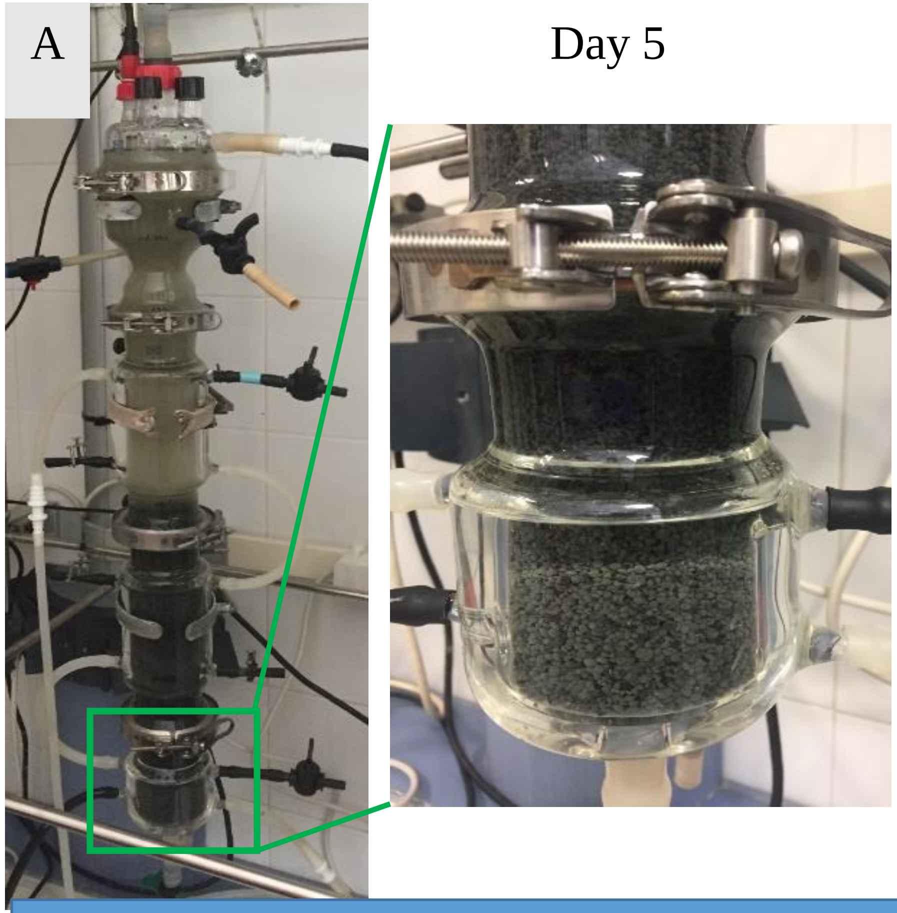


Loss of methanogens lead to a progressive degranulation of sludge and biomass loss

4. Performance results

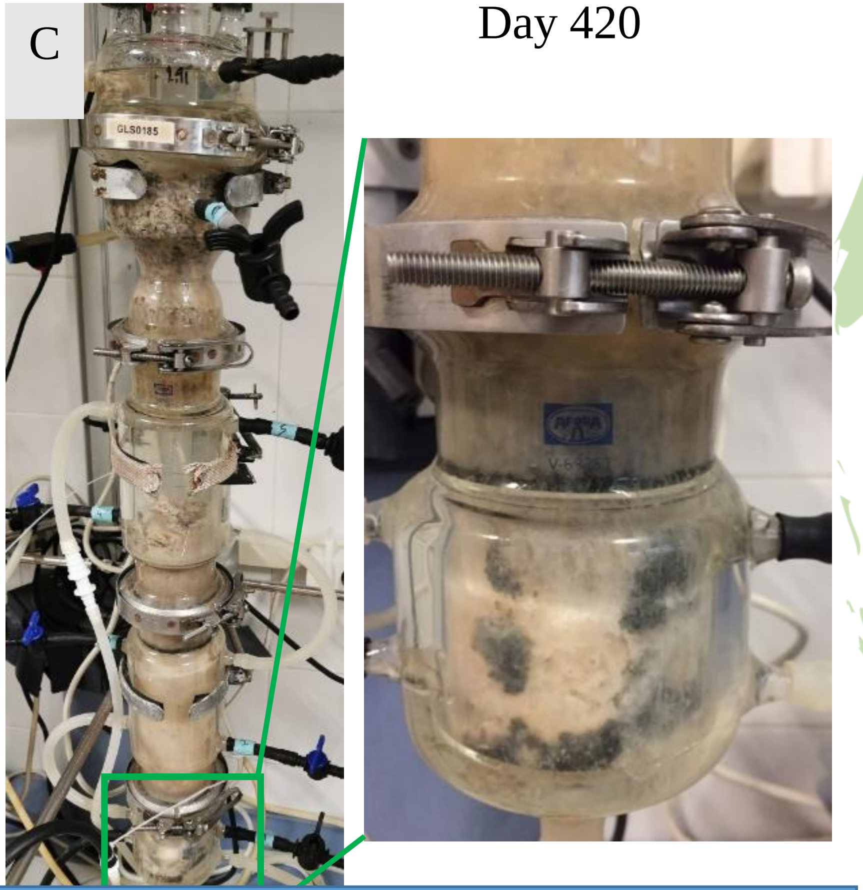
A

Day 5



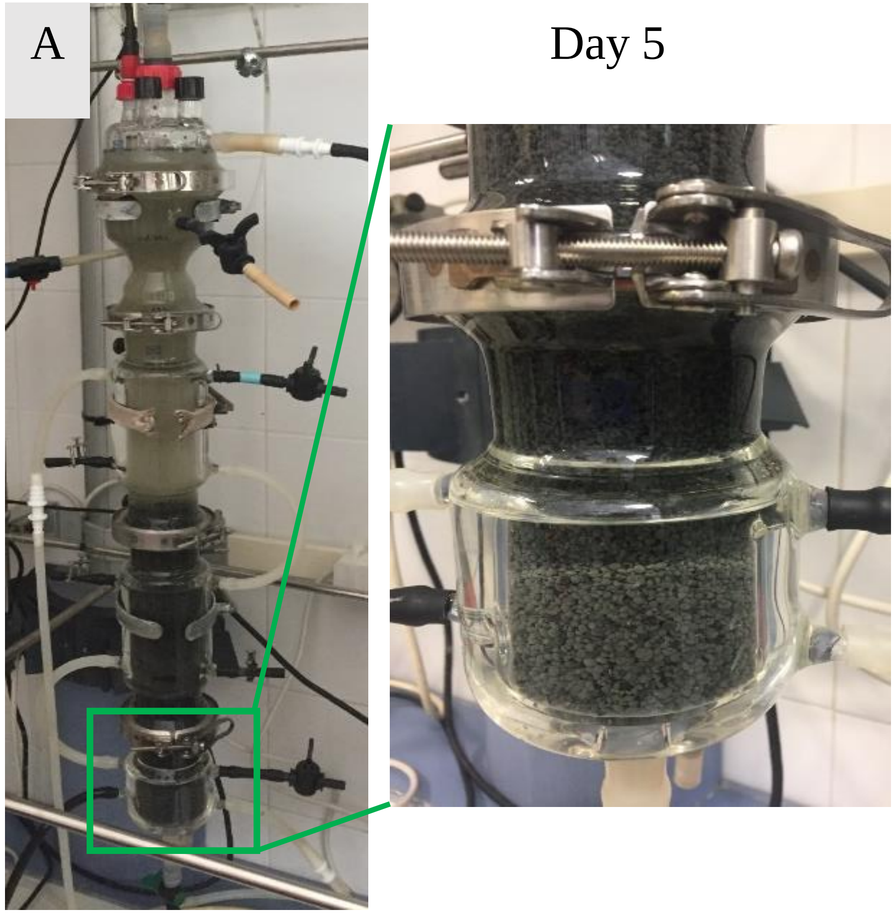
C

Day 420

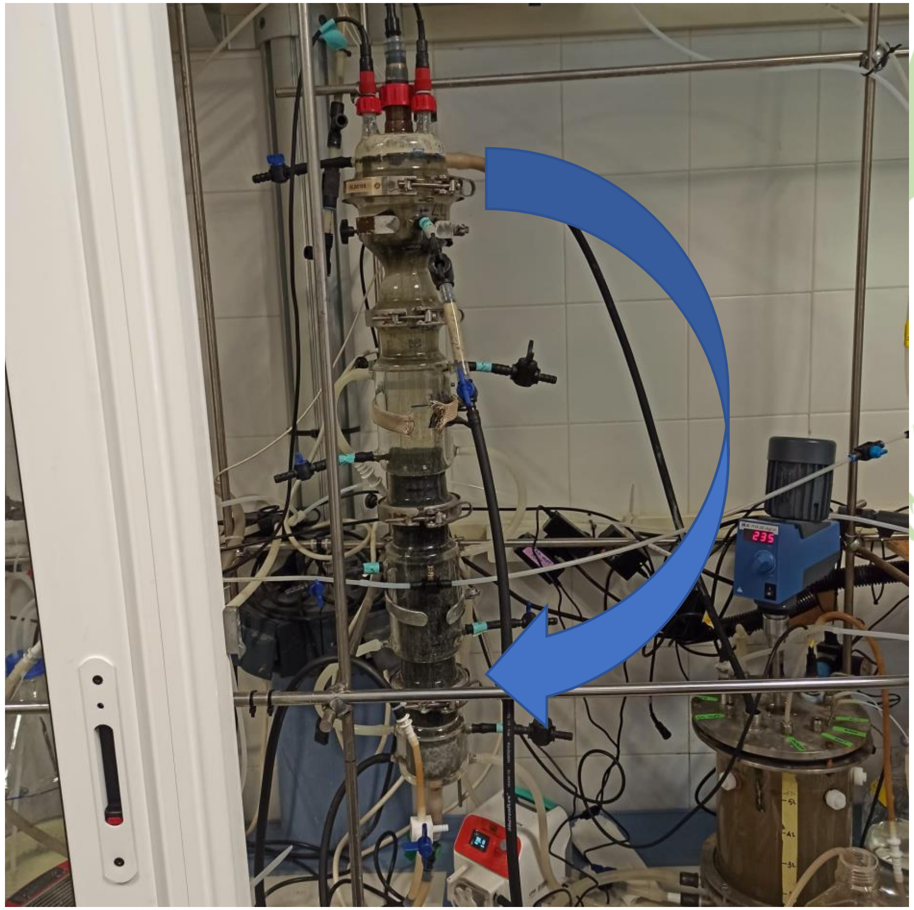


Accumuation of waxes and LCFA present in crude glycerol lead to sludge flotation and potential diffusional limitations

4. Performance results: future work



Internal Recirculation



5. Take home ideas

- ✓ **Long-term operation** of a sulfidogenic **UASB** reactor under **constant loading rate** can be achieved and lead to **highly dynamic microbial diversity changes**

- ✓ The **non-acetate degrader** ***Desulfovibrio*** was found to be the **most abundant** sulfate reducer detected and the **increase in acetate** concentration was related to the **washout of methanogens**

- ✓ **Physical and chemical** parameters and **Illumina** data **correlated** well to explain **methanogenesis dynamics**. However, microbial diversity dynamics **did not correlate well** with the **decrease** in **sulfate and TOC removal efficiencies**. Causes are still unclear

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