Anaerobic digestion of high strength bilgewater with granular sludge: confronting salinity and investigating biomass adaptation

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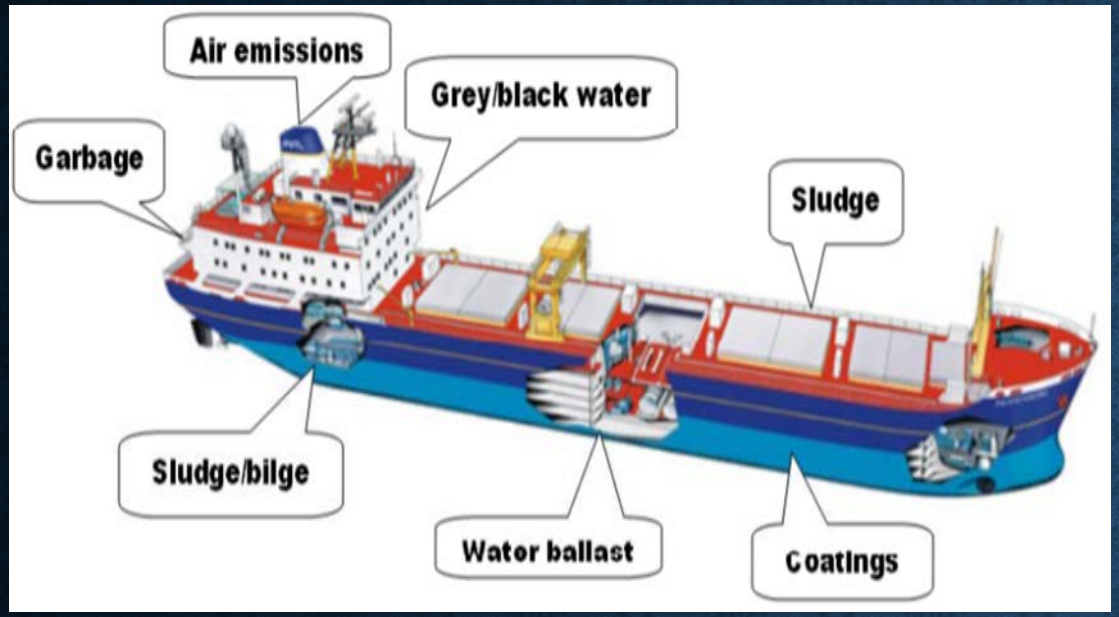
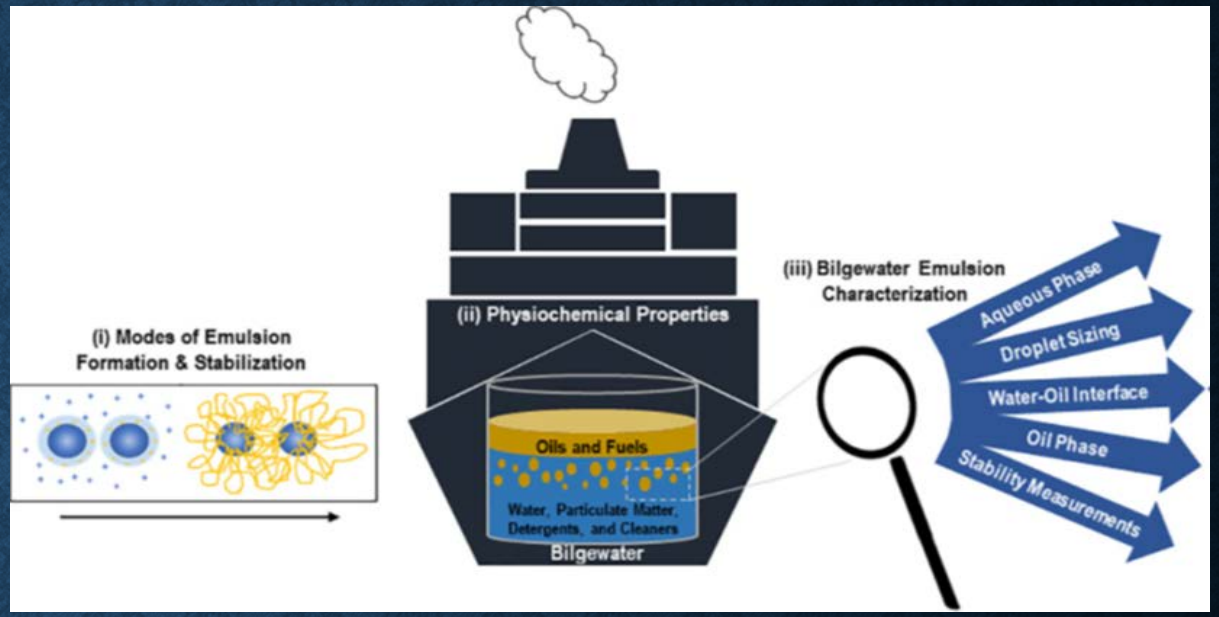
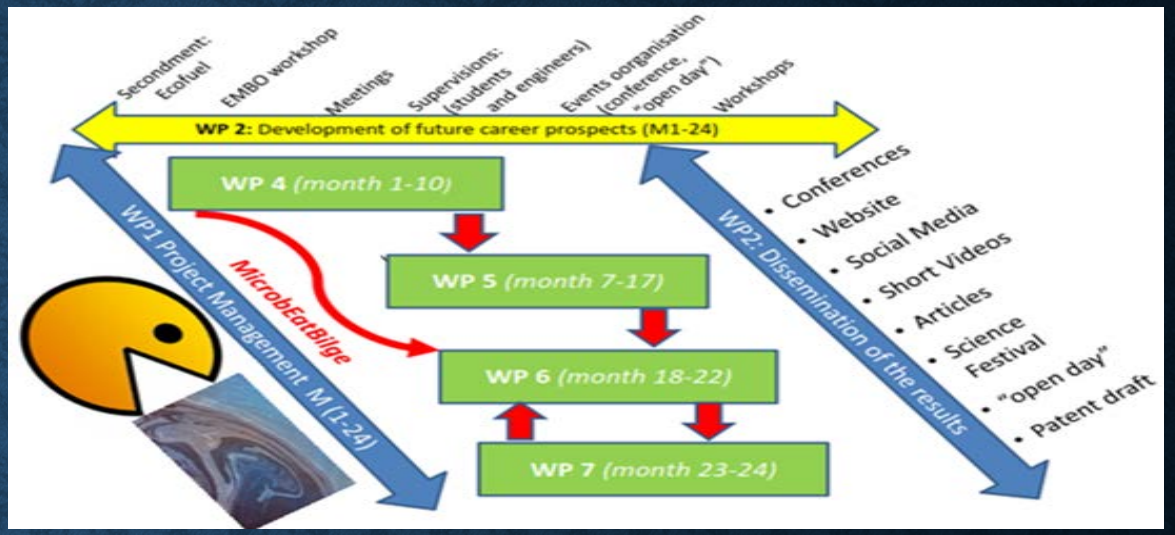
Funded Project: MicrobEatBilge





Bilgewater: saline and greasy wastewater with a high COD (> 3-15 g COD/ L) and salinity (15-35g/L).
Includes: lubricating oil, cleaning diesel oil, oily sludge, spills from the engine room, water leaks from internal pipes, and seawater filtrations and others.
Discharge of oil residue to marine environments is prohibited according to the International Maritime Organization (IMO) regulations (MARPOL 73/78) and the European directive 2000/59/EC.

Project "MicrobEatBilge"



Due to recalcitrant nature of bilge water Chemical or Physical processes are preferred (flotation, separation by centrifuge, filtration, ozonation, coagulation)

Challenge: To investigate the anaerobic treatment of bilgewater

Figure 1: Ecofuel LTD infrastructures, treatment of waste oils oily waters and petroleum waste (Cyprus)

- Objective 1: To point out the effect of salinity in anaerobic digestion of bilgewater
- Objective 2: To examine the microbial profile of anaerobic granular sludge expose for a long time to bilgewater
- Objective 3: To investigate strategies to alleviate this inhibition, such as the use of activated carbon synergistically with ZVI

MATERIALS AND METHODS

Biochemical Methane Potential

Gass composition (CH_4, H_2, CO_2, O_2)

gas chromatography used (Vardanyan et al. 2018)

- Agilent 7820A system
- ShinCarbon ST packed column
- Thermal Conductivity dDetector
- Argon as the carrier gas

Biogass volume

Analytical Methods

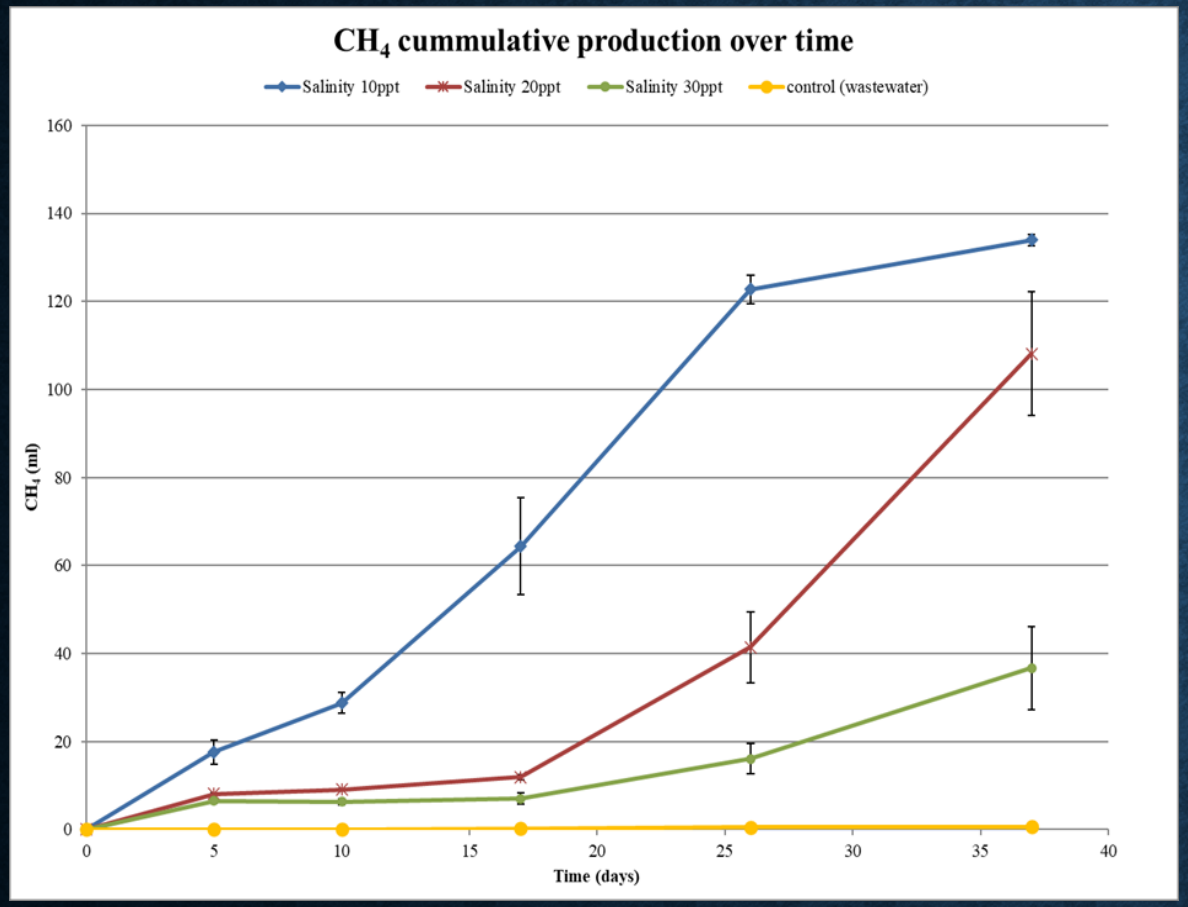
Wastewater characterization

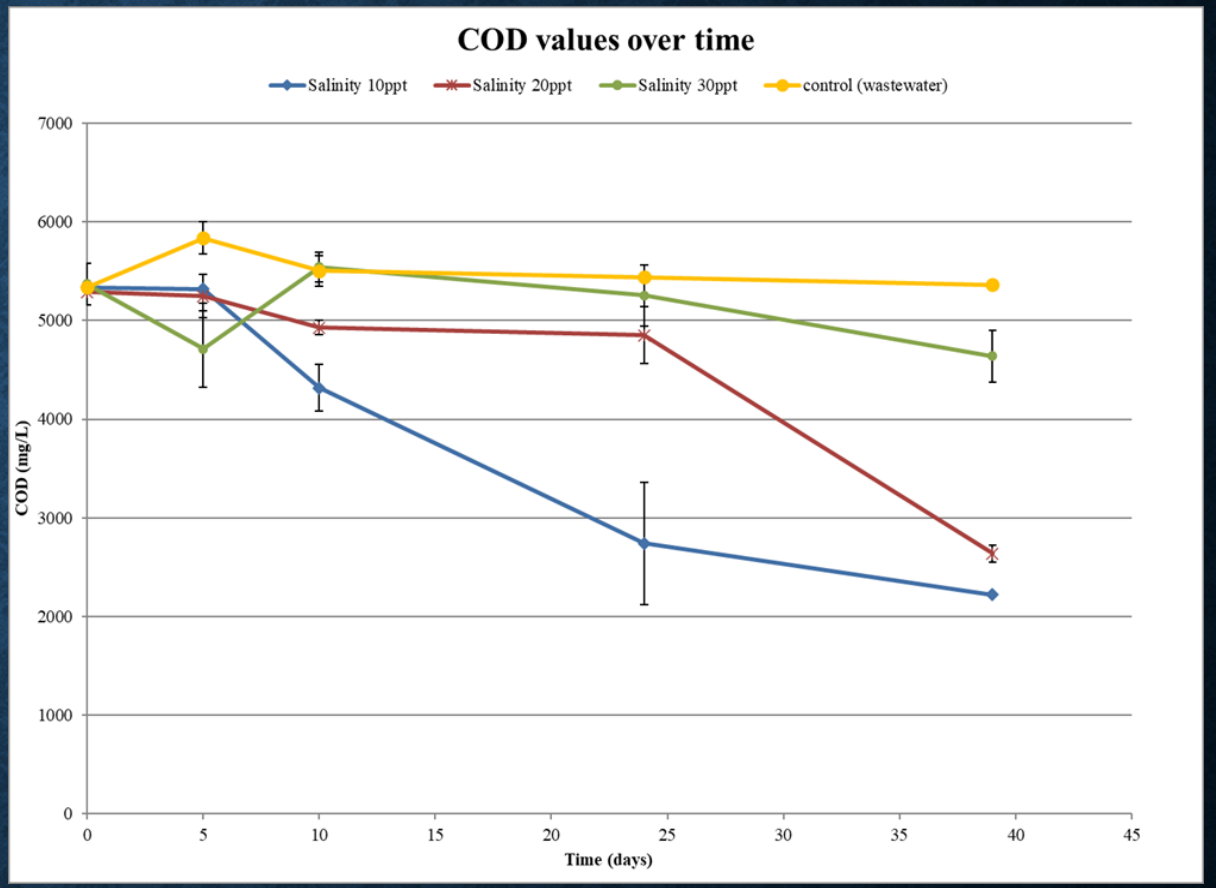
- COD with a modified method for saline samples (Mazioti et al. 2020)
- BOD_5 with a respirometric system (Lovibond BD 600)
- Salinity, pH with portable instruments (Consort C6030)

DNA extraction and sequencing

- DNA extracted with NucleoSpinDNAstool kit
- sequencing

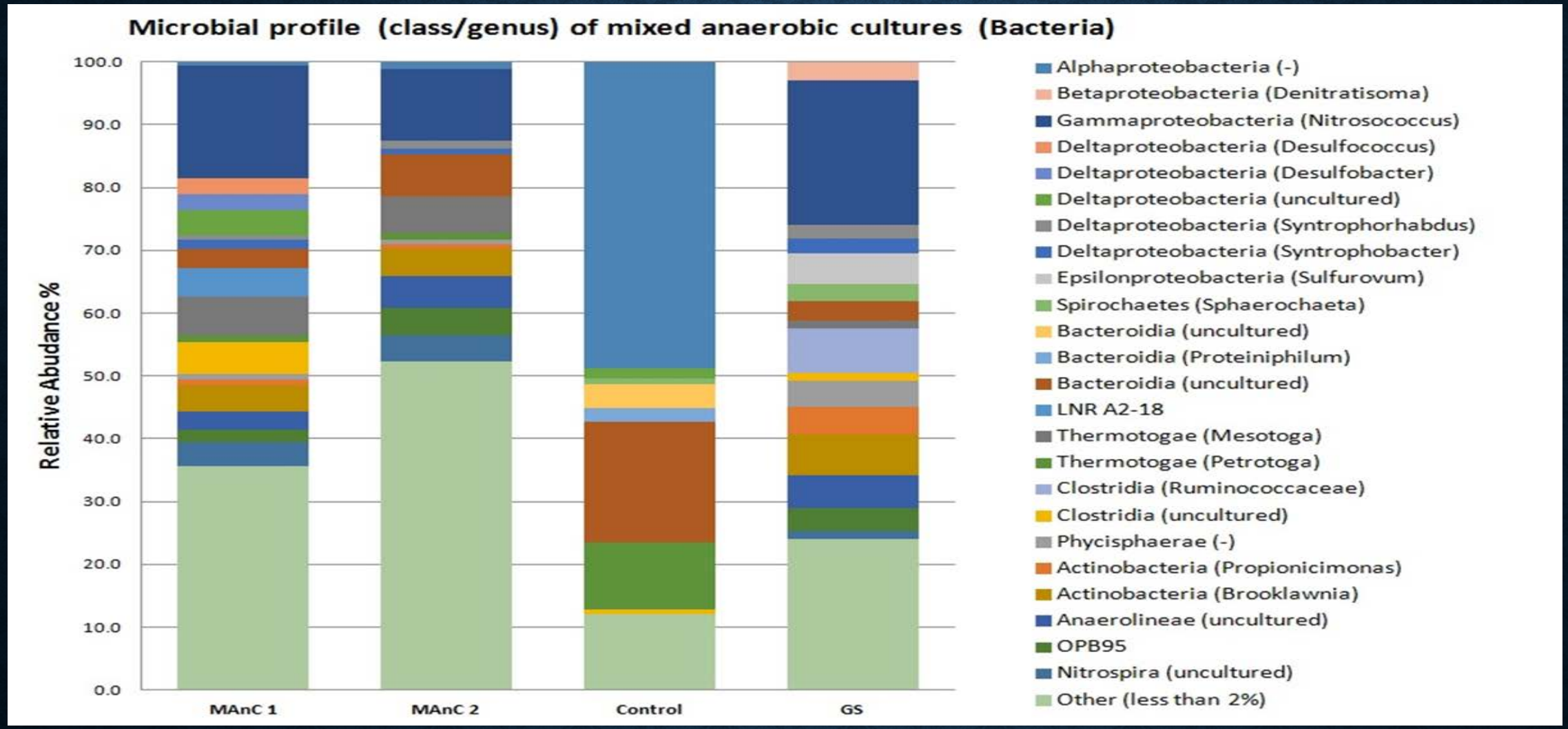
RESULTS





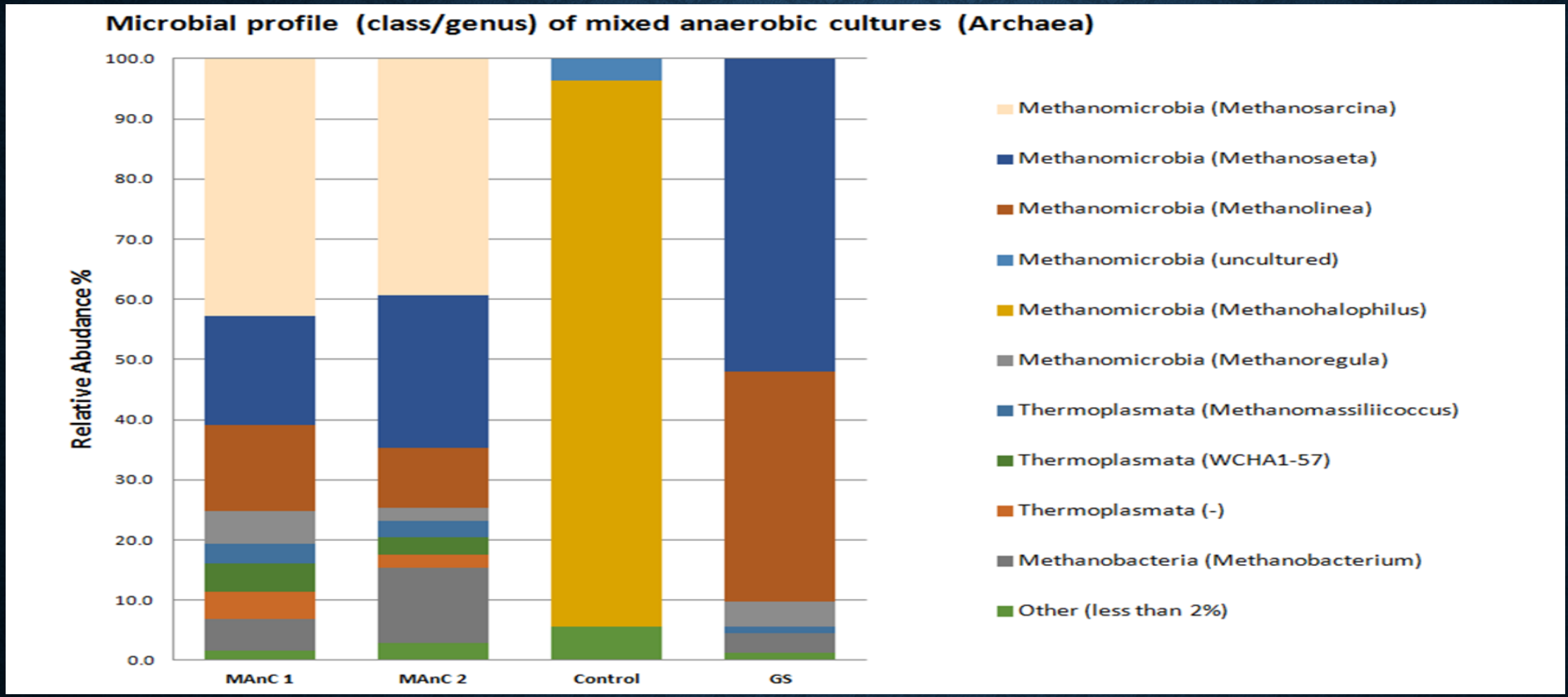
- Salinity increase from 10 → 20 gNaC/L -lag phase 17 days, 18% lower CH_4 production
- Salinity increase from 10 → 30 gNaC/L -lag phase 26 days, 72% lower CH4 production

Adaptation of anaerobic granular sludge to bilge wastewater



Microbial profile (Bacteria) of mixed anaerobic cultures in: i) mixed anaerobic culture with granular sludge, after 141 days exposure to bilge (MAnC 1), ii) mixed anaerobic culture with granular sludge, after 51 days exposure to bilge (MAnC 2), iii) suspended biomass formed after 51 days exposure to bilge (Control), and iv) Initial granular sludge used for mixed anaerobic cultures, not being exposed to bilge (GS) (2% cutoff). Class and genus classification is presented in bars and class is also presented in pie charts.

Mazioti, A.A., Vasquez, M.I. and Vyrides, I., 2021. Comparison of different cultures and culturing conditions for the biological deterioration of organic load from real saline bilge wastewater: microbial diversity insights and ecotoxicity assessment. Environmental Science and Pollution Research, pp.1-17.



Microbial profile (Archaea) of mixed anaerobic cultures in: i) mixed anaerobic culture with granular sludge, after 141 days exposure to bilge (MAnC 1), ii) mixed anaerobic culture with granular sludge, after 51 days exposure to bilge (MAnC 2), iii) suspended biomass formed after 51 days exposure to bilge (Control), and iv) Initial granular sludge used for mixed anaerobic cultures, not being exposed to bilge (GS) (2% cutoff). Class and genus classification is presented in bars and class is also presented in pie charts.

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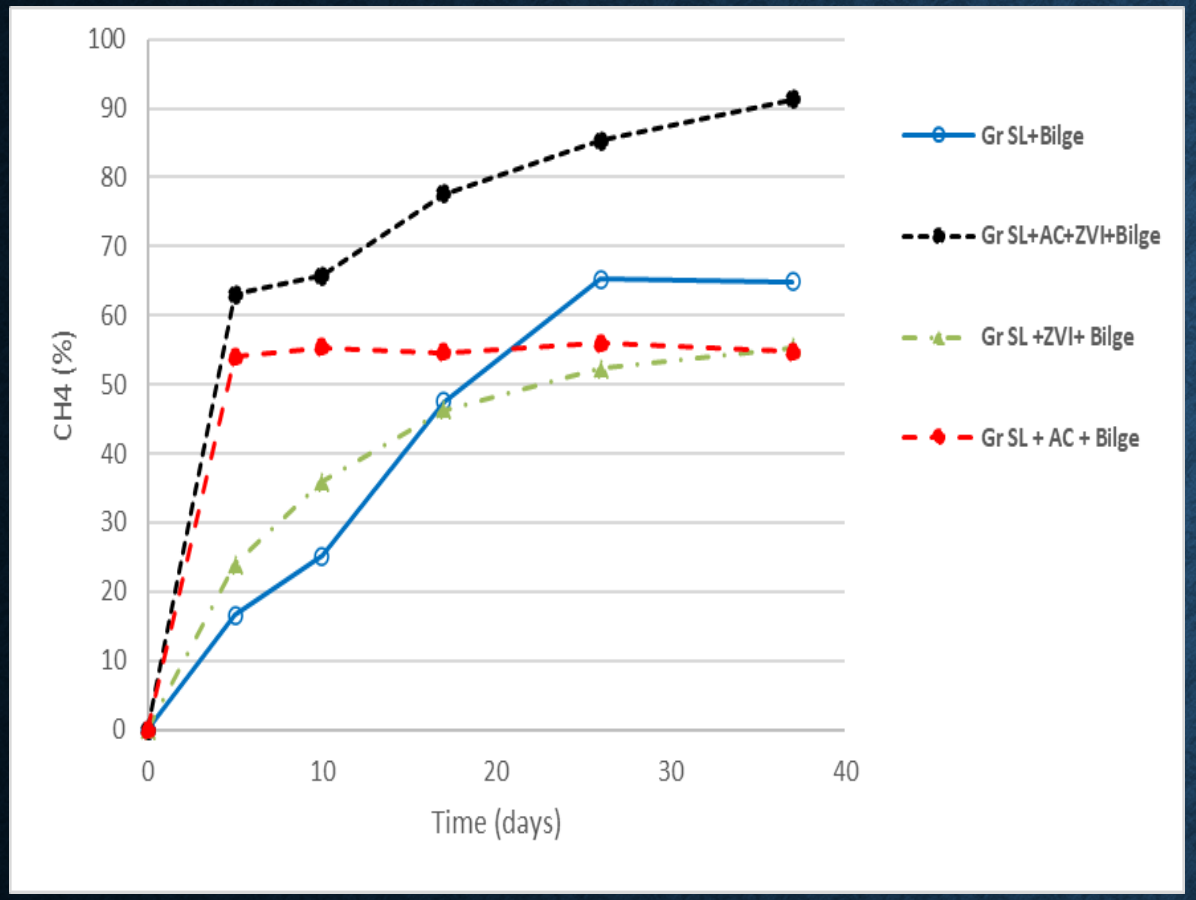

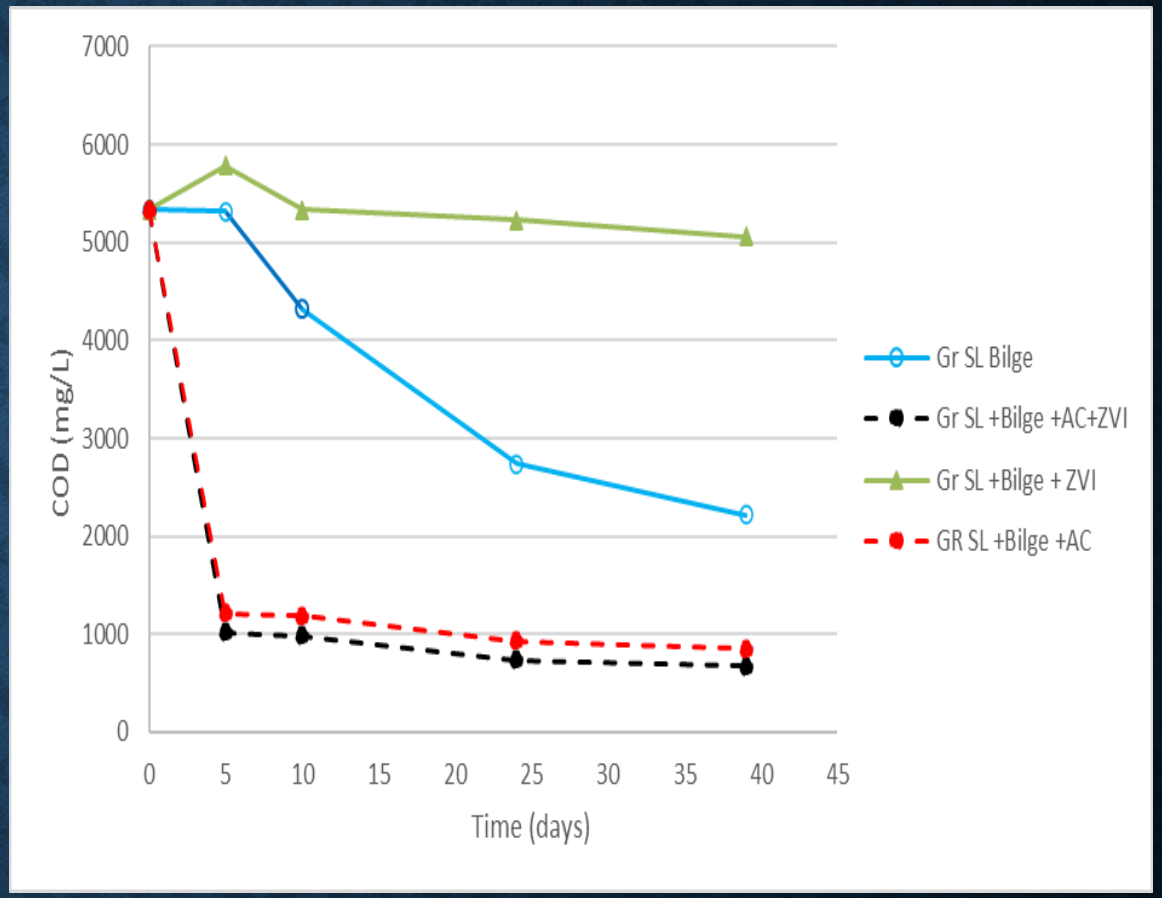

- Exposure of anaerobic granular sludge to Bilge
- Exposure of anaerobic granular sludge to Bilge + zero-valent iron +Activated carbon
- Exposure of anaerobic granular sludge to Bilge + zero-valent iron
- Exposure of anaerobic granular sludge to Bilge +Activated carbon

Why activated carbon and Zero-Valent iron positively contribute to the anaerobic treatment of bilgewater?

Many researchers explain the positive effect of activated carbon on the anaerobic process to the direct interspecies electron transfer theory. However, we should be skeptical when this is proposed as several studies proposed these based only on indirect evidence rather complex set of methods (Van Steendam et al. 2019). Many studies overlook the adsorption of organics in activated carbon that can be also a crucial factor for the performance of activated carbon in anaerobic system.

Regarding ZVI: Many studies proposed that the absence of H2 indicates direct electron transfer with the zero-valent iron in the anaerobic process. However, this could be due to the immediate utilization of H2 by hydrogenotrophic methanogens/homoacetogens compare to the production from ZVI.

Currently, we investigate the synergistic effect of ZVI and activated carbon and the mechanisms for these processes.

CONCLUSIONS

- Bilgewater with 30gNaCl/L is inhibitory to anaerobic granular sludge
- Anaerobic granular sludge exposed to bilgewater for an extended period shows a high abundance of the bacteria class *Deltaproteobacteria* a high abundance of the archaea *Methanosarcina.*
- The addition of activated carbon (3g/L) and ZVI in anaerobic granular sludge resulted in higher than 90% of CH4 and high COD removal (60%) compared to no addition

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