

Supplementary information

**Addition of granular activated carbon during anaerobic oleate degradation
overcomes inhibition and promotes methanogenic activity**

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Sample preparation for LCFA quantification

Samples for LCFA extraction and esterification were prepared following Guihéneuf et al.¹ with modification following the steps below:

- Entire bottle content after samples are taken for VFA measurement were lipolyzed using a freeze dryer at -56 °C and 0.050 mbar for 1 week using a freeze dryer L-200 basic with Edwards nXDS6iC dryscroll vacuum pump (Buchi, Mason Technology).
- Freeze dried solids were then collected and dissolved in 3 ml hexane and 0.3 ml HCl [1 M].
- 0.9 ml of the hexane sample from the previous step was taken and mixed with 0.1 ml of internal standard (5 g/L pentadecanoic acid in hexane solution), 5 ml methanol and 1 ml 8% HCl-methanol in a glass vial with PTFE protected seal caps.
- Vials were vortexed for mixing and then incubated for 1 h in 100 °C for esterification process;
- After incubation, the vials were cooled down before adding 1 ml hexane and mixed for 20 s.
- 3 ml demineralized H₂O was then added and shake vigorously to stop the reaction,
- Vials were left to stand to allow for the organic-water layer to separate.
- 1 ml from the top layer was taken and used for LCFA analysis.

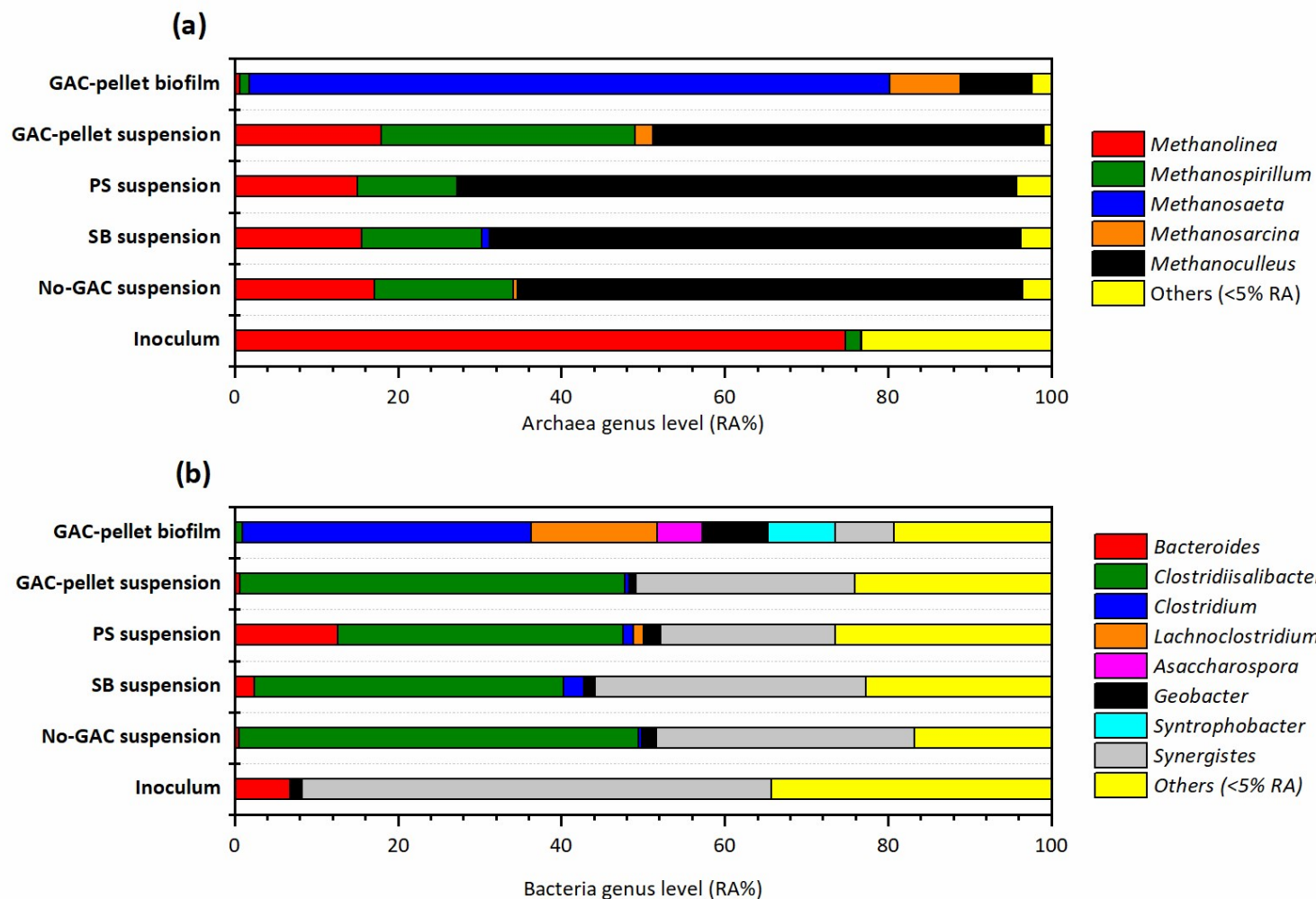
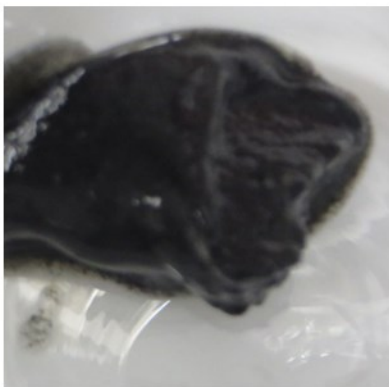


Figure S1 Relative abundance of archaeal (a) and bacterial (b) groups at genus level in the inoculum, suspensions (no-GAC, SB, PS and GAC-pellet) and GAC-pellet biofilm. Sequences with an RA < 5% were grouped together as ‘Others’.

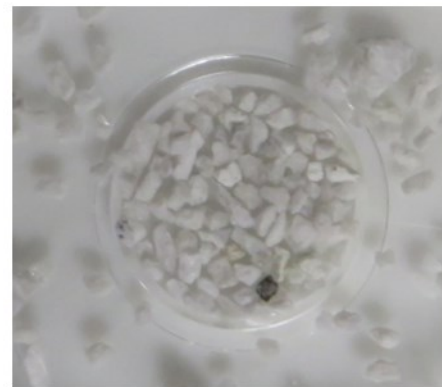
(a) Crushed inoculum



(b) Silica bead



(c) Pumice stone



(d) GAC-pellet



(e) GAC-powder



(f) GAC-slag

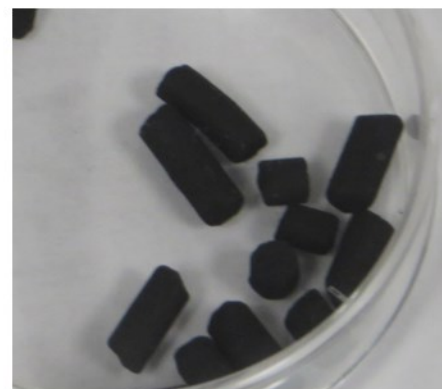


Figure S2 Digital images of inoculum and carrier materials used in this study.

References

1. Guihéneuf F, Schmid M, Stengel DB. 2015. Lipids and fatty acids in algae: Extraction, fractionation into lipid classes, and analysis by gas chromatography coupled with flame ionization detector (GC-FID), p. 173–190. *In* Stengel, DB, Connan, S (eds.), *Methods in Molecular Biology*. Springer Science, New York, USA.