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Molasses injection as a MEOR strategy:

Enrichment incubations of brine/oil from North Sea Oil Field

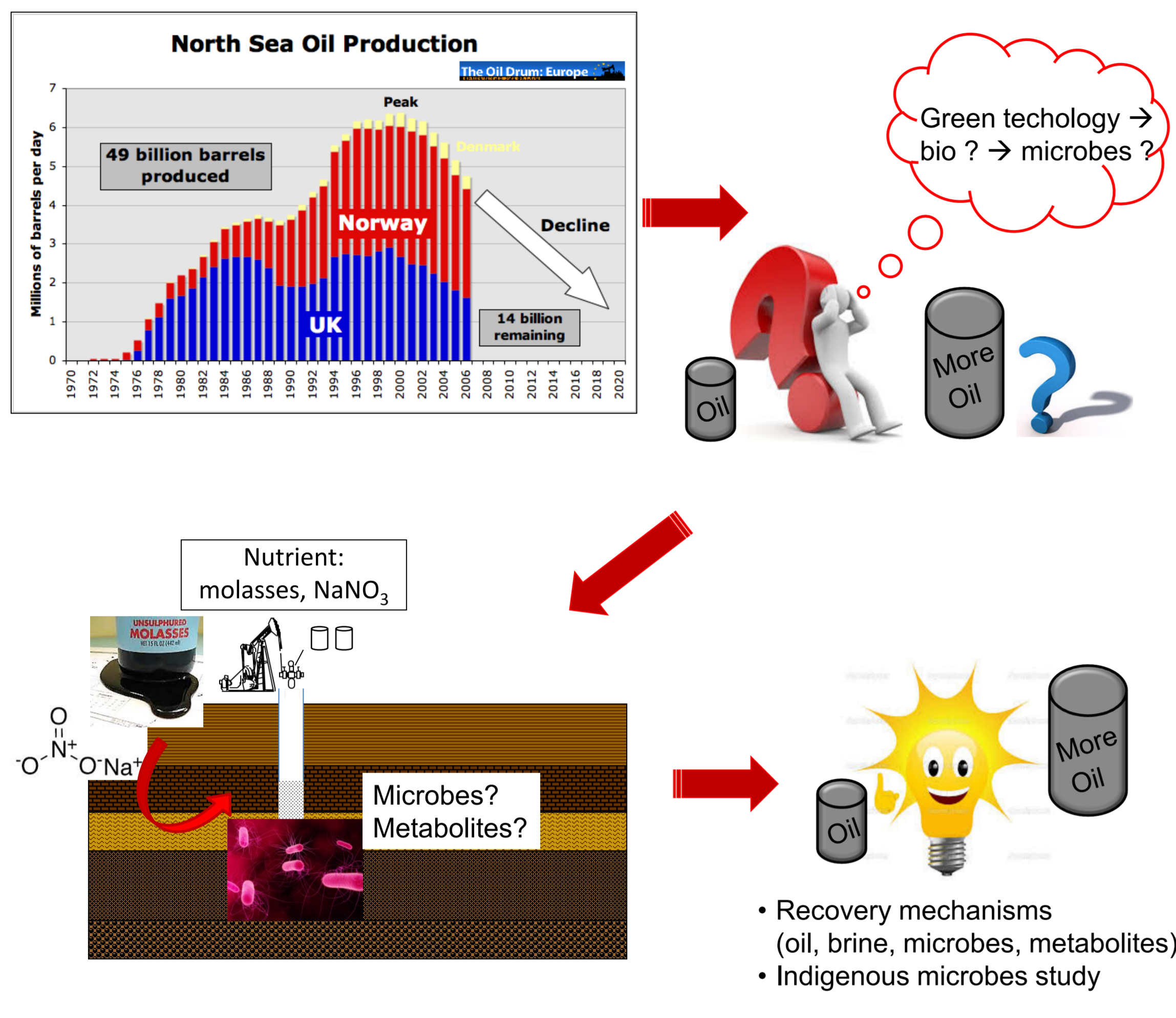
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Background and Objectives



Results

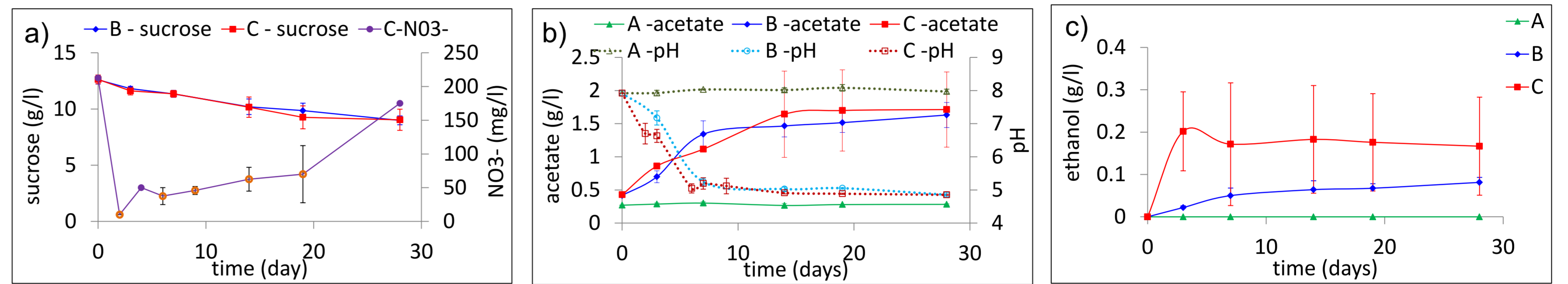


Figure 2. (a) molasses (sucrose) hydrolyses by microbes in incubation B (blue) and incubation C (red). Nitrate (NO₃⁻) was monitored in incubation C, NO₃⁻ was added at day 2, 6, 9, 14, 19 (orange circle) to keep NO₃⁻ concentration at 212.5 mg/l (2.5mM), (b) acetate production and brine pH during incubation, (c) ethanol production during incubation.

- Sucrose hydrolyses (incubations B and C) – presence of fermentative microbes. Nitrate was consumed continuously in incubation C (Fig. 2a).
- Acetate production – pH decrease (incubations B and C, Fig. 2b) and ethanol production (Fig. 2c).
- No production of succinate, lactate and glycerol on all incubations.

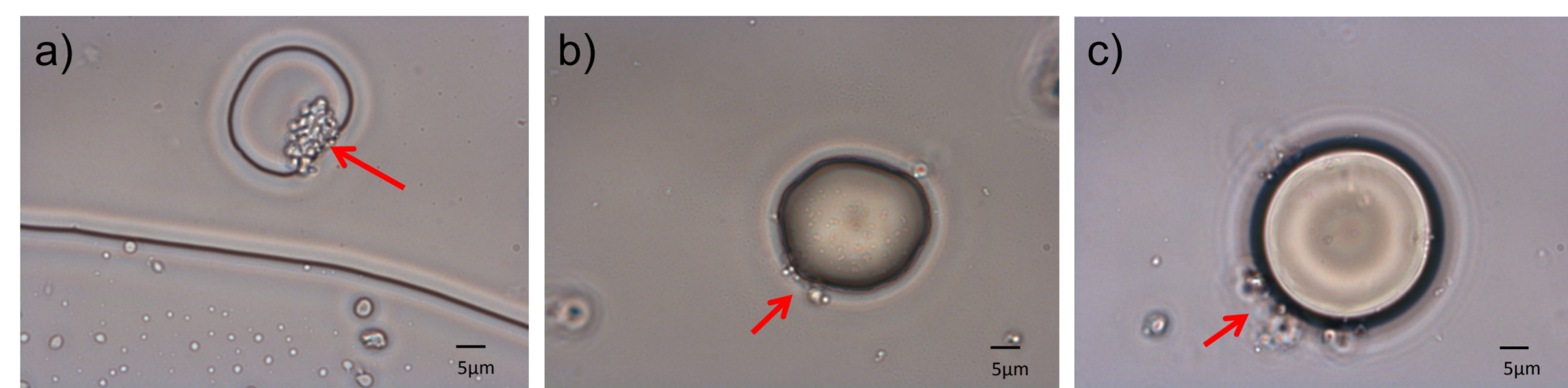


Figure 3. Microbes were present in the oil-brine interphase in all incubations (red arrow). (a) In incubation A, without molasses, microbes had a tendency to form aggregates. (b, c) In incubation B and C, with addition of molasses, microbes are present as single cell (loose particle).

Experimental Design

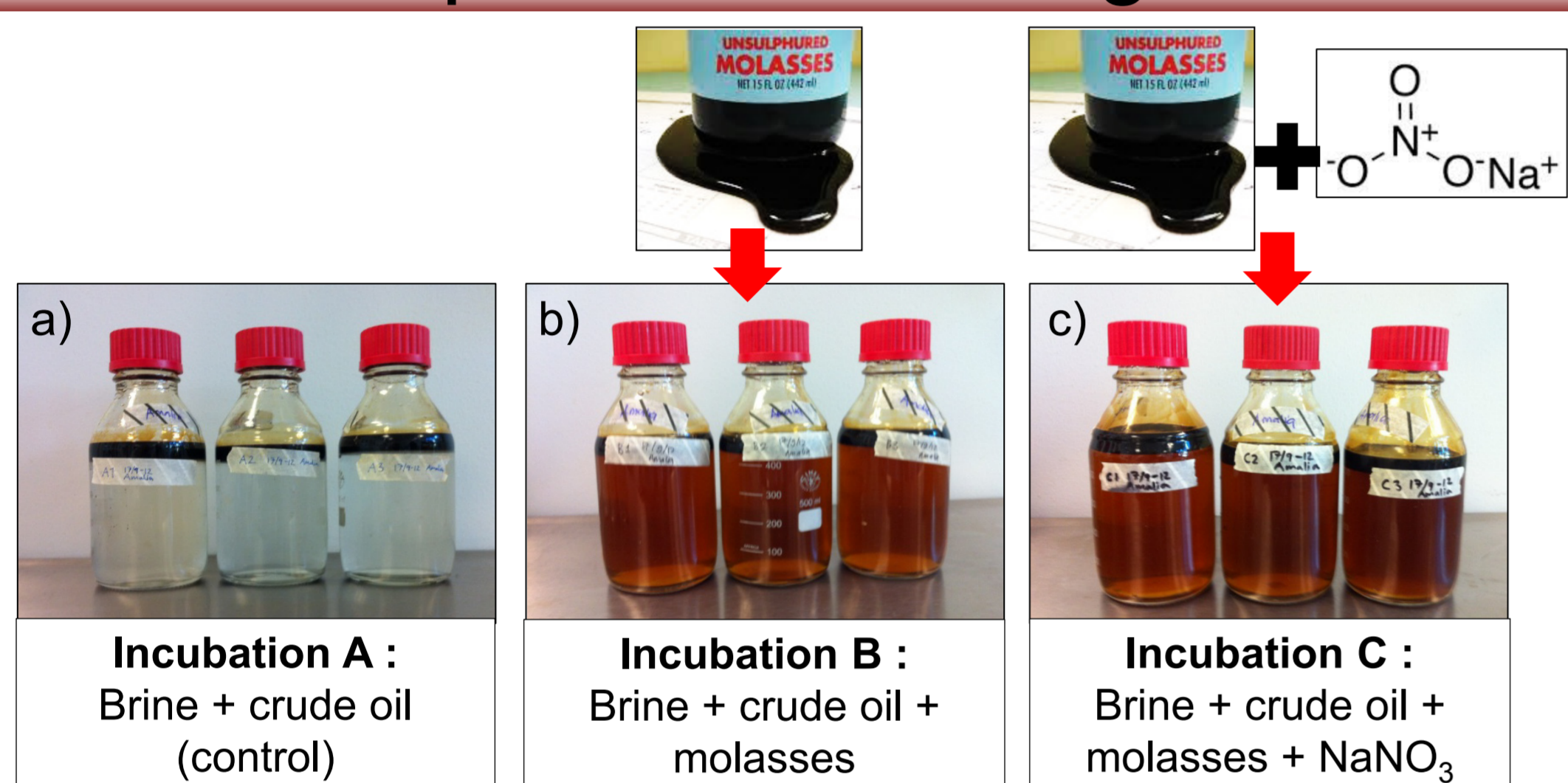


Figure 1. Anaerobic enrichment tests by using brine and crude oil from the North Sea oil field for 28 days at 55°C. Three different conditions on the enrichment test: (a) incubation A (control) – no nutrient addition, (b) incubation B with addition of 2% molasses and (c) incubation C with addition of 2% molasses and 2.5 mM nitrate.

Table 3. Interfacial tension (IFT) between the brine and the crude oil in incubation A, B and C

Sample	T0	T28 - with biomass	T28 w/o biomass
A	19.29 ± 0.12	17.70 ± 1.48	15.20 ± 0.32
B	18.79 ± 0.18	2.34 ± 0.15	21.65 ± 0.15
C	18.60 ± 0.09	2.96 ± 0.43	22.60 ± 0.88

- In chalk it is important that microbes are present as single cells (hydrophilic) to prevent blocking of pore throat.
- No significant change in oil characteristics: viscosity, density and composition (total weight fraction) in all incubations.
- Formation of gasses, emulsions and smaller oil droplets in incubations B and C.
- IFT decreases when microbes are present in oil-brine interphase (Table 3).

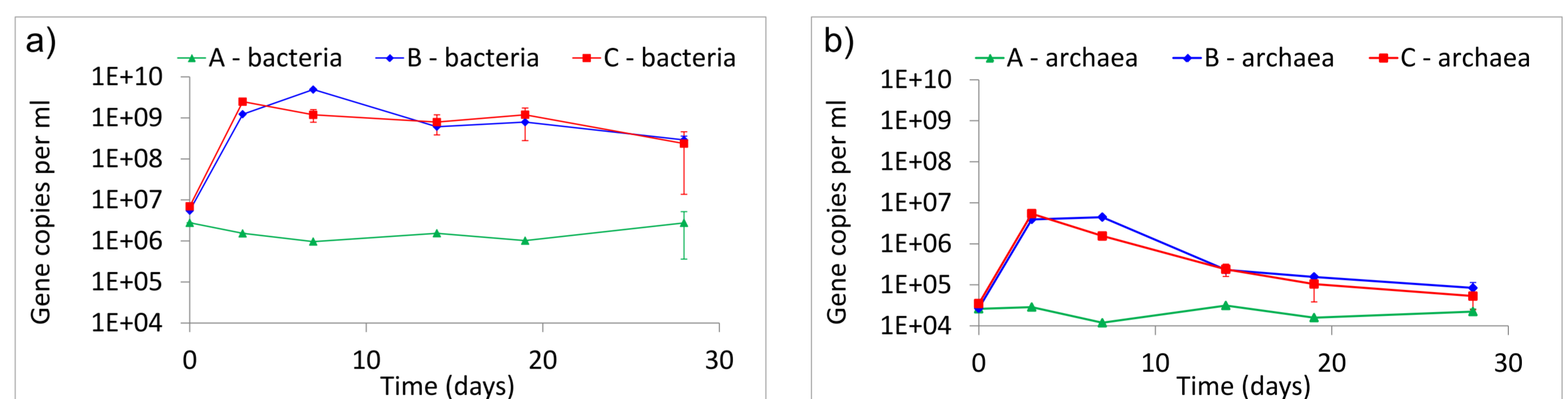
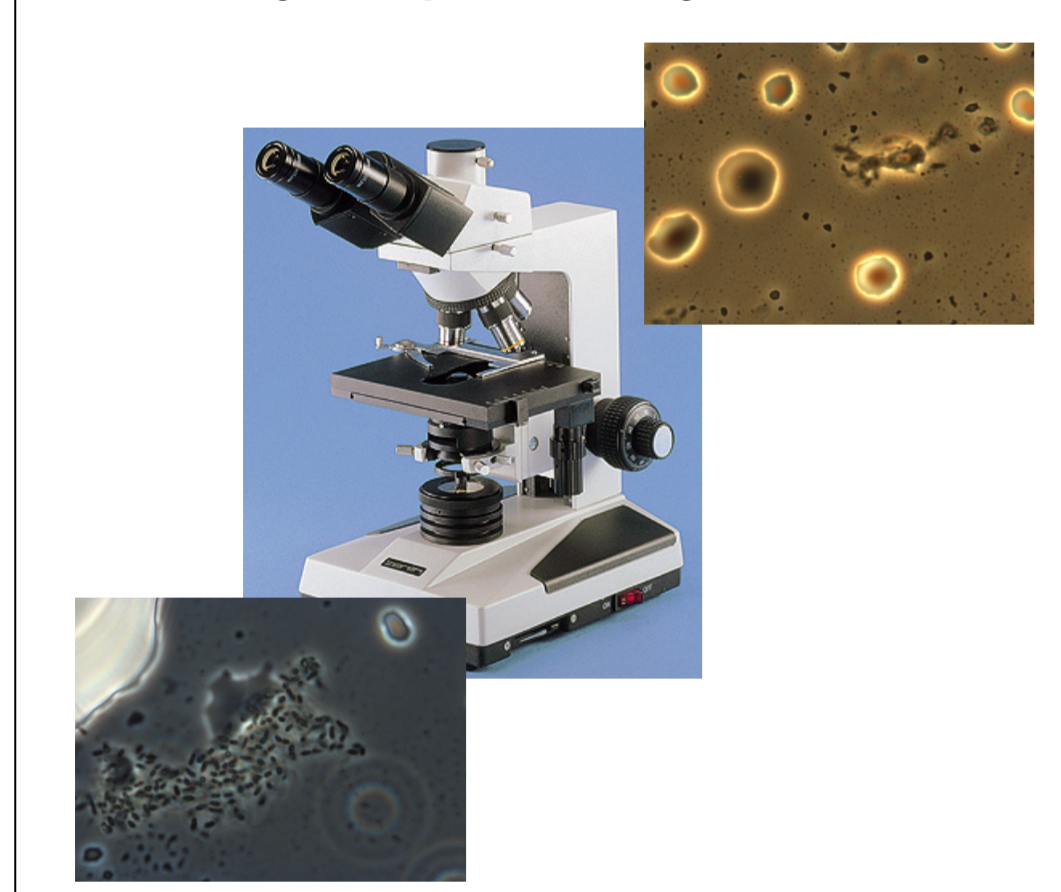
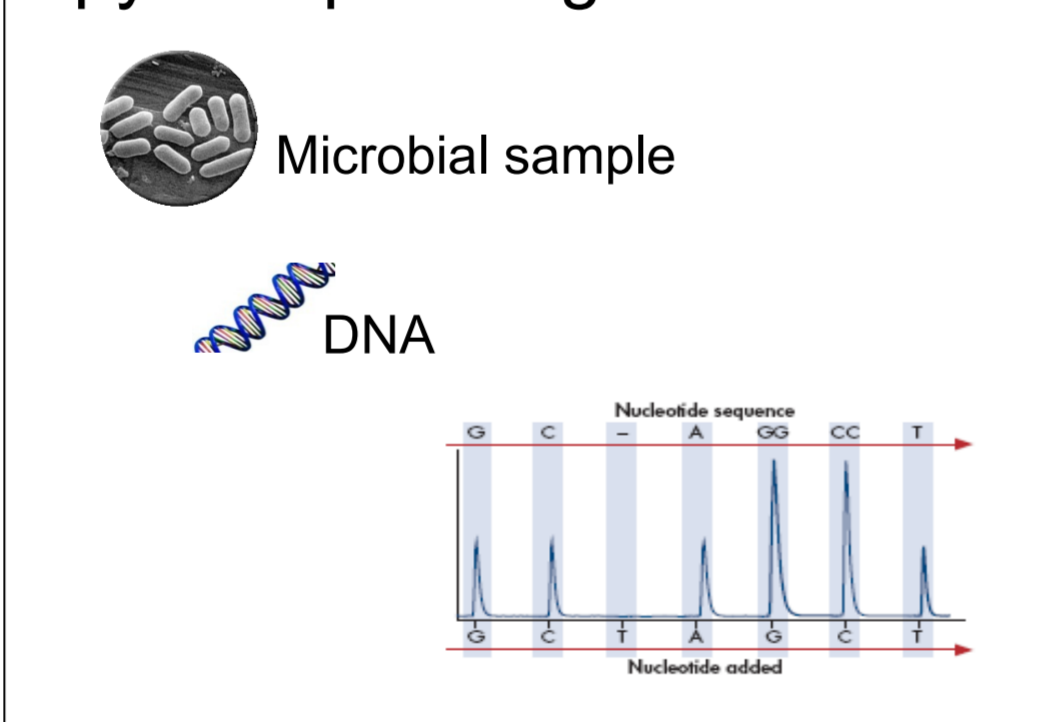


Figure 4. Abundance of 16S rRNA bacteria and 16S rRNA archaea gene copies in incubations A, B and C.

Cell hydrophobicity



How many microbes? qPCR



- Sugar hydrolysis and metabolite production
- Oil characteristics (viscosity, density, and total weight fraction)
- Interfacial tension (oil vs. brine)
- Emulsion



Acknowledgement

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Conclusion

- Production of fermentation products : acetate, ethanol and gas from sugar hydrolyses.
- No significant change in oil characteristics; but formation of emulsion, smaller oil droplets and reduction of IFT were observed due to microbial biomass when molasses was added (incubation B and C).
- Addition of molasses made microbe cells hydrophilic - prevent microbes to form aggregates.
- Molasses stimulated microbial growth in incubations B and C, as shown by increase in cell number of both bacteria and archaea (qPCR results).
- Pyrosequencing revealed that molasses stimulated the growth of *Petrotoga* and *Anaerobaculum*.
- Nitrate was consumed continuously and *Petrotoga* was favoured over *Anaerobaculum* in the presence of nitrate (incubation C).
- The growth of methanogenic archaea was potentially fueled by products from fermenting bacteria.

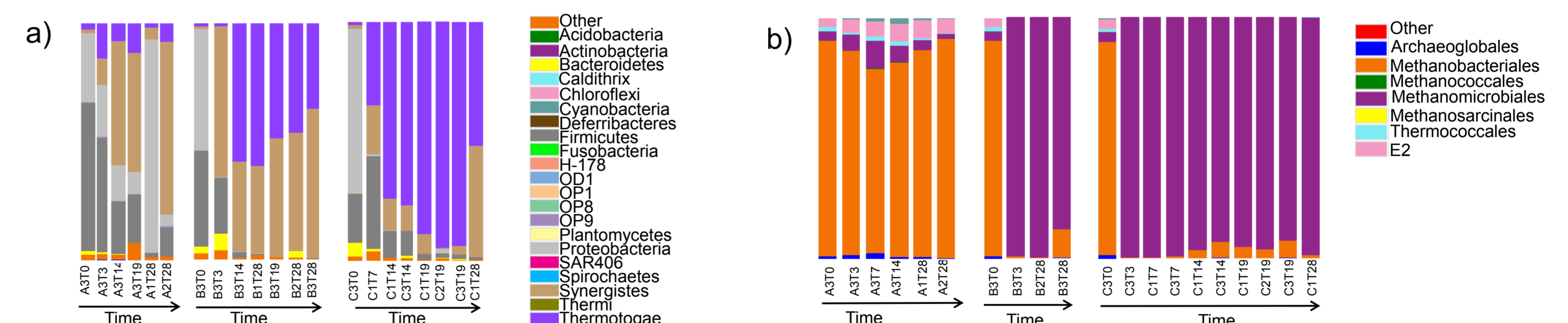
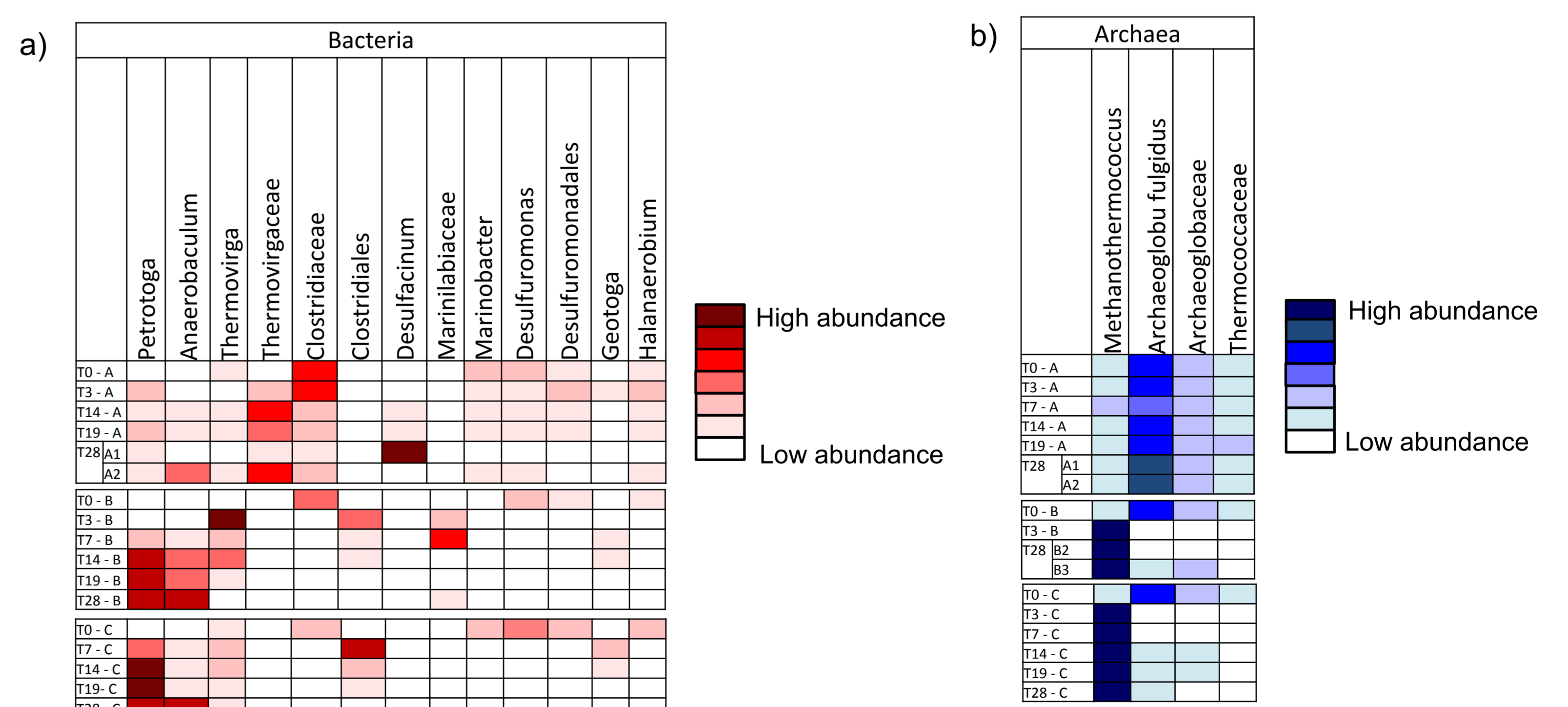


Figure 5. Development in (a) bacterial community at phylum level and (b) archaeal community at organism level in incubation A, B and C.

Table 2. Summary of heat map of the most abundant microorganisms based on sequencing of the 16S rRNA gene of (a) bacteria and (b) archaea



- Microbial growth was stimulated by addition of molasses in incubations B and C (Fig. 4).
- Sequencing revealed the population dynamics of the microbial community (Fig. 5 and Table 2).