Does the vertical distribution of cultured sulfate-reducing bacteria reflect elevated activities in deep sediments of an intertidal sand flat?

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Introduction

Fig. 1 Recovering sediment cores from intertidal flats

Sandy sediments show low organic carbon content and microbial cell numbers in comparison to fine-grained compartments. Nevertheless, previous studies on surface sediments of intertidal sand flats demonstrated high microbial activity of sulfate-reducing bacteria (SRB) [1,2]. Those studies, however, were restricted to the uppermost tens of centimeters [3].

Thus, the question arises whether activity of SRB is detectable also in deeper Wadden Sea sediments and, if so, how their community is composed and vertically distributed. Therefore, this study focuses on the *in situ* abundance of SRB that were identified by cultivation-based approaches.

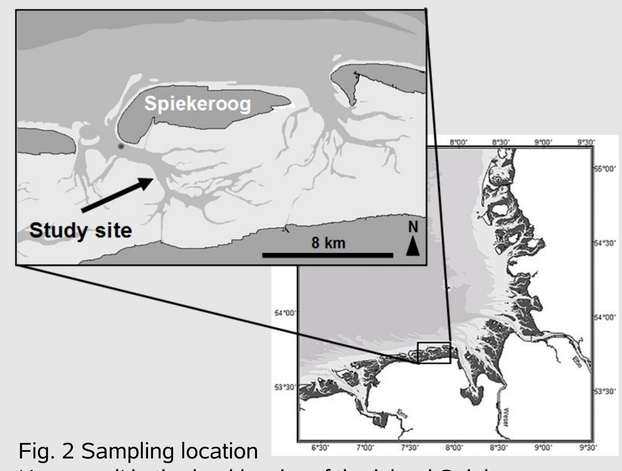

Fig. 2 Sampling location 'Janssand' in the backbarrier of the island Spiekeroog

Results

Sulfate reduction rates

Surprisingly, determination of sulfate reduction rates along a 5 m long sediment core revealed **activity peaks** in subsurface sediments. These rates reached values close to those measured in the near surface layers (Fig. 3).

In situ abundance of SRB

The relative contribution of targeted SRB to the microbial community was low at sediment surface, but increased with depth (Fig. 4).

Deeper layers were characterized by a SRB community numerically almost constant with depth and accounting for up to **7 %** of all cells detected. However, the SRB communities at single layers significantly differed with respect to their composition.

A **predominance of *Desulfotalea* relatives** was shown over the entire sediment column.

Desulfobacula-related bacteria were also found in all layers and partially dominated the SRB community. *Desulfosarcina*-related bacteria gave generally low counts of less than 1% of all DAPI stained cells.

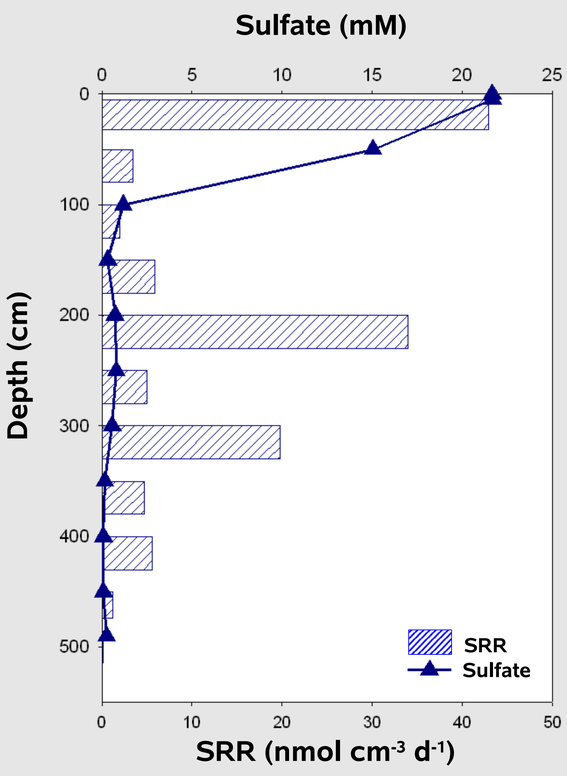

Fig. 3 Pore water concentrations of sulfate and sulfate reduction rates (SRR, determined via tracer injection and whole core incubations)

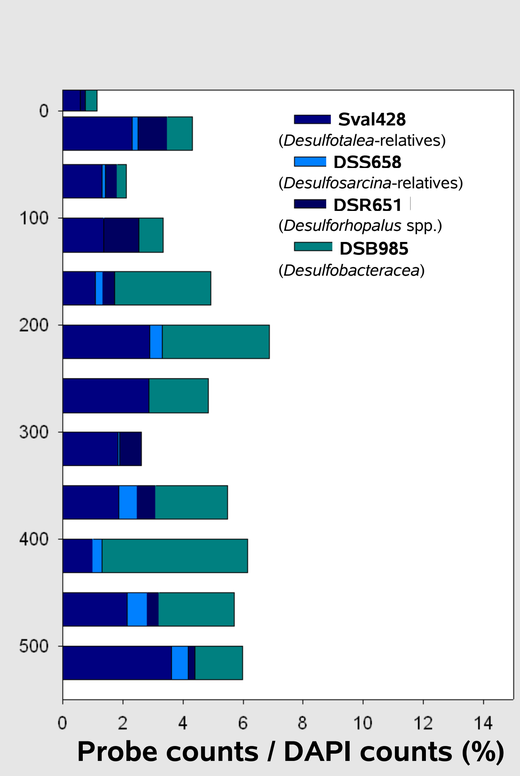

Fig. 4 Depth profiles of different phylogenetic groups of SRB as detected via CARD-FISH using specific oligonucleotide probes

Cultured SRB

Cultivation was performed in liquid dilution cultures inoculated with sediment from four different depths (50 - 400 cm). **Lactate**, **acetate, or hydrogen** served as electron donors to enrich SRB with different physiological capacities.

All enrichments showed sulfide production and, therefore, growth of sulfate reducers. In most cases, highest sulfide-positive dilutions were dominated by a single morphotype (Fig. 5).

Screening by PCR-DGGE and sequencing of selected DGGE bands revealed a variety of partial 16S rRNA genes, mostly related to known marine SRB of the *Deltaproteobacteria* (Fig. 6).

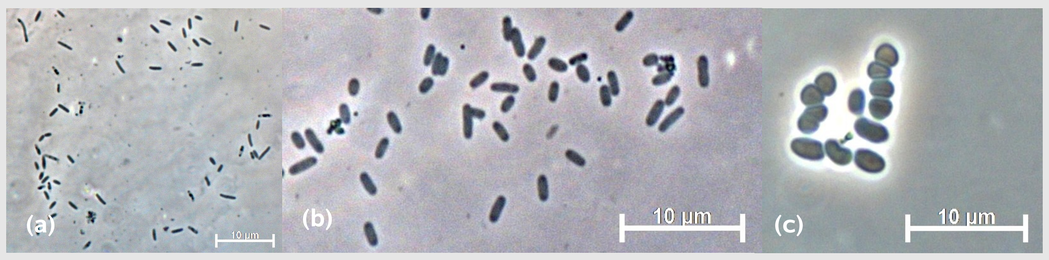

Fig. 5 Different morphotypes of SRB cultivated from Wadden Sea sediment
(a) 100 cm, hydrogen; (b) 400 cm, acetate; (c) 250 cm, acetate

Relatives to the genus ***Desulfotalea*** were frequently found and were enriched with each substrate offered. In contrast, ***Desulfosarcina***- and ***Desulfobacula***-related SRB were exclusively found in cultures containing hydrogen or acetate, respectively. Additional sequences affiliating with ***Firmicutes*** were found.

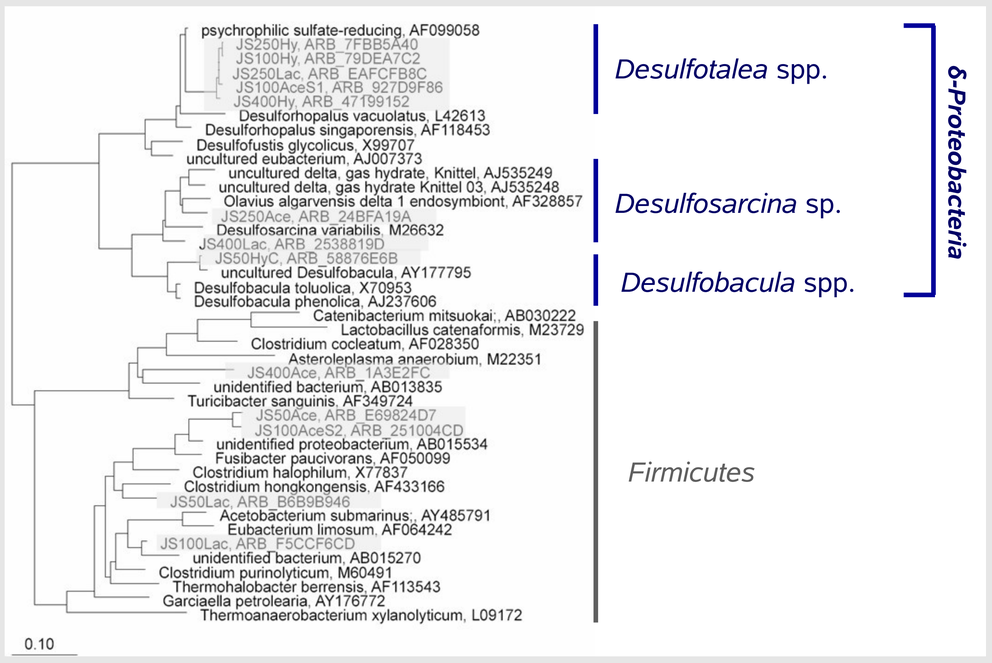

Fig. 6 16S rRNA-based phylogenetic affiliation of extracted DGGE bands received from sediment enrichments. Sequences from this study are highlighted. The scale bar corresponds to 10% estimated sequence divergence.

Conclusions

- We found elevated activity of SRB in deep sediment layers of an intertidal sand flat within the German Wadden Sea
- The presence of SRB utilizing a variety of potential electron donors along the entire sediment column demonstrates the nutritional versatility of the SRB community *in situ*
- *Desulfotalea*-related bacteria formed a dominant fraction as proven by cultivation as well as quantification of *in situ* abundance using CARD-FISH
- Our findings suggest a coincidence of local maxima of both sulfate reduction rates and active, metabolically diverse groups of SRB

References

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