

Introduction

The important role of bacteria in the energy flux and nutrient cycling on aggregates in marine and estuarine ecosystems has been reviewed recently by Simon et al. (2002). In the German Wadden Sea, a shallow water column and strong tidal currents build a highly dynamic system influencing the size and abundance of the colonized aggregates. Despite many investigations about physical and biogeochemical processes, only little is known about the microbial degradation of particulate organic matter. In this study, we used specific primer sets for PCR of 16S rRNA gene fragments and subsequent Denaturing Gradient Gel Electrophoresis (DGGE) to assess relevant species of abundant phylogenetic groups. In addition, biochemical and phytoplankton data were used to determine relationships between DGGE banding patterns and alteration of particle and algae composition.

Conclusions

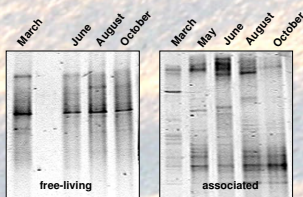
- Distinct communities of free-living and aggregate-associated bacteria in the Wadden Sea and the German Bight
- Few similarities between the German Bight and the Wadden Sea bacterial communities detectable
- No changes of the bacterial community during diurnal cycles
- Seasonal changes of particulate organic matter reflected by pelagic bacterial communities
- Some bacterial species not affected by seasonal changes

A Distinct Bacterial Communities: Free-Living and Aggregate-Associated (March - October 2002)

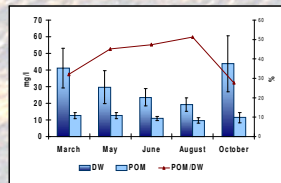
Seasonal sampling during 2002 showed seasonal changes in

- quality of particulate matter as indicated by POM/DW ratio
- bacterial abundance of free-living bacteria
- composition of aggregate-associated bacterial community

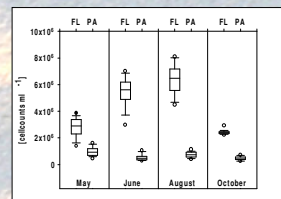
DGGE Fingerprints of 16S rRNA Gene Fragments of Wadden Sea Samples



Dry Weight, Particulate Organic Matter and POM/DW Ratio



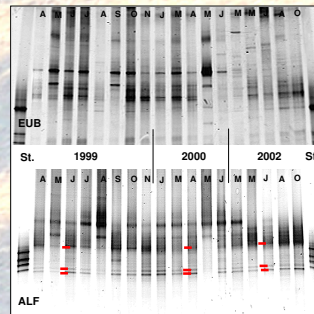
Cell Counts of Free-Living and Aggregate-Associated Bacteria



Legend: DW=Dry Weight; POM=Particulate Organic Matter; PA=Aggregate-Associated; FL=Free-Living

B Comparison of Different Years: Bands Persisting Independently from Seasonal Changes

DGGE Fingerprints of Aggregate-Associated Bacteria Using Specific Primer Sets for Eubacteria and α -Proteobacteria



Legend: EUB=Eubacteria; ALF= α -Proteobacteria; St=Standard; marked bands=bands existing in 1999, 2000 and 2002

Comparison of 1999, 2000 and 2002 showed

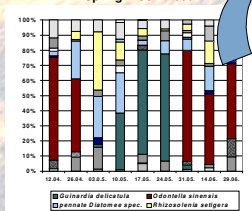
- differences between 2002 and former years for Eubacteria
- substantial similarity in banding patterns for α -Proteobacteria
- several bands of α -Proteobacteria persisting all years

C Focus: Composition Changes of Bacterial Communities During an Algal Bloom

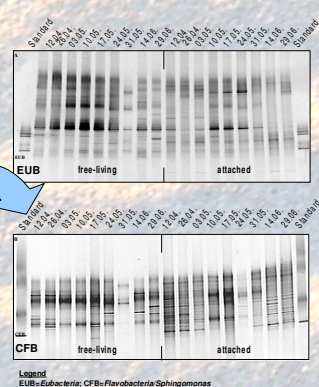
Sampling during an algal bloom showed

- significant changes of algal biomass composition
- strong effect of algal composition changes on bacterial communities of both fractions
- clear reflection by attached bacteria belonging to the CFB group

Algal Biomass Composition (%) Spring Bloom 2000



DGGE Fingerprints of Aggregate-Associated Bacteria April - June 2000



Legend: EUB=Eubacteria; CFB=Flavobacteria/Sphingomonas

D Comparison of Wadden Sea and German Bight: Few Similarities of Bacterial Communities

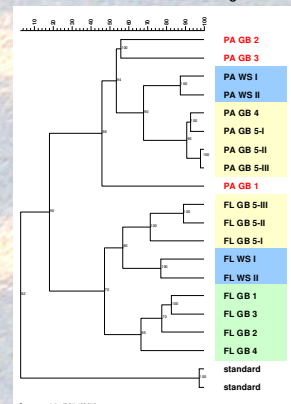
Sampling Stations in German Bight and Wadden Sea (June, 2002)



Spatial sampling in German Bight and Wadden Sea showed

- distinct clusters for free-living and associated bacteria
- subclustering of Wadden Sea samples and German Bight sample near Spiekeroog (GB 5)
- high similarity of samples taken at different times at the same station

Cluster Analysis of DGGE Banding Patterns of Eubacterial 16S rRNA Gene Fragments



Legend: PA=Aggregate-Associated; FL=Free-Living; WS=Wadden Sea Station; GB=German Bight Station; H=Samples taken at different times at the same station

Materials and Methods

Samples were taken during diurnal cycles in the backbarrier tidal flat system near Spiekeroog on board RV "Senckenberg" in March, May, June, August and October 2002 and on different locations in the German Bight in June 2002. Two fractions were separated by filtration: a) aggregate-associated bacteria on a 5.0 μ m Nuclepore filter, and b) free-living bacteria on 0.2 μ m Nuclepore filter. Bacterial cell counts were determined by DAPI staining and epifluorescence microscopy [2]. The composition of the bacterial community was investigated by DGGE of 16S rRNA gene fragments. Primer sets for specific bacterial groups (Flavobacteria/Sphingomonas [3], α -Proteobacteria [this study]) were applied to enhance the detection of relevant species. Cluster analysis of DGGE banding patterns was performed using GelCompare III (Version 2.5, Applied Maths). Calculation was generated curve-based using Pearson correlation and UPGMA. As indicator for quality changes of particulate matter dry weight (DW) and ratio of particulate organic matter (POM) were determined as described by Lunau et al. (2003).

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References

- [1] Simon, M., H.-P. Grossart, B. Schweitzer & H. Ploug (2002). Microbial ecology of organic aggregates in aquatic ecosystems. *Aquatic Microbial Ecology* 28: 175-211
- [2] Crump, B. C., J. A. Baross & C. A. Simenstad (1998). Dominance of particle-associated bacteria in the Columbia River estuary, USA. *Aquatic Microbial Ecology* 14: 7-18
- [3] Jaspers E, Nauhaus K, Cypionka H, Overmann J. 2001. Multitude and temporal variability of ecological niches as indicated by the diversity of cultivated bacterioplankton. *FEMS Microbiology Ecology* 36 (2001) 153-164
- [4] Lunau, M., B. Rink, H.-P. Grossart, M. Simon (2003). How to sample marine microaggregates in shallow and turbid environments? - problems and solutions. *Berichte Forschungszentrum Terramare*.