

The Department of Biological Oceanography focuses on the role of planktonic organisms in the carbon and energy fluxes and nutrient recycling in the North Sea and the Atlantic Ocean. Specific emphasis is put on the complex interaction between bottom-up and top-down control mechanisms in the lower planktonic food web.

In the year 2002, three cruises were executed. The final Plume & Bloom cruise in the North Sea was carried out during summer with input from MCG to obtain additional data on the role of clay and nutrient transport for the dynamics in productivity of the southern North Sea. During the MOMAP-1 cruise, also in the North Sea, growth and mortality of *Phaeocystis* was studied. An important component of this cruise was the assessment of the phyto- and bacterio-plankton viral-induced mortality. In the TRANSAT-1 cruise performed in collaboration with FYS, the diagenesis of dissolved organic matter and the accompanying changes in prokaryotic community composition was followed in the North Atlantic Deep Water (NADW). This was done along a transect from the Greenland-Island-Norwegian Sea to the Azores, covering roughly the first 50 years of NADW in the oceanic conveyor belt.

A major field campaign in the frame of the AIRWIN project, funded by the EU, was performed in the Mediterranean Sea. In this project, the biology and chemistry of the air-sea micro-layer, i.e. the first ca. 200 μm of the water column, is studied. As part of the (EU)-BIOHAB program scientists participated in the microcosm experiments in Barcelona. These experiments were designed to examine the release of toxic substances of *Alexandrium catanella* and their effect on the different trophic levels of a natural plankton community (viruses, bacteria, other phytoplankton, micro- and mesozooplankton). The EU-funded BASICS project started in fall. The main goal of this project is to perform single-cell analysis using a combination of phylogenetic and functional probes and laser confocal laser scanning microscopy to decipher the phylogenetic and functional dynamics of prokaryotic plankton communities at specific sites in European coastal waters.

MAJOR SHIFT IN BACTERIOPLANKTON UTILIZATION OF ENANTIOMERIC AMINO ACIDS BETWEEN SURFACE WATERS AND THE OCEAN'S INTERIOR

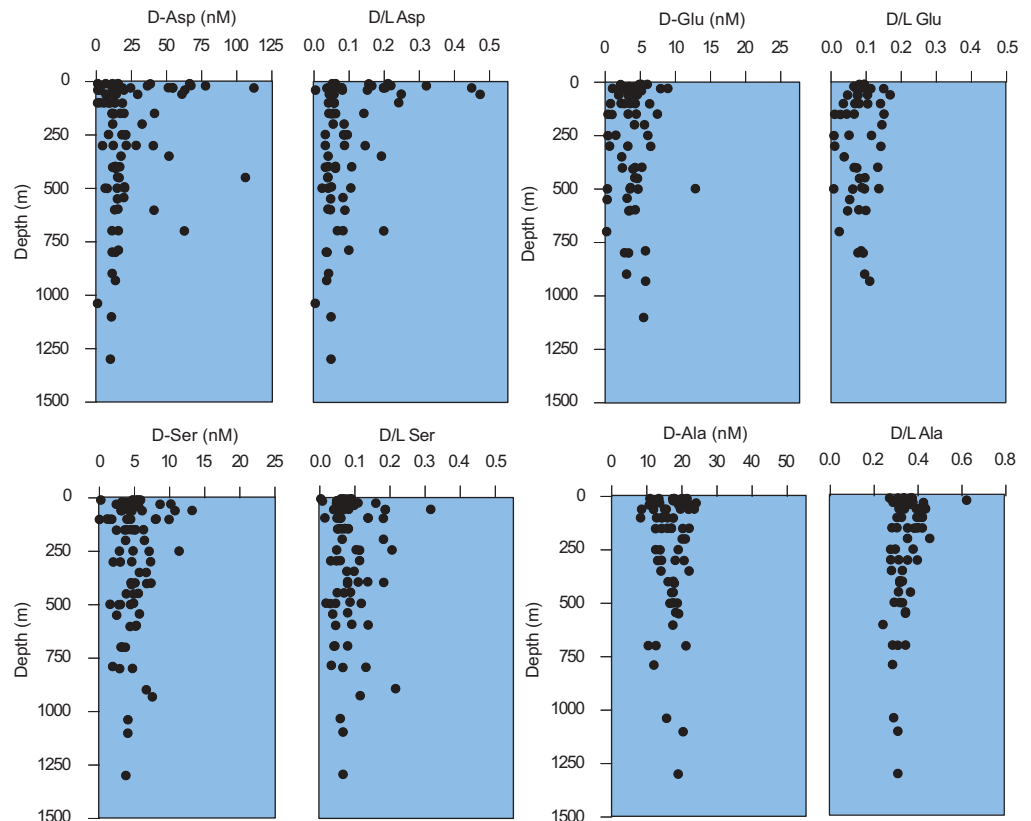
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The oceanic dissolved organic carbon (DOC) pool is considered to be mainly of phytoplankton origin. This view has been challenged by the notion that the largest oceanic biomass, the bacterioplankton, also considerably fuel this DOC pool. It has been demonstrated that bacterioplankton transform labile DOC into recalcitrant DOC. One of the most refractory compounds of the bacterial cell is its cell wall. Specific compounds of this cell wall, the peptidoglycan layer, have been shown to constitute a significant fraction of the oceanic DOC pool as indicated by the characteristic enantiomeric ratio (D-/L-ratio) of its dissolved total amino acids. L-amino acids are those where the amino group is on the L-left side in its Fisher projection of the compound, D-amino acids where the amino group is on the right side.

The main biotic source of D-amino acids in the sea is the peptidoglycan layer of the bacterial cell wall where 4 specific enantiomeric amino acids (alanine [Ala], glutamic acid [Glu], aspartic acid [Asp], and serine [Ser]) are present. Abiotically, D-amino acids are formed by racemization, which converts the L-enantiomeric form of amino acids into the corresponding D-amino acids. This racemization is a significant source of D-amino acids only over geological time scales. While the production of D-amino acids in the sea is largely restricted to bacteria, L-amino acids are produced and released into the oceanic DOC pool by a large variety of organisms but the most important source is phytoplankton. These phytoplankton-derived L-amino acids serve as an important substrate for bacterioplankton and are consequently turned over rapidly.

In contrast to that, D-amino acids are generally considered to be refractory as indicated by the increase in the ratio of D-/L-amino acids in DOC degradation experiments with surface water DOC. Therefore, it has been recently suggested that the ratio of D-/L-amino acids can be used as a diagenetic indicator of the bioreactivity of the oceanic DOC pool. Since the DOC pool becomes increasingly refractory from the surface layers to the deep waters, one would expect that the D-/L-amino acid ratio of the DOC pool increases with depth as well. However, such an increase in the D-/L-amino acid ratio with depth has not been found in the DOC fraction larger than 1,000 molecular weight which represents about 20-30% of the bulk oceanic DOC.

In this study we measured the concentrations of the 4 bacterial cell wall-derived enantiomeric amino acid species present in the DOC pool throughout the North Atlantic water column. Concurrently, we determined the uptake of D- vs. L-Asp by bacterioplankton in the different water layers. Since major shifts in the D-/L-Asp uptake ratio of bacterioplankton from the surface to the deep mesopelagic layers were found, additional laboratory experiments were performed. In these experiments, the hypothesis was tested that bacterioplankton, in the



Concentrations of dissolved total D-aspartic acid (Asp), D-glutamic acid (Glu), D-serine (Ser) and D-alanine (Ala) and the corresponding D/L ratios in the water column of the North Atlantic.

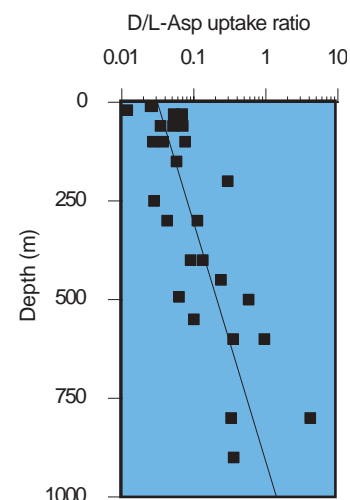
absence of other utilizable organic matter, shift from a preferential L-amino acid uptake to a more efficient utilization of D-amino acids, i.e. bacterial-derived DOC. Furthermore, the potential role of flagellates in the production of bacterial cell wall-derived dissolved amino acids was examined.

The field work for this study was done in the Faroe Shetland Channel of the North Atlantic (1°W 62°N — 5°W 60°N) during the BIOPROCS cruise with the R/V Pelagia in the summer of 1999.

The concentrations of the 4 dissolved total enantiomeric amino acid (DTEAA) species indicative for cell wall-derived DOC (Asp, Glu, Ser, Ala) exhibited no particular trend with depth in the water column of the Faroe Shetland Channel. Also, no trend with depth was discernable for the ratio of the D-/L-amino acid species. The lack of any trend with depth in the D/L ratios of these 4 DTEAA species in the water column of the Faroe Shetland Channel is in agreement with the few data available for other oceanic regions obtained with different methods.

Based on the mean D-Ala concentration of $27.9 \pm 6.3 \text{ nmol L}^{-1}$ (54 samples), averaged over all the stations and depths and assuming that all the dissolved total D-Ala originates from the peptidoglycan layer of bacterioplankton we can estimate the contribution of bacterioplankton-derived peptidoglycan to the bulk DON pool. It has been shown that the D-Ala concentration in peptidoglycan is relatively constant, at least for culturable non-marine bacteria (D-Ala-N $\times 5.7$). Based on these directly measurable hydrolyzable D-Ala and the DON concentrations (data not shown), bacterioplankton-derived peptidoglycan contributes about 2 — 2.5 % to the DON pool of the study site. There is evidence from recent NMR studies that most of the oceanic DON not recoverable on a molecular level consists probably of non-hydrolyzable amino acids. Thus, since only about 10 % of the DON pool are hydrolyzable amino acids it follows that dissolved peptidoglycan contributes about 20 — 25 % to the pool of hydrolyzable dissolved amino acids in the study area. These figures should be considered as a rough estimate only since the above calculation is based on the D-Ala concentration in peptidoglycan obtained from cultured non-marine, mostly biomedically important bacteria. The contribution of D-Ala on the peptidoglycan of marine bacteria might be more variable considering their phylogenetic and functional diversity.

In order to estimate the potential of bacterioplankton to utilize bacterial cell wall-derived dissolved D-amino acids, we compared D- and L-Asp uptake of natural bacterial communities collected from different depth layers. The ratio of bacterial D-/L-Asp uptake increased exponentially from 0.03 in the surface layers to about 1 at 900 m depth. If the other 3 amino acid species are taken up at similar D-/L- uptake ratios as Asp, then the high uptake ratio of D-/L-Asp in the mesopelagic zone indicates an adaptation of the mesopelagic bacterioplankton community



Depth dependence of the uptake ratio of D-/L-aspartic acid (Asp) by bacterioplankton in the water column of the Faroe Shetland Channel of the North Atlantic. The D-/L-Asp uptake ratio increased exponentially with depth.

to utilize D-amino acids relatively more efficiently than bacteria in the euphotic zone. In the euphotic zone, phytoplankton activity supplies mainly L-amino acids, which are taken up by bacteria efficiently and, according to our data, preferentially over D-amino acids. Thus, in the euphotic zone the supply ratio of D-/L-amino acids might be similarly low as the D-/L-Asp uptake ratio we measured. In the mesopelagic zone, however, due to the absence of phytoplankton production and the preferential use of L-amino acids in the surface layers, the bioavailable DOC, including the freshly-produced DOC supports a supply ratio of D-/L-amino acids which is considerably higher than that in the euphotic layer. In short, we speculate that the observed shift in bacterial uptake ratios of D-/L-amino acids with depth reflects the shift in the production of bioavailable D-/L-amino acids from the surface layers to the mesopelagic zone. Such a close coupling between the supply ratio and the uptake ratio would explain the rather constant D-enantiomer concentrations and D/L ratios of the 4 DTEAA species throughout the water column.

Since bacterioplankton are thought to be the main source of the 4 D-amino acids one might tentatively assume a close relation between bacterial production and D-amino acid concentration. The remarkably constant dissolved total D-Ala and D-Asp concentrations over a wide depth range are in sharp contrast, however, to the decline in bacterial production from the near-surface ($0.17 \pm 0.15 \mu\text{mol C L}^{-1} \text{d}^{-1}$) to the deep mesopelagic layer by 3 orders of magnitude. Bacterial production was positively correlated with D- and L-Asp uptake and negatively correlated with the D-/L-Asp uptake ratio. The steeper slope for L-Asp than for D-Asp uptake causes the increasing D-/L-Asp uptake ratios with decreasing bacterial production. The question remains to be solved whether these relatively higher uptake rates of D-Asp as compared to L-Asp in the deeper layers of the water column are due to specific species of prokaryotes inhabiting the mesopelagic zone.

Major shifts in the *bacterial* community composition between surface and mesopelagic waters have been reported for the Mediterranean Sea. About 50 % of all the phylotypes of Bacteria present in the mesopelagic zone are specific for this layer as determined by terminal-restriction fragment length polymorphism. Thus, there is accumulating evidence that the prokaryotic community changes significantly with water column depth and it is likely that these shifts in the prokaryotic community composition are responsible for the shifts in the uptake ratio of D-/L-Asp from the euphotic layer towards the deep mesopelagic zone.

Our findings have several important implications. For using D-/L-amino acid ratios as an indicator of the diagenetic state of DOC, caution is required since D-amino acids are more bioavailable than hitherto assumed, especially in the mesopelagic zone. For the microbial ecology of the mesopelagic realm, our results indicate that specific prokaryotic communities are present there utilizing D-amino acids as efficiently as L-amino acids. The phylogeny of these prokaryotes responsible for this efficient D-amino acid utilization in the mesopelagic environment is unknown but even surface water bacterioplankton have the physiologic capacity to utilize these D-amino acids efficiently if other organic nutrient sources are not sufficiently available. Whether this efficient utilization of D-amino acids represents a common strategy of bacterioplankton to utilize them as a supplementary carbon, nitrogen and energy source when other suitable organic substrates are scarce in the mesopelagic and deep waters remains to be investigated.