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THE INFLUENCE OF MARICULTURE
ON PLANKTONIC BACTERIAL COMMUNITIES
IN THE PELAGIC ZONE OF PIRAN BAY

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The numerous documented negative effects of mariculture on the environment are connected especially to the accumulation of uneaten food and feces in sea water and in/on the sediment. To evaluate the influence of the fishery pollution on heterotrophic planktonic bacterial communities, spatial and temporal differences in the abundance, growth rates and bacterial community structure were studied in the Gulf of Trieste (Northern Adriatic) on 18 October 2005. At the same time, differences in the chemical parameters (total suspended solids (TSS) and nutrient levels) and physical parameters (basic oceanographic parameters) were followed together with 24-hour current measurements.

Sampling was carried out at the stations around the fish cage and along the transect from the cage located inside the Bay of Piran towards the reference station located 5,400 m away in the middle of the Gulf of Trieste. A new sampling system was designed to provide accurate data at different sampling locations. Underwater samplers were constructed in a circle at a distance of 8 m around the operating fish cage (6 locations) and another circle at a distance of 20 m around the fish cage (6 locations). Each bottle (5 l) was fixed on an underwater metal frame, 5 m below the surface. All sampling bottles were opened simultaneously collecting seawater samples at all location around the fish cage. Sampling was performed before feeding and 3 hours after the feeding of the fish. The result for each parameter was expressed as a ratio ($K = C_{\text{bef}}/C_{\text{af}}$) between the values before and after feeding.

Preliminary results showed temporal and spatial distribution of bacterial abundance and growth rates as well as TSS, ammonia and nitrate concentrations, which were related to distance from the fish farm and to pre-feeding and post-feeding conditions. In the vicinity of fish cage, the abundance value of heterotrophic bacteria

was 2.5-times higher comparing with the bacterial abundance at the reference station. Significantly higher (3-times) was also the growth rate of the total bacterial population. High P/B ratio also reflects intensive bacterial metabolic process in the vicinity of the fishery.

Bacterial community compositions along the transect were determined with new molecular methods, such as FISH (*Fluorescent in situ Hybridization*) and DGGE (*Denaturing Gradient Gel Electrophoresis*). Analysis of the bacterial community compositions with FISH method has shown that bacterial groups from subclasses α -, β - and γ -*Proteobacteriae* as well as *Cytophaga-Flavobacteria-Bacteroides* group and *Vibrio* group were present in all the samples along the transect. Although all bacterial groups were present in the water samples along the transect, there was a great difference in percentage of the each group at separate stations. In the centre of the fish cage dominated the group of nitrifying ammonia-oxidizing bacteria, compared to remote stations on the transect, where *Cytophaga-Flavobacteria-Bacteroides* group and *Vibrio* group were predominant. Analysis of the bacterial community compositions with DGGE method has given similar results that also show the presence of various bacterial groups within a particular sample as well as spatial differences along the transect.

Significant decrease in bacterial carbon production was observed with distance from the pollution source according to the radioactive leucine method. The preliminary results of metabolically active bacterial groups, determined with the BrdU-DGGE method, have shown that the same bacterial groups were metabolically active at all stations along the transect.

Measurements of temporal differences in the abundance and growth rates of the total bacterial population were carried out at the stations around the fish cage before and three hours after the feeding of the fish. After the feeding, a significantly higher bacterial carbon production was measured. The results were comparable with the increased ratio between the total suspended solids (TSS) and nutrient levels measured before and after feeding. The increased bacterial carbon production could be due to the higher concentrations of the TSS and nutrient levels in the surrounding of the fishery.