SEASONAL DYNAMICS OF THE BACTERIAL COMMUNITY IN CORRELATION WITH DIFFERENT ENVIRONMENTAL FACTORS IN THE GULF OF TRIESTE (NORTHERN ADRIATIC)

Tinkara Tinta [1]* and Valentina Turk [1]
[1] National Institute of Biology, Marine Biology Station - tinta@mbss.org

Abstract
Seasonal dynamic of bacterial community was studied in the Gulf of Trieste (northern Adriatic). The bacterial abundance, productivity and community structure together with dissolved inorganic phosphate uptake by the ^{33}P orthophosphate incorporation method and nutrients factors were followed along the vertical profile. Preliminary analyses of bacterial community structure were performed.
Keywords: Adriatic Sea, Bacteria, Nutrients, Phosphorus

Introduction
The Gulf of Trieste, located in the Northern Adriatic sea, is a shallow, semi - enclosed basin, which oceanographic properties are strongly affected by water mass exchange from the Southern Adriatic, river inflows and meteorological conditions ([1]). Enrichment experiments proved that phosphorous is the primary limiting element for the growth of phytoplankton as well as for bacterioplankton in the Gulf of Trieste ([2]). Being an integral members of ecosystems, microbes are fundamental to the functioning and the health of the marine environment. Bacterial community abundance and productivity in the Gulf of Trieste has been studied in the past ([3]). However details of microbial processes in the Gulf of Trieste are still poorly understood. The aim of this study was to investigate in more details the seasonal dynamics of the bacterial community in correlation with different environmental factors in the Gulf of Trieste.

Material and methods
The seawater samples for chemical and biological parameters were collected during the period from 2007 to 2009 at standard sampling station, the oceanographic buoy, located in the middle of the Gulf of Trieste (northern Adriatic)**.** Sampling was performed biweekly at six different depths (1m, 3m, 5m, 10m, 15m and 20m) using a Niskin sampler. At the same time vertical distribution of temperature, salinity, chlorophyll *a* and dissolved oxygen was also determined using CTD probe. Samples were analyzed for nutrients, bacterial abundance and productivity as well as bacterial community structure. Bacterial abundance was determined according to standard protocol, by staining cells with 4',6-diamino-2-phenylindole (DAPI) and examining them under an epifluorescence microscope ([4]). Bacterial carbon production was measured by the incorporation of ^{3}H-leucine into newly synthesized proteins in the bacterial cells ([5]). Subsamples for nutrient analysis were filtered through glass fibre filters (GF/F) and analyzed according to standard protocols ([6]). Bacterial community structure was analyzed using two approaches: (i) by isolation of colony - forming bacteria on ZoBell media and (ii) by culture - independent genetic analysis. Subsamples for bacterial community structure analysis were obtained by filtering defined volume of unfixed seawater onto 0.2 µm polyethersulfonic PALL filters (25 mm diameter, PALL Inc.). DNA was extracted from the filters according to Böstrom ([7]) with slight modifications. Community fingerprints were determined using the denaturing gradient gel electrophoresis (DGGE).

Results and discussion
Bacterial abundance showed seasonal dynamics, with the highest values in the late spring - early summer period (6.5 x 10^8 cells L^{-1} in May) and at the beginning of the autumn period (9 x 10^8 cells L^{-1} in September). From the late autumn until the early spring we observed lower bacterial numbers and equal abundance distribution along the vertical profile. In the winter period and in the spring - early summer there were somehow higher values at the surface. On the other hand, in the late summer - early autumn period, the bacterial abundance was increasing towards the bottom of the water column. Bacterial carbon production showed seasonal dynamics as well, with the highest values measured in the summer (20 µg C L^{-1} day^{-1}) and lowest productivity in winter - early spring (5 µg C L^{-1} day^{-1}). From the late autumn to the early spring the productivity did not vary much along the vertical profile. From late spring until the middle of summer we observe higher bacterial productivity at the surface and at the bottom of the water column. At the end of the summer, on the other hand, the productivity at the surfaces in low, but increases with the depth, reaching the highest values at the bottom. Seasonally changes in dissolved inorganic phosphate uptake measured by the ^{33}P orthophosphate incorporation method were determined in total sea water and > 1 micrometer size fraction. Preliminary analyses of bacterial community structure were performed by sequencing dominate bacterial isolates and by determination of total community fingerprints using DGGE.

References
1 - Malacic V. and Petelin B., 2001. Gulf of Trieste in: Physical Oceanography of the Adriatic Sea, Past, Present and Future. Cushman-Roisin B, Gacic M, Poulain P M, Artegiani A (ed.), Kluwer Academic Press, Dordrecht, pp. 167-177.
2 - Malej A., Mozetic P., Turk V., Terzic S., Ahel M. and Cauwet G., 2003. Changes in particulate and dissolved organic matter in nutrient-enriched enclosures from an area influenced by mucilage: the northern Adriatic Sea. *J. Plankton Res.*, 25: 949-966.
3 - Turk V., Mozetic P. and Malej A., 2001. Seasonal variability in phytoplankton and bacterioplankton distribution in the semi-enclosed temperate gulf (Gulf of Trieste, Adriatic Sea). *Annales, Ser. hist. nat.*, 11: 53-64.
4 - Porter K. G. and Feig Y. S., 1980. The use of DAPI for identifying and counting aquatic microflora. *Limnol Oceanogr.*, 25: 943-948.
5 - Smith D. C. and Azam F., 1992. A simple, economical method for measuring bacterial protein synthesis rates in seawater using ^{3}H-leucin. *Mar. Microb. Food Webs*, 6: 107-114.
6 - Grasshoff K., Ehrhardt M. and Kremling K., 1983. Methods of seawater analysis. Verlag Chemie, Weinheim.
7 - Boström K. H., Simu K., Hagström A. and Riemann L., 2004. Optimization of DNA extraction for quantitative marine bacterioplankton community analysis. *Limnol. Oceanogr. Methods*, 2:365-373.