

VIRIOPANKTON DISTRIBUTION RELATED TO BACTERIAL METABOLIC STATUS DURING AN AUTUMN SURVEY IN THE NORTHERN ADRIATIC SEA

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Abstract

The study was performed during November 2004 in order to detect spatial distribution of marine viruses in the surface waters of the Northern Adriatic Sea, to assess their interaction with bacteria as their most common host, and to investigate the influence of bacterial metabolic status on viral proliferation. Our results confirmed the numerical prevalence of viral fraction within the microbial community and showed that not only the highly active but also less active or dormant bacterial cells can sustain viral proliferation, the latter resulting particularly intense at the mouth of the River Po.

Keywords: Adriatic Sea, Bacteria, Coastal Waters, Po Delta

The importance of viral infection for overall bacterioplankton mortality tends to increase with increasing bacterial abundance and/or productivity [1] with the evidence that more active bacterial cells may be more susceptible to viral infection and that a more active bacterial community may sustain higher viral abundance [2]. Since the detection of highly active cells by using CTC method [3], only partially explains the activity of bacterial community the discrimination of non-viable (dead) bacteria could further help in understanding viruses-bacteria interactions. The marked trophic gradient of water masses in the Northern Adriatic represented suitable study area for investigating differences in microbial interactions. Surface water samples were collected from 18th to 21th November 2004 along 5 transects for a total of 25 stations (Fig. 1).

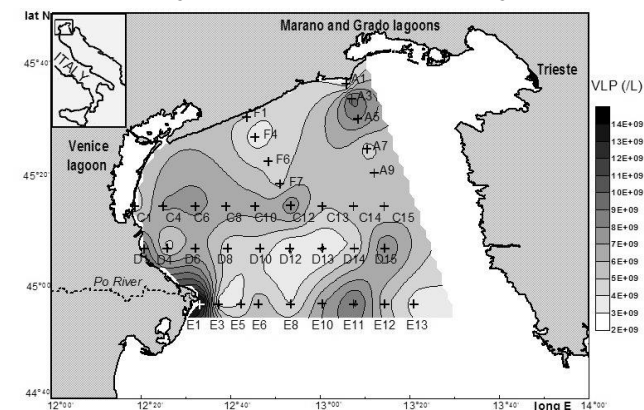


Fig. 1. Sampling stations in the Northern Adriatic Sea. Contour map indicates distribution of virioplankton abundances

Temperature, salinity, oxygen saturation and fluorescence data were obtained using CTD multiparametric probe (Itronaut Ocean Seven 316). Samples for biological analyses were collected using a Carousel water sampler carrying Niskin bottles. Viral abundance was determined using SYBR Green I protocol [4]. Total bacterial abundance was determined using DAPI staining method [5] while non vital cells were distinguished using PI nucleic acid stain and processed according the manufacturer (Molecular Probes). Metabolically active bacteria were detected by CTC incubation technique [6]. Bacterial Carbon Production (BCP) was determined by [³H]-Leucine incorporation [7] only along C and E transects (for a total of 9 stations). Seawater temperature ranged from 11.3 to 16.6°C. Minima values were found in the coastal area, increasing towards the central part of the basin. Salinity ranged between 29.7 and 38.4 with rather opposite pattern to the seawater temperature ($p < 0.01$). Dissolved oxygen, ranging between 82-93%, also resulted significantly but positively correlated with temperature ($p < 0.05$). These suggested the presence of a large amount of freshwater in the coastal belt, which matched with the highest fluorescence values. In the entire area fluorescence remained low ranging between 0.1 and 0.8 $\mu\text{g Chl a l}^{-1}$. Virioplankton abundances (Fig. 1) ranged between 3.8 and $19.9 \times 10^9 \text{ l}^{-1}$. Higher values were found near the Po River mouth (coastal stations of the E transect) and offshore the lagoons (A3 and A5 stations). Bacterioplankton displayed quite narrow variability, ranging within 0.9 and $1.8 \times 10^9 \text{ cells l}^{-1}$. The abundance of highly active bacteria varied from 1.4 to $5.9 \times 10^7 \text{ l}^{-1}$, thus remaining below 5% of total bacterial abundance. A clear coast to offshore decreasing gradient was observed with the highest CTC+ abundance and the

maximum VBR ratio of 14 near the Po River mouth. In that area also bacterial production rate was the most intense ($0.485 \mu\text{g C l}^{-1} \text{ h}^{-1}$), gradually decreasing along the C transect, whether the minimum rate of $0.026 \mu\text{g C l}^{-1} \text{ h}^{-1}$ was found in the offshore stations of the E transect. Non vital bacteria ranged between 0.1 and $9.5 \times 10^8 \text{ l}^{-1}$ and their proportion within total bacterial community strongly varied from 0.4% up to 91%. It was found quite similar but opposite distribution pattern of virioplankton and non vital cells ($p < 0.08$). Our results evidenced wide spatial variability of virioplankton and confirmed its numerical prevalence within the microbial plankton community of the N Adriatic surface waters. It is plausible that the riverine inflow increasing the trophic status of the system largely influenced viral distribution and the intensity of virus-bacteria interaction. Contrary to the results reported for the entire Adriatic [2], we found that virus-bacteria interaction can not be explained by the distribution of total bacterial community itself. Viral distribution matched quite well with bacterial production and the abundance of active bacteria, but only in the area proximal to the Po River mouth, suggesting that bacteria in this area shifted their metabolism to highly active status. The possibility of the copious enrichment in viral abundances from outside sources was also considered, but was less plausible since viruses in the sea are generally from within the system [8]. The large proportion of non vital cells was found also by other authors [9], even if they put under discussion the reliability of the PI method to indicate non active or dead bacteria. The relationship between viruses and active, non vital and total bacterial community suggests that viral proliferation is not only due to the highly active but also dormant bacterial host while the infection of non vital bacterial cells is not productive, thus further confirming the key role of bacterial activity in controlling viral dynamics.

References

- 1 - Noble R.T. and Fuhrman J.A., 2000. Rapid viral production and removal as measured with fluorescently labeled viruses as tracers. *Appl. Environ. Microbiol.*, 66: 3790-3797.
- 2 - Corinaldesi C., Crevatin E., Del Negro P., Marini M., Russo A., Fonda Umani S., Danovaro R., 2003. Large-scale spatial distribution of virioplankton in the Adriatic sea: testing the trophic state control hypothesis. *Appl. Environ. Microbiol.*, 69: 2664-2673.
- 3 - Karuza A., Del Negro P., Paoli A., Comisso S., Fonda Umani S., 2004. Highly active bacteria in the surface waters of the Gulf of Trieste (Northern Adriatic Sea). *Rapp. Comm. int. Mer Médit.*, 37: 277.
- 4 - Noble R.T. and Fuhrman J.A., 1998. Use of SYBR Green I for rapid epifluorescence counts of marine viruses and bacteria. *Aquat. Microb. Ecol.*, 14: 113-118.
- 5 - Porter K.G. and Feig Y.S., 1980. The use of DAPI for identifying and counting aquatic microflora. *Limnol. Oceanogr.*, 25: 943-948.
- 6 - Paoli A., Karuza A., De Vittor C., Del Negro P., Fonda Umani S., 2006. Daily variations of highly active bacteria in the Northern Adriatic Sea. *J. Plankton Res.*, 28(3): 325-335.
- 7 - Smith D.C., Azam F., 1992. A simple, economical method for measuring bacterial protein synthesis rates in sea water using ³H-leucine. *Mar. Microb. Food Webs*, 6: 107-114.
- 8 - Wilhelm S.W. and Suttle C.A., 1999. Viruses and nutrient cycles in the sea. *Bioscience* 49, (10): 781-788.
- 9 - Pirkner H., Pausz C., Stoderegger K.E., Herndl G.J., 2003. Simultaneous measurement of metabolic activity and membrane integrity in marine bacterioplankton determined by confocal laser-scanning microscopy. *Aquat. Microb. Ecol.*, 39: 225-233.