## MECHANISMS OF PHYTOPLANKTON SUCCESSION ACROSS STEEP SPATIO-TEMPORAL NUTRIENT GRADIENTS IN THE NORTHERN ADRIATIC.

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## Abstract

The northern Adriatic (NA) is an ideal basin to study the adaptive strategies of plankton to a variety of conditions along steep spatio-temporal gradients. Earlier studies identified P-limitation as one of the key stresses within the NA that shape the biological response in terms of biodiversity and metabolic responses. A wide range of reports support the notion that P-limitation is a globally important phenomenon in marine ecosystems. In this study, microscopic analysis allowed the determination of phosphorus (P) stress of marine phytoplankton at species level along a trophic gradient in the NA. In P-limitation all species with considerable contributions to the phytoplankton community expressed alkaline phosphatase activity (APA).

Keywords: Phytoplankton, North-Eastern Mediterranean, Phosphorus, Nutrients, Enzymes

Growing evidence suggests that P is the limiting nutrient in several coastal systems and oligotrophic oceans. Furthermore, it is believed that the open ocean, far from continental inputs of nutrients, should evolve towards P-limited conditions due to N2 fixation, while P is a non-renewable limiting nutrient. The rise in N2 fixation and the simultaneous disappearance of P stocks observed in the subtropical North Pacific Ocean between 1989 and 2004 is an illustration of this phenomenon. Therefore, the importance of alkaline phosphatase activity (APA) with regard to the transformation and turnover of organic compounds in marine environments has been investigated with growing attention. A number of studies in the northern Adriatic (NA) evidenced that this region is currently Plimited. This shallow (up to 50 m) coastal sea is characterized by significant freshwater input, mainly from the Po River. Although in the Po River waters both nitrogen and phosphorus concentrations are more than one order of magnitude higher than in the NA waters, the inorganic N:P atomic ratio provides a strongly unbalanced N versus P supply for phytoplankton requirements. Earlier studies showed that in this area organic phosphorus concentrations markedly exceeded orthophosphate (PO4) concentrations, representing an important source of P for microbial communities. In the present study an AP substrate with insoluble fluorogenic product (ELF Endogenous Phosphatase Detection Kit (E6601) (Thermo Fisher Scientific, Waltham, USA)) was used to investigate how the APA status varies among phytoplankton species along the trophic gradient across the northern Adriatic. In a previous study we found that gradients in phosphate concentrations can modify the structure of plankton communities and can constrain phytoplankton distribution. We sampled (bimonthly) phytoplankton along a transect between Rovinj (Croatia) and the River Po mouth (Italy).

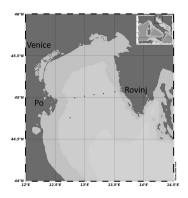


Fig. 1. The sampling area with sampling stations

We analyzed nutrient concentration, phytoplankton community structure and abindnces, bulk APA and species specific APA as well as further oceanographic conditions. Overall, in P-limited conditions all species with considerable contributions to the diatom community, with only a few exceptions, expressed APA, while only few dinoflagellate species expressed APA. Diatoms usually dominated the phytoplankton community, rendering APA to be seemingly a

very important prerequisite for success in the NA. The observation of species in very low relative abundances that nevertheless did express APA, shows that APA expressing species do not necessarily dominate the phytoplankton community. This suggests that APA is also an important strategy for species to survive and maintain active metabolic state outside of their mass abundances or blooms. This allows them to immediately react on short term nutrient availability (e.g. riverine input). This feature appears to be very beneficial in a complex and fast changing environment with steep gradients of nutrient availability and point sources for nutrient input. Findings of co-dominating species in the diatom community that did not express APA, let presume that those species store P intracellularly, especially at the western site (close to the Po plume), and at the sampling time still lived on those pools. Other species did not express APA themselves, however bacteria attached to those cells did show APA. For these species a symbiotic relationship could be supposed, where the larger host diatom cell makes use of the APA expressed by the attached bacteria. Overall we found an astonishingly high within-species homogeneity of the ELFsignal. Species that did not show any ELF-signal did actually never (within one sample) show ELF-signal. While species that did show ELF-signal did show this signal in 97% to 100% of the observed intact cells. Additional in vitro experiments show an even more complex set of metabolic adaptations. The methods employed in this study increased taxonomic resolution with respect to earlier studies and allowed cellular localization of APA. These improvements uncovered a rather complex set of strategies to compete in P-limited conditions within the marine microphytoplankton community. This study also confirms the role of P-limitation as a shaping factor in marine ecosystems.

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