



## Method paper

# Taxonomic and functional diversity of a coastal planktonic bacterial community in a river-influenced marine area



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## ABSTRACT

The Gulf of Naples is a dynamical area with intense exchanges between offshore oligotrophic and coastal eutrophic waters with frequent freshwater inputs. The Sarno River, one of the most polluted rivers in Europe, strongly contributes to the pollution of the area, discharging high amounts of heavy metals and organic wastes from heavily cultivated and industrial areas. This paper reports on the diversity and community structure of the marine residential Bacteria and Archaea of the Gulf of Naples in an area close to the river Sarno plume and investigates their small-scale taxonomic diversity and expression patterns as a proxy of potential metabolic activity using metagenomics and metatranscriptomics. Bacteria and Archaea were mainly represented by marine clades, with only minor contributors from freshwater ones. The community was dominated by *Alpha*- and *Gammaproteobacteria*, of which *Rhodospirillales*, *Pelagibacteriales*, and *Oceanospirillales* were most represented. However, *Alteromonadales* and *Rhodobacterales* were the most active, despite their relative lower abundance, suggesting that they are important for overall ecosystem functioning and nutrient cycling. Nitrification and a reversed form of dissimilatory sulfate reduction were the major metabolic processes found in the metatranscriptomes and were mainly associated to *Nitrosopumilales* and *Pelagibacter*, respectively. No clear indication of transcripts related to stress induced by heavy metals or organic pollutants was found. In general, despite the high loads of pollutants discharged continuously by the Sarno River, the microbial community did not show marks of stress-induced changes neither structural nor functional, thus suggesting that this river has little or no effect on the planktonic bacterial community of the Gulf of Naples.

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## 1. Introduction

River plumes are highly dynamic areas where environmental factors change rapidly in time and space due to dilution of freshwater into the sea. The salinity gradients are usually very steep, so as those of pH, temperature, nutrients, and organic matter, significantly challenging the physiology of the bacterial and archaeal communities (Lozupone and Knight, 2007; Lindh et al., 2013; Fodelianakis et al., 2014). In general, transitions are observed, where freshwater communities, dominated by *Alphaproteobacteria*, *Betaproteobacteria*, *Bacteroidetes*, *Verrucomicrobia*, and *Actinobacteria* (Zwart et al., 2002; Crump and Hobbie, 2005; Winter et al., 2007; Read et al., 2015) are replaced by marine communities dominated by *Alphaproteobacteria* (mainly *Pelagibacteriales* and *Rhodobacteraceae*), *Gammaproteobacteria* (mainly

SAR86 clade), *Synechococcus*, *Prochlorococcus*, and marine members of *Bacteroidetes* (Bouvier and del Giorgio, 2002; Crump et al., 2004, 2007).

Despite several studies describing microbial taxonomical diversity in river influenced marine systems, functional diversity in terms of metabolic functions and transcription of functional genes has not been investigated intensely. Investigations in the Amazon River (Brazil) plume found higher abundances of C- and N-related transcripts in the free living community as compared to the particle-attached microbes, where transcripts related to vitamin biosynthesis and S-cycle dominated. Both communities were dependent on nutrients and light availability (Satinsky et al., 2014a,b). Along a transect from the Columbia River (USA) to the ocean, transcripts related to photosynthesis and denitrification increased towards marine waters, even though the metabolic potential in terms of total transcripts was highly similar along the transect and the gene expression was independent from salinity changes (Fortunato and Crump, 2015). These studies point to a large variability in bacterial and archaeal taxonomic and functional diversity occurring in river-influenced marine areas.

The Gulf of Naples is a wide and rather deep coastal embayment. Its circulation patterns are ruled by the offshore Tyrrhenian Sea, which,

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