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#### Chemosphere

journal homepage: www.elsevier.com/locate/chemosphere



## Consequences of contamination on the interactions between phytoplankton and bacterioplankton



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#### HIGHLIGHTS

- Sediment resuspension resulted in enrichment in microorganisms and contaminants.
- Increase of microbial biomass had no effect on the pelagic microbial structures.
- Impacts of nutrients and contaminants were more marked in offshore waters.
- Pesticides and heavy metals were identified as structuring factors of the microbial community.
- Impacts were direct or direct, depending on the interactions between phytoplankton and bacterioplankton.

#### ARTICLEINFO

# Article history: Received 18 July 2017 Received in revised form 14 November 2017 Accepted 8 December 2017 Available online 15 December 2017

Handling Editor: Frederic Leusch

#### ABSTRACT

Sediment resuspension can provoke strong water enrichment in nutrients, contaminants, and microorganisms. Microcosm incubations were performed in triplicate for 96 h, with lagoon and offshore waters incubated either with sediment elutriate or with an artificial mixture of contaminants issued from sediment resuspension. Sediment elutriate provoked a strong increase in microbial biomass, with little effects on the phytoplankton and bacterioplankton community structures. Among the pool of contaminants released, few were clearly identified as structuring factors of phytoplankton and bacterioplankton communities, namely simazine, Cu, Sn, Ni, and Cr. Effects were more pronounced in the offshore waters, suggesting a relative tolerance of the lagoon microbial communities to contamination. The impacts of contamination on the microbial community structure were direct or indirect, depending on the nature and the strength of the interactions between phytoplankton and bacterioplankton.

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#### 1. Introduction

Coastal zones play a key role in the global carbon budget (Borges et al., 2005). Nevertheless, demographic pressure on these zones is increasing (Small and Nicholls, 2003) and leads to the contamination of coastal ecosystems via organic and inorganic discharge and sewage (James, 2002). Organic contaminants (including polycyclic aromatic hydrocarbons, pesticides, organometallics) and inorganic pollutants (metals) accumulate in sediments (Baumard et al., 1999), which therefore represent an important pollutant reservoir.

However, this storage is not definitive; pollutants can be released into the water column during the resuspension of sediments. Such pollutant remobilization is a major problem (Schafer et al., 2006) because, even when pollution sources are removed, pollutants stored in sediments can be released into the water column and may impact pelagic communities. Assessing the consequence of this contamination on the functioning of coastal ecosystems is a major concern for the understanding of the role of these ecosystems in biogeochemical cycles.

Bacterioplankton and phytoplankton are key players in the carbon cycle in the water column. They are involved in the production of organic matter (phytoplankton) as well as in mineralization processes (bacterioplankton). Since they represent the first

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levels of the marine food web, it is imperative to understand their functioning and their interactions within the environment to gain a better understanding of the consequences of natural or anthropic perturbations for a defined ecosystem. In coastal zone and shallowwater environments, the water column is closely associated with the sediment interface, and the functioning of the bacterioplankton and phytoplankton can be strongly affected during storm events resulting in the release of nutrients, microorganisms and pollutants into the water column (Roberts, 2012). Perturbation on phytoplankton and/or bacterioplankton as well as on their interactions may have severe consequences for the higher trophic levels (Hulot et al., 2000; Hansson et al., 2013).

The impact of pollutants can be observed i) from a structural point of view, with changes observed in the microbial community through the selection of species considered tolerant to pollutants (Pesce et al., 2009; Lekunberri et al., 2010) and ii) from a functional point of view, with modifications of the carbon cycle with, for example, stimulation of heterotrophy relative to autotrophy upon metal spiking (Rochelle-Newall et al., 2008). The structure and the dynamics of the impacted community can play a crucial role, with the appearance of populations able to tolerate pollutants and/or, the disappearance of highly sensitive populations (Dorigo et al., 2004; Pringault et al., 2008). The environmental conditions can also play a significant role in stimulating or otherwise reducing the toxic effects of pollutants. Nutrient rich environments can reduce the toxicity of Cd and As on phytoplankton while increase the toxicity of Cu (Riedel et al., 2003; Miao and Wang, 2006), The presence of nutrients can impact the bioavailability of metals and consequently their potential toxicity (Miao and Wang, 2006). Furthermore, the impacts of pollutants may also be influenced by bacterio-phytoplankton coupling. The interactions between both compartments are dependent on the trophic conditions. With tight functional coupling under oligotrophic conditions, bacterioplankton depends considerably for growth and metabolism upon dissolved organic carbon released by phytoplankton, whereas in eutrophic ecosystems, bacterioplankton species can rely on allochthonous carbon to meet their requirements (Morán et al., 2002; Pringault et al., 2009; Fouilland and Mostajir, 2011). Interactions between phytoplankton and bacterioplankton can also be observed at the structural scale, with a spatial synchrony (or temporal coherence) between both compartments (Kent et al., 2007). Phytoplankton succession may influence the dynamics of the bacterioplankton community, as differences in the phytoplankton structure lead to differences in quantity and quality of exuded organic matter that can be used by the bacterial community for growth (Kent et al., 2004, 2007; Liu et al., 2014). Consequently, effects of pollutants on phytoplankton or bacterioplankton might have indirect consequences for the counterpart, depending on the possible interactions between both compartments.

In a previous work (Pringault et al., 2016), we observed that the contamination of the water column by sediment elutriate issued from resuspension of a polluted sediment can impact strongly the functional diversity of bacterioplankton. The changes observed were explained by the release of contaminants, nutrients and dissolved organic carbon into the elutriate. The goals of the present study were to determine the impact of sediment elutriate on i) the structural diversity of phytoplankton and bacterioplankton and ii) the consequences on the synchrony between both compartments. For those purposes, diversity and dynamics of phytoplankton and bacterioplankton were investigated in microcosms containing lagoon and offshore waters contaminated with polluted sediment elutriate, incubated for 96 h. In order to discriminate the impacts of pollutants from those of nutrients and bacteria, microcosms with artificial contamination (a mix of pollutants miming those released from sediment) were also performed.

#### 2. Material and methods

#### 2.1. Study site and sampling

The study was conducted in April 2014 in southwestern Mediterranean ecosystems, the lagoon and the Bay of Bizerte; see Pringault et al. (2016) for more details on the study site and the sampling procedure. Briefly, like many Mediterranean coastal lagoons, Bizerte Lagoon (Tunisia) is a polluted ecosystem subject to agriculture, urbanization and industrialization pressures, and pressures from naval and commercial shipping harbors. Consequently, the lagoon sediments are contaminated with a wide range of pollutants (Yoshida et al., 2002; Barhoumi et al., 2014a, 2014b). Bizerte Bay is less contaminated; minor local PAH contamination has been recorded in the effluent of the oil refinery located on the shore of the bay (Zrafi-Nouira et al., 2010). Sampling was carried out in an offshore station (station O, 37°16′46.46″N 9°53′50.98″E) and a lagoon station (station L 37°12′43.96″N 9°50′79.78″E) (Pringault et al., 2016). Contaminated water was obtained from polluted sediment resuspension following the protocol described by Bonnet et al. (2000). Polluted sediment was sampled in front of a cement factory in the lagoon channel (station C, 37°15'40.22"N 9°51′30.49″E) using a Van Veen grab. More details about the organic compound and metal concentrations measured in the sediment can be found in Pringault et al. (2016). In the laboratory, sediment was mixed (1:4 w/v ratio) with channel water previously filtered through a 200-um mesh and subsequently gently stirred for 8 h. After a 12-h settling period, the overlying solution, labeled thereafter as elutriate, was gently siphoned off and stored in the dark at 4°C until use in microcosm incubations. Concentrations of chlorophyll a (Chl a), nutrients, dissolved organic carbon (DOC), heavy metals, and organic pollutants were measured in for elutriate and for in situ waters. Details of the analytical procedures can be found in Pringault et al. (2016).

#### 2.2. Incubation protocol

Seawater was incubated in 9-L glass microcosms (22.5 cm diameter and 23 cm height) covered with a quartz lid to allow full penetration of the natural sunlight, including UV radiation. A series of three microcosms was filled with 6 L of sample water (lagoon or offshore station; control microcosms, C). Another series of three microcosms was filled with 4.5 L of sample water (lagoon or offshore station) and mixed with 1.5 L of elutriate to obtain a final dilution of 25% (contaminated water microcosms: CW). A third series of three microcosms was filled in the same way as the CW treatment, but microcosms were incubated in the dark (dark contaminated water microcosms: DCW). The final series of three microcosms (artificial contaminated water microcosms, ACW) was filled with 6 L of lagoon and offshore waters and mixed with an artificial solution of pesticides (diuron, di-isopropyl-atrazine (DIA), 3,4-dichlorophenylurea (DCPU), alachlor, and linuron) and metals (Ni, Cu, Zn, Cd, As, Pb). Final concentrations of pesticides and metals were adjusted to mimic as closely as possible the concentrations observed in the elutriate; see Pringault et al. (2016) for more details. Microcosms were incubated in triplicates outdoor for 96 h under natural sunlight in a 3-m<sup>3</sup> pool where seawater was circulating (open system) to maintain the in situ water temperature. Average daily light intensity was 7800 kJ m<sup>-2</sup> d<sup>-1</sup>, with a photoperiod of about 13 h.

#### 2.3. Bacterioplankton diversity

Bacterioplankton diversity was determined for *in situ* water (Stations O and L) and the elutriate as well as at T24 and T96 in each

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