



# Evaluation of phytoplankton quality and toxicity risk based on a long-term time series previous to the implementation of a bivalve farm (Basque coast as a case study)



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## HIGHLIGHTS

- This is the first exhaustive study of phytoplankton community in Basque coastal waters.
- Essential study for the development of the new pilot-scale bivalve farm in this area.
- The observed phytoplankton community seems favorable for bivalve growth.
- The observed presence of blooms and dominance of diatoms benefit bivalve nutrition.
- Although several toxic taxa were found, their frequency was low.

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## ABSTRACT

In the last decades there has been a great development in aquaculture worldwide and, on the Basque coast (southeastern Bay of Biscay) in particular, there is a recent interest in implementing bivalve aquaculture in open marine waters. In this context, the study of phytoplankton is essential given that it is the main source of energy for bivalves and, at the same time, a main potential toxicity risk. Bivalves, as filter-feeding organisms, can accumulate phycotoxins and these can be transferred through the food-chain, posing a threat to humans. All this, together with a recently installed pilot-scale bivalve farming, motivated a study of the phytoplankton community. Here, 11-year phytoplankton time series from 16 nearshore and 3 offshore stations off the Basque Country are analyzed, as a preliminary step for evaluating the potential of this region for aquaculture development. Special attention was given to bloom events and potentially toxic taxa. A total of 32 bloom-forming taxa were detected, mostly diatoms. In regard to harmful species, all stations presented many potentially toxic taxa, mostly dinoflagellates. The diatom genus *Pseudo-nitzschia* was the one blooming in more stations. *Pseudo-nitzschia* spp. as well as the dinoflagellates *Dinophysis* spp. and *Alexandrium* spp., which might be causative of Amnesic, Diarrhetic and Paralytic Shellfish Poisoning, respectively, exceeded the abundance limits that would imply toxicity risk in several occasions, mostly during spring and summer. However, it occurred at a low frequency (in average, < 15% for *Pseudo-nitzschia* spp. and < 10% for the dinoflagellates). Overall, phytoplankton community composition and abundance, together with the low frequencies for the exceeded alert limits by the three main phycotoxin producing genera, suggest that the area presents appropriate conditions for bivalve aquaculture.

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## 1. Introduction

World aquaculture production has greatly increased in the last 60 years, from about 20 million t in 1950 to almost 150 million t in 2010 (FAO, 2012). Production of marine mollusks presently accounts for 75% of global marine aquaculture and it is expected to keep expanding given the depletion of natural stocks

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(Barg, 1992; FAO, 2014). Spain is amongst the biggest aquaculture mussel producers in a world scale and the first in the European Union. In Spain, almost the entire aquaculture mussel production is developed on the northwest coast, in Galicia. There, mussel cultivation is the most important socio-economic activity with an annual production above 250,000 t (Figueiras et al., 2002). Nevertheless, this activity has never been developed on the Basque coast (northern Spain).

Bivalves, as filter-feeding organisms, get the energy and nutrients necessary to grow from suspended microscopic food particles (e.g. Jørgensen, 1990). Regarding shellfish aquaculture, phytoplankton is the main component of the diet of suspension feeding bivalves (Shumway and Cucci, 1987; MacDonald and Ward, 1994; Grant, 1996; Petersen et al., 2008). The quantity and size of the phytoplankton can influence the recruitment of oysters for instance, as well as the survival of bivalve larvae (Robert and Trintignac, 1997; Bourlès et al., 2009). Moreover, in field studies, Wall et al. (2013) observed that the growth rates of bivalves were more related to the density of certain cellular types than to the total phytoplankton biomass. Therefore, a good knowledge of phytoplankton composition and variability is essential to assess the appropriateness of an area to sustain bivalve aquaculture.

Phytoplankton can also be harmful: the so-called “Harmful Algal Blooms” (HABs) can have deleterious effects on entire ecosystems, and even cause important economic impacts (Anderson, 2009). In fact, the increased frequency of HABs has been indicated as one of the main problems in coastal regions worldwide. In terms of harmful effects, two types of causative organisms can be considered within the phytoplankton: the high-biomass producers and the toxin producers. Although some taxa present both features, the last ones can be harmful even at very low densities (Masó and Garcés, 2006). This is because phytoplankton toxins ingested by filter feeding organisms can accumulate within their flesh (e.g. Wang, 2008) and get gradually transferred to the higher trophic levels along the food web, posing a threat to human health (Davidson and Bresnan, 2009). Examples of toxic syndromes include ciguatera fish poisoning (CFP), and paralytic, diarrhetic, neurotoxic, azaspiracid and amnesic shellfish poisoning (PSP, DSP, NSP, AZP and ASP, respectively) associated mostly with shellfish consumption (Glibert et al., 2001). Other less frequent toxins produced by microalgae have also been evidenced to produce damages to humans and/or shellfish, such as yessotoxins (Amzil et al., 2008), palytoxins (Aligizaki et al., 2011) and pectenotoxins (Fernández et al., 2006). In addition to the production of toxins, some phytoplankton species could cause mechanical stress to other organisms (Delegrange et al., 2015) implying also a damage to aquaculture.

Many studies have been carried out in the Basque estuaries regarding phytoplankton composition (Orive et al., 1998; Trigueros and Orive, 2001; Ansotegui et al., 2003; Seoane et al., 2005, 2006; Laza-Martinez et al., 2007) and potentially toxic species (Orive et al., 2010, 2013). However, very few studies have addressed the composition and the size-structure of the phytoplankton communities in open coastal waters of the Basque Country. Furthermore, there is a limited amount of information about toxic species from the neighboring areas (e.g., the French Phytoplankton and Phycotoxins Monitoring Network—REPHY Maurer et al., 2010) and, to our knowledge, only the studies of Seoane et al. (2012) and Batifoul et al. (2013) addressed it in open waters near the Basque coast.

In this context, research at the local scale is necessary in order to understand HAB dynamics and enhance the management of coastal ecosystems. This is of special concern in the Basque Country, since during the last years there is an increasing interest in developing shellfish aquaculture in open waters of this region, where a pilot-scale bivalve farming (longline system) was installed in 2012 (Azpeitia et al., 2016).

Taking all this into account, the present study aims to contribute to the evaluation of the potential of this region for the development of aquaculture activities in exposed marine areas from the perspective of the phytoplankton composition taking advantage of a long-term data series (2003–2013). For this, the specific objectives of this study are to evaluate (i) the quality of the phytoplankton to sustain bivalve growth, and (ii) the occurrence of phytoplankton species considered to have the capacity for toxin production, within open coastal waters of the Basque Country (southeastern Bay of Biscay).

## 2. Material and methods

### 2.1. Study area

The Basque coast is located in the eastern Cantabrian Sea, southeastern Bay of Biscay (Fig. 1). It extends approximately 100 km along the north of Spain. It can be described as an exposed littoral coast, mostly formed by cliffs and influenced by 12 short rivers, accounting for a total flow of about  $150 \text{ m}^3 \text{ s}^{-1}$  (annual mean). Although no large coastal plumes are formed (Diez et al., 1999), this freshwater supply modifies the chemical composition of the shelf waters and leads often to increased nutrient levels in inner shelf waters (Valencia et al., 2004; Ferrer et al., 2009). The upwelling activity is almost negligible on the Basque coast (Valencia et al., 2004). The climate of the area is rainy, temperate and oceanic, with moderate winters and warm summers. According to Köppen's classification it is described as marine west-coast and mild (Fontán et al., 2009).

### 2.2. Sampling strategy and laboratory work

In this study, data from 19 stations of the “Littoral Water Quality Monitoring and Control Network” of the Basque Water Agency were employed (Fig. 1) (Borja et al., 2004, 2016). Most of these stations (16 of them) are located in exposed coastal areas at a depth of 25–35 m. Three further stations are offshore, at 100–120 m depth.

The data set involves 11 years (from 2003 to 2013) except for two offshore stations that present a 5-year data set (RF20 and RF30, from 2009 to 2013). Two samplings per year (spring and summer) were conducted from 2003 to 2007 and four (winter, spring, summer and autumn) from 2008 to 2013.

Samples were taken in surface waters (0–1 m depth), preserved immediately and maintained in 125 ml borosilicate bottles in dark and cool conditions ( $4^\circ \text{C}$ ) until analysis. Glutaraldehyde was used for preservation until 2011 and acidic Lugol from then on. The taxonomic identification and cell counting were made on subsamples of 50 and 10 ml, depending on the density of particles settled, following the Utermöhl method (Utermöhl, 1958; Hasle, 1978; Edler and Elbrächter, 2010). Therefore, the picoplankton fraction was not recorded. Most of the diatoms and armored dinoflagellates were identified to the level of species. Smaller or more fragile forms were classified generally at the level of genus or class. The nanophytoplankton cells that could not be assigned to any taxonomic group were clumped together into a group named “unidentified forms  $<10 \mu\text{m}$ ”. A taxa list is provided as Electronic Supplementary material (see Appendix A).

### 2.3. Composition and size-structure of phytoplankton blooms

Historically, blooms have been inferred to be significant population increases. However, there is no universal criterion or specific cell abundance to define a bloom event (Smayda, 1997). In this study the cell-size-based approach defined by Revilla et al. (2009) was followed to define bloom episodes. Two different thresholds were used based on the Equivalent Spherical Diameter (ESD):  $7.5 \times 10^4 \text{ cells L}^{-1}$  for taxa  $>20 \mu\text{m}$ , and  $7.5 \times 10^5 \text{ cells L}^{-1}$

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