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ENVIRONMENTAL TOXICITY ASSESSMENT: STATE OF THE ART AND FUTURE DIRECTIONS IN A WORLD OF ARISING THREATS



Mediterranean alien harmful algal blooms: origins and impacts

Christina Marampouti¹ · Anita G. J. Buma¹ · M. Karin de Boer^{1,2}

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Abstract

Harmful algal blooms (HABs) are mostly phytoplankton blooms, which have detrimental environmental and socioeconomic impacts. The Mediterranean Sea due to its enclosed nature is of special concern since it has an enormously rich native biodiversity. Though, it is also the world's most invaded marine ecosystem and is considered at very high risk of future invasions. The aim of this review study is to explore the origins, establishment, environmental, and socioeconomic impacts of HABs caused by nonnative algal species in the Mediterranean Sea. Based on this, it is also discussed whether HABs form an increasing threat in the basin, and what could possibly be done to prevent or to minimize their impacts. The increasing rate of their introduction and the harmful impacts that they have on the environment, economy, and human health makes it important to have accurate knowledge about HABs. Anthropogenic activities and climate change are considered the main contributors of alien invasions but also the main enablers of HAB events. Mediterranean HABs are adequately studied, but there are no studies purposefully concerning invasive microalgae species in the basin. In the present study, 20 species have been identified, and an attempt has been made to collect their introduction information, as well as known or suspected impacts. Future research should be focused on data mining, current legislation updates, and monitoring of Mediterranean coastlines.

Keywords Harmful algal bloom · Invasive alien species · Mediterranean Sea · Introductions · Pathways · Toxin · Syndromes

Introduction

Harmful algal blooms (HABs) have shown an increase in frequency, intensity, and distribution in a global scale (Ben-Gharbia et al. 2016; Van Dolah 2000). These events develop mostly in coastal and sheltered areas all around the world, such as harbors, smaller bays, and coastal lagoons that can of course be attributed to the increase of monitoring programs that came with technological progress and scientific curiosity, but also with global climate change and anthropogenic impacts. These include eutrophication, habitat modification,

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² Bèta Science Shop, Faculty of Science and Engineering, University of Groningen, Nijenborgh 6, AG 9747 Groningen, The Netherlands and human-mediated introduction of no indigenous species (Kudela et al. 2015).

What are harmful algal blooms

Photosynthetic microalgae contribute in healthy aquatic ecosystems by being the foundation of the food web, fixing carbon, and producing oxygen. However, under certain circumstances, some species have the ability to form high-biomass or toxic proliferations of cells ("blooms"). These microalgae cause harm to aquatic ecosystems, including fauna and flora, but also to humans via direct exposure to water-borne toxins or by toxic seafood consumption (Ferrante et al. 2013). Most HAB species are microalgae that belong to the classes of Bacillariophyceae (diatoms), Dinophyceae (dinoflagellates), Dictyochophyceae, Prymnesiophyceae, Raphidophyceae, and Cyanophyceae (cyanobacteria). Dinoflagellate species are the most abundant HAB species and produce the majority of the toxins in the Mediterranean Sea (Arff and Miguez 2016). HABs are naturally occurring phenomena, present in nearly all aquatic environments (freshwater, brackish, and marine). The different types of marine HABs can be

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distinguished by three distinctive phenomena: anoxia (due to high biomass), intoxication of marine life, and accumulation of toxins in the food chain.

The blooms of pelagic microalgae that discolor the water have traditionally been called red tides, whether they are toxigenic or not. More specifically, in the Mediterranean, some blooms can be caused by algal species that produce toxins (e.g., *Ostreopsis ovata*) where others cause nontoxic high-biomass blooms (e.g., *Alexandrium taylorii*) (Kudela et al. 2015).

HABs from alien invasive species

Harmful microalgae that are found in tropical areas are expanding to temperate ecosystems probably due to water temperature rising (Ben-Gharbia et al. 2016). Evidently, HABs of pelagic and benthic microalgae are also increasing both in intensity and frequency in the Mediterranean Sea. It has been also observed that many alien microalgae blooms are co-occurring with native species blooms, but the mechanisms of their insurgence are poorly studied. Even though native Mediterranean phytoplankton species have always been manifesting blooms, alien species are a new parameter in a preexisting problem that only increases HABs in the Mediterranean waters (Streftaris and Zenetos 2006). A fine example of Mediterranean alien HAB event increase is given by Garcés et al. (2000) that reports the increase of HAB phenomena in the Mediterranean over a period of 50 years. By focusing on the findings of *A. taylori* (Fig. 1) that is considered alien, it is obvious that the species distribution is expanding, and thus, other Mediterranean areas are affected by its blooms.

According to the IUCN definition, agreed also by the Convention on Biological Diversity, an "invasive alien species (IAS), is an alien species which becomes established in natural or semi-natural ecosystems or habitats, is an agent of change, increases in abundance and distribution and threatens native biological diversity" (IUCN 2017). In accordance with the EU Regulation 1143/2014/EU on the prevention and management of the introduction and spread of invasive alien species, IAS are "any live specimen of a species, subspecies or lower taxon of animals, plants, fungi or microorganisms introduced outside its natural range; it includes any part, gametes, seeds, eggs or propagules of such species, as well as any hybrids, varieties or breeds that might survive and subsequently reproduce." Following the same regulation are referred to as "an alien species whose introduction or spread has been found to threaten or adversely impact upon biodiversity and related ecosystem services." A more precise difference in terminology between alien and invasive species is related to their stage of introduction and strength of impacts and expansion, as presented in Fig. 2.

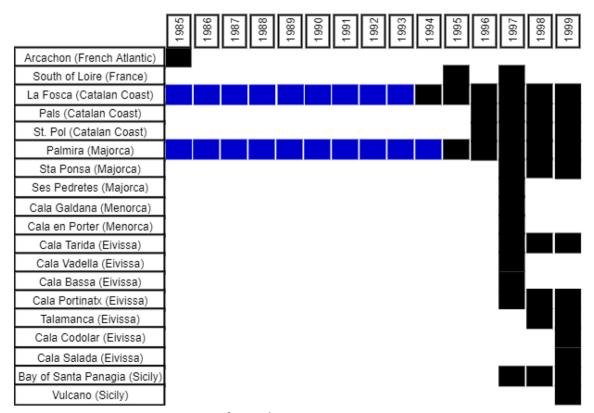


Fig. 1 Areas of Alexandrium taylori blooms with up to 10^5 cells L⁻¹ in Balearic basin (Table 3 in Garcés et al. 2000)

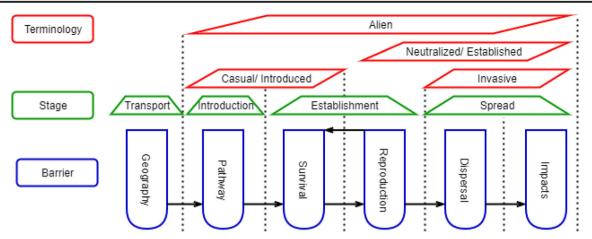


Fig. 2 Schematic representation separating alien from invasive species (terminology). Stages of introduction of alien species (stage). The paths followed by different species to reach different stages from alien to

Mediterranean Sea

The Mediterranean Sea connects to the surrounded sea bodies via three relatively narrow pathways, the Strait of Gibraltar, the Dardanelles waterway, and the Suez Canal. Its enclosed nature makes it susceptible to alien species introduction, thus threatening the already existing rich biodiversity of the basin. The Mediterranean is considered to be the world's most invaded marine ecosystem prone to future invasions as the anthropogenic activities and sea water temperatures are increasing (Ghabooli et al. 2013). Alien species invasions are one of the main causes for biodiversity loss in the basin because they alter the ecosystem dynamics of the marine environment in various direct but also indirect ways (Galil 2007; Coll et al. 2010; Otero et al. 2013). Their always facilitated and expanding introduction rate that extends to all Mediterranean regions poses a serious threat since they are causing unexpected and harmful impacts on the environment, economy, and also human health. They are considered "focal species"; thus, monitoring is essential in all Mediterranean regions to minimize those impacts (Katsanevakis et al. 2014a).

In the past, alterations of Mediterranean biodiversity were driven by geological and physical changes, and the Mediterranean Sea has been traditionally considered as being of low risk for harmful algal blooms due to its oligotrophic nature. However, nowadays, human activities are essential elements to consider as well, since they are one of the main reasons for habitat loss, degradation and pollution, overexploitation of marine resources, invasion of species, and of course climate change (Coll et al. 2010). The high population growth in coastal areas and the extraordinary increase in the number of harbors for recreational purposes have created suitable nutrient-rich environments for the proliferation of some algal species (Bravo et al. 2008).

Climate change also has a significant impact on coastal zones, as it drives the predicted rise in sea levels, sea, and

invasive species according to their geography, pathway of introduction, survival rate, reproduction ability, further dispersal, and impacts (amendment of Fig. 1 in Otero et al. 2013)

air temperatures and also changes other hydrological characteristics of the Mediterranean coasts (Katsanevakis et al. 2014a). In the last decades, as climate change became more and more threatening, the "tropicalization" of the Mediterranean has been reported (Bianchi and Morri 2003), meaning that with the water temperature increase deriving from climate change, the temperate Mediterranean climate is becoming more and more tropic. The increased temperature of Mediterranean waters is the main reason for many alien species dispersal and establishment. There have been reported cases of species established for years in a specific Mediterranean area (resting cysts) that have not managed to expand. Although in the last years, as a result of environmental change, their population has been expanding to invasive levels (Genovesi et al. 2015; Leydet and Hellberg 2015). The increase of thermophilic biota in the Mediterranean Sea would involve changes in both indigenous (meridionalization) and non-indigenous (tropicalization) species (Saiz et al. 2014).

Before the 1980s, blooms in the Mediterranean were considered rare events (Maso and Garcés 2006). Therefore, interest from the scientific community on this topic has risen over the last decades due to their increasing distribution, from tropical waters to temperate zones such as the Mediterranean Sea (Hachani et al. 2018).

Anthropogenic activities have a direct link to Mediterranean invasions and species establishment. Pollution deriving from human pressure and climate change followed by eutrophication are known to play a key role in the increase of HAB phenomena. When nutrient loads increase, phytoplankton becomes successful over bacteria due to competition, and their growth can stimulate an algal bloom. With the current expeditiously altering Mediterranean nutrient ratios, the phytoplankton dynamics are expected to shift in the area, facilitating the expansion of nonsiliceous species such as flagellates and dinoflagellates (Danovaro 2003). Consequently, together with a modification of the coastal habitats, an increase in red tide events is expected, in conjunction with the spread of toxic dinoflagellates, as already reported (Garcés et al. 2000; Vila et al. 2001). It is also safe to assume that the Mediterranean Sea is threatened from new invasions and HAB occurrences more and more (Coll et al. 2010), even though little is known about the invasion of alien phytoplankton taxa in the area (Gómez 2003). Most existing studies are focusing either on alien Mediterranean species or on HAB-causing species (native and alien) but there has not yet been a collective approach on the issue of introduced microorganisms that can cause HABs (Gómez 2010).

Research aim

The majority of alien invasive HAB-causing species has been already identified in the Mediterranean basin; however, every year, new species are found to be present. Although mostly due to the European Union (EU) and individual countries' initiatives, several databases and scientific papers exist that describe or contain information about these alien species. No collective database or paper exists to present only alien/ invasive microalgal species that can cause HABs in the Mediterranean Sea and their impacts.

The present review study is focusing on alien/invasive species that cause HABs, their origin, vector of introduction, distribution, and impacts in the basin. The main aim of the study, apart from presenting a collective information source, is also to examine if HABs derived from introduced species are actually increasing over the last decades and why. It is hypothesized that HAB Mediterranean events are indeed increasing due to the tropicalization of the area that promotes their establishment, dispersion, and successful reproduction.

Findings

After a thorough review of the literature, a list was composed of 20 microalgal species. From these, only 6 are referred to as invasive in the Mediterranean Sea and the remaining 14 as alien (Table 1). It is crucial to mention that more species than presented below were considered. However, not enough information could be found for those species or their invasiveness status is debatable; thus, further research is required (e.g., *Alexandrium leei*, *Alexandrium monilatum*, *Amphidoma languida*, *Tripos candelabrus*, *Prorocentrum borbonicum*, *Prorocentrum levis*, *Scrippsiella acuminate*). The final list of species consists of the most welldocumented cases (e.g., *Gymnodinium catenatum* and *Ostrepsis ovata*) as well as comparably new introductions (e.g., *Prorocentrum shikokuense*).

Final list of introduced harmful algal species

- 1. Alexandrium andersonii
- 2. Alexandrium ostenfeldii
- 3. Alexandrium pacificum
- 4. Alexandrium taylori
- 5. Chaetoceros bacteriastroides
- 6. Chaetoceros pseudosymmetricus
- 7. Coolia monotis
- 8. Dinophysis acuminata
- 9. Fibrocapsa japonica
- 10. Gymnodinium catenatum
- 11. Karenia mikimotoi
- 12. Lingulodinium polyedrum
- 13. Ostreopsis ovata
- 14. Ostreopsis siamensis
- 15. Prorocentrum emarginatum
- 16. Prorocentrum shikokuense
- 17. Pseudo-nitzschia multistriata
- 18. Sinophysis canaliculata
- 19. Skeletonema tropicum
- 20. Gambierdiscus sp.

Distribution and origin

Even though invasive alien species origins and distribution are relatively well documented since they pose an increasing Mediterranean problem, alien microalgae are a different category due to their small size and multiple genera. They are often misidentified, like in the case of Alexandrium pacificum that was considered Alexandrium catenella in the Mediterranean until 2005 because they are both included in the Alexandrium tamarense species complex (John et al. 2014). Data published before 2005 which may refer to A. catenella could also offer information on A. pacificum too. It is also important to mention the absence of Alexandrium minutum from the list. This is because although its blooms are thoroughly studied, its origin is uncertain since it is believed to originate from Alexandria, Egypt (WoRMS 2019), thus being native to the Mediterranean. However, it is also referred to as alien with an uncertain invasiveness status in some other cases (GISD 2019). Moreover, the origin of the Mediterranean A. pacificum strain has been debated to be derived from Australian (Guiry and Guiry 2019) or Japanese (Genovesi et al. 2015) populations. Another species that was previously misidentified is Karenia mikimotoi that was confused with the nontoxic Gyrodinium aureolum and Coolia monotis that was first described in the Mediterranean as Glenodinium monotis (Gómez 2008). Another challenging factor was the status identification of Gyrodynium catenatum, since it is considered to be invasive (Katsanevakis et al. 2014b) but also cryptogenic (Zenetos et al. 2005; Perini

Table 1 An overview o	An overview of the alien/invasive Mediterranean microal	nicroalgae, their invasiveness status	, origin, year of introduction, vector	lgae, their invasiveness status, origin, year of introduction, vector, and their Mediterranean distribution	u.
Species	Status	Origin	Year	Vector	Distribution
A. andersonii	Invasive (WoRMS 2019)	Cape cod, Massachusetts, Eastern US coast (WoRMS 2019)	1998 (Ciminiello et al. 2000)	N/A	Adriatic, Tyrrhenian Sea (Ciminiello et al. 2000)
A. ostenfeldii	Alien (Molnar et al. 2008; WoRMS 2019)	Iceland (WoRMS 2019)	N/A	Ballast water (Molnar et al. 2008)	Egypt, Spain (Molnar et al. 2008)
A. pacificum	Invasive (Genovesi et al. 2015)	Japan, Australia (Genovesi et al. 2015; Guiry and Guiry 2019)	1983 (Genovesi et al. 2015)	ballast water, shellfish aq. (Genovesi et al. 2015)	France, Italy, Spain, Western Med. (Genovesi et al. 2015)
A. taylori	Alien (Molnar et al. 2008; WoRMS 2019)	France, Atlantic Oc. (Guiry and Guiry 2019)	1982 (WoRMS 2019)	Ballast water, Gibraltar inflow currents (Molnar et al. 2008)	France, Central - Western Med. (Molnar et al. 2008; WoRMS 2019)
C. bacteriastroides	Alien (Čalić et al. 2018; WoRMS 2019)	Indian Oc. (Aligizaki et al. 2008; Čalić et al. 2018)	2010 (Čalić et al. 2018)	Levantine inflow currents (Čalić et al. 2018)	Adriatic Sea (Čalić et al. 2018)
C. pseudosymmetricus	Alien (Čalić et al. 2018)	Indian Oc. (Čalić et al. 2018)	2015 (Čalić et al. 2018)	Levantine inflow currents (Čalić et al. 2018)	Adriatic Sea (Čalić et al. 2018)
C. monotis	Invasive (WoRMS 2019)	Belgium, North Sea (WoRMS 2019)	1930s (Molnar et al. 2008)	Ballast water, oyster aq. (Molnar et al. 2008)	Widespread (Molnar et al. 2008; WoRMS 2019)
D. acuminata	Alien (Ignatiades and Gotsis-Skretas 2010; WoRMS 2019)	Norway, North Sea (WoRMS 2019)	N/A	N/A	Eastern Med. (Varkitzi et al. 2018)
F. japonica	Alien/established (Cucchiari et al. 2008)	Japan, Australia (Cucchiari et al. 2008; Gómez 2008)	1997 (Cucchiari et al. 2008)	Ballast water (Cucchiari et al. 2008)	Adriatic, Tyrrhenian Sea (Cucchiari et al. 2008)
G. catenatum	Invasive/cryptogenic (Zenetos et al. 2005; Katsanevakis et al. 2014a; Mozetič et al. 2017; Gladan et al. 2019)	Gulf of California, Pacific Oc. (WoRMS 2019)	1989 (Bravo et al. 1990)	Gibraltar inflow currents (Gladan et al. 2019)	Alborán Sea, South Med. (Katsanevakis et al. 2014a; Gladan et al. 2019)
K. mikimotoi	Alien/established/cryptogenic (Zenetos et al. 2008; WoRMS 2019)	Japan, East China Sea (Perini et al. 2018; WoRMS 2019)	1985 (AquaNIS 2015)	Ballast water (Zenetos et al. 2008)	Tyrrhenian, Aegean, Balearic Sea (Roselli et al. 2019)
L. polyedrum	Alien (Guiry and Guiry 2019)	Germany, North Sea (Guiry and Guiry 2019)	Before 1976 (Boni et al. 2000)	N/A	Adriatic Sea (Boni et al. 2000; Mozetič et al. 2017)
O. ovata	Invasive (Cohu and Lemee 2012; WoRMS 2019)	South-West Pacific Oc. (WoRMS 2019)	1979 (Gladan et al. 2019)	Shellffish aq., ballast water (Molnar et al. 2008; (Ignatiades and Gotsis-Skretas 2010)	Widespread (Streftaris and Zenetos 2006; Ghabooli et al. 2013; WoRMS 2019)
O. siamensis	Alien (WoRMS 2019)	Gulf of Thailand (Guiry and Guiry 2019; WoRMS 2019)	1979 (WoRMS 2019)	Ballast water (Ignatiades and Gotsis-Skretas 2010)	Widespread (Tichadou et al. 2010)
P. emarginatum	Alien (Zenetos et al. 2005)	Japanese Sea (Guiry and Guiry 2019; WoRMS 2019)	N/A	N/A	Greece (Ignatiades and Gotsis-Skretas 2010)
P. shikokuense	Alien (Roselli et al. 2019)		2016 (Roselli et al. 2019)		

(
Species	Status	Origin	Year	Vector	Distribution
		East China, Korean, Japanese Sea (Roselli et al. 2019)		Ballast water (Roselli et al. 2019)	South Adriatic Sea (Roselli et al. 2019)
P. multistriata	Invasive (Mozetič et al. 2017)	Japanese Sea (Guiry and Guiry 2019; WoRMS 2019)	1995 (Mozetič et al. 2017)	Ballast water (Mozetič et al. 2017)	Adriatic, Greece (Mozetič et al. 2017; Roselli et al. 2019)
S. canaliculata	Alien (Aligizaki et al. 2008)	Indo-Pacific Oc. (Aligizaki et al. 2008)	2004 (Aligizaki et al. 2008)	N/A	South-East Med. (Aligizaki et al. 2008)
S. tropicum	Alien (Mozetič et al. 2017; WoRMS 2019)	Tropics (Mozetič et al. 2017)	2002 (Mozetič et al. 2017)	N/A	Adriatic Sea (Mozetič et al. 2017)
Gambierdiscus sp.	Alien (Aligizaki et al. 2008)	Indo-Pacific Oc. (Aligizaki et al. 2008)	2003 (Aligizaki et al. 2008)	N/A	South-East Med. (Aligizaki et al. 2008)

et al. 2018). Furthermore, *Gambierdiscus* sp. is not identified to a species level in the Mediterranean (Aligizaki et al. 2008; Zenetos 2010) but some papers describing the impacts of the species are comparing it with *Gambierdiscus toxicus* (Vila et al. 2001; Aligizaki et al. 2008). Finally, *Chaetoseros bacteriastroides* and *Chaetoseros pseudosymmetricus* findings in Adriatic Sea represent the northernmost records in world's oceans and seas. For *C. pseudosymmetricus*, this is also the first occurrence in European seas (Čalić et al. 2018). Recent records of these unusual *Chaetoceros* species in the Mediterranean Sea should be considered as an example of the expansion of thermophilic phytoplankton species.

Toxins of alien/invasive species

It is imperative to know what kind of toxins the alien or invasive alien microalgae are co-introducing in the Mediterranean Sea and what syndromes these toxins can cause (Table 2). Toxins produced by HABs are associated with several syndromes, including paralytic (PSP), diarrhetic (DSP), amnesic (ASP), neurotoxic (NSP), and ciguatera (shell) fish poisoning (CFP), caused by consumption of contaminated seafood. Toxins are bioaccumulated by organisms that ingest algae, and thus transmitted through the food web up to humans (biomagnification).

Some of the toxins produced by microalgae include polytoxins (PLTXs), okadaic acid (OA), dinophysistoxins (DTXs), pectenotoxins (PTXs), yessotoxins (YTXs), azaspiracids (AZAs), saxitoxins (STXs), neosaxitoxins (NSTXs), goniautoxins (GTXs), spirolides (SPs), ovatoxins (OVTXs), cooliatoxins, and domoic acid (DA) (Table 2).

One of the most toxic alien microalgae in the Mediterranean is O. ovata that can produce putative PLTXs, ovatoxin-a, DSP-like, and ciguatoxin-like toxins and has been shown to affect Mediterranean coastal areas. The high cell concentration of O. ovata that is present in sea water can affect human health in two ways; a direct contact with affected water can cause skin and respiratory disorders and in an indirect way by bioaccumulation and/or biomagnification of the produced toxins. Most human health incidents by O. ovata are via filter-feeding organisms (mussels), making seafood consumption alarming. More specifically, between 2006 and 2009, nine O. ovata blooms were observed along the French Mediterranean coastline. Five of those events affected not only tourists but also coastline inhabitants. Symptoms presented in 47 cases varied up to mild irritations of the skin, mucous membrane and the respiratory system, and regressed without any treatment after 12-72 h. In addition, five recreational beaches were also closed to the public for a short period of time (Tichadou et al. 2010).

On the other hand, the toxic diatom's *C. monotis* Mediterranean strains does not seem to be toxic. However, Table 2 An overview of the alien/invasive Mediterranean microalgae, the abbreviation of toxins they produce, and the syndromes they may cause in the Mediterranean Sea

Species	Toxins	Syndrome
A. andersonii	STX, NSTX	PSP
A. ostenfeldii	GTX, SP	PSP, mussel toxicity
A. pacificum	STX, SP, goniodomins, gymnodimines	PSP
A. taylori	Nontoxic	N/A
C. bacteriastroides	Nontoxic	N/A
C. pseudosymmetricus	Nontoxic	N/A
C. monotis	Cooliatoxin	PSP
D. acuminata	OA, DTX	DSP
F. japonica	hemolytic compounds, brevetoxins	Fish killings
G. catenatum	STX, GTX	PSP
K. mikimotoi	1-acyl-3-digalactosyl glycerol, octadeca-pentaenoic acid	Fish and invertebrate mortality
L. polyedrum	STX, YTX,	PSP, YTX no proven effect on humans
O. ovata	PLTX, OVTX, ciguatoxin-like toxins, ostreocins	respiratory
O. siamensis	PLTX, OVTX, YTX, ciguatoxin-like toxins, ostreocin D	Clupeotoxism, respiratory
P. emarginatum	Unknown	Potentially, DSP
P. shikokuense	OA, DTX	DSP
P. multistriata	DA	ASP
S. canaliculata	N/A	N/A
S. tropicum	N/A	N/A
Gambierdiscus sp.	N/A	CFP

Paralytic (PSP), diarrhetic (DSP), amnesic (ASP), and ciguatera (shell) fish poisoning (CFP)

further studies are needed to identify the toxicity of all alien Mediterranean algae and their effects on human health and environment. These are several irregularities and lack of knowledge; for instance, little is known about the effects of YTXs on humans (Zingone et al. 2006), and even though P. shikokuense's blooms are considered toxic in Japan, no harmful effects were observed during Mediterranean blooms (Roselli et al. 2019).

Alien invasive HAB Mediterranean events

Mediterranean HAB events are mostly related to locations with limited water exchange like harbors (Vila et al. 2001), sheltered beaches, smaller bays, and coastal lagoons (Giacobbe et al. 2000). In these enclosed areas, during summer and early autumn months, the terrestrial water inputs are limited, and thus due to solar radiation subsequently evaporation, sea levels have a tendency to decrease; thus, nutrient and salinity levels are periodically increasing boosting phytoplankton concentrations and generating blooms (Armi et al. 2012). Furthermore, biological invasions are an indication of global change. These invasions are considered to be one of the major drivers of change for biodiversity. For the Mediterranean area, HAB events of alien species induce severe environmental and economic impacts affiliated with biodiversity loss, unbalanced ecosystem, and degradation of ecosystem services such as fisheries, aquaculture, and tourism (Roselli et al. 2019). The Harmful Algal Information System (HAIS) is the main initiative that focusses on accumulating data on HAB events. They provide information on harmful algal events on HAEDAT, a database that assembles most harmful algal monitoring and management systems worldwide. With the use of the current taxonomic names of harmful algae, HAEDAT also provides biogeographical and toxicological information.

In the case of the Mediterranean, most blooms seem to form in the Western part of the basin (Table 3). This could be attributed to the focus of studies in these areas and to existing monitoring stations. For the purpose of this study, a recent update of all HAEDAT HAB-recorded events of the listed alien/invasive microalgae that occur in the Mediterranean was made (Table 3). This table gives an example of the frequency of the occurrence and distribution of these microalgae, even if limited.

By taking as an example O. ovata, a study by Cohu and Lemee (2012) determined that different ecological factors could influence the abundances of the species. The temperature seems to have a severe influence, especially in Ostreopsis spp. that reported growth between 22 and 30 °C in temperate

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Table 3 An overview of all HAB events in the Mediterranean until 2019 caused by the listed alien/ invasive microalgae that are presented in HAEDAT (2019)

Species	Year	Location
A. taylori	1994; 1995; 1996; 1999 (× 3); 2000; 2001; 2004	Catalonia, Spain
	1996; 1997; 1998; 1999 (× 2); 2001; 2003 (× 2); 2004 (× 2)	Balearic Islands, Spain
G. catenatum	1987; 1989; 1999; 2012; 2013 (× 3); 2014; 2016	Andalucía, Spain
	1990	Valencia, Spain
	2000 (× 2); 2001 (× 2); 2002; 2003 (× 2); 2004 (× 2); 2006; 2010; 2012; 2017; 2018;	Alboran sea, Spain
	2011 (× 2)	Cadiz-Malaga, Spain
L. polyedrum	2009 (×3); 2010 (×2); 2004 (×3); 2005 (×2); 2009 (×5); 2010 (×10); 2014 (×8); 2015 (×4); 2016	Slovenia
	2006 (× 2)	Catalonia, Spain
O. ovata	2002; 2005	Ligurian Sea, Italy
	2010 (× 2); 2014; 2015; 2016; 2017; 2018 (× 2)	Catalonia, Spain
O. siamensis	2004; 2006	Catalonia, Spain

areas. Concerning light, *O. ovata* was abundant at very shallow depths showing a good adaptation to high intensity. Since the species is also considered one of the most invasive in the Mediterranean, it is useful to have an overview of its distribution to be able to better comprehend the range of alien species adjustment capability and dispersal.

Gladan et al. (2019) also presents an overview of temporal and spatial distribution of *Ostreopsis* sp. (*O. siamensis* included) events in the Mediterranean from 1972 to 2016, providing another fine example of Mediterranean alien HAB occurrence in the basin. In this study, it is also mentioned that the highest abundances of *Ostreopsis* sp. in the Mediterranean were after 2005 and also co-occurring during the negative phase of the North Atlantic Oscillation (NAO) index. This phase is resulting in fewer and weaker winter storms that bring moist air into the Mediterranean and has been related to changes in phytoplankton biomass, primary production, and toxic algal blooms.

Introduction vectors

The Mediterranean Sea is a semi-rigid basin connected via navigational canals through the Suez Canal (since its opening in 1869) with the Red Sea, particularly in the Levant area, the Dardanelles waterway with the Sea of Marmara and coincidentally connects to the Black Sea, and also the Strait of Gibraltar with the Atlantic Ocean. Man-made canals such as the Suez Canal are the most potent mechanisms and corridors for invasions by marine species known in the world (Table 1) (Galil et al. 2016). Molecular methods demonstrate high levels of gene flow between the Red Sea, the Sea of Marmara (Black Sea), the Atlantic Ocean, and the Mediterranean populations (Otero et al. 2013). Shipping and generally invasions by transport vector has always been an introduction vector for invasive species, even since the thirteenth century via Viking ships (Lasota et al. 2016). More specifically, ships can transport microalgal alien species in ballast water (Otero et al. 2013; Katsanevakis et al. 2014a).

Another important vector of introduction is via aquaculture. The increasing market-driven demands for exotic fish and shellfish and the decline in wild fisheries have created the need of introduced species for aquaculture. Alien microalgae are usually co-introduced with imported shellfish, or a combination of vectors such as *O. ovata* that was first introduced from the Pacific to the Atlantic Ocean through shellfish aquaculture and then into the Mediterranean through the Strait of Gibraltar (Di Cioccio et al. 2014).

In the last decades, the expansion of warm water species has been speeded up by the increase of water temperature in the Mediterranean Sea that enhances the survival of newcomers (Parravicini et al. 2015), facilitating their establishment in certain areas and under favorable conditions their expansion and invasive behavior (Čalić et al. 2018).

Impacts

Since HABs in the Mediterranean are occurring more often and also intensifying, the risk on marine ecosystems and socioeconomy (ecosystem services) is higher. This is either due to the toxicity of the blooms (toxic HABs), or due to their ability to cause anoxic environmental conditions (high-biomass HABs). Most microalgal toxins can bioaccumulate in bivalves (filter-feeding) or other marine organisms and can affect via biomagnification not only humans but also wildlife. In many cases, these toxins can cause acute illness or death. HAB events can deem seafood consumption unsafe and decrease water quality in their occurrence range causing acute and severe, or prolonged and chronic impacts (Kudela et al. 2015). The recent update of the Mediterranean distribution, frequency and intensity of HAB species and events suggests an increase since the last decades (Table 3, HAEDAT 2019), making their impacts more extensive throughout the basin. Another possibility is that the suggested increase can be due to an increasing awareness and monitoring effort in the Mediterranean.

Because of the microalgal strain variety, it is challenging, on occasion, to determine which species are causing or could cause toxic HABs in the Mediterranean Sea. For example, C. monotis strains from the Mediterranean Sea have been shown not to produce toxins (Aligizaki and Nikolaidis 2008; Aligizaki et al. 2009), unlike strains from Australia that have been found to produce cooliatoxin (Amany 2014). P. shikokuense blooms fall into a similar category, since the Mediterranean blooms did not have any harmful effects, but they are known to be toxic in Japanese waters (Roselli et al. 2019). Furthermore, previously nontoxic strains have now been detected as toxic, like in the case of the Mediterranean strain of A. andersonii that has been found in Italy (Gulf of Naples) to produce a new PSP toxin and is raising concern about possible toxic events (Streftaris and Zenetos 2006).

Socioeconomy

Human health

The toxins produced by marine HAB species affect human health through seafood consumption or by exposure to aerosolized toxins. In general, acute intoxication impacts of algal toxins are studied to a greater extent and are better documented than the impacts that derive from chronic environmental exposure to lower concentrations of the released toxins. Since chronic exposure impacts are poorly studied, they remain an arising issue (Ferrante et al. 2013).

Some of the health symptoms of the different syndromes are gastrointestinal disorders like nausea, diarrhea and vomiting, and muscular and neurological conditions. Numbness of mouth and extremities can last for months in the case of CFP. PSP intoxication can manifest paralysis of chest muscles and the abdominal area that can also lead to death. Symptoms such as dizziness, headaches, disorientation, confusion, short-term memory loss, and motor deficiency are attributed to ASP. Most of these syndromes are well-documented, but with scientific progress, more issues are arising such as azaspiracid shellfish poisoning (AZP) that was first diagnosed in 1990s (Kudela et al. 2015). The most common symptom observed in the Mediterranean area derives from algal aerosolized toxins and causes respiratory irritation (Table 2). More specifically, in Genoa, Italy, in 2005, during *O. ovata* and *O. siemensis* blooms, 200 people exhibited exhalatory problems after coming into contact with contaminated sea spray (Tichadou et al. 2010).

Aquaculture

Toxic HAB species are on the rise in many parts of the Mediterranean Sea. The produced toxins have severe effects on aquaculture because commercially important species can accumulate high levels of toxins, becoming a worldwide food safety concern for humans, in some cases causing native species mortality and also leading to farm closure (Kacem et al. 2015). For example, in the case of *G. catenatum*, the toxins are released when *G. catenatum* cells are eaten by shellfish, such as oysters, mussels, and scallops, making them poisonous to consume (ISSG 2015).

Over the past 25 years, mussels farmed along the Mediterranean coasts, mostly in the North Adriatic, have been contaminated by harmful toxins, resulting in farm closure and severe economic losses. In most cases, marine lipophilic toxins (MLTs) were the source of mussel contamination. In 1995, the DSP-related yessotoxins (YTXs) became the main lipophilic biotoxins in Adriatic shellfish (Perini et al. 2018). Most recently, in Northern Greece, at Thermaikos Gulf, the species D. acuminata was mostly responsible for the DSP-related intoxications of Mytilus galloprovincialis and long harvest closures of mussel farms over the last 15 years (Koukaras and Nikolaidis 2004; Varkitzi et al. 2018). On the other hand, nontoxic HABs caused by species like C. bacteriastroides, in high biomass, bigger individuals with thicker spines, can damage other organisms' gills (Sunesen et al. 2008), thus killing native species or causing problems in aquacultured fish. Dinoflagellate blooms of O. ovata and C. monotis have also had repercussions on the economy by negatively impacting fisheries. During O. ovata blooms, shellfish and Arbacia sp. mortality has been detected on Marina di Massa reefs, and further studies have mentions of DSPlike and ciguatoxin-like toxin's presence (Streftaris and Zenetos 2006).

Recreation

Some HAB events can cause water discoloration that negatively can affect the esthetic value of coastlines of areas that are economically depending on tourism and recreation (Kudela et al. 2015). In the western Mediterranean (Balearic Island, Sicily, Catalan, and Italian west coast), an increase on the dinoflagellate *A. taylori* blooms has been detected over a period of 15 years in the early 2000s. Even if the occurring blooms are nontoxic, economic losses have been reported for the impacted regions. The occurrence of high-biomass HABs during summer months can prove economically catastrophic since the deterioration of water quality damages severely the touristic industry. Similar bloom events with water discoloration and water deterioration that have been observed in the Eastern part of the Mediterranean in 2004 are also alarming (Streftaris and Zenetos 2006). A study performed by Oliveri Conti et al. (2011), focusing on O. ovata blooms in the Italian coastline of Ionian Sea, states that there is no imminent threat towards coastline residents, recreation, or fisheries deriving from the presence of HABs in the reported study area. However, the presence of toxic HAB species detected could be of potential future concern. On the other hand, in a later study by Funari et al. (2015), guidelines are being suggested in Italy, to minimize O. ovata impacts concerning recreational activities. Accidental water digestion, consumption of contaminated food, and inhalation of aerosolized toxins are the main dangers that a person can face during recreational activities. The need to put guidelines in place to facilitate recreational activities is a strong indication of an already existing and increasingly threatening problem in the Mediterranean.

Environment

Especially the enclosed coastal areas are becoming more and more afflicted by the increase of harmful phytoplankton species. Eutrophication as well as the intentional or unintentional alteration of water circulation dynamics (dredging activities, navigational channels), the excessive use of coastal zones, and the facilitation for alien species distribution via ship ballast water contribute to the presence, frequency, and intensity of HABs making Mediterranean coastal ecosystems extremely susceptible to degradation (Armi et al. 2012). High-biomass HABs can cause ecosystem damage due to suffocation of the marine organisms via gill damage (C. bacteriastroides) or anoxic environmental conditions "dead zones" (A. taylori). Marine organism mortality can also be caused by toxin-producing HAB species (Kudela et al. 2015). For instance, the toxins produced by G. catenatum and Alexandrium spp. do not only accumulate in bivalves but also affect the bivalves' shell movement and feeding behaviors (Bricelj et al. 1996; Escobedo-Lozano et al. 2012). G. catenatum toxins that have been studied along the Iberian Peninsula during blooms are various fish species. Sardines and horse mackerels, planktivorous, and zooplanktivorous fish, respectively, that are eaten by top predators have been shown to transfer biotoxins up the food chain, affecting negatively an entire ecosystem (Costa et al. 2010). Next to bioaccumulation and biomagnification of toxins, also, the high biomass of G. catenatum toxic blooms have been shown to affect other marine organisms via negatively affecting their environmental living conditions (Katsanevakis et al. 2014a).

Discussion

Strength of evidence

Identifying and describing the origin and impacts of alien phytoplankton species in the Mediterranean Sea have been proven challenging. Although there is vast literature of alien phytoplankton species in European seas (Gómez 2008 and references therein; databases such as AquaNIS, DAISIE, and EASIN), Gómez (2008) suggested that there is usually no supporting research with valid data about alien species behind this literature. This is due to a decrease in taxonomic expertise, under investigation, under sampling, or an insignificant dispersal of species. All the above components contributed in the exceptionally low number of confirmed invasions concerning phytoplankton species in the Mediterranean Sea (Mozetič et al. 2017).

In an always expanding world, it is difficult to pinpoint all new invasions. Especially for microalgal species, it is difficult to identify their origin and vector of introduction because their taxonomic differences are not individually undetectable with a naked eye. Climate change and anthropogenic activities contribute to the fast alteration of species status from introduced to alien, to established, to invasive. Some scientists also use the term cryptogenic to attribute a status to a species that means that there are no sufficient data or knowledge to indicate whether this species is native or alien in a specific area. With biological invasions developing into a rising hazard for biodiversity and socioeconomy, more parties are getting interested in cataloging the issue. Thus, there are a lot of existing databases and scientific papers on international and European level concerning the Mediterranean invasions. Their presence can be very useful in order to organize information about the species involved; however, these databases tend to be incomplete and not up to date since bioinvasions are such a rapidly progressing problem. Microalgae are often excluded or lacking information in comparison with other taxa because they are hard to observe and to identify. Identifying a species' way of introduction is a highly difficult task due to the fact that some species have multiple possible ways of introduction, and the identification of the transport vector is considered to be based on the scientific knowledge of the researcher. Impact assessments are usually based on unclear data such as distribution of alien species. For species that can cause HABs, it is probably easier to identify their toxins and impacts in both socioeconomy and biodiversity. However, the severity of the impacts is up for debate for species of which their Mediterranean distribution is uncertain or lacking information. This problem exists because the more area a species occupies, the more its impact range is expanding (Katsanevakis and Moustakas 2018).

It is important to mention that this list is not complete concerning all monitoring efforts that have been done but were not published in journals or reports. The list only describes some of the most researched alien HAB-causing species in the Mediterranean Sea, and it demands further study to determine the missing information, which goes beyond the scope of this thesis.

Ways of establishment

Biological invasions are enabled the last decades due to the rapid globalization and the always increasing human pressure that derives from shipping trade, traveling, and transport (Katsanevakis et al. 2014a). Due to several key characteristics, alien algal species can colonize new environments more successfully than other organisms. Their ability to tolerate variations of environmental conditions, the lack of natural predators in their newly introduced environment in combination with being r-strategist, and opportunistic feeding patterns constitutes them challenging to regulate (Otero et al. 2013). For example, the extensive human pressure from fish and mussel (Mytilus galloprovincialis) aquaculture, overfishing, and inadequate sewage waste disposal that takes place in the Greek coastal areas can facilitate not only alien species establishment but also extensive bloom formation (Varkitzi et al. 2018). In general, in the Mediterranean basin, the presence of tropical alien species with high bloom formation can lead to the tropicalization of the basin, by altering its natural dynamics, especially in the southern parts (Coll et al. 2010).

What about the future?

Recognizing, assessing, and measuring the seascape-wide impacts of alien phytoplankton organisms are essential for successfully designing and implementing mitigation measures for the prevention of new invasions. The high increase of human population and activities in the last century especially in coastal and developing countries, alleviating the impact of HABs, is becoming more and more critical (Berdalet et al. 2015). The anthropogenic (exponential) need to exploit marine resources will result in acceleration of invasions and bloom formations of HAB species in the imminent future. Such needs can cause changes in temperature gradients, ocean acidification, light depletion, acute stratification, irregularities in nutrient dynamics, etc. The lack of sufficient knowledge of the processes induced by invasive alien HAB species works as a barrier that prevents predicting their future pervasiveness (Wells et al. 2015). Although forecasting the future possible impacts that derive from HAB presence is challenging under constantly altering parameters, such as species behavior and new introductions, current evidence suggests that their expanding distribution patterns and changes in algal community dynamics can distress previously unaffected areas in the Mediterranean and also in a global scale (Kudela et al. 2015).

The increase in artificial environments (ports, breakwaters, and semi-closed beaches) in highly populated coastal areas multiplied those habitats favorable for HAB species facilitating their establishment (Maso et al. 2003). The water discharges caused by anthropogenic activities in Mediterranean coastal areas could have an impact on the phytoplankton species composition, leading to an increase in the abundance of opportunistic species, like alien-introduced HAB species (Jenhani et al. 2019). Plastic pollution was suggested to also play a role in microalgal dispersal. Resting cysts of unidentified dinoflagellates and both temporary cysts and vegetative cells of A. taylori, Ostreopsis sp., and Coolia sp. were found on plastic debris (Maso et al. 2003). Thus, future research on the matter is suggested in order to mitigate alien microalgae dispersal in the Mediterranean. Another factor that facilitates not only microalgae establishment but also further dispersal and introduction in the basin is of course shipping. Shipping vessels are considered to have introduced one-fifth of the alien species found in the Mediterranean. The regionwide increase in shipping activities can directly be connected with biological invasions. By developing new trade patterns that can result in new shipping routes improve the water quality in port environments; managing ballast water, decreasing combining introduction vectors when is possible and cannot be avoided, and rising awareness and research possibilities, HAB Mediterranean events could be easier to monitor and mitigate (Coll et al. 2010).

Measures to reduce risk

As biological invasions are an expanding problem, and HABs in the Mediterranean Sea are increasing in frequency and dispersal, measures are needed to be taken in order to protect the biodiversity and the neighboring countries' populations from HAB events. Most legislations, initiatives, and projects are initiated by the EU but also from Mediterranean countries and researchers individually.

Efforts have been made worldwide to prevent and mitigate the negative effects of HABs in the last few decades. The most successful advances have been achieved through coordinated HAB monitoring and scientific research, integrated with end users and management through effective and informed policy decisions. Constant monitoring, modeling and prediction, prevention by efficiently improving coastal water quality, further research, and international coordination are the key factors in better understanding, predicting, and mitigating HABs, not only in the Mediterranean but worldwide. More specifically, measures to reduce risk could vary from observational approaches of water discoloration, to predetermined constant monitoring of smaller commercial vessel's ballast water. More recently, several technological approaches have been made for the early detection of HABs. Satellite remote sensing, empirical, and numerical models can be put in place, but also ferry box systems, automated water sampling equipment, etc. are tools that can be equipped on smaller commercial vessels to help monitor more closely HAB occurrences, since these untreated vessels play a big role in HAB species dispersal (Anderson et al. 2017).

Unfortunately, HABs cannot easily be eliminated or prevented, but they can be monitored and predicted, and their potentially negative impacts can be managed and mitigated. Changes in human activities and behavior could also contribute to prevent or minimize certain HABs and their effects (Kudela et al. 2015).

Since HABs are a natural phenomenon, it could be assumed that there are natural ways of mitigating them. Some organisms, such as the invasive copepod *Acartia tonsa*, have been shown to be able to change energy and matter flows between pelagic and benthic compartments, modify trophic structure of invaded ecosystems, and thus even serve as a potential biological control of algal blooms (Katsanevakis et al. 2014b). *Crepidula fornicata*, an invasive Mediterranean sea snail, is another example since its feeding activities may prevent blooms of harmful algae as well (Cebrian et al. 2012).

Current legislations

The Convention on Biological Biodiversity (CBD) understands the need for the "compilation and dissemination of information on alien species that threaten ecosystems, habitats, or species to be used in the context of any prevention, introduction and mitigation activities," and calls for "further research on the impact of alien invasive species on biological diversity" (CBD 2000). The objective set by Aichi Biodiversity Target 9 is that "by 2020, invasive alien species and pathways are identified and prioritized, priority species are controlled or eradicated, and measures are in place to manage pathways to prevent their introduction and establishment." This reflects Target 5 of the EU Biodiversity Strategy (EU 2011). The Marine Strategy Framework Directive (MSFD; EU 2008) identifies alien marine species as a major threat to European biodiversity and ecosystem health, compelling member states to establish strategies to achieve the "Good Marine Environmental Status" (Katsanevakis et al. 2014a). In 2004, the International Maritime Organization (IMO) released the International Convention for the Control and Management of Ship's Ballast Water and Sediments (BWM Convention 2004). This action supervises the management of ballast waters, as they are the primal vector for the dispersion of harmful marine organisms. This regulation is regarding all potentially harmful alien species, cryptogenic but also all impacting endemic marine species and pathogens (HAOP) in the entirety of marine environments around the world. Apart from the BMW Convention, the EU Marine Strategy Framework Directive (2008/56/EC) and the EU Regulation on the prevention and management of the introduction and spread

of NIS (Regulation (EU) n. 1143/2014) are also policies focusing on the oversight of endemic species (Roselli et al. 2019). The MSFD pursues an environmental perspective, by referring to the detrimental impacts that derive mostly from anthropogenic activities and result in marine water deterioration and eutrophication. A proper assessment of eutrophication involves associating data on algal species composition and biomass, but also HAB occurrence in accordance with nutrient levels availability. As a primary ecological indicator, phytoplankton is also one of the "Biological Quality Elements" (BQE) of the Water Framework Directive for the coastal water quality assessment (WFD 2000/60/EC) providing preliminary data for an area's environmental health assessment (Varkitzi et al. 2018).

Other worth mentioning initiatives include the Mediterranean Action Plan of UNEP where 21 Mediterranean countries are collaborating to meet the challenges of protecting the marine and coastal environment while boosting regional and national plans to achieve sustainable development, and the International Society for the Study of Harmful Algae (ISSHA).

Conclusion and recommendations

The Mediterranean Sea is undoubtedly highly impacted by alien invasions. Climate change that is causing the "tropicalization" of the Mediterranean and anthropogenic activities in coastal areas is degrading water quality, shifting temperature ranges and altering the nutrient ratio leading to ecological imbalance. These circumstances are allowing alien algal species not only to get established but also to produce high-biomass HABs. Such blooms are responsible for biodiversity and socioeconomic loss by impacting negatively marine organisms and human health.

This ecological imbalance in the Mediterranean not only provides favorable conditions for future algal introductions and bloom events but also is considered responsible for the increase of alien species. On the other hand, it is important to mention that scientific focus has shifted towards HABs in the last years because of their harmful nature, and that could also be a reason for the increase of the documentation of Mediterranean HABs.

No experimental or applied research has been found that considers all changing environmental conditions attributed to climate change that affect the increase of HABs. However, the increase of temperature in connection with the increase of HABs in every research examined is presented as a fact. Logically, that is the case, but this fact in the present review was deducted from the examined studies (e.g. Garcés et al. 2000). It is mentioned that alien-recorded HAB events seem to be increasing after 1980s (Maso and Garcés 2006). Without excluding the fact that more intense scientific research has also taken place, the other logical deduction is that since higher temperatures facilitate the expansion of alien algal species, their abundance is also increasing. Adding alien HAB events to already existing native ones is a way of facilitating on occasion their bloom cooccurrence, frequency, and density. By considering the above, the hypothesis has been verified and alien HABs are indeed an addition to an already existing situation, making it an actual problem.

For future research on changing biodiversity under changing environmental conditions, it is imperative to closely monitor alien algal species and to carry out mesocosm experiments in the field with natural water samples. Further knowledge is required in order to predict whether, where, and when changes should be expected in HAB frequency and density or reformations in community structure aiming to design efficiently mitigation and management measures. Primarily focus should be given on invasion pathway/vector management to minimize the risks of new introductions (Ojaveer et al. 2015); thus, monitoring programs should be always present in Mediterranean ports (Magaletti et al. 2018), which are common liable areas for alien and invasive species (Roselli et al. 2019).

The invasion of alien HAB species will continue to change the biodiversity of the Mediterranean Sea (Coll et al. 2010) and requires a more organized scientific approach, concerning mostly data that are scattered in various databases; update of existing data and the creation of one updated database that concerns Mediterranean alien microalgae species could prove useful in future scientific research.

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