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Toxic phytoplankton in eutrophic regional seas: An overview

Michael Karydis*
Laboratory of Environmental Quality and Geospatial Applications, Department of Marine Sciences, University of the Aegean, GR-81100 Mytilene, Lesvos, Greece
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*to whom all correspondence should be addressed: e-mail: mkar@aegean.gr
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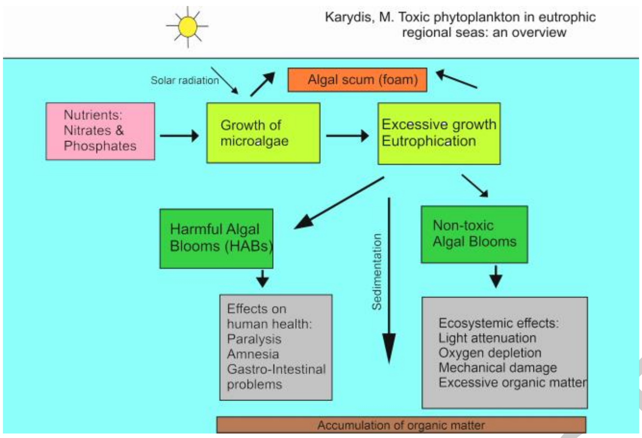
Graphical abstract



Abstract

Toxic algal blooms have become a major environmental problem over the last few decades because of their impact on fisheries, leisure activities, national income and human health. It is known that Harmful Algal Blooms (HABs) can occur naturally but the frequency of occurrence as well as their geographical distribution are alarming. HABs, beyond the scientific interest, have been an issue of concern for policy makers due to the high cost for implementing management practices. Unlike other marine environmental problems, the causes of HAB formation are not known so far with certainty and a high degree of uncertainty remains, regarding possible triggering mechanisms. Various factors, apart from nutrient concentrations, seem to be connected with this phenomenon: abundance, presence and absence of phytoplankton species, presence of grazers, weather conditions, seawater temperature and water mass circulation patterns, have already been reported in the scientific literature as potential factors. However, there are strong indications that eutrophic conditions play a paramount role in HABs formation. Machine learning methods, applied over the last few years to predict HAB's occurrences, have also confirmed the role of nutrients. In the present work, toxic algal blooms in regional seas characterized by eutrophic conditions that is the Mediterranean Sea, the Black Sea, the Baltic Sea, the North Sea, Wider Caribbean Region and the South China Sea are reviewed. Relevant issues including the drivers of eutrophication triggering HAB's events as well as effects on ecosystem services and socio-economic consequences are also considered.

Keywords: Eutrophication, HABs, marine environment, environmental protection, management, policy making

1. Introduction

It is known since a long time that Harmful Algal Blooms (HABs) can occur as a natural phenomenon (Prakash *et al.*, 1971). The term HABs refers to excessive algal growth of toxin producing microalgal species, causing negative impacts to other organisms. However, it is also possible that an algal bloom event can be dominated by non-toxic species. Even in that case, negative effects are recorded due to mechanical damage to other organisms or even impact at ecosystemic level, usually due to oxygen depletion, light attenuation and excessive production of dead organic matter after the end the bloom (Pinet 2016; Sanseverino *et al.*, 2016). However, over the last few decades the occurrence of HABs has increased regarding frequency, intensity and geographical distribution (Hallegraeff 2003; Gu *et al.*, 2022). These days, it is a matter of concern for government agencies, scientists, and the public; HABs incidences have received increased attention in the public media including press and electronic media. HABs cause serious health problems to humans, usually through the food (Anderson *et al.*, 2002). Increasing or decreasing trends in frequency and intensity do not follow a uniform, reasonable global pattern, in spite of increased observational efforts and modeling applications. In spite of the fact that check-lists of causative species are already available and the biological characteristics of many species are already known, HAB's events show different ecological characteristics and at the same time variable responses to environmental changes (Hallegraeff *et al.*, 2021). In addition, health and economic impacts due to toxic microalgae regarding seafood poisoning, death of finfish, aquaculture impact and tourism, differs between regions. This situation is getting worse as overexploitation of living marine resources functions as a natural multiplier of the impact of HABs (Hallegraeff *et al.*, 2002).

Toxic effects are observed when some organisms such as mussels, clams, oysters and scallops, known as filter-feeders, retain toxic microalgae. An interesting review article on toxicity aspects of the cyanobacterium *Microcystis* spp. by Chen *et al.*, gives a good account of toxicity issues on various animal groups. These effects have to be taken into account in HAB's management practices in the marine environment, although cyanobacteria are mainly responsible for HABs in freshwater systems and to a lesser extent in marine environments. Their toxins have effects on the liver (hepatotoxins), the kidneys (cytotoxins), the nervous system (neurotoxins) and the skin (dermotoxins) (Sanseverino *et al.* 2016). Accumulation of toxins due to the presence of algal species can be lethal to humans or even to marine animal species (Hallegraeff *et al.*, 2003; Anderson *et al.*, 2002). Although the shellfish can be marginally affected, the stored toxin can be ample to kill humans.

Beyond the toxic effects to humans, toxic algae cause damage to aquaculture operations and indirectly, they can have negative effects on marine ecosystems. High microalgal biomass results into significant light attenuation down the water column, shading submerged vegetation, eventually affecting the benthic biota. In addition, changes in irradiance can affect nutrient uptake, altering the competitive abilities of species according to Litchman *et al.* (2004). The authors working on a model on nutrient competition proposed by Droop (1974) have found that light dependent nutrient uptake may change competitive relationships under conditions of light fluctuation. These changes, although they seem to have an indirect effect, they have to be taken into account when toxic algal formation mechanisms are considered. The same authors suggested that fluctuations in light irradiance could be also applicable in competition between phytoplankton and bacteria.

The role of interactions between microalgae and bacteria in marine harmful algal blooms has already been reported (Gajardo *et al.*, 2023). It is known that heterotrophic bacteria surrounding algae and getting attached to algal cells, can uptake and catabolize nutrients and metabolites. This algae-bacteria association may be an additional causative factor for bloom formation as symbiotic or antagonistic interactions may contribute to the development or decline of algal blooms. Another algal-bacterial relation may be phagotrophy on bacteria by dominant mixotrophic dinoflagellates through phagocytosis, a phenomenon that needs further work in order to evaluate a possible role of phagotrophy in bloom formation. In addition, high biomass formation leads to foam and scum accumulation and when the bloom decays, oxidation of organic matter depletes the dissolved oxygen, causing mass mortality to fish and shellfish Pedde *et al.*, 2017)

A lot of descriptive information has been collected and published by marine biologists all over the world but the causes of mass phytoplankton outbursts so far, have not been sufficiently understood. Five possible explanations have been proposed: (a) relatively recent scientific interest and awareness of toxic species (b) extensive development of aquaculture facilities in coastal areas (c) stimulation of phytoplankton growth due to excessive supply of nutrients, mainly from terrestrial sources (d) climate change and (e) transportation of microalgae through the ballast water or translocation of shellfish stocks to new sites (Glibert, 2020; Gobler, 2020; Karydis *et al.*, 2013)

Among the factors mentioned above, cultural eutrophication seems to play a predominant role (Anderson *et al.*, 2002; Davidson *et al.*, 2014). Recent work (Tamvakis *et al.*, 2021), using machine learning techniques has shown the connection between nutrient concentrations and the stimulation of toxic phytoplankton genera. Nutrient increase in the marine environment due to human activites had a remarkable impact on toxic algal bloom formations. Phosphorus supply into the oceans has shown a 3-fold increase compared to pre-industrial levels, whereas nitrogen has increased even more over the last four decades (Caraco, 2004; Smil, 2004). Smayda (1989) using available databases worldwide came to the conclusion that the increase of some HAB's species in algal blooms was the result of coastal eutrophication. It has been observed that frequent outbursts of toxic phytoplankton species occured in regional seas that face excessive nutrient supplies from terrestrial sources such as areas in the Mediterranean Sea (Karydis and Kitsiou, 2012) or the Baltic Sea (Karydis and Kitsiou 2014). These blooms continue, in spite of measures adopted by the United Nations Regional Seas Programme that covers 13 regional seas and 149 countries that participate as signatory states. Several regional sea conventions have been adopted for the protection of the marine environment. Marine eutrophication and HABs formation are priority objectives in some of the international conventions (DiMento and Hickman, 2012).

In the present work, the connection between eutrophication and toxic algal bloom formations is examined. Furthermore, seven regional seas facing marine eutrophication problems (DiMento and Hickman, 2012; Karydis and Kitsiou, 2020) were selected as case study areas regarding toxic phytoplankton bloom phenomena. These regional seas were The Mediterranean Sea, the Black Sea, the Caspian Sea, the Baltic Sea, the North Sea, the Wider Caribbean Region and the South China Sea. The selection criteria were mainly based on the accessibility of scientific information, the severity of the impact and the relevant environmental policy.

2. Toxic algae and eutrophication

2.1. Principal groups of microalgae causing bloom formations

These days many thousands phytoplanktonic species have been identified in the marine environment (Sournia *et al.*, 1991). Nevertheless, it is only a limited number of microalgal species, about 300, reaching from time to time so high numbers that cause discoloration of the sea-surface, a phenomenon known as "red tides". Not all species associated with bloom formations are toxic. However, even the non-toxic species, under extreme conditions especially in sheltered bays, can form algal blooms so dense that often cause indiscriminate kills of invertebrates and fish, usually through oxygen depletion (Hallegraeff *et al.*, 2003). The number of species that have been found to be toxic is about 200 (Lundholm), most of them dinoflagellates and diatoms. There are also some toxic species belonging to the group of the "blue-green" algae (cyanobacteria), most of them being freshwater species and only a few live in the marine environment

The dinoflagellates, unicellular microalgae, contribute significantly to primary production but they are also responsible for blooms formations. They are characterized by a high degree of trophic adaptations: they can be autotrophic, heterotrophic or even mixotrophic. The dinoflagellates produce toxins that may affect public health upon consumption of contaminated food or exposure to HABs. Frequently occuring dinoflagellate toxic species belong to the genus *Dinophysis*. The toxic character of the species *D. acuta, D. fortii* and *D. norwegica* has been reported (Hallegraeff, 2003). The genus *Alexandrium* comprises of various toxic species. *Alexandrium* species, commonly reported in HAB events are *A. catenella, A. fundyense* and *A. tamarense* forming what is known as the *A. tamarense* complex (Janson and Hayes, 2008). Although toxin producing species of the genus Alexandrium have been studied extensively, it is possible that new taxa can still be found (Murray *et al.*, 2014). Despite the fact that a large number of Alexandrium species, namely 34, have been classified, only a subset of these species has been traced in ecological studies due to the limited resolution of morphological features and the workload required in ecological studies. In a recent work by Ding *et al.* (2023), performed metabarcoding of the 18S rDNA V4 region to ascertain both biodiversity and spatial-temporal dynamics of *Alexandrium* species in an area in Qingdao, China. The results showed that eight *Alexandrium* species were identified in this area, compared to only four species that had been reported until then. Metabarcoding analysis seems to be a promising tool for studying diversification and dynamics in toxic phytoplankton species. Encouraging results, including the presence of *Alexandrium*, using metabarcoding methodology in the Black Sea have also been reported (Dzhembekova *et al.*, 2020) as well as in the Gulf of Mexico (Gaonkar *et al.*, 2023). Information on *A. catenella*, using molecular techniques has also been provided (Yu *et al.*, 2021).

There are many toxic *Prorocentrum* species divided into two groups: (a) the benthic species (*P. lima, P. arenarium, P. maculosum* and *P. convacum*) and (b) the bentho-planktonic species (*P. micans, P. minimus, P. mexicanum* and *P. panamensis*). The species of the genus *Karenia* include *K. brevis* (Florida), *K. papilionacea, K. selliformis* and *K. bicuneiformis* (New Zealand). Several species of the genera *Amphidinium, Cohlodinium* and *Gyrodinium* have been identified as toxic.

The diatoms, unicellular photosynthetic organisms, contribute about 40% of primary productivity and 20% of the global carbon fixation. Diatoms are one of the largest algal groups. A number of species between 10,000 and 12,000 is accepted by many scientists, although their real number is probably much higher. In addition, they contribute to the ocean cycles of carbon, nitrogen, phosphorus and silica. However, some diatom species produce toxic compounds. Their effects on human health and their geographic distribution are presented in the next section. The diatom *Pseudo-nitzschia* encompasses many toxic species. The species *P. multiseries, P. pungens, P. pseudodelicatissima, P. australis, P. seriata* and *P. delicatul*a are known for their toxic effects

The cyanobacteria or "blue-green algae" is a diverse group of oxygenic photosynthetic bacteria. Their toxic effects have been studied since the 70's when it was documented their acute poisoning on domestic animals. Although cyanobacteria are widely spread in fresh waters, some species occur in marine ecosystems. Large populations of the cyanobacteria *Anabaena* and *Aphanizomenon* occur in brackish waters including the Baltic Sea. Various species of *Anabaena* are present in the Baltic Sea, the species *A. lemmezmannii* typically occur in the open sea, whereas *A. inequalis* is found in the near-shore zone where land-based sources of nutrients are remarkable. *Nodularia* is a genus of heterocyst-forming cyanobacterium. Some *Nodularia* species form extensive blooms, particularly in brackish waters. Three species of *Nodularia* have been reported for frequent bloom events: *N. baltica, N. litorea* and *N. spumigena*. *Trichodesmium* is a nitrogen fixing cyanobacterial genus, abundant in tropica oceans.

2.2. Effects of toxic algae on human health

The impact of HABs is not limited in the marine ecosystem but is also having a serious effect on human health through poisoning. Poisoning due to toxic phytoplankton can happen either by consuming food contaminated with toxins or through skin contact with contaminated seawater. Toxic symptoms can also appear by inhaling aerosolized biotoxins. Furthermore, fragments derived from cells disrupted by waves, as they move onshore, can have toxic effects. The toxic syndromes are classified into five categories according to health symptoms that can be amnesia, paralysis, gastrointestinal problems as well as respiratory irritation. The main clinical syptoms are presented in Table 1.

The toxins produced by marine phytoplankton can be acutely lethal. They can cause a wide range of acute symptoms to humans as well as to other species. The main problem with these toxins produced by phytoplankton is that they are tasteless, heat-resistant and acid-stable. Ordinary cooking methods, therefore do not protect consumers from intoxication once the food has been contaminated. In populations where seafood is the basic part of their diet and the toxic-borne diseases are endemic, the number of cases due to toxins derived from phytoplankton is high. The groups of people that is likely to be poisoned by marine algal toxins are: (a) workers along the production line dealing with harvesting, transportation, processing; (b) seafood consumers, usually in restaurants; (c) laboratory staff involved in sample collection and analytical work; (d) holiday makers spending their leisure time on or near the water and (e) indigenous populations mainly relying on seafood as their main diet.

Paralytic Shellfish Poisoning (PSP) is caused by consumption of molluscan shellfish contaminated by some dinoflagellate species (Table 1) producing toxic compounds known as saxitoxins. PSP is a life-threatening syndrome, the symptoms being purely neurological. Flagellates connected with PSP occur in both temperate and tropical waters. Before the 70's, PSP outbreaks were considered as an endemic phenomenon in North America, Europe and Japan. Later, PSP occurrences were also documented in South America (mainly Chile), Australia, SE Asia and India (Hallegraeff, 2003). These days PSP outbreaks have been observed in many more areas around the world.

The PSP toxin syndrome occurs after consumption of bivalve shellfishes like clams, mussels and scallops contaminated with saxitoxin that tends to accumulate in the hepatopangreas (digestive gland) of many filter-feeders. It has also been found that consumption of coral reef crabs and gastropods have caused paralytic poisoning (PSP). The main sources of PSP toxins are dinoflagellates of the genera *Gymnodinium, Alexandrium* and *Peridinium*. The biological activity of these toxins is connected with the blockage of nerves and skeletal muslce fibers. In addition to humans, other mammals, birds and fishes can be poisoned. The symptoms appearing often, are provided in Table 1. Human deaths in outbreaks in Europe and U.S.A. vary between 2 and 14 % of the infected people. The frequency of mortlaity is also depended on the availability of emergency hospital care in the area of outbreak events

Diarrheic Fish Poisoning (DFP) is a comparatively mild seafood poisoning. The symptoms are mainly gastrointestinal (Table1). DFP toxins have been identified and all of them are derivatives of the okadaic acid. Causative organisms are some dinoflagellates (*Dinophysis* spp., *Prorocentrum lima*), characterized by a widespread distribution mainly in Europe and Japan, but also in S. America, S. Africa, New Zealand, Australia and Thailand (Van Dolah, 2000). The symptoms of DSP are gastro-intestinal such as diarrhea, nausea, vomiting and abdominal pain. People infected usually recover completely within a few days. Although DSP poisoning is not life threatening, *Prorocentrum* spp. and *Dinophysis* spp. produce additional toxins that have been characterized a hepatotoxic and immuno-suppressive. There is also a risk factor for cancer in humans

Amnestic Shellfish Poisoning (ASP) is the only case of intoxication that is known to be caused by a diatom. It was first recorded during 1987 in Prince Edwards Island in Canada. Domoic acid, which is the toxic agent, was first identified in California during 1991 when pelicans and cormorants were poisoned by eating contaminated anchovies. Domoic acid is an excitatory neuro-transmitter. The causative microalga belongs to the genus *Pseudo-nitzschia* as many of its species are toxic. ASP symptoms include various neurological effects such as dizziness, discoloration, lethargy, seizures and short-term memory deficits (Table 1). Toxins produced by *Pseudo-nitzschia* species have shown a wide geographic distribution: Their presence has been recorded in New Zealand, Denmark, Spain, Scotland, Japan and Korea (Van Dolah, 2000)

Neurotoxic Shellfish poisoning (NSP) is a form of intoxication resulting from consumption of mussels contaminated with brevetoxins. As causative species, Karenia brevis and Gymnodinium brevis (both those species are marine dinoflagellates) seem to be connected with most of NSP outbreaks. NSP usual symptoms include nausea, severe muscular ache and loss of motor control (Table 1). NSP poisoning is not considered as a fatal intoxication in humans. NSP occurrences have been documented in the Gulf of Mexico and the west coast of Florida extending as far as North Carolina possibly due to warm Gulf Stream waters (Van Dolah, 2000). However, an unprecedented NSP outbreak was recorded in 1993 in New Zealand. The toxins causing NSP belong to a group of polyether compounds called brevetoxins. These toxins tend to bind to the receptor site of the sodium channel. The usual symptoms include temperature-sensation reversals, myalgia, vertigo and ataxia. Brevetoxins can also cause respiratory distress as well as eye irritation.

Ciguatera Fish Poisoning (CFP) is a seafood intoxication, frequently reported in various marine areas around the world (Friedman *et al.*, 2017). The symptoms are gastro-intestinal, often followed by a variety of neurologic symptoms, including muscle and joint aches, headache, itching, tachycardia, hypertension, blurred and paralysis (Van Dolah *et al.*, 2000). Ciguatera poisoning is rarely fatal. Humans contact CFP by consuming finfish containing a category of toxins known as ciguatoxins. Many commercial fish species have been identified as "ciguatoxic fish species". Among them, barracuda, snapper, grouper and jacks are particularly notorious for high ciguatera toxin loads. The causative microalga is the dinoflagellate *Gambiediscus* spp. This microalga is usually located as a bottom-dweller, sometimes attached to seaweeds, corals or even other substrates in shallow tropical and subtropical waters. Carnivores fishes associated with coral reefs are often a source of ciguatoxins. On the other hand, oceanic fishes derived from open seawaters are less susceptible to the accumulation of ciguatoxin. CFP poisoning is not prevented during food preparation and cooking of toxic fishes. The toxins are heat and acid stable; they are also stable for at least six months at commercial freezing temperature (Friedman *et al.*, 2017). Every year a number of cases between 50,000 and 500,000 is recorded globally. Chinain *et al.* (2021) have recently published an interesting review on ciguatera poisoning at a global scale. An account of CFP occurrences of French people living overseas, describing symptoms and clinical practices, has been given by the French Poison Control Centre Network (de Haro *et al.*, 2021); the report deals with 130 cases occurred from 2012 through 2019. Clinical studies (Gatti *et al.*, 2018) regarding CFP due to the gastropod Tectus niloticus characterized as a vector for a mass poisoning in the French Polynesia, describes clinical investigations as well as actions to alert local authorities about the potential risk of this gastropod

The geographic dispersion of microalgae causing ciguatera seems to expand between the 35° northern and the 35° southern latitudes. CFP events have mainly been recorded in the Caribbean Sea, the Indian Ocean and the Pacific Ocean (Wang *et al.*, 2022). A hypothesis has been supported that changes in sea-surface temperatures connected with El Niño events, often favor an increase of ciguatera cases in the Asian Pacific (Hales *et al.*, 1999). However, over the last two decades, ciguatera seems to become endemic even in previously unaffected areas such as Europe and the West African coasts. An explanation is connected with climate change: global warming favors the growth of dinoflagellates (Mattei *et al.*, 2014). An emerging threat of ciguatera poisoning in Europe is associated with fish caught in the Canary Islands (Spain) and Selvagens Islands (Portugal) (Godinho *et al.*, 2023). Godihno *et al.* (2023) are concerned that ciguatera's threats in the Canary and Selvagens Islands may be the "gateway to Europe". Ciguatera poisoning was also reported along the West Africa coast (Boada *et al.*, 2010). These findings suggest that Atlantic endemic regions are emerging (Solino and Costa 2020). Some coastal areas in the Mediterranean Sea are also emerging as ciguatera "spots". Ciguatoxin type of substances have been identified in edible fish on the Eastern Mediterranean since the year 2007 (Bentur *et al.*, 2017). Table 1 shows some toxic or potentially toxic algal species connected with CFP.

Table 1. Examples of frequenty occurring toxic algae, their toxins and toxic syndrome

Harmful algal species	Toxins produced (1)	References	Toxic syndromes (2)
	(a) Paralytic shellfish poisoning (PSP)		
Alexandrium spp.	Saxitoxins and derivatives	(Berdalet *et al.*, 2016)	Nausea, vomiting, diarrhea, numbness of lips, mouth, face and neck
A. tamarense		(Band-Schmidt *et al.*, 2019)	
Pyrodinium bahamense		(Band-Schmidt *et al.*, 2019)	
Gymnodinium catenatum		(Anderson *et al.*, 1989)	
	(b) Diarrheic shellfish poisoning (DSP)		
Dinophysis acuminata	Okadaic acid and derivatives (dinophysistoxins)	(Kat, 1983)	Nausea, severe diarrhea, vomiting, abdominal cramps, respiratory distress
D. caudata		(Krock *et al.*, 2003)	
Prorocentrum lima		(Berdalet *et al.*, 2016)	
	(c) Amnesic Shellfish Poisoning (ASP)		
Nitzschia pungens	Domoic acid and its isomers	(Bates *et al.*, 1989)	Nausea, vomiting, headache, diarrhea, confusion, dizziness, memory deficits
Pseudonitzschia spp.		(Tas *et al.*, 2017)	
P. calliantha		(Caroppo *et al.*, 2005)	
P. delicatissima		(Zingone *et al.*, 2021)	
	(d) Neurotoxic Shellfish Poisoning (NSP)		
Karenia brevis	Brevetoxins	(Berdalet *et al.*, 2016)	Nausea, muscle weakness, vertigo, parenthesias of the mouth, lips, tongue, dizziness
Gymnodinium breve		(Ibrahim, 2007)	
	(e) Ciguatera Fish Poisoning (CFP)		
Gambierdiscus spp.	Ciguatoxin,Maitotoxin	(Friedman *et al.*, 2000)	Nausea, vomiting, diarrhea, numbness of mouth, neurological symptoms
G. toxicus		(Ibrahim, 2007)	
Ostreopsis siamensis		(Gu *et al.*, 2022)	
Coolia monotis		(Abdennadher *et al.*, 2020)	
Amphidinium carterae		(Yang *et al.*, 2023)	

Sources: (1) Toxins produced: Berdalet et al. (2016), (2) Toxic syndromes Friedman et al. (2017); Berdalet et al. (2016).

2.3. The nature of eutrophication

Excessive nutrient concentrations in coastal areas and regional seas enhance the growth of phytoplankton, a phenomenon known as marine eutrophication (Karydis and Kitsiou, 2020). It looks like a simple phenomenon but in fact it is a complex ecosystemic procedure. This has led working groups of scientists as well as policy makers to set up a number of definitions, depending on the point of view that eutrophication is considered. One of the first definitions given by Steele (1974) is fairly short and simple: "*Eutrophication is the increase of growth rate of the algae, following a faster rate of nutrient increase in the marine environment as well as the consequences*". Other authors (Gray, 1992; Nixon, 1995) as well as international organizations (OSPAR) and legal authorities (Clean Water Act PL 92-500/1992, the European Union Directive: "Urban Waste Water Treatment 1991") have also proposed definitions on eutrophication. The definitions differ as far as the causes and/or effects of eutrophication that are used to formulate the definitions. For example Nixon (1995) defines eutrophication as an increase in the rate of supply of organic matter to an ecosystem. Nixon avoids to mix causes and effects and is limited on the outcome (effects) which is the production of biomass. On the contrary Vollenweider's definition, mentioned above, had emphasized the causative factor which is nutrient supply to the ecosystem, inducing higher growth rates. Any definition of eutrophication is not just expressing a different scientific point of view but also leaves substantial room for interpretation in a court of law. This is possibly the reason that national and international organizations provide definition on eutrophication within their legal weaponry.

The task group of eutrophication, having worked within the framework of the European Marine Strategy Framework Directive (MSFD), proposed a definition of eutrophication after considering eutrophication definitions in the scientific literature as well as the need for providing guidance for developing descriptors. This task group (Ferreira *et al.*, 2011) proposed the following definitions for eutrophication: "*Eutrophication is a process driven by enrichment of water by nutrients, especially compounds of nitrogen and/or phosphorus, leading to: increased growth, primary production and biomass of algae; changes in the balance of organisms; and water quality degradation. The consequences of eutrophication are undesirable if they appreciably degrade ecosystem health and/or the sustainable provision of goods and services".* More information on definitions of eutrophication having adopted specific aspects of this phenomenon can be found in published work (Ferreira *et al.*, 2011; Kitsiou and Karydis, 2011).

The objective of the MSFD group was to propose quality descriptors relevant to marine ecosystem quality. Three points were taken into account by the group of experts (Ferreira *et al.*, 2011): (a) eutrophication is mainly caused by nutrient enrichments from terrestrial sources as sewage outfalls, agricultural fertilizers and some types of industrial effluents that tend to modify the marine ecosystem. Additional nutrient sources are connected with deforestation and the release of nitrogen oxides in the marine environment. Loss of "pristine conditions" in the marine environment is an issue of major concern. (b) in wider marine areas such as regional seas, spatial variability due to water mass circulation and the topography of the area should be taken into account , the third point; (c) is that nutrient enrichment can occur through natural processes that function independently of nutrient supply due to anthropogenic activities. It is therefore important to discriminate between "autochthonous eutrophication" and eutrophication from human activities that is "cultural eutrophication".

2.4. Eutrophication assessment: a thorny issue

Eutrophication may sometimes be due to natural processes as the main natural sources may be coastal upwelling, sediment resuspension (as the result of the circulation of water masses) as well as nutrient deposition from the atmosphere (UNEP, 2003). River waters enriched with nutrients end up in the marine environment or adsorbed by sediment particles. To all these processes, nutrient and phytoplankton seasonality has to be taken into account. The complexity of the natural processes makes the discrimination between autochthonous eutrophication and cultural eutrophication a "thorny issue" in assessing eutrophication.

Cultural eutrophication has been identified as a serious environmental issue in some seas since the early '80s (Gray, 1992; NRC, 2000) but nowadays it tends to be a worldwide problem. Although the final effects of eutrophication can be detrimental for the marine ecosystems, the environmental impact in the sea is getting built-up in a stepwise manner. Gray (1992) has identified five levels of marine eutrophication. At the initial stage (known as the enrichment stage), elevated nutrient concentrations enhance algal, benthic and fish growth. Furthermore, (second stage) nutrient enrichments change the structure of phytoplankton community. These impacts are known as "primary effects". The secondary effects (stage three) includes serious underwater light attenuation down the water column, behavioral changes in animal life and frequent formation of algal blooms. Further deterioration (extreme effects) that is the fourth stage is characterized by toxic effects and species mortality. In addition, mass growth of some macroalgal species such as species of *Ulva* and *Cladophora* can often occur. The final stage (ultimate effects) is characterized by anaerobic conditions and extinction of most of the species. This five-step scale, introduced by Gray forms a semi-quantitative way for assessing eutrophic levels.

As the impact of eutrophication on ecosystem's health and the economy depends on the severity of the problem, quantitative scales characterizing eutrophic conditions have been developed. These scales are useful for water quality assessment as well as for the management of coastal waters. Many different scaling procedures have been proposed. They are based either on causative variables (nutrient concentrations for example) or on effect variables (variables expressing biomass increase) These eutrophication scaling systems include nutrient concentrations (Ignatiades *et al.*, 1992), chlorophyll concentrations (Giovanardi and Tromellini, 1992), productivity values (Ignatiades, 2005) and ecological indices (Karydis and Tsirtsis, 1996). A wider range of bioindicator species and environmental indicators can also be found in review articles (Kitsiou and Karydis 2011; Karydis, 2009). However simple a scheme for outlining levels of eutrophication can be, there are built-in problems (Kitsiou and Karydis, 2011; Karydis, 2022). Seasonal variations of nutrient concentrations, species succession and difficulties in discriminating water types from the trophic point of view in the natural environment due to variable overlapping (Giovanardi, 1992) are the main difficulties in setting up ranges for each level of eutrophication. The overlapping of variables expressing typical oligotrophic, mesotrophic and eutrophic conditions in the proposed scales of eutrophication is the most common problem. An additional problem is the definition of reference conditions, used for comparisons between water masses to be accessed and the baseline concentrations regarding the parameters used in eutrophication assessments. By the term "pristine states" is meant unimpacted sites characterized by "*insignificant impact of pressures on ecosystem functioning and thus an approximation of the natural environment*" (Karydis and Kitsiou, 2020). In addition, pristine waters are not easy to be found these days and it is rather unrealistic when semi-enclosed regional seas are under examination. The need to find pristine waters for research purposes usually arises when pollution problems in an area are already established (Nielsen *et al.*, 2007). The situation is further complicated in shallow coastal waters where autochthonous eutrophication occurs due to sediment resuspension. This complex situation increases the degree of difficulty to understand relationships between toxic algal bloom outbursts and marine eutrophication, as the degree of uncertainty is getting remarkable.

2.5. Effects of toxic algae on human health

In spite of the well-established belief that HAB's formation is closely connected with anthropogenic nutrient enrichment, there are some doubts regarding this relationship (Gowen *et al.*, 2012): "*given the widespread enrichment of many coastal regions of the world and the putative global increase in HABs, it is not difficult to see why anthropogenic nutrient enrichment of coastal waters is thought by some to be one of the main drivers for the apparent global increase of HABs. The issue of whether anthropogenic nutrient enrichment has caused or influenced the occurrence, frequency of occurrence and spatial and temporal extent of HABs and HAB species is complex, and the nutrient enrichment HAB hypothesis has been widely debated in the Scientific Literature by among others, Hallegraeff (1993), Richardson (1997), Smayda (2008), Anderson etal.,* (*2002, 2008*) *and Sellner et al. (2003)* " According to Richardson (1997), the fact that HABs are not new phenomena, supports the view that nutrient enrichments of anthropogenic origin are not a prerequisite for their occurrence. An example is the presence of *Karenia mikimotoi* blooms in the NE Atlantic coastal waters where no apparent links to anthropogenic activities have been observed so far Davidson *et al.*, (2009).

However, most researchers, like Anderson in his 1989 paper (Anderson 1989), hold that "*it is now firmly established that there is a direct correlation between the number of red tides and the extent of coastal pollution*". Konovanova (1989) who supports a similar opinion has also expressed the view: "*undoubtfully, the frequency and concentration of red tides is directly connected with increased eutrophication of coastal waters under the influence of anthropogenic factors*". Smayda (1989) accepted as a fact that "*the implicit concept of an anthropogenic trigger seems to be the favored notion*" although he was not quite ready to embrace this view. The reservations referring to connections between red tides and eutrophication may be partly due to the fact the red tide formation is a complex process with adaptation mechanisms by some cyanobacterian species and dinoflagellates where their dominance involves many factors; these include temperature, circulation, competition between species of phytoplankton and grazing rates.

The potential of cyanobacteria to utilize atmospheric nitrogen, practically an unlimited source, and convert it into bioavailable nitrogen, is influencing widely accepted views regarding nutrient-phytoplankton dynamics. In regional seas like the Baltic Sea, where international efforts aiming at reducing nitrogen loads, fixation of atmospheric nitrogen by nitrogen fixing cyanobacteria, (often toxic like *Nodularia spumigena*, *Dolichospermum* spp. and *Aphanizomenon* spp.) (Barker *et al.*, 2000) can affect nutrient balances. The fixed nitrogen in the Baltic Sea is a major contributor to the overall nutrient budget. It has been estimated that the fixed atmospheric nitrogen accounts for 20 to 50 percent of the new nitrogen (Munkes *et al.*, 2021).

Later, Smayda reckoned that frequency and dynamics of phytoplankton blooms seem to be connected with nutrient enrichments at a global scale. Another factor favoring bloom formations that has not received enough attention in HABs is connected with the Redfield Ratio. Some species can exploit specific environmental conditions, especially when nutrients are not in what is known as the balanced Redfield proportion. These species, under favorable conditions, can become even more toxic (Glibert *et al.*, 2017). Another adaptation mechanism is based on the grazing of dinoflagellates on picoplankton. Laboratory experiments showed that when *Karenia brevis* is grazing on *Synechococcus*, the growth rate of *K. brevis* is enhanced, a fact that can furthermore sustain blooms (Glibert *et al.*, 2009). Smayda (2010) has also published work on adaptations of dinoflagellate species. A number of 27 dinoflagellate species that bloom in upwelling systems, showed morphological, physiological, toxicological and ecological adaptations. Adaptive strategies of dinoflagellates in the Baltic Sea based on allelopathy have drawn the attention of researchers (Suikkanen *et al.*, 2011). The reason was that although Baltic waters were usually dominated by diatom blooms, dinoflagellate-dominated bloom spring events showed high frequency in the Baltic over the last years. Recent studies on the trophic functionalities of dinoflagellates have further elucidated the selectivity of their mixotrophic mechanisms (Garcia-Oliva and Wirtz, 2022) as well as their interaction mechanisms with other species (symbiosis or parasitism) (Bartual *et al.*, 2022). The information collected so far suggests that the continuous changes of the physicochemical conditions driven by human pressures induce adaptations in phytoplankton community in a decadal timescale.

Work by Hodgkiss and Ho (1997) on changes of N:P ratios in coastal areas indicated that nutrient ratios were the key to understand the increasing number of phytoplankton bloom incidences. These authors specifically expressed the view that "*significant changes in phytoplankton species occurrences, biomass and productivity, as well as shifts in predominance, occurring in regions as far apart as the North Sea and Hong Kong support the hypothesis that phytoplankton blooms are increasing in coastal waters on a global scale and they are linked to long term increases in coastal nutrient levels*". Hodgkiss continued working (Hodgkiss and Ho 1997) using nutrient ratios and in a more recent work (Hodgkiss and Lu 2004) confirmed the role of nutrient ratios in harmful algal bloom formations. At the same period, it was observed (Glibert *et al.*, 2005) that increased use of fertilizers in China coincided with a 20fold increase, regarding the occurrence of algal blooms. Similar patterns were found by the same authors in the United States:"*a relationship between increased nutrient loading from the Mississippi River to the Louisiana Shelf and increase abundance of the toxic diatom Pseudo-nitzschia pseudodelicatissima has been dominated*". A "roundtable discussion" organized by the U.S. Environmental Protection Agency (Heisler *et al.*, 2008), finally, unanimously adopted seven statements connecting eutrophication to harmful algal blooms. Through these statements, they accepted the fact that increased nutrient loading promotes HAB's formation. It was further stated that continuous nutrient supply is required for the algal blooms to be sustained; this nutrient delivery can be either chronic or episodic. It is interesting that the role of nutrients in HAB's formation was confirmed in three Large Marine Ecosystems (LMEs) bordering China (Wang *et al.*, 2021). It was observed that nitrogen loading was increasing much faster than phosphorus. The authors found that the critical threshold in the N:P molar ratio was the number 25. Once the N:P ratio was above this limit, something that was observed during the 80's, changes in ecosystems' functioning were observed, namely increased primary productivity, loss of biodiversity and more frequent occurrence of Harmful Algal Blooms.

2.6. Understanding and predicting HABs: the machine learning approach

It is well known by now that HAB's formation depends on both biotic and abiotic variables (Loureiro *et al.*, 2011). Abiotic variables include salinity, temperature, inorganic nitrogen and phosphorus concentrations. Among the biotic variables, the presence of zooplankton, bacterial cell number, phytoplankton abundance and species interactions seem to be the more prominent (Hallegraeff *et al.*, 2003). A promising data analysis methodology, known as machine learning, is being used over the last few years with increasing intensity with main objective the prediction of algal blooms (Cruz *et al.*, 2021). The number of publications of machine learning methodology in connection with algal blooms increases exponentially: according to the Web of Science, the annual number of publications during the period 2006-2019 is just about 3 to 4 papers per year, whereas this number exceeds 25 publications per year since 2020.

Machine learning (ML) is a branch of artificial intelligence that uses data to improve computer's performance. The general idea is to use part of a dataset as "training data" for the computer to make predictions without having been programmed to do so. ML methodology is a collection of algorithms of different types. These algorithms include Decision trees, Multinomial logistic regression, Forest random decision trees and Randomization classifiers, just to mention a few. More information on the algorithms used for prediction of HABs can be found in a recent review article by Cruz *et al.* (2021).

Work (Tamvakis *et al.*, 2021) using a number of machine learning techniques has been applied using a number of abiotic variables: temperature, salinity and nutrients as input variables and the presence/ absence of 18 genera of phytoplankton, characterized as potentially harmful algae by IOC-UNESCO (2021), as output variables. Nutrients seemed to play a significant role in HAB's formation, especially the phosphates. The role of nutrients in general, was confirmed by most of the machine learning methods used in this particular work. Correlation between the presence of algal toxins and nutrient concentrations was also confirmed in aquaculture facilities in the NE Atlantic area (Fernandes-Salvador *et al.*, 2021). Furthermore, application of the Dense Forest technique, showed that incorporation of nutrient fluxes of terrestrial origin, improved model performance supporting the view about the role of nutrients in HABs events (Cheng *et al.*, 2021).

3. Sources of nutrients and toxic algal blooms worldwide: an outline

3.1. Wastewater

Anthropogenic activities in coastal areas stimulated a massive increase of nutrient loading in the coastal marine environment. Among the nutrient sources enriching coastal waters, industrial waste, fertilizers, aquaculture facilities and run-off from agricultural land, contribute to phytoplankton growth and consequently to increase the frequency of algal blooms events. However, the most important source of nutrients, especially nitrogen and phosphorus, is wastewater. Estimates have been recorded in literature regarding inputs of sewage in U.S. areas. The contribution of nitrogen from wastewater to Long Island Sound is as high as 67% of the total nitrogen inputs (Nixon *et al.*, 1983). Similar estimates in Narragansett Bay, showed that sewage contribution accounted for 41% of the total nitrogen; sewage contribution in the South San Francisco was almost 100%. Studies on the relation between nutrient coastal enrichment and algal blooms (Howarth *et al.*, 2008), using a model (the SPARROW model), indicated that the contribution of wastewater in nine estuaries and bays in the U.S., ranges between 8 and 63% with an average value 34.1%, whereas the average contribution of atmospheric depositions was 19.5 and 30.6% respectively. These results indicate the primary role of sewage derived nutrients in HABs formation.

A nutrient source of primary importance is also the Submarine Ground Water (SGD). Relatively recent studies in Daya Bay, in China (Wang *et al.*, 2018) on SGD have shown that the N:P ratio increased to 37.0, changing therefore the nutrient regime of the coastal waters. Regarding primary productivity, the authors evaluated that SGD accounted for 30% of the total production. These findings indicated the primary role of SGD as a key source of nitrogen and phosphorus in coastal waters. Although this N:P ratio of 37.0 is generally considered as a rather high value, it has been supported by Klausmeier *et al.* (2004) that the Redfield ratio of 16 is not a universal optimal value but is rather expressing an average. The authors working on Droop's model, found that the optimal N:P ratios can range between 8.2 and 45.0, depending on ecological conditions. In addition, physiological studies have shown that the N:P ratio seems to be species specific. The latter is important when growth conditions of toxic algal species are considered. Research on the assessment of optimal N: P ratio of the most common bloom forming species could provide valuable information towards methodologies towards the prediction of algal blooms, a relatively recent issue of ecological and economic importance (Davidson *et al.*, 2016; van den Bergh *et al.*, 2002).

3.2. Atmospheric deposition

Airborne nutrient transportation and deposition is one more physical mechanism connected with nutrient eutrophication and HABs formation (Karydis and kitsiou 2012; Herut *et al.*, 1999). Nitrogen in the form of nitrates and ammonium is present in rainwater, whereas the gaseous forms of nitrogen are N_2, NH_3 and HNO_3. Measurements of NO_x, NH_3 and total emissions in the Mediterranean Sea, have been carried out during the year 1999: the quantities were approximating 1,800, 2,300 and 4,200 kt respectively (Tarrason *et al.*, 2000). Phosphorus transportation in the marine environment is limited due to low solubility (about 30%) of the available phosphorus (Bergametti *et al.*, 1992). Globally, the use of nitrogen in fertilizers outpaces by far phosphorus as a fertilizer (Heisle *et al.*, 2008). A significant proportion of nitrogen, mainly in the form of ammonia, is deposited in the marine environment through atmospheric transportation (Glibert and Burford, 2017). Nitrogen favors the growth of phytoplankton species, some of them being dominant in HAB's formation

Work in the East China Sea (Mackey *et al.*, 2017), has provided direct experimental evidence that atmospheric deposition increases the N:P ratio, opening new niches for bloom forming diatoms and dinoflagellates. The role of atmospheric deposition has been indicated by Paerl (1997); the author maintains that direct atmospheric deposition of nitrogen may impact HAB's dynamics of coastal and pelagic waters. However, later work by Carstensen (2005) concluded that direct nitrogen inputs through atmospheric deposition in the Kattegat area could not be linked with any bloom events. In a recent paper, dealing with nitrogen atmospheric deposition in the southwestern part of Florida, the authors expressed the opinion that although direct atmospheric deposition of nitrogen is not likely to initiate HABs, it is possible to sustain existing blooms (Barcan *et al.*, 2023). Blooms in the Yellow Sea of China have shown an increase in frequency of occurrence over the last decades; this fact has been attributed to atmospheric deposition in addition to direct nutrient runoff (Zhang *et al.*, 1994)

3.3. Fertilizers

The effects of fertilizers for the formation of algal blooms has also been an issue of concern (Chakraborty *et al.*, 2017; Fisher *et al.*, 2016). The quantitative relation between fertilizers and HAB's formation has been studied using mathematical modeling (Chakraborty *et al.*, 2017). Three factors were taken into account: input of fertilizers from agricultural activities, other sources of eutrophication apart from fertilizers and overfishing. The model indicated that rapid algal growth was closely connected with increasing use of fertilizers. Recently, Hou *et al.*, (2022) have carried out similar work: in addition to nitrogen and phosphorus contribution, different environmental stressors such as climate change and different land management practices were included in the model. The model indicated that between the two main nutrients, nitrogen and phosphorus, phosphorus concentrations were found more effective in reducing HABs risks. This finding can be helpful for managers and policy makers as mitigation measures for the use of phosphorus may reduce the frequency of HABs formations. Urea has attracted the interest of researchers as a potential factor in HABs formation (Fisher *et al.*, 2016). The main anthropogenic source of urea is in the form of nitrogen fertilizers, accounting for 60% of the world's consumption of nitrogen fertilizers. In addition, urea can be found in many herbicides and pesticides used in agriculture. It is known that some algal species using urea, are prone in causing toxic algal blooms (Camargo and Alonso 2006)

3.4. Overfishing

Research on the connection of HABs formation with overfishing has received very little attention. It has been reported by Vasas *et al.* (2011) that overfishing of planktivorus fishes is acting synergistically with nutrient enrichments, promoting HAB species and therefore leading to HAB's formations. Other authors (Walsh *et al.*, 2011) maintain that the top down marine ecosystem control from piscivorus fishes to phytoplankton has been reversed; overfishing of piscivorus species and the subsequent growth of jellyfish biomass, fed on copepods, has allowed the expansion of phytoplankton, favoring the formation of HABs. Maximal landings of sardine *Sardinops sagax* within the Benguela current has been reported for the year 1965 (Grindley and Nel 1970). The response to this overfishing was a population explosion of the red alga *Gonyaulax catenella* that caused serious health problems to humans by contamination of shellfish contaminated by saxitoxin (Cerrato *et al.*, 2004). As a management practice to reduce HAB formations, it has been proposed that bivalve restoration could combat algal blooms (Cerrato *et al.*, 2004). A summary of the nutrient sources connected with HABs formation and selected literature is provided in Table 2.

Table 2. Nutrient sources connected with HABs formations

Nutrient Source	Short description	References
Wastewater	Enormous quantities of sewage inputs, treated or untreated, increase nutrient concentrations in the coastal zone, boosting algal blooms.	(Howarth *et al.*, 1996; Boynton *et al.*, 1995; Nixon and Pilson 1983; Wang *et al.*, 2018)
Atmospheric deposition	Atmospheric deposistion favors nitrogen forms, especially ammonium and urea. Increased N/P ratios provide advantages to some species with a dominant role in HABs.	(Herut *et al.*, 1999; Mackey *et al.*, 2017; Carstensen *et al.*, 2005)
Fertilizers	Fertilizers are among the primer cause in HABs formation. Modelling ecosystemic processes has confirmed the role of N and P. Mitigation practice are recommended, especially decrease of P as fertilizer.	(Chakraborty *et al.*, 2017; Fisher *et al.*, 2016; Camargo *et al.*, 2006)

4. Regional seas: a review on toxic phytoplankton

4.1. Environmental policy and governance in the regional seas

The term "regional seas" has been adopted by the United Nations during the UN Stockholm Conference on the Human Environment in 1972 (Mead, n.d.). The Stockholm Conference also gave birth to United Nations Environmental Programmes (UNEP); UNEP's Governing Council designated 18 seas as one of the organization's priorities and led to the development of the Regional Seas Programme (RSP). Since then the RSP is UNEP's most important regional mechanism for the protection of the marine environment (UNEP, 2023). The Programme encompasses 18 regional seas; in 13 out of the 18 regional seas, their environmental governance is administered by UNEP. There are Action Plans, which are the executive arm of UNEP. This Programme has been signed by 149 states (95% of the World's States). It is the biggest regional seas Programme for marine environmental protection; programme's objectives, *inter alia*, include guidelines and actions for the control of marine pollution and for the protection of aquatic resources. The impact of pollution on human health, marine ecosystems, amenities as well as management of marine and coastal resources are also within the Programmes' objectives (Kaniaru 2000). The first International Convention for marine environmental protection within the UNEP framework was signed for the Mediterranean Sea in 1974. The BARCON Convention that is the Barcelona Convention formed the "model" for a number of similar agreements signed for the regional seas that are mentioned in the present work (Karydis and Kitsiou 2014; DiMento *et al.*, 2012).

The strategies to be used for the protection and development of the regional seas, according to the decision of the Governing Council of UNEP in 1978, include guidelines and actions for the control of marine pollution, protection of aquatic resources, environmental impact on human health, ecosystems' health and protection of amenities. It is obvious that all the objectives mentioned above are directly or indirectly connected with toxic algal formations (DiMento *et al.*, 2012)

The key role in marine environmental governance is the effectiveness. The effectiveness can be addressed in practical terms by adopting appropriate criteria (DiMento, 2003; Miles *et al.*, 2001; Boehmer-Christiansen and Kellow 2002; Oberthur, 2006). In simple terms the effectiveness "*denotes a solution of an environmental problem that brought together international actors to confront it*" (DiMento *et al.*, 2012). Effectiveness can be realized by combining institutional output with expert's advice. The knowledge of physical parameters is indispensable together with economic and political relations. However, the problem according to Kirk (2008) is that "*the states at times rely on imperfect knowledge and information when attempting to meet their obligations and consequently, compliance is not always achieved even when states have employed their best efforts to do so*". There is always a degree of uncertainty about the nature and severity of the problem. This is the case regarding HABs. As HAB's formation is an unexpected event in space and time, relevant field information may fail to be collected. Even if a monitoring program is in progress, which is the rule for most of the regional seas as part of their action plans, HABs phenomena might take place at a much shorter time-scale that is during the time interval between two successive field surveys. In addition, even when data are available, data interpretation is not always possible or easy and therefore, lack of understanding makes any measures to be proposed rather doubtful regarding their effectiveness.

Within these constraints, the problem of HABs will be reviewed in the seven regional seas already mentioned. On the other hand, there is sufficient published work for marine eutrophication and toxic algae in the selected regional seas. In these seas, HAB formation seems to be a serious ecological and socio-economic problem closely connected with eutrophication. This is the reason that toxic algal bloom formations should be considered within a framework that includes the physiography of the regional sea, environmental pressures, water quality and marine policy. This is the approach that has been followed in the present work

The criteria used for the selection of the case study areas were: (a) accessibility of relevant scientific information and conditions (water quality and ecosystem's health) of the sea (b) sources and causes of environmental impacts (c) information on the governance schemes (d) geographical dispersion of the case study areas (e) eutrophication status (f) information on toxic algal events (g) policy schemes: international conventions – Strategy - Action Plans – Proposed solutions – Assessments

4.2. The Mediterranean Sea

4.2.1. Physiography and environmental pressures

The Mediterranean Sea is an evaporation basin, which means that evaporation rates are higher than the rates of precipitation (rain, snow, hale) and river inflow (Conovaro, 2013). This semi-enclosed basin is exchanging water masses with the Atlantic Ocean through the Straits of Gibraltar, being about 15 km wide (Figure 1). The coastline of the Mediterranean is about 46,000km, bordered by 22 countries. The Mediterranean Sea is also connected with the Black Sea through the Straits of Dardanelles (maximum width 7 km and average depth 55 m) and since 1889 with the Red Sea through the Suez Canal. The length of the Mediterranean basin from east to west is 3,800km and the maximum distance from north to south about 900km. The surface area is about 2.5 million km^2, which is equivalent to 0.82% of the total area of the world oceans. More information on the physiography of the Mediterranean has been given in previous studies (Karydis and Kitsiou 2012,2014). Due to the high inhomogeneity of the Mediterranean environment, the Mediterranean basin has been subdivided into ten sub-basins (Cruzado, 1985 and UNEP/MAP, 2012), shown in Figure 1.

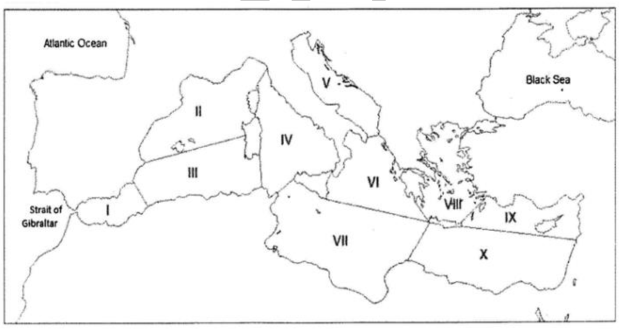


Figure 1. Sub-basins of the Mediterranean. I Alboran Sea, II Northwestern basin, III Southwestern basin, IV Tyrrhenian Sea, V Adriatic Sea, VI Ionian Sea, VII Central basin, VIII Aegean Sea, IX North Levantine Sea and X South Levantine Sea. Source UNEP/MAP (2012), modified

The main driver of environmental pressure in the Mediterranean Sea is the continuous population growth. While the population of the coastal states was 246 million in 1960, these days the population has exceeded the number of 500 million people. It has been estimated that at least one third of the population lives in the coastal zone. Mass tourism in the Mediterranean has grown exponentially after the Second World War. Many areas along the Spanish, French and Italian coasts as well as many Aegean Islands are suffering from overtourism. Mediterranean is the biggest holiday resort in the world accounting for 30 percent of the international tourist arrivals. The number of tourists is expected to exceed 350 million tourists by the year 2025. Agricultural activities have to be added to the environmental pressures: plant cultivation, irrigation, dairy farming and pasture. In addition, aquaculture production follows an exponential trend. The result of all these pressures is marine eutrophication, mainly along coastal areas and gulfs. Oil and heavy metal pollution seem to be the second on the list, although their footprint is continuously eliminated over the last decades due to measures adopted by international conventions and administrative practices.

The main legal instrument for the protection of the Mediterranean marine environment is the Barcelona Convention. Environmental law, environmental protection policy, protocols referring to specific forms of pollution, marine monitoring programs and implementation measures, they all make up the "Barcelona System". This system is characterized by the legal estate, the policy estate and the science estate (Price 1965) and has been the "model" for a number of conventions for other regional seas; some of them will be mentioned in subsequent chapters. The Strategic Action Plan (SAP) for the Mediterranean, functioning within the UNEP framework, has catalogued 131 pollution hot spots most of them characterized by eutrophic conditions (EEA 1999). More information on the physiography of the area, environmental pressures and the governance of the Mediterranean have been given in previous work (Karydis and Kitsiou, 2012).

4.2.2. Trophic conditions in the Mediterranean Sea

The Mediterranean ecosystem is characterized by very high diversity. The list of marine species includes 8,500 species (Karydis, 2021). This number accounts for 7.5% of the species living in the world ocean. There are estimates that the total number of species in the Mediterranean is about 17,000 (Donovaro, 2013). Mediterranean marine habitats support 28 animal phyla. Thirteen out of the 28 are endemic in the marine environment (Karydis, 2021). The Mediterranean Action Plan provides guidelines to the signatory states of the Barcelona Convention for their protection. The Mediterranean Sea is subdivided into ten major sub-basins (Karydis and Kitsiou 2014) and this affects the heterogeneity of the distribution pattern of diversity. For example, the Adriatic Sea hosts 38% of the total species number, the Central Mediterranean 35%, the Aegean 44% and the Levantine Sea 28% (Karydis, 2021). The western part of the Mediterranean (Alboran Sea) is influenced by the Atlantic through the Straits of Gibraltar. The Northern Adriatic Sea characterized by high nutrient concentrations and minor temperature and salinity values, presents a floristic and faunistic regime similar to the Black Sea. It is beyond any doubt that the role of climate change and nutrient enrichments in the Mediterranean are factors affecting biodiversity (Karydis, 2021). However, marine scientists should not ignore that the phenomenon of species composition changing is not new in the Mediterranean, influenced by different climatic regimes that prevailed in the area over the last ten million years. A recent article on biodiversity changes in the Mediterranean Marine Environment has been published and all these problems are presented in detail (Karydis, 2021). The present-day problem regarding biodiversity changes in the Mediterranean is that this phenomenon has been accelerated due to additional causes: destruction of habitats, fishery practices, pollution and invasive/ alien species are factors of anthropogenic origin. All these factors affect ecosystem's stability and dynamics.

Table 3. The most common toxic or potentially toxic algae in the case study areas. The most common areas recording for bloom forming are also provided.

Regional Sea	Toxic or potentially toxic microalgae	HAB events occurrence and their references
Mediterranean	Alexandrium catenella, A. minutum, A. tamarense, Prorocentrum lima, Chaetoceros, Dinophysis sacculus, D. tripos, D. caudata, Ostreopsis ovata, Prorocentrum minimum Pseudo-nitzschia verruculosa and Karenia selliformis,	Moroccan Coasts (Leblad *et al.*, 2020) Annaba Bay (Hadjadji *et al.*, 2020), French coasts (Tichadou *et al.*, 2010), Tunis area (Aissaoui *et al.*, 2014), Adriatic Sea (Roselli *et al.*, 2023), Lebanon (Hassoun *et al.*, 2021), Alexandria (El-Hadary *et al.*, 2022), Spanish coastal area (Busch *et al.*, 2016)
Black Sea	Dinophysis rotendata, D. caudata, D. accuminata, D. hastata, D. fortii and Prorocentrum lima, Alexandrium andersonii, A. minutum and A. ostenfeldii, Dinophysis acuta, D. acuminata, Prorocentrum cordatum	Azov Sea (Atayan *et al.*, 2022), Black Sea Basin (Borysova *et al.*, 2005), shores of Caucasus (Vershinin *et al.*, 2005), NE Black Sea (Silkin *et al.*, 2019; Vershinin *et al.*, 2006), W-NW Black Sea (Dzhembekova *et al.*, 2022)
Caspian Sea	Pseudo-nitzschia seriata, Nodularia spumigena, Gonyaulax polygramma, Nodularia spinifera	Southern part (Makhlough *et al.*, 2019; Nasrollahzadeh *et al.*, 2011), Eastern-Middle part (Pautova *et al.*, 2017)
Baltic Sea	Nodularia spumigena, Aphanizomenon sp., Lepidodinium chlorophorum, Tripos, spp., Karenia mikimotoi, Prymnesium polyleptis	Baltic Sea Basin (Davidson *et al.*, 2014; Luckas, *et al.*, 2005; Telesh *et al.*, 2016), Bothnian Bay (Olofsson *et al.*, 2020), Swedish coasts (Karlson *et al.*, 2021)
North Sea	Phaeocystis globosa, Dinophysis sp., Pseudo-nitzschia sp., Alexandrium minutum, A. ostenfeldii, Gonyaulax spinifera, Karlodinium veneficum and Prorocentrum cordatum	Southern North Sea (Karlson *et al.*, 2021), English Channel (Lefebvre *et al.*, 2020), Belgian coasts (Castagna *et al.*, 2021)
Wider Caribbean Sea	Margalefidinium, Prorocentrum, Mesodinium, Gonyaulax and Phaeocystis, Gymnodinium catenatum, Pyrodinium bahamense, Dinophysis ovum, Pyrodinium bahamense	Coast of Florida (Turley *et al.*, 2022; Accoroni *et al.*, 2020), Caribbean Sea (Cuellar-Martinez *et al.*, 2018), Yucatan coastal zone (Enriquez *et al.*, 2010), Cuba (Moreira-Gonzalez *et al.*, 2013)
South China Sea	Noctiluca scintillans, Phaeocystis globosa, Skeletonema costatum, Scrippsiella trochoidea, Heterosigma akashiwo, Chatonella marina, Karenia mikimotoi, Phaeocystis globosa,	South China area (Wang *et al.*, 2008), Mirs Bay (Dai *et al.*, 2022), Daya Bay (Tian *et al.*, 2021; Yu *et al.*, 2007), Guangdong (Li *et al.*, 2019)

4.2.3. Toxic phytoplankton

A growing threat for the Mediterranean is the invasion of alien species, some of them toxic. They invade mainly through the Suez Canal but also through the Straits of Gibraltar (Karydis, 2021). The main concern for species invasions is the negative effects that have been observed in most cases on ecosystem's function and ecosystem's services (Katsanevakis *et al.*, 2014). Beneficial effects from invasive species on ecosystem services and biodiversity are scanty (Vimercati *et al.*, 2020). This phenomenon is mainly due to the combination of two factors: trade at a global scale and climate change that allowed a number of invasive species to settle. There are many different ways for species to enter the biogeographic space of the Mediterranean Sea and settle (Zenetos *et al.*, 2005). The abundance of alien species in the Levantine Sea is so high that this sub-basin is considered as a different biogeographic area (Karydis, 2021). Alien species once settled, they affect ecosystems' dynamics and the new equilibrium may favor HABs formation. This situation may act as a potential mechanism for the formation of algal blooms. A more serious mechanism is due to alien harmful algal species. Marampouti *et al.* 2021 have found out, after a literature review, that at least twenty harmful algal species have been introduced in the Mediterranean. These include microalgal species from the genera *Alexandrium, Chaetoceros, Dinophysis, Ostreopsis, Prorocentrum and Pseudo-nitzschia.*

Blooms in the Mediterranean had not received the interest of the scientific community until the 70's and they were generally considered as rare events (Maso and Garces, 2006). The frequency of HAB events in the Mediterranean and the subsequent impact on the economy has convinced both governments and international agencies to support monitoring and research. Tsikoti *et al.* (2021) have mined publications regarding eutrophication and related algal blooms, providing a graph that shows an exponential increase of relevant papers for the Mediterranean, mainly over the period 2000-2020. A short review will be given below for each biogeographic area of the Mediterranean. The description of the presence and expansion of algal blooms with emphasis on toxic microalgae will follow the sub-basin division of the Mediterranean Sea as shown in Figure 1. The most common toxic or potentially toxic species in the Mediterranean Sea and the areas they usually occur are given in Table 3.

The Alboran Sea (Figure 1, sub-basin I) is the westernmost sub-basin of the Mediterranean characterized by an intense circulation pattern. Geographically is defined by the Moroccan coastline in the south and southern coastline of Spain in the northern part of the Alboran Sea. There is a strong inflowing surface current from the Atlantic. Eutrophication and harmful algal blooms have been observed along the coastal waters of Morocco (Leblad *et al.*, 2020 and Aboualaalaa *et al.*, 2022). Urban, industrial and agricultural development are the main causes of eutrophic trends especially in the Lagune de Nador, the largest lagoon in Morocco (UNEP/FAO/WHO 1996).

Studies performed along the Moroccan coastal zone (Leblad *et al.*, 2020) have also focused on phytoplankton. The number of taxa found were 92 and numerous HAB species tend to appear in a regular pattern round the year. The presence of *Dinophysis* species and *Prorocentrum lima* showed the highest frequency in the samples collected. Recent work published in 2022 (Aboualaalaa *et al.*, 2022), based on field work between 2018 and 2019 showed that *Pseudo-nitzschia* spp., a producer of domoic acid was present in many samples. The species *Dinophysis caudata*, *Prorocentrum lima, Gonyaulax spinifera, Prorocentrum reticulatum and Karemia selliformis* were also identified. The authors ended with the conclusion that the observed toxic algae were the source of toxins found in mollusks.

The southwestern basin (sub-basin II, Figure 1) is mainly affected by the Algerian coastal line (Hadjadji *et al.*, 2020). The surface inflowing Atlantic water, poor in nutrients and the gyre in the Algerian waters, shape the nutrient regime in the area. Environmental disturbance includes agriculture, heavy urbanization, industrial activities and aquaculture. However, information on nutrients and phytoplankton is scanty (Karydis and Kitsiou 2014). Work in the Annaba Bay (Algeria) has shown that cysts of *Alexandrium pacificum* when germinate they provide populations with remarkable physiological plasticity with up to 30 strains of *Alexandrium pacificum* (Hadjadji *et al.*, 2020). The authors maintain that this physiological plasticity can render the microalga, able to respond in the surrounding environment. This is possibly the reason that this species (*A. pacificum*) is distributed and expanded worldwide.

The North Western sub-basin (sub-basin III, Figure 1) is extending over the Mediterranean coastal zone of Spain and France (Karydis and Kitsiou 2014). Eutrophication is mainly due to agriculture, aquaculture, tourism, intense urbanization and industrialization (Vila *et al.*, 2005). Along the Spanish coastline HABs occurrences have been reported with dominant species the microalgae *Alexandrium catenella, A. minutum, A. tamarense, Ostreopsis ovata* as well as species of the genera *Dinophysis* and *Karlodinium* (sikoti *et al.*, 2021). Diatom blooms were dominated by species of *Pseudo-nitzschia*. The Gulf of Lions in the French coastal zone is the most eutrophic system in the area (Karydis and Kitsiou 2014). Eutrophic conditions in the Gulf of Lions stem from municipal waste and industrial effluents through the Rhone River. Among the dinoflagellates involved in HABs, species of *Dinophysis* and *Alexandrium* showed the most frequent occurrence. Species of *Ostreopsis* and the species *Prorocentrum minutum* were identified. Among the diatom blooms species of *Pseudo-nitzschia, Skeletonema* and *Leptocyndridus* have been reported (Belin *et al.*, 2021).

The northern coastal areas of Tunis, the Northern Sicilian coastal area, the western coasts of the Italian peninsula and the eastern coastal areas of Corsica and Sardinia define the sub-basin IV, the Tyrrhenian Sea (Figure 1). It exchanges water masses with the NW Mediterranean as well as the Central Mediterranean sub-basin (VII). Eutrophication events in the Tyrrhenian Sea are rather episodic, not widespread and with insignificant secondary effects. However, areas in Campania are suffering from anthropogenic pressures that is domestic sewage, industrial waste and agricultural activities (Karydis and Kitsiou 2014). HABs in the Gulf of Tunis have been dominated by the dinoflagellates *Dinophysis sacculus, Ostreopsis siamensis* (Accoroni *et al.*, 2016) and *Peridinium quinquecorne* (Aissaoui *et al.*, 2014).

The Adriatic Sea (sub-basin V, Figure 1) is an elongated system extending from north to south over a distance of 800km. The width varies between 100 and 200km. The Adriatic communicates with the Ionian Sea through the Straits of Otrando, 75km wide. It is a shallow water system, especially in the northern part of the Adriatic, with a depth of about 70m. Due to hydrodynamic reasons, water mass circulation is reduced (Karydis and Kitsiou 2014). Eutrophication in the Adriatic is rather serious. Phenomena of anoxia have been reported and the benthic population has been reduced (Karydis and Kitsiou 2014). Adriatic Sea is known for frequent blooms causing health hazards and affecting esthetic values with impact on the economy. The organisms causing blooms are mainly dinoflagellates and diatoms. Phytoplankton abundance reaching $40x10^6$cellsl^{-1} has been reported (Penna *et al.*, 2006). Among the dinoflagellates, *Dinophysis tripos, D. sacculus, D. caudata, Alexandrium minutum, Gonyaulax spp., Prorocentrum sp.* and *Ostreopsis ovata* are the most common potentially toxic species. *Skeletonema marinoi*, *Chaetoceros* spp. and *Cylindrotheca closterium* are the most common diatoms causing algal blooms (Penna *et al.*, 2006; Cerino *et al.*, 2019).

The Ionian Sea (sub-basin VI, Figure 1), extends over the central part of the Mediterranean Sea, located between the Italian and the Balkan Peninsula. The Ionian Sea is connected with the Adriatic through the Straits of Otranto and with the Tyrrhenian Sea through the Strait of Messina. The physiography of the Ionian Sea has been described in a previous paper (Karydis and Kitsiou 2014). Regarding trophic conditions, there is a gradient from the north (mesotrophic conditions) to the south (oligotrophic conditions). HAB events have been recorded in gulfs and coastal areas. The dominant presence of the raphidophyte *Pseudochatonella verruculosa* was recorded in Amvrakikos Gulf (Greece) during 1998, causing fish kills (Kountoura *et al.*, 2013). The presence of *Prorocentrum minimum* and *Alexandrium insuetum* have also been found in blooms that caused water discoloration and nuisance to fishery and recreation (Nikolaidis *et al.*, 2005).

The Central Basin (sub-basin VII, Figure 1) is defined by the southern coasts of Sicily, the eastern coast of Tunisia and the coastal area of Libya. Information referring to trophic conditions, especially from Libya is limited. However, it is known that the main water mass of the Central Mediterranean sub-basin is highly oligotrophic (Karydis and Kitsiou 2014). Most of the information regarding the trophic status and phytoplankton regarding Tunisia comes from the Gulf of Gabes (Drira *et al.*, 2008, 2010). The coastal zone characterized by high nutrient concentrations was dominated by *Dictyocha fibula*, an opportunistic species. Work carried out within the UNEP framework (UNEP/MAP, 2007) has identified eight toxic dinoflagellates. The most dominant species among them was *Karenia* c.f. *selliformis* accounting for 40% of the total dinoflagellate cells.

The Aegean Sea (sub-basin VIII, Figure 1) is the third major sea in the eastern Mediterranean. An irregular coastline, many islands and rocks characterize the Aegean Sea. The northern Aegean communicates with the Black Sea through the Straits of Dardanelles. It also receives fresh waters by several rivers. The southern Aegean Sea communicates with the main water masses of the Mediterranean through the eastern straits of the Aegean Arc and the Cretan Arc. Information on the topography and circulation has been provided in a previous work (Karydis and Kitsiou 2014). Regarding the trophic state of the sea, the Northern Aegean is mesotrophic, whereas the Southern Aegean is ultra-oligotrophic. However, there are gulfs and coastal waters near urban areas, characterized by eutrophic conditions (Karydis and Kitsiou 2014).

Numerous HAB incidents have been reported, mainly dominated by dinoflagellates, diatoms and to a lesser extent by haptophytes (Ignatiades *et al.*, 2007). Red tides caused by *Alexandrium minutum* have been recorded in the Ismir Bay (1983 and 1988), whereas widespread blooms caused by *Prorocentrum* ssp. and *Prorocentrum micans* occurred during 2015. Along the eastern coastline (Asia Minor), the dinoflagellates *Gymnodinium catenatum, Gonyaulax fragilis* and the diatom *Skeletonema costatum-complex, Thalassiosira sp., Cylindrotheca closterium* and *Pseudo-nitzschia pungens* have also been identified (Aktan *et al.*, 2005). In a review article by Ignatiades *et al.* (2007), it has been found that the most common microalgal species in HABs in the eastern, central and southern Aegean were *Dinophysis accuminata, Karenia Brevis, Alexandrium insuetum, Prorocentrum minimum, Phaeocystis puchetti* and *Noctiluca scintillans*. All these species are potentially toxic. Detailed information on HABs in the Aegean has been given in a recent work (Tsikoti and Genitsaris, 2021).

The North Levantine Sea (sub-basin IX, Figure 1) is bordered by the south coasts of Turkey as well as by the Syrian and Lebanese coastline from the east. The western area of the North Levantine is the Island of Crete. The south coasts of Cyprus are the southern limit of the North Levantine sub-basin. The main water mass of the area is characterized as oligotrophic. However, some coastal areas show eutrophic conditions. The physiography in the area and trophic conditions have been presented in previous work (Karydis and Kitsiou 2014).

Algal blooms of *Prorocentrum micans* and *Heterocapsa triquetra* have been reported in the Mersin Bay (Eker *et al.*, 2000). There is a deficit of information regarding eutrophication and phytoplankton along the southern coasts of Turkey, Cyprus and Lebanon. A recent work in 2021 (Hassoun *et al.*, 2021) was focused on biotoxins produced by algae with negative effects on gastropods, bivalves and fishes. Samples were collected from three sites, Beirut, Tripoli and Tyre. High abundance of *Pseudo-nitzschia* spp., *Gymnodinium* spp. and *Alexandrium* spp. had been observed. They were considered as the source of domoic acid, gymnodimine and spirolid that were detected in the gastropods, bivalves and fishes.

The South Eastern Mediterranean Sea (sub-basin X, Figure 1), is affected by the coastal line of Egypt and Israel. The Egyptian part of the coast has been characterized as a "eutrophication hotspot" due to both, human pressures and the runoff from the Nile (EEA, 1996). The total annual input of dissolved nitrogen input is about 700kt/year and of dissolved phosphorus about 85kt/year (Tsikoti and Genitsaris, 2021). This eutrophic regime has stimulated many toxic algal blooms and a number of toxic algae, between 29 to 38 toxic species, have been reported so far. *Alexandrium minutum* shows the highest frequency resulting in many fish kills. The most serious HAB events that happened in Alexandria where the community was dominated by *Alexandrium minutum* (Ismael *et al.*, 2014), *Prorocentrum triestinum* (Gharib *et al.*, 2006) and *Eutreptiella* belonging to class Euglenophyceace reaching a biomass of 17x106cellsl^{-1}. These genera may have been introduced via ballast water (Heneash *et al.*, 2015). Apart from the *A. minutum,* the species *A. ostenfelddii* dominated in a HAB during 2007 that caused fish mortalities. During the same year, a HAB dominated by *Peridinium quinquecorne* had an impact on the internal tourism. *Gymnodinium* spp., *Prorocentrum* spp., *Pseudo-nitzschia sp.* and *Skeletonema costatum*-complex have also been mentioned in the area (Tsikoti and Genitsaris, 2021).

4.3. The Black Sea

4.3.1. Physiography and environmental pressures

The Black Sea is a semi-enclosed basin surrounded by six countries: Russia, Ukraine, Romania, Bulgaria, Turkey and Georgia. The total area of the Black Sea is about 460,000km^2, the length of the coastline is about 4,020-4,340km (WOA 2021) and the maximum depth 2,200m. The Black Sea is connected with the Mediterranean through the Straits of Dardanelles (Figure 2). The Black Sea is characterized by a very large catchment area (about 1,900,000km^2), receiving fresh waters from 23 countries. Most of the fresh water inflows into the Black Sea through three main rivers: the Danube, Dnieper and Don rivers. This sea is characterized by excess of fresh water and there is therefore an outflow into the Mediterranean Sea. The northwestern region that covers about 25% of the total area of the Black Sea, has a rather shallow continental shelf, the depth being less than 200m. The three rivers accounting for 85% of the total riverine input into the Black Sea and cover an area larger than two million square kilometers; the impact on the northwestern basin is therefore remarkable (Kideys *et al.*, 2002).

Environmental pressures in the Black Sea have changed a highly diversified system into a eutrophic water mass. A number of 17.5 million permanent inhabitants plus 6 to 8 million tourists inhabits the coastal zone of the basin. Additional nutrient loading comes from the rivers. River runoff is the biggest nutrient source for the basin. Nitrogen fluxes have been estimated to about 760,000ty^{-1} during 2000-2005, whereas the phosphorus fluxes during the same period were estimated about 70,000ty^{-1} (Karydis and Kitsiou 2020). More information on water quality of the Black Sea basin and eutrophication status have been given by Karydis and Kitsiou (2014). Fish species have been reduced in number: from 26 species of commercial interest, only 5 species are present these days. In addition, an alien species, *Mnemiopsis leidyi*, has significantly affected the marine ecosystem (Vespremeanu and Golumbeanu 2018).

Figure 2. The Black Sea

The eutrophic areas in the Black Sea are located in the Azov Sea, the north-western part of the basin as well as the estuaries of Danube and Dnieper rivers. According to UNEP (2006), the most critical environmental issue in the Black Sea is eutrophication. Eutrophic trends in the Black Sea were observed from the early '60s (Bakan and Buyukgungor, 2000). More information on the environmental status and eutrophication of the Black Sea has been provided by Vespremeanu and Golumbeanu, (2018). Hypoxic water masses rich in organic matter trigger and maintain frequent algal blooms that are fairly often dominated by toxic algal species. These events are very common over the last thirty years. Dam constructions along the rivers have reduced the supply to silica and therefore the ratios N:P:Si affect phytoplankton community structure, favoring the presence of dinoflagellates. But even when changes in the N:P:Si ratios are milder, it is still possible that diatom succession will be induced. Example: the large diatom *Thalassiosira oestrupii*, in silica deficient conditions is replaced by the small diatom *Cyclotella choctawhatcheeana*. Dominance of dinoflagellates often favors toxic species, causing inter alia, hypoxia and fish death (Strokal and Kroeze, 2013). Phytoplankton community showed a succession over the last twenty years. The spring bloom is considered as the turning point of the annual cycle of phytoplankton succession in the Black Sea marine ecosystem. The dominant species during the spring was *Pseudo-nitzschia pseudodelicatissima* followed by the small diatom *Chaetoceros curvisetus* (Silkin *et al.*, 2019). These diatoms had been succeeded by coccolithophores that had gained dominance during late spring. A hypothesis, widely accepted, is that this dominance is coupled with Si deficiency (Egge, 1992). Silicate seems to be the key factor in species succession in the Black Sea. During the summer, the diatoms *Proboscia alata* and *Pseudo-solenia calcar-avis* are usually the dominant species. The diatom *P. alata* is also the dominant species during the autumn period. The winter period is characterized by the dominance of various large diatom species and coccolithophores. Dominance of the coccolithophore *Emiliania huxleyi* had been observed between 2005 and 2010; its relative abundance was sometimes exceeding 90 per cent. Dominance of the diatom *Chaetoceros curvisetus, Pseudosolenia calcar-avis* and *Dactyliosolen fragilissimus* was observed (2007-2009) in the offshore area (Silkin *et al.*, 2014).

4.3.2. Toxic phytoplankton

According to a recent work by Dzhembekova *et al.* (2022), 27 operational taxonomic units were used and they were assigned to 18 potentially toxic phytoplankton species. The most diversified group in terms of toxic species was the group of dinoflagellates. Among the dinoflagellates, three species of *Alexandrium* were identified that is *A. andersonii, A. minutum* and *A. ostenfeldii*. The species *Gonyaulax spinifera* and *Karlodinium veneficum* showed dominance in certain sampling sites. The species *Dinophysis acuta, D. acuminata, Prorocentrum cordatum, Amphidoma laguida, Phalacroma rotundatum*, and *Gymnodinium catenatum*, potentially toxic microalgae, have also been reported. Among the diatoms, three species of *Pseudo-nitzschia*, potentially toxic, were detected: *P. calliantha* present in 80% of the samples as well as *P. delicatisima* and *P. pungens* that showed sporadic presence. The most common toxic or potentially toxic species in the Black Sea and the areas they usually occur are given in Table 3.

The northeastern part of the basin as well as the central regions are less affected by nutrient loading because the influence by rivers is weak due to the smaller catchment area. Phytoplankton biomass is therefore lower in the northeastern part, although peaks in phytoplankton abundance and red tide events have been observed. When nutrient concentrations are high, a phenomenon is often happening during the spring that is diatoms characterized by their small cell size such as *Pseudo-nitzschia pseudodelicatissima* tend to prevail. On the contrary, when high phosphorus and silica concentrations are coupled with low nitrogen concentrations, the coccolithophore *Emiliania huxlei* prevails. Diatoms of a larger size like *Pseudosolenia calcar-avis*, dominate during the summer time (Silkin *et al.*, 2019). Published work on toxic phytoplankton in the northeastern basin is generally scarce and sporadic. The presence of *Alexandrium tamarense* has been recorded along the northeastern coast of the Black Sea during 2001 (Vershinin *et al.*, 2006).

A phytoplankton-monitoring project was carried out in the northeastern Black Sea along the shores of Caucasus during 2000-2002. A number of 93 species was identified, belonging to seven classes. Thirteen out of 93 species were potentially toxic (Vershinin *et al.*, 2005). Among the toxic diatoms, the species *Pseudo-nitzschia pseudodelicatissima* and *P. pungens* were identified. Among the dinoflagellates, the potentially toxic species *Dinophysis rotendata, D. caudata, D. accuminata, D. hastata, D. fortii* and *Prorocentrum lima* were recorded. In addition, the authors have found the ichthyotoxic dinoflagellate *Margalefidinium polykrikoides* which is considered as a risk factor in aquaculture facilities (Scholin *et al.*, 2003).

4.4. The Caspian Sea

4.4.1. Physiography and environmental pressures

The Caspian Sea located between the eastern side of Caucasus Mountain and the southwestern area of Europe, is a landlocked sea. The area of the sea is about 400,000km^2 and the volume 80,000km^3 (Figure 3). The catchment area is about 3,500,000km^2. The Caspian Sea has been characterized as one of the most polluted regional seas. Sources of marine pollution in the Caspian Sea are discharges of agricultural and industrial wastewaters, domestic sewage effluents and atmospheric deposition of various toxic compounds. However, the main source of pollution comes from drilling, extracting, transporting and refining hydrocarbons. Baku is the largest city and capital of Azerbaijan with population exceeding 2,000,000 people. Baku is the economic center of the country for petroleum production and is an important tourist resort as well. Other Caspian cities causing environmental problems are Astrakhan (Russia) built on the banks of the Volga River, Makhachkala (Daghestan), Enzeli and Babol (Iran) and two regional centers Aktau and Atyran (Kazakhstan). Ranking the pollution problems, oil pollution is on top of the list followed by eutrophication. More information on the physiography of the sea, pollution sources and eutrophic conditions have been given in a previous work (Karydis and Kitsiou, 2020).

Figure 3. The Caspian Sea

The pattern of phytoplankton distribution in the Caspian Sea is also influenced by the hydrographic conditions. In addition, large fresh water masses from rivers in the northern part decrease salinity favoring specific species (Karpinsky, 2005), therefore decreasing diversity. An example is the high dominance of *Rhizosolenia calcar-avis*; as this diatom is not easily grazed by zooplankton, is deposited on the bottom and contributes to anoxic conditions. Sampling carried out during March 2001 in the southern Caspian Sea (Iranian coasts) and the eastern Caspian Sea (Kazakstan), showed average abundance 40,000±35,000 cellsl^{-1}. A total of 45 taxa were found, 20 of them being diatoms and 17 dinoflagellates. The remaining taxa belonged to minor groups (Kideys *et al.*, 2005). A more recent work (Makhlough *et al.*, 2019) on phytoplankton abundance along the Iranian coastline of the Caspian waters, during a four season bloom period, showed minimum-maximum values 73±31 and 501±55 million cells per m^3 respectively.

4.4.2. Toxic phytoplankton

The diatom *Actinocyclus ehrenbergii* and the pyrophyte *Exuviella cordata* are also showing dominance. Invasive microalgae also cause disturbance of the phytoplankton community dynamics. The presence of the alien diatom species, *Chaetoceros peruvianus, Cerataulina pelagica* and *Pseudo-nitzschia seriata* have been recorded (Pautova *et al.*, 2021). The occurrence of a potentially toxic invasive species, *Gonyaulax polygramma* was recorded as well as blooms during the summer of 2010 and 2013 when *G. polygramma* was the dominant species (Pautova *et al.*, 2017). The most common toxic or potentially toxic species in the Caspian Sea and the areas they usually occur are given in Table 3.

Algal blooms have often been observed. The first report on algal blooms of *Nodularia spinifera* with a relative abundance of about 90%, in the southern part of the Caspian Sea was first reported in 2005 (Ramezanpour *et al.*, 2011). *Rhizosolenia calcar-avis* has dominated in a bloom by 95%, affecting sturgeon fishing. In the same area (the Southern Caspian Sea), algal blooms were recorded during 2013-2014. The species *Stephanodiscus socialis, Binuclearia lauterbornii* and *Thalassionema nitzschioides* were observed in blooms during spring, summer and autumn, whereas the harmful species *Pseudo-nitzschia seriata* was recorded during winter time (Makhlough *et al.*, 2019). The cyanophyte *Nodularia spumigena* was the dominant species in a bloom event in the Southern Caspian Sea during 2009, causing a risk to marine water quality (Nasrollahzadeh *et al.*, 2011). *N. spumigena* showed the highest abundance values in some cases approaching 99%.

There is a recent paper on phytoplankton community structure on the eastern part of the Middle Caspian Sea (Kurochkina *et al.*, 2023). The top layer (0-50m) was the most productive, biomass values ranging between 98 and 109 mg/m^3. The most dominant phytoplankton species were *Rhizosolenia calcar-avis, Anabaena bergii, Exuviella cordata* and *Binuclearia lauterbornii*. Studies on bloom development in the Caspian Sea (2010-2013) showed blooms associated with the seasonal upwelling system in the eastern Middle Caspian area, dominated by the potentially toxic dinoflagellate *Gonyaulax polygramma*. Although it was distributed all over the Caspian, most of the events were located in the Middle Caspian region. The concentration of *G. polygramma* accounted for 75 to 99% of the total community (Pautova *et al.*, 2017). The mean phytoplankton abundance was $4.5x10^6$ $cells/m^3$.

4.5. The Baltic Sea

4.5.1. Physiography and environmental pressures

The Baltic Sea is an almost landlocked regional sea communicating with the North Sea only with the Straits of Kattegat and Skagerrak. Basin's area is about $350,000km^2$ and the total length of the coastline is about 40,000 km (WOA 2021), the average depth being about 50m in more than 50% of the area (Figure 4). The basin is surrounded by 14 states. The hydrology of the Baltic Sea is influenced by 250 streams and rivers, contributing about $660km^3$ of fresh water per year. Oder River, Vistula River, Neman River, Daugava River and Neva River are the major rivers in the area. The hydrological budget, precipitation-evaporation is positive. Water excess is driven into the North Sea through the Danish Straits. Although there are many pollution sources in the area, eutrophication is the main environmental issue in the Baltic Sea. The Helsinki Commission (HELCOM) has adopted programs to mitigate eutrophication by reducing nitrogen by 25%. More information can be found in textbooks for the sea conditions and the governance of the Baltic (DiMento and Hickman 2012) as well as for the eutrophication issue (Karydis and Kitsiou 2020). A detail account of the eutrophication issue in the Baltic Sea and the policy measures have been published by HELCOM (2014, 2017).

The largest group microalgae in the Baltic Sea is dinoflagellates, being dominant during the spring because stratified waters favor their presence; this may be also due to the fact that they can choose the optimal water depth. The second important group in the phytoplankton community in the Baltic Sea is the group of diatoms. Turbulent waters facilitate diatom growth as they keep them suspended. Dinoflagellates, diatoms and cyanobacteria are the three functional groups dominating on a seasonal basis in the Baltic Sea (Tramm *et al.*, 2004). Phytoplankton composition on a long-term basis has been analyzed in the Western and Central Baltic Sea (Wasmund *et al.*, 2011). The results have shown that the Baltic Sea is not a uniform water body. The authors found long-term oscillations between diatoms and dinoflagellates. They have stressed the importance of monitoring with frequent sampling, so that short-lived blooming populations could be observed.

Figure 4. The Baltic Sea

4.5.2. Eutrophic conditions

Phytoplankton growth and community structure is a combination of two driving forces: nutrient concentrations and hydrological conditions. Chlorophyll measurements during 2001-2006 showed substantial deviation from the reference conditions set by HELCOM. The same problem was also observed in the Gulf of Riga and the Northern Baltic Proper. During winter time there is nutrient abundance but as the system is light-limited, excessive phytoplankton growth is confined. The upper mixed layer is well illuminated during springtime, favoring phytoplankton outburst. A spring bloom of the diatom *Skeletonema costatum* appears often, especially in the Southern Baltic Proper in March (Tramm *et al.*, 2004). The end of this bloom is followed by various phytoplankton species, *Mesodinium rubrum, Dictyocha speculum* and various dinoflagellates, capable of vertical migration and therefore they can use nutrients from the deeper layers. The dinoflagellate *Peridiniella catenata* is often the dominant species during spring blooms along the Central region of the Baltic Sea. The most common toxic or potentially toxic species in the Baltic Sea and the areas they usually occur are given in Table 3.

The summer time (July and August) where nutrient limitation appears is often characterized by nitrogen fixing cyanobacteria, supplying the fixed nitrogen to other phytoplankton organisms. The species *Nodularia spumigena* and *Aphanizomenon* sp. are in most cases the dominant species among the cyanobacteria. After the summer bloom, large dinoflagellates, like *Ceratium* sp., develop slowly, forming the algal bloom of the autumn. During November and December, thermal convection causes mixing, bringing nutrients to the surface layer. Flagellates of small size, favored by the low light conditions, dominate over other phytoplankton species, during winter. A trophic classification scheme regarding nutrient concentrations and phytoplankton primary production-biomass has been given by Wasmund *et al.* (2011).

4.5.3. Toxic phytoplankton

Harmful algal blooms in the Baltic occur frequently, affecting aquaculture, fisheries, tourism and recreation. The most common blooms in the brackish waters in the Baltic Sea are caused by cyanobacteria (Olofsson *et al.*, 2020). Cyanobacterial bloom events were reported every year during 1987-2019 (Karlson *et al.*, 2021). The most bloom forming cyanobacterium is the cyanotoxin nodularin (NOD) producer, *Nodularia spinifera*, mainly occurring in the Baltic Proper, in the Gulf of Finland and over the last few years in the Bothnian Bay. The presence of NOD has already been detected in fish and shellfish.

Environmental impacts in the area are rather significant. The presence of high cyanobacterial biomass in bathing waters leads to beach closures due to concerns about human health; bathing restrictions due to algal blooms are also recommended in the Bathing Water Directive (2006/7/EC). This causes a serious impact on tourism. The cyanobacterium *Dolichospermum* sp. occurs throughout the brackish waters of the Baltic Sea, producing toxins. A common bloom forming cyanobacterium in the area, *Aphanizomenon flos-aquae* is among the potentially toxin producing organisms. The endotoxins produced, once ingested can damage liver and nerve tissues in mammals. However, even blooms of *A. flos-aquae* that are not toxic can have negative effects due to high biomass accumulation. These effects are having an impact on leisure and tourism (Sanseverino *et al.*, 2016). Algal blooms connected with harmful effects are the species *Lepidodinium chlorophorum* (dinoflagellates), producing blooms in the Western Kattegat and *Tripos*, spp. (dinoflagellate) that appeared at high densities on the Swedish coast of Kattegat; these events resulted in low oxygen concentrations near the bottom water layer.

Species of *Dinophysis* that produce Diarrheic Shellfish Toxins (DST) appear in dense subsurface layers in stratified waters in the Baltic. Cell toxicity of *Dinophysis* cells has been shown to vary depending on cell density (Lindahl *et al.*, 2007). The phototrophic dinoflagellate *Karenia mikimotoi* (first known as *Gyrodinium aureolum*, commonly known as a fish-killing species) (Davidson *et al.*, 2009) appears frequently. Blooms of *K. mikimotoi* were observed sporadically along the Swedish Skagerrak coast, although in these days are rather rare. The phototrophic flagellate *Dictyocha* spp., with siliceous skeleton, widely distributed in the Northern Europe, has been found in the Baltic. In addition, *Octactis speculum*, a fish-killer, affecting aquaculture, was reported in the Southern Baltic during 2004. A bloom of *Pseudo-chatonella* sp., a flagellate belonging to Dictyophyceae was recorded in the Arhus Bay, in the Danish part of Kattegat. Among the twenty species of *Prymnesium*, *P. polyleptis*, was observed in the Kattegat during May-June 1998. The bloom impact on the ecosystem was serious; it affected plankton communities, the benthic ecosystem and caused serious fish mortality (Olofsson *et al.*, 2020; Karlson *et al.*, 2021).

Apart from the bloom forming species mentioned above, a number of potentially toxin producing species have been observed. The species *Halamphora coffeaeformis, Pseudo-nitzschia calliantha, P. delicatissima, P. seriata, P. pungens, Phaeocystis globosa, P. puchetti, Prymnesium polyleptis, Amphidinium carterae, A. operculatum, Dinophysis acuminata, D. acuta, D. mervesia, D. tripos, Alexandrium minutum, A. ostenfeldii, A. pseudogonyaylax, A. tamarense, Gonyaulax spinifera, Lingulodinium polyedra, Prorocentrum reticulatum, P. lima, Karenia mikimotoi, Larlodinium polyedra, Prorocentrum lima, Azandinium spinosum, Fibrocapsa japonica* and also a number of cyanophyceae have been reported (Karlson *et al.*, 2021).

4.6. The North Sea

4.6.1. Physiography and environmental pressures

The North Sea is a marginal sea of the Atlantic Ocean (Figure 5). It communicates with the Atlantic through the Norwegian Sea in the north and the English Channel in the south. The length of the North Sea is about 970km and the width about 580km. The area of the North Sea is 570,000 km^3 and the maximum depth 700m. It receives freshwater from the British Isles as well as from European watersheds, the largest being the Elbe and the Rhine-Meuse watersheds (Karydis and Kitsiou 2014). More information about the physiography and the environmental pressures of the North Sea have been give elsewhere (Karydis and Kitsiou, 2020). The North Sea is subdivided into two sub regions (WOA, 2021): (a) a shallow eutrophic coastal system along the southeastern part and (b) a deeper oligotrophic system in the open sea. Forty million people surround the North Sea. In addition, the states surrounding the North Sea are heavily industrialized; therefore, the environmental pressures are severe. Pressures coming from the tourist industry during the summer months should be added to the existing pressures. Nitrogen loadings have been estimated between 1,400 and 2,000 kt per year. A percentage of this quantity, about 30% enters the North Sea through the atmosphere (OSPAR, 2007). Although nitrogen inputs in the Greater North Sea have been stabilized over the last two decades and nutrient inputs from the Bay of Biscay as well as the Iberian coast have followed a downward trend, the problem of nutrient enrichments remains.

Figure 5. The North Sea

Common procedures applied by the OSPAR contracting parties for assessing levels of eutrophication, have concluded that eutrophication is still a problem (OSPAR, 2017). Eutrophic waters cover about seven per cent of the North Sea area mainly along coastal waters (Karydis and Kitsiou, 2020). In the Greater North Sea, an area, which is estimated about 100,000 km^2, has been identified as eutrophic, extending over the coastal waters of Belgium, the Netherlands, Germany, Denmark and Sweden. Satellite images in the North Sea (Karydis and Kitsiou, 2014), have shown mesotrophic to eutrophic characteristics. Eutrophic areas in the southwestern area extended offshore during winter and spring. On the contrary, northern offshore areas showed mesotrophic trends. High concentrations of nitrogen and phosphorus have led to silicon deficiency. This deficiency increases the risk for bloom formations dominated by non-diatom species. These can be toxin producing species but even if they are not toxic they may disrupt coastal ecosystems. Phosphorus reduction has also been observed, leading to an overall change in the N:P ratios. The 45:1 ratio in 1990, became 80:1 in 2014 (OSPAR, 2017), P being the limiting nutrient. The benthic-pelagic coupling is important for the functioning of the ecosystem because shallow waters extend over wide areas.

It is known since a long time (Moll *et al.*, 1986) that the annual cycle of phytoplankton dynamics is strongly influenced by nutrient availability and zooplankton. Although zooplankton grazing should not be neglected, it seems that nutrient depletion is the key factor in phytoplankton succession. A strong phytoplankton spring peak is observed followed by a decline coupled with decaying biomass until the autumn. Two pronounced peaks are observed in the Central and Eastern North Sea, one during the spring and one during the autumn. In the southern part of the North Sea, a shrinking is observed regarding the spring and autumn maxima, depending on the summer state. The fact that phosphorus is the limiting nutrient during the spring and summer time is known since a long time ago (Riegman *et al.*, 1990). In a recent work (Meszaros *et al.*, 2021), changes in spring bloom dynamics were studied in the Dutch coastal waters and was found that over the last twenty five years and in spite of the prevailing eutrophic conditions, phytoplankton spring characteristics were impacted by climatic conditions. The particular bloom characteristics were the early start and the longer spring bloom duration. It has also been observed that mixing in shallow waters in the North Sea favored stronger spring blooms (Silva *et al.*, 2021).

4.6.2. Toxic phytoplankton

Regarding HABs in the North Sea, it has been found that the presence of some dinoflagellate species showed pronounced variations in the southern and eastern part of the North Sea (Edwards *et al.*, 2006). Although the role of eutrophication in HABs formation is known, climate oscillations in the North Sea seem to play a principal role as well (Belgrano *et al.*, 1999). Long-term changes in the functional groups of phytoplankton are driven by hydroclimatic changes and it has been observed that this regime encouraged HABs growth since the 80's. Their presence is connected with an environment that favors earlier succession (Edwards *et al.*, 2004). In addition, an increase in the ratio of dinoflagellates over diatoms in the southern North Sea has been observed (Hickel *et al.*, 1998). It was found that the dominance of dinoflagellates was related to milder winter conditions. Temperature is also favoring dinoflagellate growth indirectly through an earlier stratification of the surface layer. The abundance of the genera *Ceratium, Dinophysis, Protoperidinium* and *Prorocentrum* seem to be related to hydrodynamic changes recorded in the Norwegian coast. It is likely that these changes are also enhanced by nutrient export to marine waters and may further intensify the formation of HABs in the Skagerrak area (Gjosaeter *et al.*, 2000).

The role of nutrients in bloom creation was observed along the French coast of the eastern English Channel where *Phaeocystis globosa* was competing with diatoms; for example, it was competing with *Pseudo-nitzschia* spp. for resources, especially for nitrogen, phosphate and light. This was happening when silicate was available (Karasiewicz *et al.*, 2022). The concentrations of silicates played a key role for the stability and duration of the diatom community, which in turn influenced *Phaeocystis globosa*. Three toxin producing species along the French coastal waters were identified: *Alexandrium sp., Dinophysis* sp. and *Pseudo-nitzschia* sp. The dinoflagellate *Phaeocystis globosa*, a species with deleterious effects on benthic and pelagic ecosystems were found in the Eastern English Channel and the Southern North Sea. The species *P. globosa* was a good species-indicator when dealing with eutrophic trends (Lefebvre *et al.*, 2020). During work carried out in the Belgian coastal zone during 2018-2019 several toxin producing species were reported (Castagna *et al.*, 2021): the dinoflagellates *Alexandrium minutum, A. ostenfeldii, Gonyaulax spinifera, Karlodinium veneficum* and *Prorocentrum cordatum*; the raphidophytes *Fibrocapsa japonica* and *Heterosigma akashiwo*. Among the species mentioned above, *Prorocentrum cordatum, Pseudo-nitzschia delicatissima* and *P. pungens* were the most abundant.

4.7. The Wider Caribbean Region

4.7.1. Physiography and environmental pressures

The Wider Caribbean Region (WCR) covers an area of about 15,000,000 km^2. WCR encompasses the Caribbean Sea, the Gulf of Mexico, the southeastern US Continental Shelf and the marine areas surrounded by the Central American States and the marine areas of the northern part of the Latin America (Figure 6). The WCR includes, in addition to continental countries, 28 islands. Three large rivers, Amazon, Orinoco and Magdalena outflow in the Caribbean region. There are also many other small rivers outflowing in the WCR.

Figure 6. The Wider Caribbean Region

The natural marine environment is characterized by extensive coral reef formations extending over an area of about 50,000 km^2, accounting for 9% of the world's coral area. The number of coral species is about 70 and more than 500 species of fishes have been identified in the area. In addition, there are many lagoons favoring the development of seagrasses. Numerous lagoons and estuaries are surrounded by mangrove formations. There are also 90 species of sea-mammals in the area (Karydis and Kitsiou, 2020). Due to the abundance of the endemic flora and fauna, the Caribbean has been characterized as one of the world's "biodiversity hotspots".

The population inhabiting coastal areas is estimated to exceed 100 million people. Most of them are heavily dependent on the Caribbean Sea for economic prosperity and human well-being. The coastal and ocean economy in the area includes tourism, fisheries, oil production and shipping. These sectors generate remarkable revenues for the Caribbean countries and provide livelihoods and employment (State of the Cartagena Convention Area – Report 2020). However, the WCR economy is highly oriented towards tourism, accounting for the 18% of the GDP (Gross Domestic Product) of the countries in the region. The growing coastal development is the main cause of marine deterioration. Solid waste is an emerging problem. Various industrial activities produce liquid discharges: food industries, distilleries and sugar refining factories are the main contributors to organic marine pollution. At the same time, oil pollution is a serious threat: oil transportation, offshore production, oil terminal and refineries are the main sources of chronicle oil pollution. The area is among the largest producers of crude oil, as the oil production per year exceeds the amount of 170,000,000 tons (DiMento and Hickman, 2012).

In addition to pollution sources mentioned above, eutrophication is a prevalent and widespread problem in the WCR. The main problem of eutrophication is due to insufficient sewage water treatment (Gonzalez-Rivas *et al.*, 2020). Developing agricultural activities including overuse of fertilizers, intensive husbandry of domestic animals, aquaculture expansion, urbanization of coastal areas and accelerated erosion to watersheds, increase of nitrogen and phosphorus loads are all adding to the problem of eutrophication (Karydis and Kitsiou, 2020). Eutrophic conditions favor the formation of algal blooms, including toxic species of phytoplankton. The blooms cause fish mortalities and provide a suitable background for tropical diseases; deterioration of water quality and decrease of marine biodiversity are also serious problems (Lapointe, 2019 and Sassenhagen *et al.*, 2018).

4.7.2. Toxic phytoplankton

During the last decades climate change and the concomitant hydrologic changes seem to be an additional factor for the boosting of HABs formations. Warming of the surface and therefore the intensification of vertical stratification, favors the growth of cyanobacteria. Along the Mexican coast of the Gulf of Mexico (MC-GoM) it appears that nutrient enrichments of anthropogenic origin are the main drivers of eutrophication, which in turn seems to favor blooms of *Karenia brevis, Chatonella marina, C. subsalsa, Glenodinium pseudostigmosumum* and *Chaetoceros holsaticus*. Some of these species are toxic, other are potentially toxic (Ulloa *et al.*, 2017). The most common toxic or potentially toxic species in the Wider Caribbean Region and the areas they usually occur are given in Table 3.

Temporal and spatial distribution of these species are not well known, neither the cause of the appearance and their origin. Cysts contained in ballast water, seem to explain the presence in the estuary of Barberena and Garrapatas Tideland, the presence of the rapidophyte *Chatonella marina* and *C. subsalsa* as well as of the dinoflagellate *Glenodinium pseudostigmosum*. Other mechanisms regarding cyst's survival are connected with the offshore benthos or their ability to be dormant on surface sediments, even for a period of decades. This may be the case of *Karenia brevis*; molecular techniques have shown that there was no generic difference between the Florida and the Texas of *K. brevis* populations, suggesting a common source of these blooms. A reasonable explanation is a migratory process that may take several successive generations (Waters *et al.*, 2015 and Steidinger, 2009). The upwelling generated by currents in the Yucatan Channel, is pushing nutrient-rich water uphill following the steep continental slope that favors the development of algal blooms and results in food web enhancement, supporting many species of fishes (Enriquez *et al.*, 2010; Steidinger, 2009).

It is known that more than 80% of HAB's events in the Caribbean were dominated by dinoflagellates and to a lesser extent by ciliates, rapidophytes, diatoms, haptophytes and silicoflagellates. The genus *Gymnodinium* showed the highest relative abundance (38%), followed by *Pyrodinium* (26%), *Gambierdiscus* (20%), *Alexandrium* (13%) and *Dinophysis* (3%) (Sunesen *et al.*, 2021). The number of genera involved in harmful events was greater compared to the number of genera related to toxic cases. The genera *Margalefidinium, Prorocentrum*, *Mesodinium*, *Gonyaulax* and *Phaeocystis* were often appearing in harmful events (Sunesen *et al.*, 2021). In the coastal area of Colombia six blooms were recorded between 2010 and 2017, three of those dominated by *Margalefidinium* spp. and the other three by *Mesodinium* cf *rubrum* (Malagon *et al.*, 2013). Seagrass meadows have hosted a number of dinoflagellate species, including the frequently occurring genera *Prorocentrum* and *Ostreopsis*. The seagrass *Thalassia testudinum* also known as turtlegrass, favored the occurrence of *Prorocentrum lima* (Arbelaez-Merizalde *et al.*, 2016).

In the marine area of Cuba (Cienfuegos Bay), about 20 potentiallyy harmful species were recorded between 2007 and 2009. Apart from *Gymnodinium catenatum, Pyrodinium bahamense, Dinophysis ovum* that were observed for the first time (Moreira-Gonzalez *et al.*, 2013), a bloom of *Heterocapsa circularisquama* occurred in 2009. Municipal discharges and limited water circulation in the Cienfuegos Bay, favored HAB formations. Outbreaks of *Phaeocystis* sp., *Chatonella* sp. and *Gambierdiscus*, were also recorded (Diaz-Asencio *et al.*, 2019. Blooms of *Pyrodinium bahamense* and high concentrations of saxitoxins in shellfish were recorded during 2011 and 2012 in El Salvador. A bloom of *Alexandrium peruvianum* during 2012 has been connected with sea turtle deaths, whereas a bloom of *Margalefidinium polykrikoides* produced abundant scum and caused serious fish mortalities. Turtle deaths of the species *Chelonia mydas* (green turtle) and *Lepidochelys olivacea* (olive ridley turtles), were observed during 2013 in El Salvador. These deaths were associated with *Peridinium bahamense*. These findings were confirmed from the gastrointestinal contents of affected sea turtles (Amaya *et al.*, 2018).

Apart from the WCR, algal blooms with impact in marine ecosystems, aquaculture, human health and economy have been recorded in the whole Latin America. Latin America Countries (LAC) have strengthened their cooperation over the last decade to manage the problem better. They have set up a "*Regional Monitoring and Response Network for Marine Resources and Coastal Environments in Latin America and Greater Caribbean*". This project, inter alia, records HAB events, toxic species and in addition, is focusing on biotoxin indentification (Cuellar-Martinez *et al.*, 2018).

4.8. The South China Sea

4.8.1. Physiography and environmental pressures

The South China Sea, a marginal sea of the Pacific Ocean is covering an area of 3,500,000 km^2. It is bordered in the north by the southern Chinese coasts and Taiwan whereas in the west by the Peninsula of Indochina. The South China Sea is bordered by Malaysia and the eastern part of the sea is bordered by the Philippines (Figure 7). It communicates with the East China Sea via the Taiwan Strait and the Philippine Sea via the Luzon Straits. There are also the Straits of Malacca as well as the Karimata and Bangka Straits.

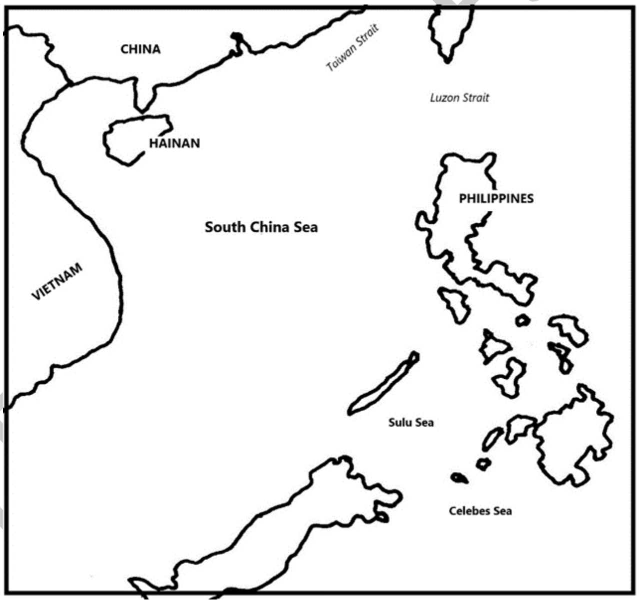


Figure 7. South China Sea

The South China Sea is a region of outstanding economic and geostrategic importance. One-third of the international maritime shipping passes through the sea including 10 million barrels of crude oil per day. It has been estimated that the area accommodates one third of the entire marine biodiversity. The South China Sea ecosystems are distinctive. Unfortunately, fish stocks have almost been depleted due to overfishing (Karydis and Kitsiou, 2020; UNEP/COBSEA, 2010). Environmental pressures in the area are rather intense, causing serious environmental problems. The high population in some areas in conjunction with the economic growth are the reasons of severe and in some cases destructive environmental impact. Environmental impact is due to urban development, land reclamation, destruction of mangrove belts, environmentally non-friendly aquaculture practices, outflow of domestic sewage, industrial effluents and tourist development in the coastal marine environment. The pollution in the coastal area is from land-based sources, as more than 60% of the population lives in the coastal zone of the East Asia territory. The most heavily polluted marine areas are near the cities of Bangkok, Jakarta and Manila (DiMento and Hickman, 2012). The population in those cities increases rapidly because of high birthrates as well as because of immigration from inland areas. The high growth rates of the economy in these areas, increases pollution levels by 10-20% every year. This is because these states prioritize development at the expense of environmental quality (DiMento and Hickman, 2012).

As a whole, the South China Sea has been characterized as a moderately productive system, the primary productivity ranging between 150 and 300gC$m^{-2}y^{-1}$ (WOA, 2021). However, the risk of eutrophication is high but limited to the coastal margins of the sea (Sun, 2017). There are hot spots of seasonal hypoxia and toxic algal bloom events, mainly in the vicinity of major river deltas connected with substantial urban development. This is particularly prominent between April and September during the wet season when 80% of river discharge occurs (Yin, 2001). Eutrophication is the most serious among the problems in the area. The major impact is from sewage disposal into the sea. The inadequate sewage systems release untreated human feces into the sea; in addition to algal growth, the risk of health problems cannot be neglected. Many millions of tourists visiting every year these sites enhance this awkward situation. There is also a considerable amount of organic waste into the sea from poor husbandry of farm animals. It has been estimated that land sources contribute by 77% to marine pollution (UNEP, 2000). The use of chemical fertilizers by China to respond to the food demand of the ever-increasing human population has been escalated over the last decades. It has been estimated that China has increased its use of nitrogen fourfold since the 70s; the same pattern has been followed by the other Southeast Asian states, using in total half the world's nitrogen supply (Glibert, 2013).

In addition to algal blooms another impact of eutrophication is the increase in the jellyfish abundance and blooms. It has been reported that three jellyfish species (*Aurelia aurita, Cyanea nozakii* and *Nemopilema nomurai*) tend to form extensive blooms. The frequency of those blooms has been increasing since the 50's. Published work on offshore areas of the South China Sea is scanty. Most of the studied areas are in estuarine systems and coastal waters where environmental problems are severe.

4.8.2. Toxic phytoplankton

There are frequent HAB occurrences along the Chinese coastal areas causing severe losses to the economy and threaten ecosystems' health. Work carried out from 1990 to 2019 based on relevant statistical data and remote sensing imagery has shown that 1557 HABs were recorded during that period (HI *et al.*, 2021). A number of 249 HAB events was observed in the South China Sea, many of those in offshore areas. The spatial and temporal characteristics of HAB's events were also recorded during 1980-2016 along the coastal area of Guangdong (Canton), a coastal province on the north shore of the South China Sea (Li *et al.*, 2019). The registered population of the Guangdong province is about 80 million permanent residents and about 30 million migrants (2010 census), a number of people that exercise significant pressures in the marine environment. The total number of HABs recorded in the area was 337. Most of the HABs were observed in Mirs Bay, Zhejiang (Pearl) River estuary and the Daya Bay. High frequency of occurrence was shown by the HAB species *Noctiluca scintillans, Phaeocystis globosa, Skeletonema costatum* and *Scrippsiella trochoidea*, that are usually blooming between March and September (Li *et al.*, 2019). These species were responsible for 75 algal blooms, accounting for 20% of the observed HAB events. Blooms of the species *N. scintillans*, a cosmopolitan species, were favored by stable weather, without heavy rains, a fact that explains why *N. scintillans* is blooming during April, instead of during the period between June and September. On the contrary, the June-September period, characterized by relatively high surface water temperature and heavy rainfalls, favored the blooming of most of the species mentioned above (Mohanty *et al.*, 2007). The most common toxic or potentially toxic species in the South China Sea and the areas they usually occur are given in Table 3.

The Hong Kong area has been monitored and HABs were recorded between the 1980s and 1990s (Ho *et al.*, 2022). The author maintained that the major cause for those blooms was nutrient enrichments as well as critical N:P ratio changes in the seawater. These conditions favored excessive growth of selected species of diatoms and dinoflagellates. In addition to the bloom frequency, the duration period was extended as well as the fact that the blooms were covering a wide area. This complex phenomenon was attributed to influences of outflows from the Chinese mainland. The increasing influences were interpreted as the result of the intense social and economic growth that happened over the last twenty years.

Ichthyotoxic blooms in the Guangdong coastal waters were common and were associated with catastrophic fish losses to both mariculture and fisheries. The most common ichtyotoxic species were *Heterosigma akashiwo, Chatonella marina, Karenia mikimotoi* and *Phaeocystis globosa.* Among these species *Karenia mikimotoi* (formerly described as *Gymnodinium mikimotoi*), has been observed for the first time in the coastal Chinese waters since 1988. A big bloom that occurred in the Guangdong coastal area between March and April 1998, caused serious mortalities of numerous economically significant species: *Seriola sp., Pagrosomus major* and *Epinephelus epistictus* were the most important losses (Qi *et al.*, 2004). The toxicity of *K. mikimotoi*, produces various toxins, including hemolytic toxins, cytotoxic polyesters and reactive oxygen species. However, the hemolytic toxins seemed to have a key role in most of the incidences. The species *Chatonella marina* in the Guangdong coastal area has caused economic damage in aquaculture and fish seedlings. In spite of the severity of the problem, toxicity mechanisms in *Chatonella* are still controversial.

The causes for triggering, developing and sustaining algal blooms in the Guangdong area are: (a) anthropogenic nutrient loading: large quantities of waste and raw sewage are released into the sea. Mariculture is also contributing large amounts of nutrients in the marine environment and is a source of concern. Intensive aquaculture causes pollution due to excess of feeding, fish feces and aging water (b) meteorological and oceanographic conditions. Wind direction and current movement seem to be an important element affecting the direction of bloom movements as well as bloom expansion and (c) Climate change and temperature. The monsoons blowing from March to May are the beginning of the rainy season and wind velocity is weakened. These conditions favor algal blooms. Precipitation is also a factor reducing salinity to brackish levels and in combination with high nutrient levels and increased temperature induces bloom outbreaks (Qi *et al.*, 2004). A summary of the most frequenty occuring toxic or potentially toxic phytoplankton species and the places where toxic bloom events usually occur is given in Table 3.

5. HABs in the Regional Seas: matching Science with Policy

5.1. Ecosystems' health and ecosystems' services

The rapidly increasing demand for space in the marine environment either for expanding existing activities (aquaculture, recreation) or for establishing activities based on new technologies such as energy production from renewable sources, has been an almost global trend. The current policy by many states and international organizations connects economic developments with the ecosystem-based approach that will in turn contribute to sustainable development and growth of coastal economies. The sustainable use of marine resources was adopted for the first time as a legal instrument in the Rio Declaration. The Brundtland Report described sustainable development as a process that "*meets the needs of the present without compromising the ability of future generations to meet their own needs*" (Birnie *et al.*, 2009). However, it was not until the year 2002, in the Johannesburg Declaration, where the three pillars of sustainability were placed: economic development, social development and environmental protection. The principle of sustainable development has been adopted by European Union Directives, namely the Marine Strategy Directive (EU, 2008) and the Framework Directive for Maritime Spatial Planning (EU, 2014).

It is obvious the sustainable development requires healthy ecosystems, which in turn provide ecosystems' services (Karydis and Kitsiou, 2020; Culhane *et al.*, 2020). The main services are: (a) regulating services including prevention of coastal erosion, climate control, limiting impacts on coastal flooding and maintenance of water quality (b) supporting services. These are related to biodiversity, primary production and nutrient cycling (c) provisioning services: raw materials, food supply and pharmaceuticals (d) cultural services including recreation and aesthetic values. Coastal eutrophication is a threat for those services, especially when toxic algal blooms are formed with devastating effects on marine biota and marine resources. Management for good ecosystems' health is the link between science and policy, being in an interactive mode.

5.2. Economic consequences

Human health impacts, beyond their significance regarding human safety and welfare, they involve societal costs that are not always easy to assess. Direct coasts referring to medical expenses are rather easy to estimate (Sanseverino *et al.*, 2016). However, indirect costs such as lost wages, lost vacation's time and loss of various personal activities, is difficult to calculate. As marine monitoring for HABs and their toxins is necessary, a partitioning of monitoring expenses in the different sectors of economic impacts would increase the cost estimates. Although there is a plethora of scientific articles and reports related to toxic algae, their toxins and their effects, there is limited information regarding impact assessments on the economy. A systematic review based on known databases (Medline EBSCO, Scopus, ScienceDirect, PubMed and the Cochrane Library), published in 2019, estimated health costs including expenses on healthcare and medication, loss of income (illness), cost of suffering as well as cost of death (Kouakou *et al.*, 2019). The cost of digestive illness for mild, moderate and severe cases was found per person \$86, \$1,015 and \$12,605 respectively. The cost for respiratory illness were \$86, \$1,235 and \$14,600 respectively. Other authors (Martino *et al.*, 2020; Moore *et al.*, 2020) have recently carried out similar calculations.

Commercial fishery impacts are not of minor importance for areas depending on this resource. There is a two-fold impact: direct economic losses in the fish market due to absorption of HAB's toxins by fishes and fish death due to anoxia if oxygen depletion occurs as the result of extreme algal bloom growth. In addition, there are indirect impacts of HABs on fishery economy: consumers are reluctant to buy fishes from areas where HAB episodes occur or near the time of toxic algal bloom formations. An early study (Habas *et al.*, 1974) concluded that a red tide incident in the Southwest Florida cost \$1.5 million but the total losses including tourism were as high as \$20 million. Shellfish production is a significant economic source, especially in rural areas where limited opportunities are available. A threat has emerged over the last decades from HABs and their toxins as their production is exacerbated, mainly due to eutrophication and the global climate change. Work performed in Scottish farms (Martino *et al.*, 2020), has shown that an increase in biotoxin production by 1% causes a shellfish decline by 0.66% and an economic loss of £1.37 million per year. Economic impacts due to HAB's blooms have also been observed in finfish aquaculture facilities.

The economic impact on tourism may be of primary importance in areas relying on tourism, which is based on seafood and recreation. It has been found that brevetoxins (substances mainly produced by *Karenia brevis*), in the form of marine aerosols can cause skin and eye irritation (Fleming *et al.*, 2005). Any negative effects from bloom presence also affect recreational fishes and recreation of tourists along the beaches. This in turn, cause a drop in attendance in hotels, restaurants as well as the attendance in holiday homes. The media may amplify all these negative effects. Sequential impacts entail that it is difficult to get a good estimate on the overall economic losses.

5.3. HABs: monitoring and management practices

Many countries mainly try to protect human health, fisheries and ecosystems. In addition, they are trying to mitigate economic losses. For this purpose, they have adopted monitoring programs. A well-organized monitoring program should include regular water sampling for water quality assessment and qualitative/ quantitative analysis of the phytoplankton community. Furthermore, basic elements of a monitoring program are also sampling of shellfish and fish, recording of fish kills and anomalous animal behavior, data analysis and evaluation of the results, dissemination of information, regulatory actions with main objectives mitigation measures.

Once the principal factors have been understood regarding bloom dynamics and the mechanism producing the impact, mitigation strategies can be planned and applied. These include reduction of nutrient inputs into the marine system, moving fish cages from the HAB infected area, reducing the quantity of food to reduce fish vulnerability to HABs. As measures of nutrient reduction are costly and this may affect other sectors of the economy, it is necessary before the adoption of any relevant measures "*to be proven that human pollution is in fact responsible for proliferation of HAB algae in that area"* (Anderson *et al.*, 2001). Another measure to mitigate the possibility of HABs formation is the control over ballast water. It has been estimated that 80% of the global cargo transportation is carried out by sea and the ballast volume is at least 10 billion tons per year. The number of species carried out intercontinentally every day is at least 3,000 species. Ballast water is usually released near harbors or inshore areas where aquaculture facilities are located. Increased sea traffic, shortening of trip duration and higher speeds provide a better survival chance to the organisms. The increased size of ships that means more oxygen availability, is an additional factor favoring alien species invasions. Alien species (macroalgae and associated fauna) are also introduced in coastal waters through aquaculture activities. Live dinoflagellate cells are often transferred in aquaculture shipments. Bivalves exposed to toxic blooms can contaminate unaffected areas if transferred to uncontaminated places for toxic depuration: feces of bivalves containing toxic algae can serve as un algal inoculum.

5.4. HABs and the Socio-Ecological System

This very short presentation of economic and health effects due to HAB blooms, shows that ecosystemic, social and economic factors are not independent of one another but they form a socio-ecological system (Van Dolah *et al.*, 2016; Borbor-Cordova *et al.*, 2018; Walter *et al.*, 2004). This concept provides the framework for assessing the resilience of the system and its capacity for change. Any policy towards HAB's management should take into account the resilience of coastal communities to these natural disasters. By the term resilience, we mean "*the capacity of a social-ecological system to absorb or withstand perturbations and other stressors such that the system remains within the same regime, essentially maintaining its structure and functions. It describes the degree to which the system is capable of shelf organization, learning an adaptation*" (Walter *et al.*, 2004). The socio- ecological systemic approach allows a firm examination of the ability of this type of system to respond and recover from severe impacts caused by HABs. The final point is whether it can lead to the development of solutions that will enhance social and ecological sustainability (Bauer *et al.*, 2010).

5.5. Future outlook

Any management practices assume reliable technologies that can be applied on a routine basis. Over the last twenty years, molecular techniques for detection of toxin producing organisms have been developed (Galluzzi *et al.*, 2004; Penna and Galluzzi, 2013). These methods provide reliable evidence of the toxic potential of the bloom and enable implementation of targeted measures (Sanseverino *et al.*, 2016). This way coupling of molecular techniques and environmental parameters will contribute towards a better understanding of the ecological processes connected with blooms and will make more feasible the prediction of harmful conditions at an earlier stage. In addition to scientific work and the application of monitoring protocols, a greater public awareness of HAB's episodes is necessary as well as estimations of the impact on the economy would make up a system that could predict HAB's manifestations as much as possible and mitigate their effects. Advances in modeling including all the aspects mentioned above could pave the way towards the success of this effort.

6. Conclusions

It is well established by now that toxic algal bloom events have increased their frequency over the last three decades. In many cases, the duration of these blooms is prolonged. Their negative impact on human health, fishery, aquaculture, tourism and recreation is remarkable. There are also negative effects on ecosystem's health and ecosystem's services. The problem with HAB occurrences is also expanding geographically, appearing in most of the regional seas. It is not any more an issue for scientists but also for economists and managers as their impact does not simply cause ecological damage but in some cases, it can be catastrophic.

The exact mechanism for HAB's creation is not well understood yet, in spite of the generous research funding by some states facing this issue. Advanced methodologies are employed for toxic species identification – enumeration as well as for the extraction and analysis of seafood toxins. Monitoring programs have been deployed in many regional seas, not only for collecting relevant information but also to be used as an early warning system. In addition, numerical modelling developments are underway for a better understanding of HABs mechanisms and for forecasting HAB"s events at a longer time scale: risk assessment type models, ecosystem based models as well as coupled observational – modelling systems are among the most common numerical approaches. A relatively new tool in this field, machine-learning methods, has been added over the last years in the HABs methodological weaponry for forecasting algal blooms. All these efforts have come to a common outcome: irrespective of the applied methodology, a considerable degree of uncertainty remains.

The uncertainty, as the outcome of the data analysis methods used, may be due to the experimental design applied in marine monitoring: HABs duration and geographic characteristics may not be detectable by the standard monitoring practices. There is a scientific consensus nowadays that the principal factor for triggering and maintaining a HAB is nutrient availability (eutrophication), followed by climate change. Although information regarding nutrient, phytoplankton and temperature is collected on a routine basis, other factors can contribute to algal bloom formations. Hydrodynamic conditions, individual responses of some species to nutrient regimes, species interactions among the microalgal community but also interactions between bacteria and phytoplankton as well as grazing, although they may be important, they are not monitored on a routine basis.

Although there is a lot of published information on HAB's biology and ecology, studies on the socio-economic cost of HABs impact, with the exception of USA, are rather limited. If the need for socio-economic impact studies is widely accepted, policy makers will get a better idea of HAB's consequences and would probably finance further research efforts for HABs forecasting and management.

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