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Review

Non-indigenous Marine Species in the Greek Seas and the Role of Ballast Water in their Dispersal: A Mini-integrated Review

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Abstract: The Greek seas as a part of the Eastern Mediterranean Sea, have been considered as a hotspot for the entrance of non-indigenous species (NIS). Ballast water functions as a major pathway for the spread of NIS in new environments, posing significant threats to both the ecosystems and human health. Nine non-indigenous fish species, originating from the Red Sea, have been introduced to the Greek seas since 1925. Despite the implemented laws for limiting the spread of NIS and the subsequent impacts, current global environmental issues, such as climate change and microplastic pollution, could result in a rapid spread and establishment of NIS in hot-spot regions, including the Greek seas. A more systematic use of advanced tools for the systematic monitoring of all NIS in the Mediterranean Sea is necessary.

Keywords: non-indigenous species; ballast water; Greek seas; Mediterranean Sea

1. Introduction

The spread of exotic, or else non-indigenous species (NIS) is a worldwide phenomenon that affects the integrity and function of ecosystems in different ways (Bonanno et al., 2019). The presence of NIS can disturb the naturally occurring ecosystems by establishing populations in areas where they did not previously occur (Kumschick et al., 2015). Consequently, NIS is potentially a threat for the environment, the economy (Kourantidou et al., 2021), as well as human health (Çinar et al., 2014). In this context, the Mediterranean Sea is an area of primary importance, as it is considered a NIS hotspot hosting more than 660 established marine NIS according to a December 2019 report (Zenetos et al., 2020). Factors that contributed to this high number of NIS in the area are (a) the opening of the Suez Canal (1869) that connects the Mediterranean to the Red Sea, (b) aquaculture - related activities, such as packaging and stock movement and (c) the vessels (ships and platforms) through hull fouling, i.e. the attachment of organisms to ships' submerged appendages, and ballast water and sediments (Ojaveer et al., 2014).

Ballast water has been used since the late 19th century for ships to operate with safety, stability, propulsion and maneuverability. It is also used to balance the weight of the fuel consumed, and to prevent any excessive mechanical stress (Takahashi et al., 2008). Despite its necessity, ballast water poses a severe environmental threat, because of its role as a major pathway for the introduction and spread of non-native species. According to Altug et al. (2012), harmful NIS including pathogens are the fourth biggest threat worldwide for the aquatic ecosystems. When ships discharge these species through ballast water, they may establish themselves in a new region and thus threaten the integrity of the local ecosystems. A successful establishment of NIS requires both a method of transport of these species to a new area and suitable conditions, such as lack of natural predators and availability of resources, in the new environment. Under these conditions, NIS successfully compete with indigenous organisms for restricted resources, modify habitats and reduce biodiversity, often causing the extinction of native species. (Galil 2008, Levine 2008).

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Concerning the Mediterranean Sea, the hydrological conditions in the eastern and western basin are a significant parameter for understanding the successful spreading of many Lessepsian species in the Mediterranean Sea (Mavruk and Avsar, 2008), however the key determinant of their successful dispersal is species ecology (Lasram et al., 2008). The term Lessepsian species refers to species that have passed from the Red Sea through the Suez Canal after its opening and were established in the Eastern Mediterranean (Por, 1978). The entrance of Lessepsian immigrants into the Greek seas is a core issue for the ecosystem stability and function, the local economy and public health. The latter is threatened when, among others, people consume NIS as food, being unaware of their toxic ingredients, such as tetrodotoxin (TTX). TTX is a potent neurotoxin, found in exotic fish species, responsible for many human intoxication and fatalities each year. It is a heat-stable substance, that prevents the transmission of electrical signals in skeletal muscles by blocking sodium channels, thus potentially resulting in even death by muscular paralysis, respiratory depression, and circulatory failure (Katikou et al., 2009; Isbister et al., 2005). The minimum lethal TTX dose in humans is estimated to be 2 mg/50 kg (Noguchi et al., 2001).

The total reported cost (observed and potential) of NIS in Greece over the period 1990-2017 is estimated at 78,82$ million (Kourantidou et al., 2021). Considering the climate scenarios predicting a significant warming of the Mediterranean Sea in the next decades (Shaltout and Omstedt, 2014 and Pisano et al., 2020), the impacts of NIS are probably expected to be broader in the near future (Schindler et al., 2018). Due to the environmental, economic and mainly public health significance of NIS, the aim of this mini-integrative review was to search the available scientific sources regarding the presence in the Greek seas of non-indigenous fish species that threaten public health, to provide evidence about the current situation. Additionally, we aimed to summarize the evidence supporting the importance of ballast water as the major cause facilitating the transport of NIS.

2. Materials and Methods

This mini-integrative review was initially focused on searching academic sources for identifying and reporting aquatic species that meet the following criteria: They are non-indigenous fishes that have been officially recorded in the Greek seas, and they can be potentially harmful for human health. We initially identified two official bodies dealing with the issue of aquatic non-indigenous species in Greece, the Hellenic Network on Aquatic Invasive Species (ELNAIS) and the environmental organization iSea. On the iSea website (https://isea.com.gr/) we were able to locate information about the species that meet the criteria we set. After identifying the NIS reported in the Greek seas, we conducted an online literature search in Scopus and Google Scholar from June to September 2021, including the names of the aforementioned species in the keywords. More specifically, the key terms used were: ¨non-indigenous species in Greece and their impact on human health, environment and economy¨, ¨non-indigenous species in Greece and ballast water¨ and ¨Lessepsian migration¨ and ¨*Lagocephalus sceleratus*; first record in Mediterranean Sea and in Greece¨ and ¨*Lagocephalus sceleratus*; impact on human health¨ and ¨*Lagocephalus suezensis*; first record in Mediterranean Sea and in Greece¨ and ¨*Lagocephalus suezensis*; impact on human health¨ and ¨*Sphoeroides pachygaster*; first record in Mediterranean Sea and in Greece¨ and ¨*Sphoeroides pachygaster*; impact on human health¨ and ¨*Lagocephalus guentheri*; first record in Mediterranean Sea and in Greece¨ and ¨*Lagocephalus guentheri*; impact on human health¨ and ¨*Torquigener flavimaculosus*; first record in Mediterranean Sea and in Greece¨ and ¨*Torquigener flavimaculosus*; impact on human health¨ and ¨*Pterois miles*; first record in Mediterranean Sea and in Greece¨ and ¨*Pterois miles*; impact on human health¨ and ¨*Sargocentron rubrum*; first record in Mediterranean Sea and in Greece¨ and ¨*Sargocentron rubrum*; impact on human health¨ and ¨*Plotosus lineatus*; first record in Mediterranean Sea¨ and ¨*Plotosus lineatus*; impact on human health¨ and ¨*Siganus rivulatus* and *Siganus luridus*; first record in Mediterranean Sea and in Greece¨ and ¨*Siganus rivulatus* and *Siganus luridus*; impact on human health¨ and ¨Ballast Water regulations¨.

3. Results

3.1. Literature search results

124 papers were initially found, after an online search for academic resources. 38 of them were rejected because they did not cover any of the review's aims at least partially. We finally included 86 papers in our mini-review, 8 of them referring to ballast water and their rest related to the NIS in the Greek seas.

3.2. Non-indigenous fish species in Greece and their impact on human health

A total of nine NIS have been reported in the Greek seas during the period 1925 - 2017: Lagocephalus sceleratus, Lagocephalus suezensis, Sphoeroides pachygaster, Lagocephalus guentheri, Torquiner flavimaculosus, Pterois miles, Sargocentron rubrum, Siganus rivulatus, and Siganus luridus (https://isea.com.gr/). One additional species, Plotosus lineatus, not yet recorded in the Greek seas, but found in their contiguous zones (Doğdu et al., 2016) was deemed appropriate to be mentioned. All the species are Lessepsian (Kasapidis et al., 2007, Ben-Abdallah et al., 2011, Peristeraki et al., 2006, Al Mabruk et al., 2020, M. Corsini-Foka et al., 2006, Golani et al., 1992, Farrag et al., 2018, Temraz et al., 2013). L. sceleratus, L. suezensis, S. pachygaster, L. guentheri and T. flavimaculosus are not edible species as they contain TTX in their tissues (Kosker et al., 2019, Nagashima et al., 2018, Kosker et al., 2016, Hwang and Noguchi, 2007). The greatest concern is for L. sceleratus and T. flavimaculosus as they contain toxic concentrations of the particular neurotoxin (Katikou and Vlamis, 2017; Kosker et al., 2018). Official reports for the years 2015, 2016, and 2017 revealed that the main TTX intoxication incidences occurred in Greece (4 in 2015, 4 in 2016 and 5 in 2017) and in Spain (6 in 2015, 7 in 2016 and 11 in 2017) (Bedry et al., 2021). The species P. miles, S. rivulatus, S. luridus and P. lineatus have venom glands (Galil, 2018), however their removal makes them suitable for human consumption. S. rubrum is also a potentially harmful species due to its likely high concentrations of heavy metals (Papagiannis et al., 2004).

Lagocephalus sceleratus (Gmelin, 1789)

Lagocephalus sceleratus (Gmelin, 1789), also known as the silver-cheeked toadfish, is a widely spread species, which originates from the tropical Indian and Pacific Oceans (Boustany et al., 2015). It was first detected in the Mediterranean Sea in Turkey in February 2003 (Akyol et al. 2005). In Greece it was first reported two years later, in July 2005 in the Cretan Sea (Kasapidis et al., 2007). *L. sceleratus* rapid spread owes its success to its capacity to tolerate different environmental conditions and to utilize food resources, as well as to its high reproduction and growth rate and the absence of natural predators (Yaglioglu et al., 2011). It is one of the largest members of the family Tetraodontidae, with an elongated body that reaches up to 110 cm in length (Boustany et al., 2015). This fact is significant given that there is a positive correlation between body size and toxicity levels. *L. sceleratus* has amounts of TTX that can be lethal after consumption of this species (Boustany et al., 2015). Since 2003, several episodes of death and serious illness have been reported since neither fishers nor the consumers can identify the new species (Coro et al., 2018).

Lagocephalus suezensis (Clark & Gohar, 1953)

Lagocephalus suezensis (Clark & Gohar, 1953) is a fish species commonly known as Suez pufferfish (Manasirli et al., 2020). It lives in benthic, sandy and muddy habitats of up to 40 m depth (Ben-Abdallah et al., 2011) and was first recorded in the Mediterranean, in the Lebanese coast (Mouneimne, 1977). In Greece, it was first detected in the Aegean Sea, close to the island of Rhodes (Corsini et al., 2005). According to other reports, the species has also been found in the coasts of Spain (Izquierdo-Munoz and Izquierdo-Gomez, 2014), Israel, Turkey and Syria (Basusta et al., 2013). *Lagocephalus suezensis* is also a member of the family Tetraodontidae and it can be toxic for humans because of TTX (Giusti et al., 2018). Additionally, *L. suezensis*, as a bottom feeder, contains high levels of heavy metals, and especially Zn, Pb, and Cd, that make it a potential threat to human health (Meltem et al., 2017).

Sphoeroides pachygaster (Müller & Troschel, 1848)

The blunthead puffer *Sphoeroides pachygaster* (Müller & Troschel, 1848) originates from both sides of the Atlantic Ocean. The first confirmed record of *S. pachygaster* in the Mediterranean Sea was in 1979 from Cala Ratjada, Maiorca, in the western Mediterranean Sea (Oliver, 1981). In Greece, the first report was made in 1992, in Lindos, Rhodes (Zachariou-Mamalinga et al., 1994). *S. pachygaster* is a member of the Tetraodontidae family, too. It has been shown that the particular species can accumulate TTX after a long time exposure to it (Nagashima et al., 2018), which explains why its consumption by humans can result in severe poisoning cases (Rahman et al., 2014).

Lagocephalus guentheri (Ribeiro, 1915)

Lagocephalus guentheri (Miranda Ribeiro, 1915) is a species from the Indo-Pacific Ocean, commonly known as diamondback puffer. It is a newly identified invader species, as *L. guentheri* was recorded for the first time in the Mediterranean in the Syrian coastal waters (Levantine Basin) in November 2016 (Sabour and Masri, 2018). A year later, it was detected for the first time in Greece, on the coast of Pachi, Attiki (Kleitou et al., 2019). Although this species is a relatively new entry into the Mediterranean Sea, its geographic expansion in the basin seems to follow a similar pattern as most Lessepsian migrants. The species is a potential threat for public health due to its content in TTX and saxitoxin, mainly concentrated in the intestine, liver, gonads, and skin (Kosker et al., 2019).

Torquigener flavimaculosus (Hardy and Randall, 1983)

Torquigener flavimaculosus (Hardy and Randall, 1983) also known as yellow spotted puffer, is a pufferfish distributed in the western Indian Ocean from the Red Sea to the Persian Gulf and south Madagascar (Al-Mabruk et al., 2018). It is an alien species in the Mediterranean Sea, first recorded in Haifa Bay in Israel, in 1987 (Golani et al., 1987). In Greece, this pufferfish was first recorded during the winter 2006-2007, near the island of Rhodes. More specifically, three individuals of *T. flavimaculosus* were collected by trawlers in Yalissos, Trianda Bay, a coast in NW Rhodes. Regarding the species' impact on human health, Kosker et al. (2018) showed that its tissues have a higher concentration of TTX than the dose considered lethal to humans according to Hwang and Noguchi (2007). The latter supports that, *T. flavimaculosus* is a highly toxic pufferfish, dangerous if consumed by humans, and thus its consumption should be avoided under any circumstances (Kosker et al., 2018).

Pterois miles (Bennett, 1828)

The lionfish *Pterois miles* (Bennett, 1828) has a native distribution range in the Indian Ocean (Kleitou et al., 2021), while the first detection of the species in the Mediterranean Sea occurred in Israel, in 1991 (Golani & Sonin, 1992). After two decades, two specimens were identified as *P. miles* in a lebanese coast (Bariche et al., 2013). In Greece, a single lionfish was first recorded in the middle of July 2015 in Kallithea, Rhodes. Additional recordings followed in a short time in the same area. Apart from being one of the most successful invasive aquatic species at a global scale (Crocetta et al., 2015), *P. miles* is also venomous with a venom apparatus containing 18 spines and the venom glands (Halstead, 1988). Envenomation produces intense pain and swelling and, depending on the amount of venom, the symptoms may continue for several hours. Although there are no recorded injuries by lionfish in the Mediterranean Basin, the species' presence along the coastlines is a significant public health hazard (Mazza et al., 2014).

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Sargocentron rubrum (Forsskål, 1775)

The Indo-Pacific squirrelfish *Sargocentron rubrum* (Forsskål, 1775) is one of the earliest Red Sea immigrants to the eastern Mediterranean (Golani and Ben-Tuvia, 1985). The first record in the Mediterranean Sea occurred in Israel (Haas and Steinitz, 1947) and a year later, the first specimen was recorded in Greece as *Holocentron rubrum* (Laskaridis, 1948). Although *S. rubrum* is an edible species, its consumption may carry health risks because of the accumulation of heavy metals in its tissues. (Papagiannis et al., 2004). A study in Bangladesh (Baki et al., 2018) showed that the tissues of this fish contained higher levels of Cd and Pb than the upper limit set by the European Community legislation (EC, 2001). Long-term or high-dose exposure to Cd may cause kidney failure, osteomalacia (softening of bones) (Vannoort and Thomson, 2005) and prostate cancer (Gray et al., 2005). High dose exposure to Pb can result in neurotoxicity, nephrotoxicity and many other adverse health effects (Garcia-Leston et al., 2010).

Siganus rivulatus (Forsskål, 1775) and Siganus luridus (Rüppel, 1829)

The most successful invaders in the Mediterranean Sea include the two rabbitfishes, *Siganus rivulatus* (Forsskål, 1775) and *Siganus luridus* (Rüppell, 1829). Both are Indo-Pacific species (Bariche et al., 2009). The marbled spinefoot *S. rivulatus* was firstly recorded in the Mediterranean in 1924 (Steinitz, 1927) while the dusky spinefoot *S. luridus* was recorded for the first time in 1955 (Ben-Tuvia 1964). The first occurrence of *S. rivulatus* in Greece was in 1925 around the Dodecanese Islands. *S. luridus* was firstly recorded in Greece in 1964 in the same area (Kavallakis, 1968). The two species have venomous gill species. Although their venom is not life-threatening to adult humans, it causes severe pain and swelling lasting several hours. Furthermore, patients have complained suffering tremors, nightmares, muscle cramps, hallucinations, agitation, anxiety and nausea, symptoms that lasted between 12 and 30 hours before being completely resolved (Raikhlin-Eisenkraft and Bentur, 2002).

Plotosus lineatus (Thunberg, 1787)

The striped eel catfish *Plotosus lineatus* (Thunberg, 1787) is an Indo-Pacific species and its occurrence in the Mediterranean Sea is a result of migration from the Red Sea via the Suez Canal. It was recorded for the first time in the eastern Mediterranean, in the area between Ashdod and Ashqelon, Israel, in November 2001 (Golani et al., 2002). The species' subsequent expansion followed with records in Lebanon (Bitar, 2013), Syria (Ali et al., 2015) and Turkey (Doğdu et al., 2016). There are no officially confirmed recordings in Greece yet, but due to its rapid population increase, the detection of its occurrence is extremely possible in the near future. *P. lineatus* is a venomous fish species that has been identified as one of the major potential threats against native ecosystems and human well-being throughout the Mediterranean and within EU marine waters (Galanidi et al., 2019).

A summary of information about the origin, the first detection and toxicity of the NIS found in Greece is provided in Table 1. Additionally, Figure 1 shows the regions the above NIS were first recorded in the Mediterranean Sea and in Greece.

3.3. Ballast water and relevant legislation

Vessels conducting maritime transport use ballast water to ensure their stability and safety during their sailing (Takahashi et al., 2008). Since ballast water is a main source of aquatic non-indigenous species in a region, including the Mediterranean Basin and Greece (Ojaveer et al., 2014), a major challenge of disruption is put on the integrity and function of ecosystems with potential harmful effects on the environment, resources, and human health at a global scale (David et al., 2015). Andaloro et al. (2012) provide an indicative example through a study conducted in Italy. The authors reported that the plankton from ballast water and sediment collected in the Gulf of Trieste and Naples included species never recorded in the Italian seas before, among which several were potentially toxic. Additionally, characteristics such as wide temperature range, degraded habitats, high volume of shipping traffic and high occurrence of aquaculture make the Mediterranean an ideal area for the installation of NIS (Flagella & Abdulla, 2005). Another main factor is constructions, namely the Suez Canal, which allowed the movement of tropical species from the Red Sea into the Mediterranean (Zenetos, 2017). In Greece, Saronikos Gulf, including the industrial zone of Elefsis Bay and the Port of Piraeus, as one of the most developed and industrialized areas of the Eastern Mediterranean, is considered a major hotspot of NIS introductions in the country (Zenetos et al., 2020).

Table 1. Summary information about the area and year of first detection and the toxicity of NIS found in Greece.

Species	Common name	Area and year of first record in the Mediterranean Sea	Area and year of first record in Greece	Poisonous or Venomous	Suitable for consumption
Lagocephalus sceleratus	Silver-cheeked toadfish	Turkey (2003)	Crete (2005)	Poisonous/TTX	No
Lagocephalus suezensis	Suez pufferfish	Lebanon (1977)	Rhodes (2005)	Poisonous/TTX	No
Sphoeroides pachygaster	Blunthead pufferfish	Spain (1979)	Rhodes (1992)	Poisonous/ TTX	No
Lagocephalus guentheri	Diamond- back puffer	Syria (2016)	Attiki (2017)	Poisonous/TTX	No
Torquigener flavimaculosus	Yellow spotted puffer	Israel (1987)	Rhodes (2006-7)	Poisonous/TTX	No
Pterois miles	Lionfish	Israel (1991)	Rhodes (2015)	Venomous/Venomous glands	Yes
Sargocentron rubrum	Squirrelfish	Palestine (1947)	Dodecanese (1948)	Venomous/Heavy metals	Yes
Siganus rivulatus	Rabbitfish (marbled spinefoot)	Israel (1924)	Dodecanese (1925)	Venomous/Venomous glands	Yes
Siganus luridus	Rabbitfish (dusky spinefoot)	Israel (1955)	Dodecanese (1964)	Venomous/Venomous glands	Yes
Plotosus lineatus	Striped eel catfish	Israel (2001)	No official record	Venomous/Venomous glands	Yes

Figure 1. Map showing the location of first detection of the non-indigenous fish species that have been recorded in Greece in the Mediterranean Sea (white circles) and the Greek seas (black circles). 1: *Lagocephalus sceleratus*; 2: *L. suezensis*; 3: *Sphoeroides pachygaster*; 4: *L. guentheri*; 5: *Torquigener flavimaculosus*; 6: *Pterois miles*; 7: *Sargocentron rubrum*; 8: *Siganus rivulatus*; 9: *S. luridus*; 10: *Plotosus lineatus*. The arrow shows the location of the Suez Canal (Map source: Google Maps).

The above given, in 2004, the International Maritime Organization (IMO) drafted the Ballast Water Convention, an international treaty, that requires party nations to implement limits to the discharge of viable organisms including specified indicator microbes, harmful to human health, known as the IMOD-2 discharge standard. This Convention entered into force in September 2017 (Cohen et al., 2017). In Greece, Law No 4470/2017 ensures the approval of the International Convention on the Control and Management of Ballast Water and Sediments from Ships (2004) and other provisions.

4. Discussion

The primary goal of this mini-integrative review was to provide updated information regarding the presence of non-indigenous marine fish species in the Greek seas, their origin and toxicity for human consumption.

Our findings showed that five out of ten identified species (*L. sceleratus, L. suezensis, S. pachygaster, L. guentheri, T. flavimaculosus)* are inedible because of the significant amounts of TTX they contain in their tissues. The rest of the NIS (*P. miles, S. rubrum, S. rivulatus, S.luridus, P. lineatus)* are intended for human consumption under conditions, because of their venomous glands or the elevated amounts of toxic heavy metals. All the species were introduced to the Mediterranean Sea through the Suez Canal, and they were initially recorded in its eastern basin (Table 1, Figure 1). In the Greek seas, most of them have been identified in Dodecanese/Rhodes (Table 1, Figure 1).

Apart from the present mini-review, other studies have also confirmed the entrance of NIS in the Mediterranean Sea, and they have also shown the magnitude of the phenomenon. The Israeli coast, for instance, is a hotspot for NIS since 121 new NIS were recorded in the 2010-2020 decade: 38 fish, 30 mollusks, 20 crustaceans and 33 macroalgae. Their entrance occurred mainly through the Suez Canal (Galil et al., 2021). On the other hand, there are cases, such as Italy, where the abundance of non-indigenous fish seems to be still low. The latter probably indicates the necessity for more intense monitoring processes with the application of new tools, for an accurate view of the biodiversity status in the Mediterranean Sea to be achieved. (Tiralongo et al., 2019).

Furthermore, in our mini review, we underlined the significance of ballast water as a major factor contributing to the spreading of non-indigenous species of different taxa and to the subsequent risks for the environment and human health. The problem of NIS becomes worse, if other contributing factors, and particularly the biophysical traits of a region, are considered.

To limit the negative effects from the use of ballast water, laws and regulations have been enacted by local and international authorities. Naik et al. (2019) claim that despite the current measures, more attention should be given to ballast water, given the growing concern about the role of the microplastics it contains as vectors of invasive species, harmful chemicals, metals and associated bacterial pathogens across continents.

Another reason the ecological problems caused by ballast water require immediate management is that new invasive species are likely to pose an additional risk of impact to ecosystems because of the ongoing climate change. According to Vilizzi et al. (2021), who screened 819 non-native aquatic species around the world and classified them into high-, medium- and low-risk categories under different climate conditions, a significant proportion of them will likely turn into invasive in the near future, mainly due to their broad thermal tolerances.

In conclusion, nine aquatic fish non-native species have been reported in Greece until 2020. NIS are posing a constant and developing threat against native ecosystems and consequently to local economies and public health, due to the broad use of ballast water and the geography and physical conditions of particular regions, such as the Mediterranean Sea. The current global environmental challenges of plastic pollution and climate change require tighter and systematic screening procedures that will allow the effective management of NIS and will prevent their uncontrolled spread and the significant disturbances they cause in the local ecosystems.

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