Research Article

Modeling the risk of introducing non-indigenous species through ship hull biofouling: case study of Arzew port (Algeria)

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Abstract

Biofouling of ship hulls is one of the most important vectors for the transfer of aquatic invasive species. These species cause widespread impacts to native environments and ecological communities, in addition to imposing financial costs for industry. Targeted surveillance and effective adaptive management require knowledge on the likelihood of new introductions of non-indigenous species (NIS). We develop a model of the likelihood of introduction and invasion of NIS for the port of Arzew (Algeria), based on the length of stay of vessels in the ports of call, the latitude of these ports, the geographical distance from the port of Arzew, ship's speed, effectiveness of the antifouling system and antifouling strategy used in port of origin. We identified areas that represent a source of high risk species invasion, according to the environmental similarity of the ports of origin with the Arzew port using the Mahalanobis distance. We show that over one year, 738 trips have been made at the port of Arzew, inflicting a very high risk of invasion, in particular from six coastal ecoregions, (the Western Mediterranean ecoregion, the Northern and Central Red Sea, the South European Atlantic Shelf, the Ionian Sea ecoregion, the North Sea, and the Aegean Sea). These results can be used for invasive species management purposes, such as: the application of specific regulations to high-risk vessels and ports in order to minimize the transfer of these species. The methods and models developed here are transferable to any region around the world with similar data availability.

Key words: ecological modelling, probabilistic model, non-indigenous species, maritime traffic, ecoregion, exotic species

Introduction

The transfer of aquatic non-indigenous species (NIS) can threaten freshwater, brackish and marine environments; human, animal and plant life, as well as economic and cultural activities (Narščius et al. 2012). Part of the problem of NIS lies in the difficulty of eradicating them from the environment, particularly for NIS that possess invasive qualities. For these species, mitigation of further introductions is the one of the most effective ways to combat biological invasions (Pyšek and Richardson 2010). The best chance comes from early detection, decisive action and continued support (Ferguson 1999). Targeted surveillance and effective adaptive management of NIS require knowledge on the dynamics of the spread of potential future invaders and thus on the distribution of species, their invasiveness and the likelihood of new introductions. This is particularly important in global industries that directly lead to the spread of NIS such as shipping (Saebi et al. 2020; Seebens et al. 2013).

The intensification of shipping has accelerated the process of global homogenization of biota. The mechanisms and processes governing the adaptation and biological responses of NIS to new environments remain poorly understood, and efforts to minimize the risk of bioinvasion by shipping have been hotly debated over the past decade. This discussion has been boosted by an International Maritime Organization (IMO) convention (IMO 2004) that calls for the treatment of ballast water on board all ships, followed by guidance in 2011 and 2012 on the treatment of biofouling on ships (IMO 2011, 2012). With the emergence of novel technologies, several techniques for managing the fouling of ships' hulls have emerged, from early TBT-based antifouling paints to new silicon-copper-based copolymers. With the evolution of robotics, ROVs (Remotely Operated Vehicle) have been developed for cleaning ship hulls in water (Morrisey and Woods 2015). Some IMO member countries such as New Zealand and the United States of America have introduced specific regulations on the management of ship biofouling (Georgiades et al. 2018), however few local policies have been proposed in many countries, and remains non-existent in Algeria. In Algeria, 70 species across 7 taxonomic groups have been identified as exotic, vagabond or cryptogenic species, many of which are widening their range (Bensari et al. 2020; Grimes et al. 2018). Therefore, it is critical to evaluate the risk of introduction of NIS through biofouling accumulation on ships' hulls throughout this region.

In this study, we develop a model of the introduction and invasion of NIS using the movements of the world merchant fleet, with combining available data from the Arzew port company (APC), and terrestrial AIS (Automatic Identification System). The biofouling of ship hulls is a complex series of sequential events that begins with the adsorption of organic, protein, mineral and osidic compounds, forming a primary film on hard submerged surfaces on which a microbial biofilm attaches itself. This is followed by the colonization of larger sedentary organisms such as algae and invertebrates, and then of mobile organisms such as fish and decapods. Previous models of the spread of NIS by ships have focused on ballast water transport (Bouda et al. 2016; Cope et al. 2015; MacIsaac et al. 2002; Seebens et al. 2013; Sieracki et al. 2014). In comparison, fewer models have been proposed for biofouling (Bouda et al. 2017; Muirhead et al. 2015;

Saebi et al. 2020), although this vector is considered to be responsible for the same or even higher number of NIS introductions: 74% in Hawaii (Eldredge and Carlton 2002), 87% in New Zealand (Kospartov et al. 2008), 78% in Port Phillip Bay, Australia (Hewitt et al. 2004), 50% in the North Sea (Gollasch 2002), and 70% on the North American coast (Fofonoff et al. 2003). Biofouling has been extensively studied because of its economic and ecological importance, particularly in the shipping (Adland et al. 2018; Coutts et al. 2010b; Demirel et al. 2017; Moser et al. 2017) and aquaculture (Bannister et al. 2019) sectors.

We can utilize this knowledge base to ascertain risks of introductions from ship hull biofouling accumulation. To do so, we focus on estimating two key processes relevant to the introduction of NIS through biofouling from commercial vessels: *I*) the magnitude of biofouling that accumulates on a vessel in a port of origin and *II*) the probability that biofouling organisms survive the voyage from a port of origin to a port of destination. The fouling accumulation rate in each donor port can then be combined with environmental similarity to estimate the risk of invasion for each maritime trajectory towards the port of interest. The final stage of the study was devoted to the elaboration of several scenarios for the treatment of biofouling on ships' hulls according to some IMO guidelines and a projection of the invasion risk on the horizon 2050 to assess the effect of changing environmental parameters on hull fouling invasion.

Materials and methods

We used data on the change in fouling coverage to generate a starting model that estimates the percentage of fouling coverage as a function of vessel hull immersion over time, at the stationary state of the vessel in each port of origin (PC_i) (Figure 1). We then integrated into this model an exponential decay function that gives the probability of survival of the fouling as a function of the speed of the vessel as well as a third parameter explaining the efficiency of the vessel's antifouling system (A^t) and a last factor *Pr* relating to the different antifouling strategies used in the port of origin *i* to model the probability of introduction of the fouling from port *i* to port *j* ($P_{ij}(Intro)$). Second, the probability that the NIS is non-native was estimated according to the geographical distance of each port of origin from the port of Arzew (*Pij(Alien)*). The third parameter of the bioinvasion model was calculated by environmental similarity using the Mahalanobis metric (Mahalanobis 1936), then transformed into the probability of establishment of the fouling (*Pij(Estab)*). Finally, the risk of invasion of each port was estimated by combining the three probabilities $P_{ij}(Intro)$, *Pij(Alien)* and *Pij(Estab)*.

After estimating the risk of introduction and invasion of fouling species through the hulls of the vessel in the case of non-treatment of the vessel in

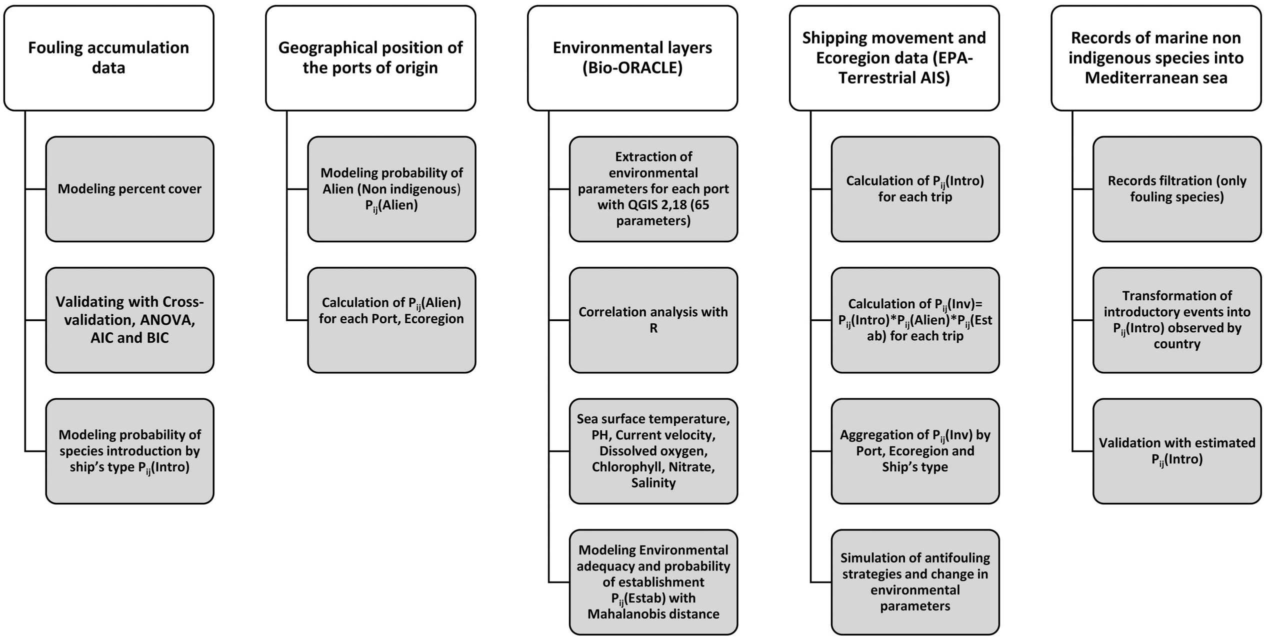

Figure 1. Risk construction diagram. The diagram shows the construction stages and the data used to estimate the risk invasion and introduction of NIS.

the ports of origin (*Pr* = 1), we produced invasion risk maps by port and by ecoregion, using Arc Map GIS. In addition, we modelled the scenarios of biofouling treatment in these ports and the impact of the change in environmental parameters on the risk reduction rate in the port of Arzew. The wetted surface (WSA) of ships is the primary cause of the introduction of fouling, providing the necessary substrate for the adhesion of marine organisms. We consequently use this parameter to analyze the variation of the WSA as a function of the probability of invasion (*Pij(Inv)*). To summarize, the input parameters of the bioinvasion model were: the latitude of the port of origin, the duration of stay in each donor port, the antifouling strategy used in this donor port, the efficiency of the antifouling system, the average speed of the crossing, the geographical distance from the port of Arzew, the environmental similarity between the donor ports and the port of Arzew. More details on the methods, calculations and data sources are given below, while more comprehensive material on the steps of validation, model construction and environmental data analysis are provided in the Supplementary material.

Data source

We incorporated into the bioinvasion model: data on maritime traffic in the port of Arzew, environmental data, and data on maritime ecoregions. Then we validated these results using a database of records of introductions of NIS in the Mediterranean (Figure 1). The data for the validation from 1972 to 2020, have been filtered to include only species suspected to be introduced by ship hulls. We transformed the introduction events of NIS in 13 Mediterranean countries into the observed probability of introduction, by dividing the number of introduced fouling species in each country per the total number of introduced fouling species. We then compared these introduction events with the estimates of the *Pij(Intro)* introduction model. The idea is to find a correlation between the introduction events observed in these countries and the risk of NIS introduction predicted by the model from these countries.

Shipping movement data

We used the shipping movement data to apply the bioinvasion model. These data were obtained from the Arzew Port Authority (EPA) for the year 2016. The database includes all vessels that have called at this port and their respective duration of stay. The EPA also records information on the vessels, such as: IMO number, gross tonnage, tonnage landed, tonnage embarked, date of entry and date of exit from the port. A total of 279 vessels called at the port of Arzew with 738 voyages during the year 2016, coming from 126 ports. This database was supplemented via the Vesseltracker network: https://cockpit.vesseltracker.com/#/cockpit/live/vessels, with other information relating to the vessels, such as the last ports of call during the year 2015–2016, the duration of stay in each port of call, the length between perpendiculars, the tonnage moved by the vessel, ..., the distance traveled between two ports of call, the time and date of entry and exit of each port; transmitted by the terrestrial AIS. This information allowed us to estimate the average speed of each trip to the port of Arzew. We downloaded the data individually for each vessel (research by IMO number) in CSV format, then extracted to the base file containing the 279 vessels.

Environmental data

We accessed the environmental layers from Bio-ORACLE: https://www.bio-oracle.org/downloads-to-email.php, as described in (Tyberghein et al. 2012) and (Assis et al. 2018). We used the All.Present.Surface.BOv2_1.asc (70N-70S Real Values) package, which included 65 raster layers at a resolution of 5 arcmin. Data extraction for each of the donor ports was done with QGIS 2.18 software using the "Point Sampling Tool" extension. These data were extracted for all 126 ports and then analyzed using R software to eliminate variables causing collinearity. For the simulation of environmental variables change, we used the package: All.2050AOGCM.RCP26.Benthic.Mean.Depth.BOv2_1.asc containing forecasts to 2050. These datasets were used for the analysis of the environmental suitability and the probability of establishment of the fouling species. A total of eight variables were selected to implement the *Pij(Estab)* model: mean sea temperature, PH, mean current velocity, mean dissolved oxygen, mean salinity, minimum sea temperature, mean chlorophyll and mean nitrate. We were unable to consider the specific environmental conditions of each trip to the Arzew port, which are known to influence the survival of the fouling.

Ecoregions data

Ecoregions are geographic regions of the ocean that share sets of species because of a common evolutionary history. We used the Marine Ecoregion of the World (MEOW) data (Spalding et al. 2007) to characterize the probability of invasion by ecoregion of origin. The World seas layer IHO-V3 of the Flanders Marine Institute was used as a basic shapefile to create the risk maps.

Fouling accumulation data

We used data on percent change in biofouling coverage from studies that tracked the accumulation of fouling from temperate and tropical coastal sites over time in different parts of the world: eight global biogeographic regions (Australia, Brazil, Japan, Chile, England, Sweden, Portugal, Italy) (Canning-Clode et al. 2010), three mussel farms in Port Phillip Bay Australia (Sievers et al. 2014), two coastal ecoregions of the U.S. coast (Chesapeake Bay and San Francisco Bay) (Davidson et al. 2020), Saltholmen marina in the Kattegat Sea (Oliveira and Granhag 2020), and the marina of Quinta do Lorde in the Madeira archipelago in the Atlantic Ocean (Ferrario et al. 2020).

Mediterranean records of marine alien and cryptogenic species

Katsanevakis provide a dataset on non-native and cryptogenic species in the Mediterranean Sea presents georeferenced data on NIS for the assessment of the spatio-temporal dynamics of biological invasions (Katsanevakis et al. 2020). This dataset offers 5376 records of NIS from 1972 to 2020. This database was used to validate the model that gives the probability of introduction of fouling.

Risk assessment method

In this section, we describe the construction of the probabilistic model of bioinvasion of NIS at the port of Arzew in the western Mediterranean, and the method of its evaluation. First we describe the construction of the risk of introduction between each pair of ports (port of origin and Arzew port) for different voyages. Then we will calculate the probability of establishment of biota attached to the hull (*i.e.*, biofouling) and the probability that the species is non-native (*i.e.*, alien) by biogeographic dissimilarity. Finally, we explain the simulation steps to evaluate the effect of different antifouling strategies, and to predict invasion risk under a future climate change scenario.

NIS Spread

The extent of biofouling on a vessel can be characterized in several ways, including the number of species that have colonized an area (species richness), the biomass formed by the adhesion of these species, the abundance and density of these fouling species, or the extent to which these species cover the available area (e.g., percent cover). In this study we develop a model to estimate the percentage of cover and not the species richness, because species richness as the only measure tells us very little about biological fouling, e.g. 10 species of macroalgae versus 10 species of microalgae represent a richness of 10, but are very different in terms of their impact on shipping and invasion. On the other hand, the coverage rate includes both species richness, the number of individuals (propagule pressure) and the extent to which the species occupy a given area. Biomass would be an ideal measure for ship biofouling because it represents the excess weight that hinders the ship's progress by increasing the shear forces of the water, thus causing over-consumption of fuel and increased greenhouse gas emissions (Schultz 2007). However, the difficulty of obtaining this information and the difficulty of converting it from a surface does not allow us to use it.

Successful bioinvasion depends on several factors, and may be estimated from: a) the probability that a species succeeds in being introduced by a vector beyond its natural range, b) the probability that this NIS is indeed non-native to the receiving environment, and c) the probability that this species establishes itself in the receiving ecosystem (Bouda et al. 2016; Saebi et al. 2020; Seebens et al. 2013) . We used the fouling accumulation data to estimate the percentage of fouling coverage on vessels and translate this percentage into the probability of introduction *Pij(Intro)* (Figure 1). We then estimate the probability the species is alien (*Pij(Alien)*) by biogeographic dissimilarity (Thieltges et al. 2009; Tuomisto et al. 2003), and finally the calculation of the probability of establishment using the environmental data *Pij(Estab)*. Assuming statistical independence among the three probabilities (described in detail below), the invasion probability for a trip between port *i* and port *j* is calculated as:

$$P_{ij}(Inv) = P_{ij}(Alien) \times P_{ij}(Intro) \times P_{ij}(Estab) \quad (1)$$

<u>The probability to be alien or non-indigenous</u>

According to biogeographic studies, there is a characteristic geographic scale at which species composition does not change (Spalding et al. 2007), agreeing to (Seebens et al. 2013):

$$P_{ij}(Alien) = \left(1 + \frac{\gamma}{d_{ij}}\right)^{-\beta} \quad (2)$$

Equation 2 describes the probability that a native species in donor port *i* is non-native in recipient port *j*. This probability is estimated by biogeographic dissimilarity (Thieltges et al. 2009; Tuomisto et al. 2003) and is assumed to increase sigmoidally with the geographic distance *dij* between sites (Tuomisto et al. 2003). Where, $\beta > 0$ is a shape parameter and γ is a characteristic geographic scale on which species composition does not change (Spalding et al. 2007). The probability *Pij(Alien)* is a crucial element of this model and takes into account the fact that the probability of new introductions increases with the dissimilarity between donor and recipient communities. In particular, this term ensures that the risk of invasion between two nearby ports is negligible, which reinforces the hypothesis that the surroundings of a port should primarily contain species that are already present in the port (Seebens et al. 2013). Note that *Pij(Alien)* identifies the probability that a species may be non-native, but does not differentiate whether or not that species has a negative impact on the receiving ecosystem. With γ =1000 km for marine macroinvertebrate (Seebens et al. 2013). This model does not account for the phenomenon of secondary spread, where shorter distances favor biological invasion between adjacent ports. The largest geographical distance from the ports of origin of the vessels that have called at Arzew is 1.83×10^4 km, and the smallest 34.67 km. We assigned a probability of the Alien ~ 0 for the smallest distance (34.67 km), which is in the same coastal ecoregion of the port of Arzew (Alboran Sea) in order to derive the shape parameter β and adjust it to our data. β = 3.39.

$$\beta = -\frac{ln(P_{ij}(Alien))}{ln\left(1+\frac{\gamma}{d_{ij}}\right)} \tag{3}$$

The introduction probability via Biofouling

The risk of introduction through biofouling was calculated according to five key factors: 1) the latitude of the port of origin *i*, 2) the duration of the vessel's stay in port *i*, d_i, 3) the survival of the fouling species throughout the voyage from port *i* to port *j*, 4) the effectiveness of the antifouling system used, which varies according to the type of vessel $A^{(t)}$, and 5) the antifouling strategy used in the port of origin *i* (*Pr*). The relationship between biofouling accumulation (measured as percentage of fouling coverage PC_i) and the length of stay d_i in port *i* (measured in days) was derived from data taken from studies that tracked fouling accumulation as a function of time in several latitudes: tropical, subtropical and temperate (Canning-Clode et al. 2010; Davidson et al. 2020; Ferrario et al. 2020; Oliveira and Granhag 2020; Sievers et al. 2014). We averaged the percentage coverage per site and then applied 1st –5th order polynomial regression models to the percentage coverage data as a function of the residence time of the plates in the water (Figure S2, Figure S3). The AIC ranked the 3rd order polynomial model as the best for estimating the percentage cover as a function of immersion time, while taking into account 1st, 2nd, 3rd, 4th order polynomial models for temperate and tropical data. The ANOVA test (F-test of Overall Significance in Regression Analysis) for 3rd, 4th order models confirmed that the addition of higher order coefficients has no significant effect on the dependent variable (percentage of coverage; Table S1). In addition to time, we also took into account the use of antifouling paints and other antifouling systems depending on the type of vessel. Antifouling paints reduce the colonization and adhesion of biofouling organisms on the surface of ships. The effectiveness of the paint can vary depending on the type of paint, the depth of application and the time elapsed since the last application. Based on our data, the two models that estimate the percentage of coverage of fouling (*PCi*) in the temperate and tropical regions are:

$$PC_i = 1.674 \times d - 1.075 \times 10^{-2} \times d^2 + 2.27 \times 10^{-5} \times d^3, Temperate \quad (4)$$

$$PC_i = 1.668 \times d - 9.88 \times 10^{-3} \times d^2 + 1.524 \times 10^{-5} \times d^3, Tropical \quad (5)$$

In order to generalize these two models (4 and 5), we added a parameter A^t that represents the efficiency of the ship's antifouling system, and a factor *Pr*, relating to the different treatment of the fouling in the port of origin *i*. A survey of commercial ships in California found that the likelihood of a ship having one or more functioning anti-fouling systems varied according to the type of ship (Scianni et al. 2013). Based on this study, the proportion of ships equipped with a functioning anti-fouling system was as follows: container ships 0.19, car carriers 0.20, oil tankers 0.30, passenger ships 0.31, bulk carriers 0.42, and "general" ships 0.53, and all other commercial ships 0.60.

Considering the length of time the ship has been in port *d*, the likelihood of the presence of an anti-fouling system A^t, and the antifouling strategy used *Pr*, we estimated the percentage of fouling coverage (PC_i) on a ship in donor port *i* as follows:

$$PC_i = A^t * P_r * (B_{Tr1} * d + B_{Tr2} * d^2 + B_{Tr3} * d^3), \text{Tropical} \quad (6)$$

$$PC_i = A^t * P_r * (B_{Tp1} * d + B_{Tp2} * d^2 + B_{Tp3} * d^3), \text{Temperate} \quad (7)$$

Where PC_i: percent coverage of fouling in a ship coming from port *i*, *d*: length of stay of ship in port *i* (in days), A^t: refers to the proportion of ships of a given type without an operational anti-fouling system, and *Pr*: a factor related to the different antifouling strategies used by ships, this factor can be defined according to different ship movements, ports or ship types and thus allows modelling different biofouling treatment scenarios. If, for example, the biofouling of the hull is treated equally at each port of call for ship *j* and *Tr* describes the rate of reduction of fouling after a single treatment, the risk reduction over the entire trajectory from *i* to *j* is given by $P_r = 1 - T_r$. Where B_{Tp1} =1.674, B_{Tp2}= −1.075 × 10^{-2}, B_{Tp3} = 2.27 × 10^{-5} are the temperate coefficients, and B_{Tr1} = 1.668, B_{Tr2} = −9.88 × 10^{-3}, B_{Tr3} = 1.524 × 10^{-5} are the tropical coefficients. We used experimental data from Davidson et al. (2020); (Oliveira and Granhag 2020) to infer the reduction rate *Tr* for each antifouling strategy used: a) Antifouling coating with biocide AF (InterswiftVR 6800HS, AkzoNobel), *Tr* = 48.88%, b) Free-release coating FR (IntersleekVR 1100SR, AkzoNobel), *Tr* = 34.9%, c) Antifouling coating AF (Ecofleet 290), without hull cleaning: Tr = 19.30%, *d*) Free-releases coating FR (Sigma Glide 1290), without hull cleaning: Tr = 12.22%, e) AF (Ecofleet 290) with monthly hull cleaning: Tr = 23.38%, f) FR (Sigma Glide 1290) with monthly cleaning of the hull: Tr = 20.58%, g) AF (Ecofleet 290) with bi-monthly cleaning of the hull: Tr = 23.53%, h) FR (Sigma Glide 1290) with bi-monthly cleaning of the hull: Tr = 22.58%. (Figures S5, S6, S7, Table S5). Here we have explicitly considered antifouling paint, using two types of antifouling paints with different hull cleaning frequencies (monthly and bi-monthly). Other antifouling systems include MGPS (electrolytic systems), chemical dosing systems, ultrasonic systems and electrochlorination, and their use is much more variable depending on the type of vessel. Other biofouling management systems have been proposed by IMO and could not be included in this scenario simulation, including the use of biofouling management plans and a biofouling record book.

The survival of fouling organisms during the journey from port i to port j depends on the speed of the trip, V_{ij}. We used experimental results from Coutts et al. (2010a) to adjust an exponential decay function to express the change in probability of survival as a function of vessel speed. Their results show a significant reduction in the percentage of biofouling coverage at displacement speeds above 18 knots (85% reduction at 18 knots):

$$P_{survival}{}^{V} = e^{-\gamma v_{ij}} \quad (8)$$

Where, v_{ij} is the average speed of vessel V from port i to port j, measured in kilometers/day, and $\gamma = 0.0024$. This model (also see Figure S4) estimates a survival rate of 0.15 at 18 knots (or 800.064 kilometers per day), which is similar to the survival rate experimentally found in Coutts et al. (2010a).

Taking into account the accumulation and survival of fouling species, the length of stay at port i, the antifouling strategies used in port i, the latitude of each port i, we estimated the risk of introduction for each trip between port i and j by:

$$P_{ij}{}^{(V)} = P_{ij}(Inro) = A^t * P_r * (B_{Tr3} * d^3 + B_{Tr2} * d^2 + B_{Tr1} * d)\, e^{-\gamma v_{ij}}, \text{Tropical} \quad (9)$$

$$P_{ij}{}^{(V)} = P_{ij}(Intro) = A^t * P_r * (B_{Tp3} * d^3 + B_{Tp2} * d^2 + B_{Tp1} * d)\, e^{-\gamma v_{ij}}, \text{Temperate} \quad (10)$$

Where, $Pij^{(V)}$: probability of introduction of fouling by vessel V from port i to port j, d: length of stay of the vessel in port i (in days), A^t: refers to the proportion of vessels of a given type without an operational anti-fouling system. Pr is a factor relating to the different antifouling strategies used by vessels, $B_{Tp2} = -1.075 \times 10^{-2}$, $B_{Tp3} = 2.27 \times 10^{-5}$ are the temperate coefficients. $B_{Tr1} = 1.668$, $B_{Tr2} = -9.88 \times 10^{-3}$, $B_{Tr3} = 1.524 \times 10^{-5}$ are the tropical coefficients. V_{ij} is the average speed of vessel V from port i to port j, measured in kilometers per day, and $\gamma = 0.0024$. Here the probability of introduction refers only to the fact that the species is brought back by the vessel from port i to port j, without taking into account the probability that the species may or may not become detached from the hull to propagate in the receiving environment, and does not take into account the phenomenon of secondary spread. We focused on modelling the risk of biofouling introduction as a function of parameters that: 1) could be predicted from our global marine and environmental dataset and 2) could be estimated using empirical data. Other parameters that could influence the risk of biofouling introduction could not be included in this study, including travel conditions (i.e., the change in environmental parameters during the route from *i* to *j*), and 2) the passage of ships in dry docks or the duration of the last application of antifouling paint. Further, all the last ports of call before the Arzew harbor break-in could have been taken into consideration, we only focused on macro-invertebrate and algae and we do not consider marine growth prevention systems that can further reduce fouling loads on ships or other areas beyond the hull that can contain biofouling such sea chests and internal water pipes (McDonald 2012).

The probability of establishment

We calculated probability of establishment on the basis of environmental similarity between the donor ports and the port of Arzew, using the Mahalanobis distance in a 8-dimensional ecological space (Mahalanobis 1936). This metric is consistent with the ecological niche theory, which suggests the existence of optimal environmental conditions for a species in addition to maxima and minima, outside of which the species cannot exist (Hutchinson and MacArthur 1959), but it also explains better than other metrics (Euclidean distance) the gradual change in environmental conditions (variance-covariance matrix) to which fouling is subject throughout the ship's passage. The formula is given by the following equation:

$$D^2(x_m) = (x_m - \bar{x})^T S^{-1}(x_m - \bar{x}) \quad (11)$$

Where, D^2: Mahalanobis distance, $x_m = [x_{m1}, x_{m2}, x_{m3}, \ldots, x_{mn}]$: vector of environmental variables of the different ports, $\bar{x}$: vector of environmental variables for Arzew port. *S*: variance-covariance matrix of the 126 ports. Twenty-eight raster layers were selected for the calculation of the environmental adequacy between the donor ports and the port of Arzew. The environmental parameters were documented from previous studies on the influence of abiotic factors on the distribution of fouling species (Kocak et al. 2019; Lin et al. 2017; Martin et al. 2020; Mhaddolkar Sonali et al. 2019). A correlation matrix was constructed from these data using the 126 donor ports as variables. Layers with a correlation greater than 0.75 or less than −0.75 were excluded from the environmental suitability calculation (see Figure S1). These layers were considered to cause collinearity and provide the same information at the Mahalanobis distance and only one of the layers was considered. Of the 28 layers used, eight variables were selected for the calculation of environmental suitability: mean sea temperature, PH, mean current velocity, mean dissolved oxygen, mean salinity, minimum sea temperature, mean chlorophyll and mean nitrate.

The conversion of this distance into the probability of establishment *Pij(Estab)* was carried out using the cumulative distribution function of inverse chi-square (Etherington 2019), $F_{\chi_n^2}^{-1}(x) = P(\chi_n^2 > x) = 1 - P(\chi_n^2 \leq x)$ which has 1 at the optimum. The formulas are given with the following equations:

$$P_{ij}(Estab) = F_{\chi_n^2}^{-1}(D^2) = P(\chi_n^2 > D^2) = 1 - P(\chi_n^2 \leq (x_m - \bar{x})^T S^{-1}(x_m - \bar{x})) \quad (12)$$

$$P_{ij}(Estab) = \int_{D^2}^{\infty} \frac{1}{2^{\frac{n}{2}}\Gamma(\frac{n}{2})} e^{-x/2} x^{\left(\frac{n}{2}\right)-1} dx \quad (13)$$

Where, χ_n^2 : chi-squared random variable with n degrees of freedom, Γ : gamma function, $F_{\chi_n^2}^{-1}$: cumulative distribution function of inverse chi-square. This technique is a corrected technique of the original model that estimates the probability of belonging to the fundamental niche using the chi-square cumulative distribution function $F_{\chi_n^2}(x) = P(\chi_n^2 \leq x)$, which has 0 at the optimum. Using this corrected technique means that the probability $P(\chi_n^2 > D^2)$ indicates locations with D^2 lower than would be expected by chance and are therefore more likely to belong to the Arzew port niche. The closer the distance a donor port is to the port of Arzew, the more likely it is that species from that port belong to the basic niche, and the more likely they have the capacity to establish in that receiving port. It is important to recognize that the use of this Mahalanobis metric does not take into account species with high ecological valence (e.g., euryecium species).

The probability of invasion *Pij (Inv)*

The total probability of invasion along the route between port i and j is obtained by combining the three probabilities for the specific stages of the invasion. Our calculations show that these three probabilities are only weakly correlated (Figure S8). Assuming statistical independence, we can calculate the probability of new invasions from a particular ship movement by taking the product:

$$P_{ij}(Inv) = 1 - \prod_{ij}[1 - P_{ij}(Alien)P(Intro)_{ij}P_{ij}(Estab)] \quad (14)$$

The final invasion probability between port i and port j for all voyages from i to j was calculated by aggregating all invasion probabilities calculated individually for each of the voyages from i to j. This aggregation can be applied to ship types, ecoregions as well:

$$P_j(Inv) = 1 - \prod_i[1 - P_{ij}(Inv)] \quad (15)$$

Likewise total introduction probability from port i to port j:

$$P\ (Intro)_{ij} = 1 - \prod_{ij}(1 - P_{ij}{}^{(v)}) \quad (16)$$

Wetted surface area calculation

The wetted surface area (WSA) was used to characterize the probability of invasion as a function of the wetted area (substrate for fouling species) for different types of vessels. WSA of the vessels calling at the port of Arzew was calculated using the HOLTROP-MENNEN formula (Holtrop 1977):

$$WSA_{max} = L_{WL}(2T + B) * \sqrt{C_w} * (0.530 + 0.632C_b - 0.360(C_w - 0.5) - \frac{0.00135L_{WL}}{T} \quad (17)$$

Where; WSA_{max}: Maximum wetted area (m^2) (which corresponds to the summer draft), T: Summer draft, L_{WL}: length of the ship on the waterline (m), B: Breadths at midship (m), C_w: midship area coefficient of the underwater hull (from Bouda et al. 2017), C_b; block coefficient. Here, the maximum wetted surface of the vessels is used to estimate potential biological invasions. Further parameters would improve the accuracy of such estimates, including the surface area of the niches, the wear of the antifouling coating, the contact of the ship with the seabed in the port waters, the working speed of the ship, the passage in dry dock, and the last cleaning operation of the hull. Niche areas of vessels, such as side thruster tunnels, sea chests, and propellers, are often hot spots for the accumulation of biofouling organisms, a potential source of aquatic invasive species. Moser et al. (2017) quantified the extent of this area as 10% in the global fleet (120,252 vessels), but this varies dependent on vessel type. We were unable to estimate the niche area as it requires data on maximum hull bearing stress, propeller diameter, volumetric flow rate of the seawater intake, maximum flow velocity through the grating.

Results and discussion

We first present the results of the environmental adequacy and the probability of establishment between the donor ports and the port of Arzew, and then the results of the invasion according to proposed scenarios.

Environmental similarity and probability of establishment

The port of Arzew is part of the Alboran Sea Ecoregion (MED-36), the Mediterranean Sea Province and the North Atlantic Temperate biogeographic realm. The coastal ecoregions that represent a high risk of establishment of NIS are distributed primarily in the Mediterranean basin and the Ecoregions of the Lusitanian Biogeographical Province (Figure 2).

The Mediterranean basin

The ecoregions (MED-30 to MED-36) were at very high risk, represented respectively by: the Aegean Sea ecoregion with a maximum establishment probability of 0.97, the Western Mediterranean ecoregion (0.99), the Levantine Sea (0.64), the Adriatic Sea (0.64) and the Ionian Sea ecoregion from Italy and the island of Malta (0.91) (Figure 2). The biota of these areas

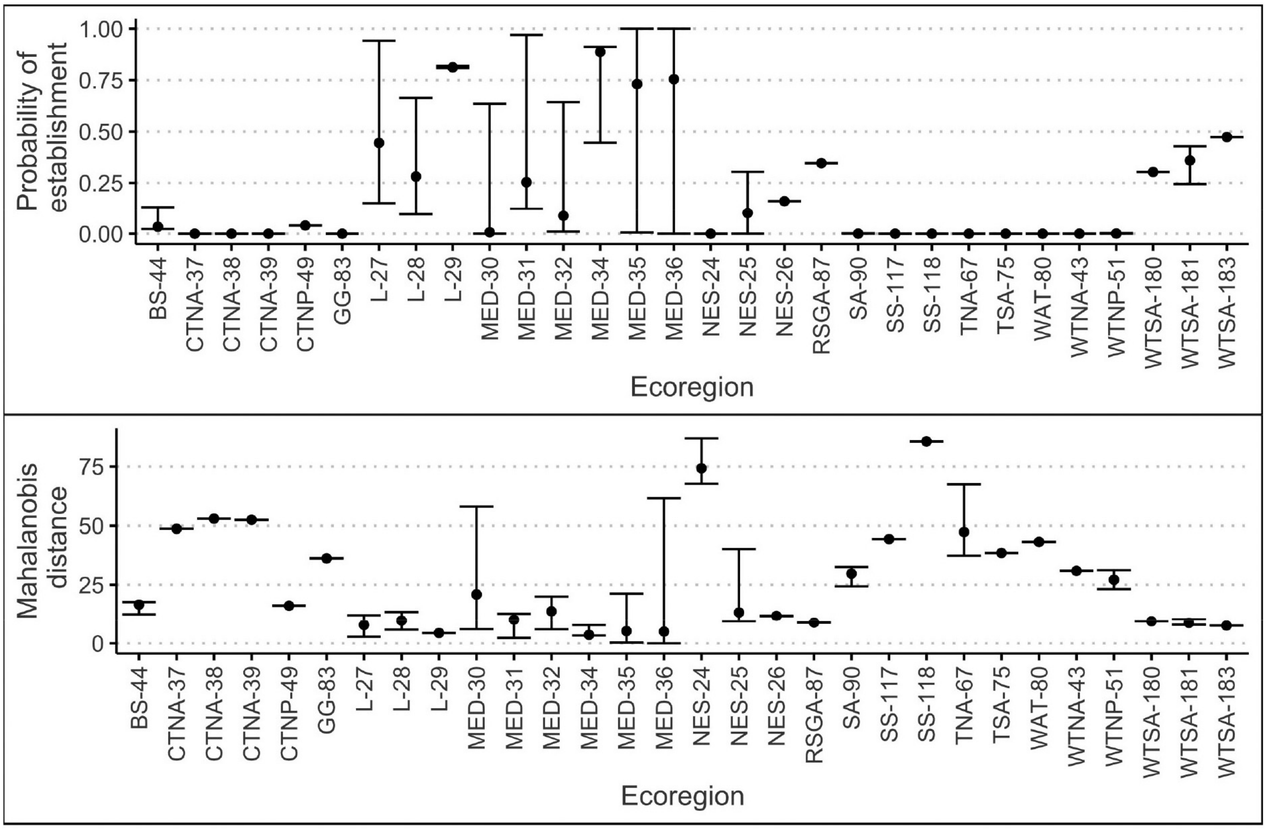

Figure 2. Environmental similarity between donor ecoregion and Arzew port shown as probability of establishment of non-indigenous species and Mahalanobis distance. Error bar showed for the maximal and minimal Mahalanobis distance and probability of establishment.

is very similar to the composition of the Western Mediterranean, leading to the observed environmental similarity (Figure 2). Fouling communities of sessile affinity transiting through these regions will adapt easily if they survive to the ship's drag, all the more so since most commercial ships navigate close to the coast where environmental changes are less significant, which could increase the chance of survival of these non-native species, thus enabling them to invade the receiving environments. Like the case of the cumaceous crustacean *Eocuma sarsii*, which was reported by Grimes et al. (2018) in the Port of Arzew, and by Corbera and Galil (2016) in the Levantine Sea, this cryptogenic species could be native to the Asian Seas. The Levantine Sea is an important transitory for the introduction of Lessepsian species via the Suez Canal; 70% of the primary introductions of NIS reported in this region come from this canal (Nunes et al. 2014), which is in connection with the Asian and Middle Eastern Seas via the Red Sea.

Ecoregions of the Lusitanian Biogeographical Province

The Canary Azores Madeira ecoregion (L-29) has high environmental similarity with the Alboran Sea ecoregion, with an average distance of Mahalanobis = 4.47, and an average settlement probability of 0.81 (Figure 2, Table S2). Similarly, the two coastal ecoregions L-28 (Saharan Upwelling) and L-27 (South European Atlantic Shelf) both have a mean Mahalanobis

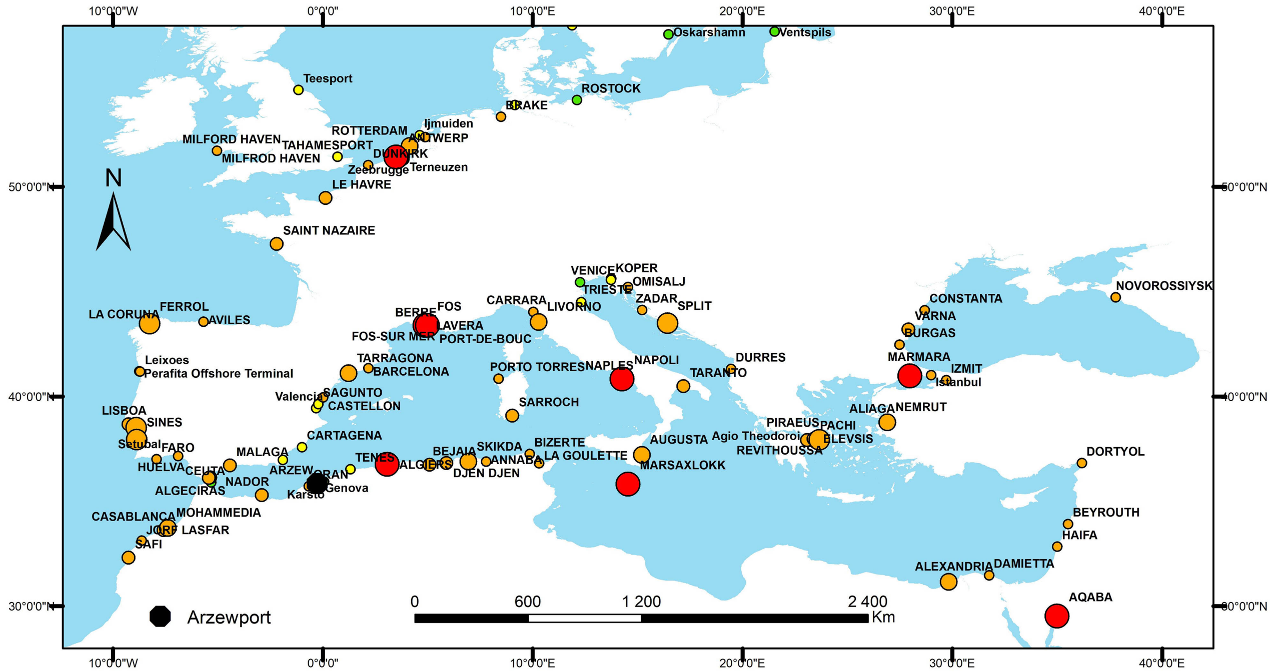

Figure 3. Map showing the probability of invasion through biofouling of the ship hulls for all donor ports in the Mediterranean and North Sea during the year 2016, expressed as the relative risk of each port to the Arzew port. The size of each point indicates the relative contribution $P_{ij} = P_{ij}(Inv)/\sum_i P_{ij}(Inv)$ of source port *i* to the total invasion risk of all ports. Green indicates low risk ($0 \leq P_{ij}(Inv) < 10^{-9}$), yellow moderate risk ($10^{-9} \leq P_{ij}(Inv) < 10^{-4}$), orange high risk ($10^{-4} \leq P_{ij}(Inv) < 10^{-2}$), red very high risk ($P_{ij}(Inv) \geq 10^{-2}$).

distance of 9.77 and 7.88, respectively, and a mean settlement probability of 0.66 and 0.94, respectively. Coastal shipping routes from these areas favor the survival of species introduced from these ecoregions, and environmental conditions would be favorable for most taxa that could become invasive. This result is consistent with the observations of Bensari et al. (2020) at the port of Arzew concerning the species *Apseudopsis adami*, a non-native and unrecorded species in the Mediterranean, originating from southern Portugal (L-27).

Risk of invasion by port of provenance

In this section, we present the bulk of our results, with additional results presented in the supplementary information. The invasion risk calculated individually for each of the maritime trajectories was aggregated by port of origin (Figure 3). The Mediterranean ports, namely the port of Naples, Marsaxlokk, Algiers, Fos, Marmara, and the port of Bouc in France, present a probability of Alien that is more or less low, given their distance from the port of Arzew (Figure 3). However, the high number of ships coming from these ports, the environmental similarity with the western Mediterranean, the length of stay of ships in these ports (0.05–25 days), and the low speed of ships crossing from these areas result in a very high probability and risk of invasion (*Pij (Inv)* ≥ 0.01). Several Mediterranean ports are hubs, such as Marsaxlokk in Malta, Valencia in Spain, and Piraeus in Greece. These hubs receive a considerable number of ships, mainly from Asia. Propagule pressure is consequently high. Additionally, the Mediterranean exhibits high environmental similarity with some Asian seas (Bouda et al. 2017). The probability of species introduction is increased. Thus, an introduction occurring around these hubs will inevitably affect the port of Arzew, through the process of secondary introduction. This is a widespread phenomenon along the Algerian coasts.

Risk of invasion by ecoregion

From the 30 donor ecoregions, 6 are at very high risk of hull fouling invasion, 10 at high risk, 9 at moderate risk and 5 at low risk (Figure 4). Indeed, the RSGA-87 "Northern and Central Red Sea" ecoregion is classified as a very high risk area, with an invasion probability of $Pij\ (Inv)$ = 0.012 based on only 15 trips. The average speed of vessels coming from this area is 8.83 knots, and the average duration of stay of vessels is 2.62 days. A probability of survival of 0.40 was estimated for the fouling transported from this zone and a probability of the alien of 0.46. The accumulation of fouling in the ports of this ecoregion is moderate due to the short stay of the ships, but the reduced speed and coastal navigation of the ships coming from the Red Sea and the environmental similarity with the Alboran Sea have increased the probability of survival and therefore the probability of invasion is increased. Our predictions for this bioregion are consistent with several studies that have dealt with biological invasion through Lessepsian immigration. For example, *Galaxaura rugosa*, a species of rhodophyte algae of Indo-Pacific origin, established in the Mediterranean would probably be introduced by the hulls of ships from the Red Sea which is connected with the Asian seas (Hoffman and Dubinsky 2010).

The Mediterranean Sea bioregions MED-34, MED-31 and MED-35, respectively: "Ionian Sea", "Aegean Sea" and "Western Mediterranean" have a very high probability of invasion ($Pij\ (Inv)$ = 0.015, 0.03 and 0.07 respectively), with a relative contribution of 54.9% ($P_{ij} = P_{ij}(Inv)/\sum_i P_{ij}(Inv)$) in 402 trips. The average speed of vessels from these three ecoregions is 7.05 knots, and the average stay of vessels is 2.56 days. The Mediterranean ecoregions show a great environmental similarity between them, thus favoring the establishment of biota introduced by the process of secondary introduction. The environmental similarity between these three ecoregions and the Alboran Sea is very high (D^2 = 3.64, 10.16, 5.25 respectively) (Figure 4), to which is added the high probability of survival of the fouling carried by the hulls of vessels ($0.11 \leq P_{survival}{}^{V} \leq 0.99$). These results can be interpreted as follows: if an invasive species has been introduced through the hull of a ship at one of the Mediterranean ports and succeeds in becoming established, it will soon invade the whole Mediterranean in a short time, particularly the Alboran Sea. This is supported by Ferrario et al. (2019) who found 33 introduced fouling species

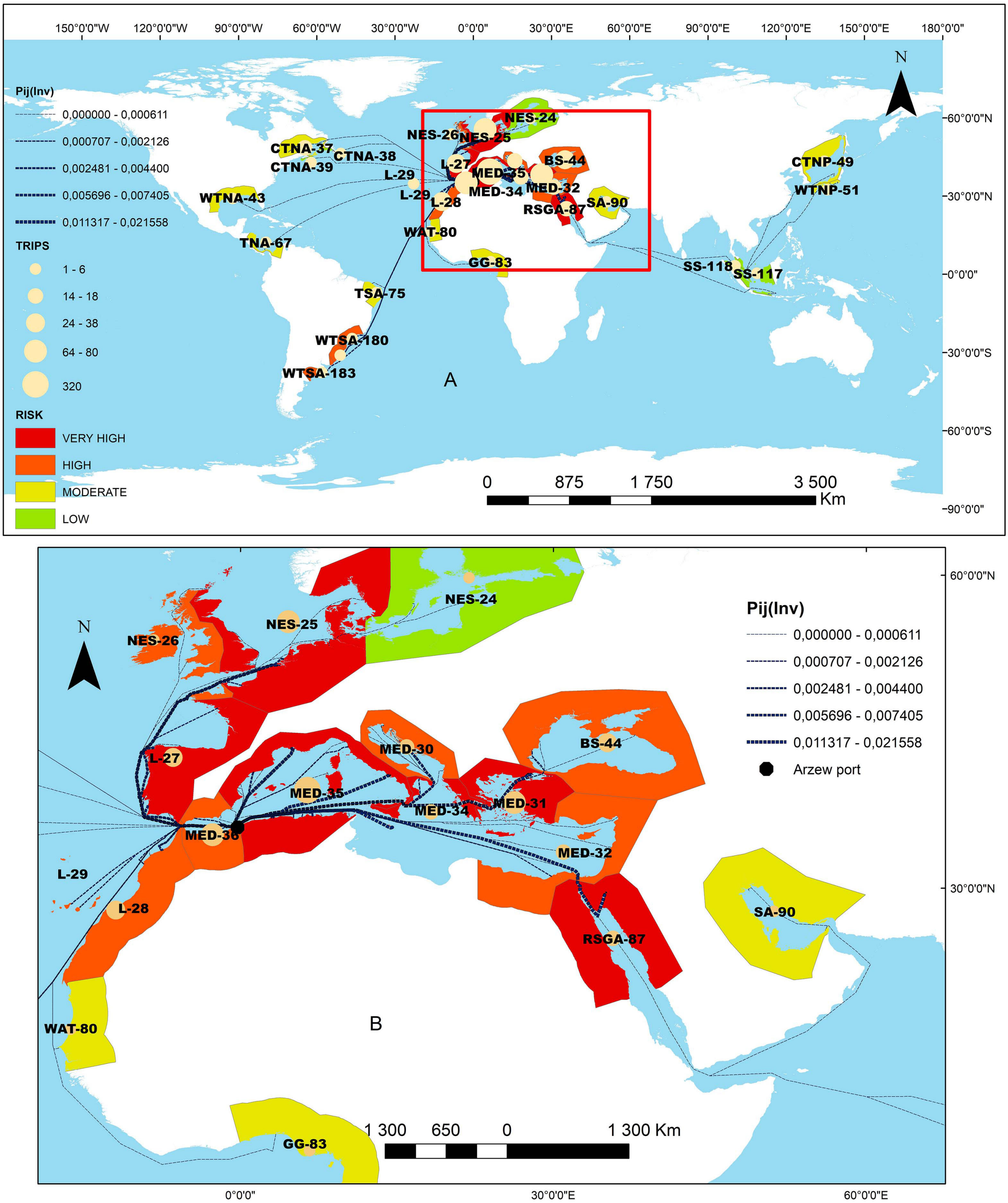

Figure 4. A: Risk of invasion by biofouling in the ship's hull for all donors ecoregion in the word. The pink circles represent the number of trips from each ecoregion. The lines show the risk of invasion by donor port *i*. The ecoregion number and the initials of the biogeographic province coded Ecoregions. B: Zoom in for Mediterranean Sea, North Atlantic Sea, Red sea and Gulf of Aqaba. Green indicates low risk ($0 \leq P_{ij}(Inv) < 10^{-9}$), yellow moderate risk ($10^{-9} \leq P_{ij}(Inv) < 10^{-4}$), orange high risk ($10^{-4} \leq P_{ij}(Inv) < 10^{-2}$), red very high risk ($P_{ij}(Inv) \geq 10^{-2}$).

at 20 marinas across France, Italy, Malta, Greece, and Turkey; species that could be introduced through the hulls of commercial vessels. The ports of the bioregions MED-30 to MED-36 are known as hubs, receiving arrivals from Asia. Taking into account the environmental similarity of the Mediterranean

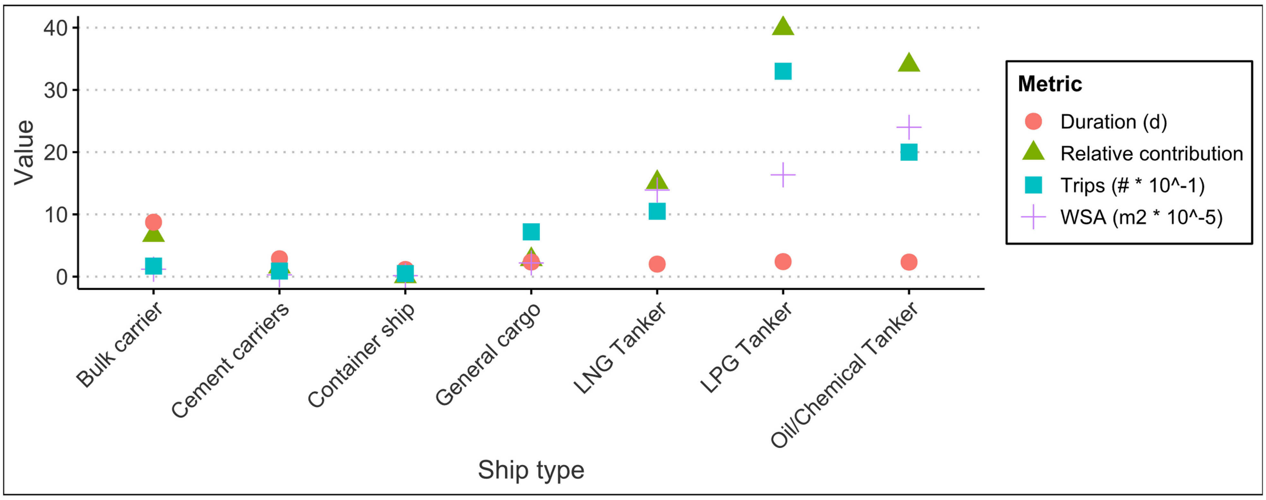

Figure 5. Risk invasion by biofouling for different ships types. The red circles shows the average duration of stay in the ports of origin, the blue square represents the number of aggregated voyages by type of ships, the cross represents the maximum wetted surface area accumulated from the different ports and the green triangle represents the relative contribution of each category of vessel to the risk of invasion $P_{ij} = P_{ij}(Inv)/\sum_i P_{ij}(Inv)$.

with some Asian seas (e.g., Japan Sea and Gulf of Aqaba), and the process of secondary introduction of NIS, these ecoregions could constitute a major risk for the Western Mediterranean, especially the island ecosystems and marine protected areas of the region. This classification is in line with Zenetos et al. (2020) who classified the Gulf of Saronikos in Greece as a hotspot of bioinvasion in the Mediterranean, with 89 NIS including 55 established species and 8 invasive species present in the region. The classification of these areas as bioinvasion hotspots is also consistent with Bouda et al. (2016); Bouda et al. (2017) for ballast water and the wetted surface of vessels.

Risk of invasion by ship type

The risk of invasion has been aggregated by vessel type (Figure 5). The analysis of invasion risk by vessel type shows that the three ship categories LPG Tanker, Oil/Chemical Tanker and LNG Tanker present the highest risk of invasion through biofouling (Figure 5, Table S3), with invasion probabilities: *Pij(Inv)* = 0.08, 0.07 and 0.03, respectively. Figure 5 shows that the risk of invasion is proportional to the wetted area of the vessels; the larger the wetted area of a vessel, the greater the risk of invasion through biofouling of the hull. These three categories of vessels accumulated the largest wetted area compared to the other categories of vessels (WSA = 23.99 × 10^5 m² for Oil/Chemical Tanker, WSA = 16.35 × 10^5 m² for LPG Tanker and WSA = 13.91 × 10^5 m² for LNG Tanker). Another category of vessels that represent a significant risk of invasion by fouling are "Bulk carriers", which have the greatest length of stay in ports (8.74 days). Proportionally, the contribution of each vessel category to the risk of invasion is: LPG Tanker (39.87%), followed by Oil/Chemical Tanker (34%), LPG Tanker (15.14%) and Bulk carriers (6.65%). This can be explained by

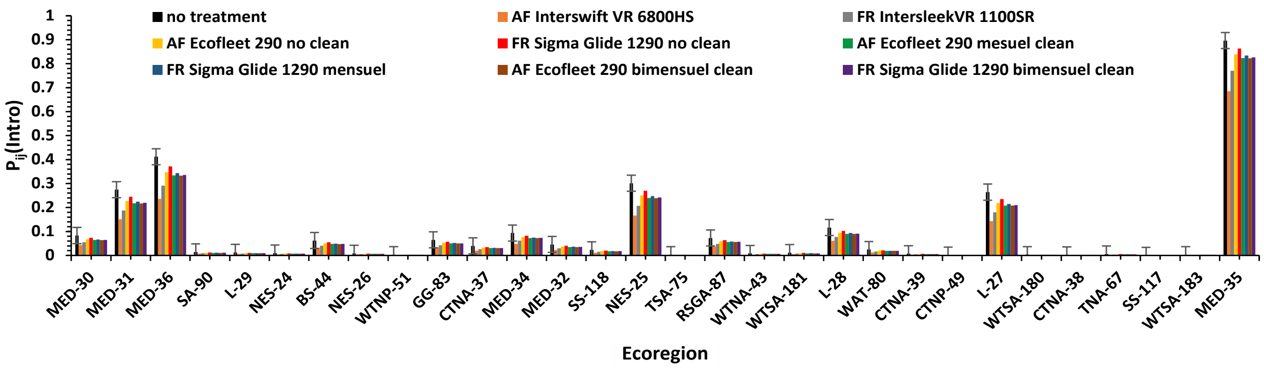

Figure 6. Treatment effort by different antifouling strategies and the reduction in probability of introduction of NIS by biofouling for the different ecoregions. FR: Free-releases coating, AF: Antifouling coating with biocides.

the increased fouling accumulation of these categories of vessels, given their high gross tonnage which gives them a larger wetted area and the high number of trips these vessels take. Indeed, LPG Tanker made 330 trips to the port of Arzew, followed by Oil/Chemical Tanker (200 trips), LNG Tanker (105 trips) and Bulk carriers (17 trips). This difference can also be interpreted by the efficiency of the antifouling system which varies according to the type of vessel, with bulk carriers having a higher efficiency compared to tankers and container ships.

Risk of invasion by antifouling strategy

Treatment of biofouling in each port of origin can influence the likelihood of introduction. By changing the *Pr* coefficient, several scenarios can be modeled (Figure 6).

By complying with the standards proposed by IMO (2011, 2012), we tested the effect of antifouling strategies on the reduction of the probability of introduction. The use of AF coating (InterswiftVR 6800 HS, AkzoNobel) in a donor port can reduce the risk of ecoregions which causes a great risk of introduction to the Arzew port by: 24% for MED-35, 43% for MED-36, 45% for NES-25, 45% for MED-31, and 46% for the South European Atlantic Shelf ecoregion; 48% for Saharan Upwelling. The FR coating (IntersleekVR 1100SR, AkzoNobel) shows a reduction of 14% for the West Mediterranean ecoregion, 29% for MED-36, 31% for NES-25, 31% for the Aegean Sea ecoregion, 32% for L-27, and 34% for L-28 (Figure 6, Table S4). Under conditions of no hull cleaning, the AF coating (Ecofleet 290) reduces the probability of introduction by 7% for MED-35, 16% for MED-36, 17% for NES-25, 17% for MED-31, 17-19% for both ecoregions L-27 and L-28, 18.79% for RSGA-87. For its part, the FR coating (Sigma Glide 1290) without cleaning the hull can reduce the probability of introduction by 12% for the whole maritime trajectories. For monthly hull cleaning, the AF (Ecofleet 290) coating offers a 22% reduction in total risk per ecoregion compared to a 20% reduction for the FR (Sigma Glide 1290) coating. In the case of bi-monthly hull cleaning, the AF (Ecofleet 290) coating reduces the

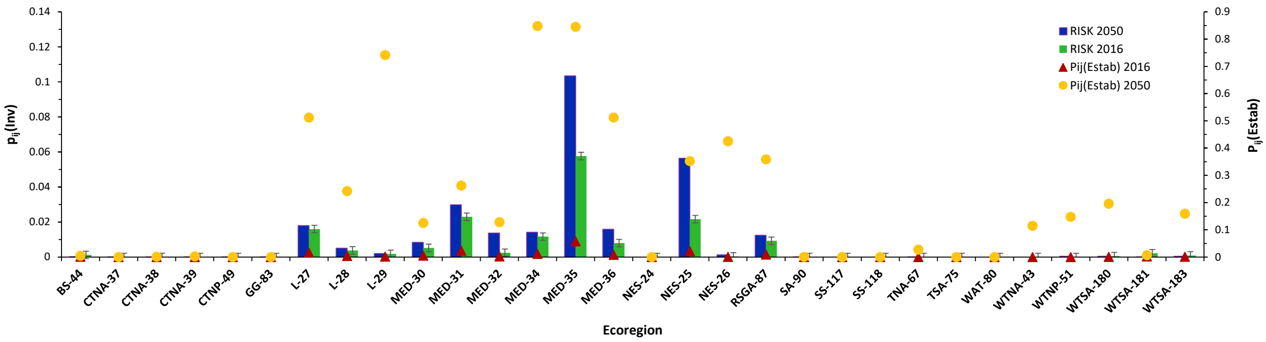

Figure 7. Risk invasion by biofouling of ships for all ecoregions, shown for 2016 and 2050. Black and green circles show the probabilities of biota establishment for 2050 and 2016 respectively.

total risk by 22.5% per ecoregion versus a reduction of 22% for the FR (Sigma Glide 1290) coating. Our calculations show that the risk of global invasion towards the port of Arzew could be reduced by 21% if the fouling is treated with an effort of 45% by the AF coating (InterswiftVR 6800HS, AkzoNobel) at the 10 ports presenting the highest risk and by 100% if it is treated with the same effort in the 6 most threatening ecoregions.

Effect of environmental variables on risk of invasion

Species, due to their evolutionary history and common characteristics, are separated by biogeographic barriers. These barriers are essentially governed by abiotic parameters, such as temperature and salinity. Alteration of these parameters allows species to either expand or shrink their natural range. Based on these fundamental assumptions of species distribution, we test the survival capacity of the fouling introduced by the hulls of ships in the Alboran Sea, using 2050 predictions. The Mahalanobis distance calculated for three environmental variables (temperature, salinity and current velocity) will be expressed as the probability of establishment of the biota to analyze the effect of its change on the probability of invasion (Figure 7).

We identify a very large increase in the probability of invasion at the port of Arzew for the year 2050 (Figure 7). This increase is proportional to the increase in environmental parameters. The increase in the probability of establishment accelerates the phenomenon of biota homogenization. Figure 7 shows an increase in the risk of invasion of the ecoregions for the same shipping effort in Arzew: 12.47% for ecoregion L-27, 33.82% for L-28, 9.95% for L-29, 60.52% for MED-29, MED-30, 90% for MED-31, 455.83% for MED-32, 21.45% for MED-34, 79.32% for MED-35, 78.77% for NES-24, 160.73% for NES-25, 241.77% for the Celtic Sea ecoregion NES-26 and 33.45 for the Red Sea ecoregion. Some ecoregions such as Black Sea "BS-44", Gulf of Central Guinea "GG-83" show a reduced risk of invasion. These results are consistent with fundamental theories of global biota homogenization and predictions of Seebens et al. (2020), which predict a 37% increase in the number of NIS globally by 2050. The increase in environmental dissimilarity plays an important role in species survival, particularly fouling species. The passage of ships through leaching areas that are environmentally dissimilar to the Alboran Sea will cause environmental shock and stress to organisms attached to the hull, thus contributing to the mitigation of the risk of invasion.

Conclusions

Biofouling of ship hulls is an important source of NIS spreading into new environments. A greater mechanistic and quantitative understanding of the risks of invasion are desperately needed. We have used an innovative approach to estimate the risk of this vector in the port of Arzew as a case study location. We show that for a total of 738 voyages, the risk of invasion is considerable. The results presented in this paper can be used by international governmental authorities such as the International Maritime Organization and by local authorities of IMO member countries. In particular, our results are useful for Algerian authorities to establish specific laws and regulations on large tonnage vessels such as LNG and LPG Tanker, Oil/Chemical Tanker, in order to minimize the transfer of NIS around the world. This modeling approach developed from experimental data from several global studies can be applied to ports around the world by adjusting certain parameters and adding other factors known to influence fouling elsewhere. Better knowledge of biofouling rates on ship hulls and the composition of biofouling communities will improve the accuracy of models in the future. In addition, the acquisition of a large set of vessel traffic data and environmental data with high resolution, will greatly assist in better understanding the extent of the risk of introduction and invasion of fouling species. We demonstrate that the treatment of fouling in a few high-risk donor ports could reduce the risk of overall invasion significantly. Similarly, the passage of vessels in areas with a large environmental difference with the receiving environment will significantly reduce the rate of fouling introducing NIS. The choice of a good antifouling system is also a crucial element in reducing hull fouling. For example, cleaning the hulls of ships in the water, without going to the shipyard, will save both cost and time for the ship manager and operator and can be effective in reducing the rate of fouling. For vessels operating in the same areas, the risk is much higher because of the great environmental similarity between adjacent areas. For example, the introduction of regulations at regional level, forcing vessels calling at several Mediterranean ports at the same time to clean their hulls by passing them through leaching areas (such as Black and Baltic sea) at regular intervals, or by regular underwater hull cleaning, would have a significant effect on fouling and invasive species. Overall the study has contributed to the understanding of the impact of ship-borne fouling, is consistent with work on NIS in the Mediterranean, and provides a framework for similar work in other regions around the world.

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Authors' contribution

All authors contributed to the study conception and design. AC, AB, MS, BB, FB, LN, NB – material preparation, data collection and analysis. The mathematical modelling was done by AK and supervised by AB, NB, LN and FB. The R script was generated by AK and MS. The English was revised by MS. All draft of the manuscript was written by AK and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Supplementary material

The following supplementary material is available for this article:

Table S1. Synthesis of polynomial models generated for temperate and tropical regions.
Table S2. Calculation of *Pij (Estab)* and Mahalanobis distance by Ecoregion (Figure 2).
Table S3. Data used to develop Figure 5.
Table S4. Data used for build Figure 6.
Table S5. Calculation of *Fr* reduction rate (antifouling paint).
Figure S1. Correlation of environmental variables.
Figure S2. Polynomial regression applied to temperate data.
Figure S3. Polynomial regression applied to tropical data.
Figure S4. Biofouling survival modelled as a function of velocity.
Figure S5. Biofouling cover with varying in-water durations and different coatings.
Figure S6. Percentage of fouling coverage for different antifouling coatings (AF) and (FR).
Figure S7. Proportion of fouling for AF and FR coatings according to the cleaning of the hull.
Figure S8. Correlation matrix between *Pij(Inv*), *Pij(Alien)* and *Pij(Intro)*.

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