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-RESEARCH ARTICLE-

The Factors Increasing of Invasiveness Potential of Five Pufferfishes in the Eastern Mediterranean, Turkey

Halit Filiz[1*], Sercan Yapıcı[1], Gökçen Bilge[1]

[1] Muğla Sıtkı Koçman University, Faculty of Fisheries, 48000, Kötekli, Menteşe, Muğla

Abstract

The Aquatic Species Invasiveness Screening Kit (AS-ISK) has been used in order to exhibit the factors increasing the invasiveness of Pufferfishes on the Muğla coast. Basic Risk Assessment (BRA) scores were calculated as 28.0 (*Lagocephalus guentheri*), 31.0 (*Torguigener flavimaculasus*), 33.0 (*Lagocephalus suezensis*), 38.0 (*Lagocephalus spadiceus*) and 40.0 (*Lagocephalus sceleratus*) and these scores were indicated a high risk of invasiveness for the species. The factors increasing overall AS-ISK scores were; high climate match, tolerance of a wide range of environmental conditions, flexibility in utilizing food resources, high fecundity, small size at maturity, high reproductive effort, reliable data about parasite transmission and high invasiveness potential elsewhere while factors decreasing scores were; no hybridization with native fish, and no parental care. This information is expected to allow managers and agencies that are responsible for risk assessment and management of lessepsian/invasive species to perform a better decision-making.

Keywords:

AS-ISK, pufferfish species, biological invasion, Muğla coast

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Introduction

Migration of biota from Red Sea to Mediterranean waters via Suez Canal was named Lessepsian migration (Por, 1978); it is ongoing with both the positive and the negative effects (Farrag et al., 2016). According to most recent data, there are 165 non-indigenous marine fish (N-IMF) species

[*] *Corresponding author: Halit Filiz, e-mail: halit.filiz@mu.edu.tr*

in the Mediterranean (CIESM, 2017). Turkey, on the other hand, has 52% (85 sp.) of these N-IMF species in its ichthyofauna, and seven of them were pufferfishes (family Tetraodontidae) (Filiz et al., 2017a). Pufferfishes are mostly found benthic habitats ranging from 0 to 274 m on sandy and muddy substrate (IUCN, 2017; Froese & Pauly, 2017).

The application of species-risk analysis was on potentially invasive freshwater fish (Copp et al., 2005) and is known by the abbreviation FISK (Fish Invasiveness Scoring Kit). This tool has been replaced by a generic decision-support tool for screening all plants and animals in marine, brackish and fresh waters: The Aquatic Species Invasiveness Screening Kit (AS-ISK: Copp et al., 2016) that has been developed to incorporate the 'minimum requirements' (Roy et al., 2014) for the assessment of species with regard to the recent EU Regulation on the prevention and management of the introduction and spread of invasive alien species (European Commission, 2014). AS-ISK and FISK v2 have been used successfully for the screening of non-native and translocated fish species in the freshwater systems (e.g. Tarkan et al., 2014; 2017). Risk screening practices are part of risk assessment systems and used to estimate a potential invader colonizing into a new environment (Daehler et al., 2004). Risk screenings are based on a synthesis of information about the biological, ecological and developmental characteristics of a target organism and the bio-geographical region in which it is found or may be found (Pheloung et al., 1999). Risk scanning tools have a wide range of use and application strategies. These systems can be particularly useful in distinguishing between a number of potential invasive and non-invasive alien species with a fast and efficient way (Baker et al., 2007). In addition, risk screening tools can also be very useful in recognizing shortcomings in the quality and reliability of data in the literature (Copp et al., 2009), which plays an important role in determining management and research priorities. Even in some countries, risk scans are used when the importation status of some non-native species has been decided (Pheloung et al., 1999). More commonly, risk scans make initial assessments of the species studied and then decide whether to conduct further risk analysis and management actions (Gordon et al., 2008).

The invasion of pufferfishes in the Mediterranean Sea resulted important ecological and socio-economic impacts. The aim of the present study was therefore to assess the invasive potential of five Lessepsian pufferfish (*Lagocephalus spadiceus*, *Lagocephalus sceleratus*, *Lagocephalus suezensis*, *Lagocephalus guentheri* and *Torquigener flavimaculosus*) distributed on the Muğla coast using a tool recently developed The Aquatic Species Invasiveness Kit (AS-ISK). Notably, the outcomes of the present study are expected to assist local environmental managers and stakeholders in the implementation of suitable policies for the prevention and management of existing and potential Lessepsian pufferfishes in the Mediterranean Sea.

Material and Methods

Before starting the risk screening, through literature reviews were carried out to obtain all available information on bio-geographical and historical traits (AS-ISK Section 1) and biological and ecological characteristics (AS-ISK Section 2) of puffer fish, together yielding Basic Risk Assessment (BRA) score. Peer-viewed publications were priority sources in retrieving this information, with Internet databases, dissertations and, occasionally, reports used whenever necessary to fill in gaps in the peer-reviewed literature. For Climate Change Assessment (CCA) section (AS-ISK Section 3), Demir et al. (2008), Poursanidis (2015) and Turan et al. (2016) that provided several likely future scenarios for the Mediterranean region were used. Notably, most scenarios in these paper predicted 0.5 and 1˚C increase of air temperature in near future (i.e. next

fifty years). Likely future change in water temperature was calculated from this prediction based on the relationship between water temperature (T_w) and air temperature (T_a) as per Erickson & Stefan (1996): $T_w = 3.47+0.898T_a$.

Using AS-ISK v1 (available at https://www.cefas.co.uk/nns/tools/), the Muğla coast was identified as the risk assessment (RA) area. Assessment was carried out by the second author of the study, who is knowledgeable in the Lessepsian/invasive marine fish fauna in the eastern Mediterranean. As each response in AS-ISK for a given species is allocated a confidence category (1= low; 2=medium; 3=high; 4=very high), a confidence factor (CF) was computed as:

$$\sum(CQi)/(4 \times 55)\ (i = 1, \ldots, 55)$$

where CQ*i* is the certainty for question *i*, 4 is the maximum achievable value for certainty (i.e. 'very certain') and 55 is the total number of questions comprising the AS-ISK tool. The CF therefore ranges from a minimum of 0.25 (i.e. all 55 questions with certainty score equal to 1) to a maximum of 1 (i.e. all 55 questions with certainty score equal to 4).

Results

The highest BRA-scoring species *L. sceleratus* also achieved the highest CCA score, and was followed by other high risk species such as *L. spadiceus*, *L. suezensis*, *T. flavimaculosus* and *L. guentheri* (Table 1). This suggests that risks of entry, dispersal and establishment, impact on biodiversity, ecosystem structure and socio-economic factors of pufferfishes in the eastern Mediterranean under predicted future climatic conditions are highly likely.

Statistics	**Scores**				
	L. sceleratus	*L. spadiceus*	*L. suezensis*	*L. guentheri*	*T. flavimaculosus*
BRA Score	40.0	38.0	33.0	28.0	31.0
CCA Score	52.0	48.0	45.0	40.0	43.0
Score partition					
A. Biogeography/Historical	**13.0**	**13.0**	**13.0**	**13.0**	**13.0**
1. Domestication/Cultivation	2.0	2.0	2.0	2.0	2.0
2. Climate, distribution and introduction risk	2.0	2.0	2.0	2.0	2.0
3. Invasive elsewhere	9.0	9.0	9.0	9.0	9.0
B. Biology/Ecology	**27.0**	**25.0**	**20.0**	**15.0**	**18.0**
4. Undesirable (or persistence) traits	11.0	10.0	6.0	8.0	10.0
5. Resource exploitation	7.0	6.0	5.0	2.0	2.0
6. Reproduction	2.0	2.0	2.0	2.0	2.0
7. Dispersal mechanisms	4.0	4.0	4.0	3.0	4.0
8. Tolerance attributes	3.0	3.0	3.0	0.0	0.0
C. Climate change	**12.0**	**12.0**	**12.0**	**6.0**	**12.0**
9. Climate change	12.0	12.0	12.0	6.0	12.0
Confidence factor	0.80	0.80	0.80	0.79	0.80

Table 1. AS-ISK (v1) scoring outputs for pufferfishes on the Muğla coast.

The factors increasing overall AS-ISK scores of the pufferfishes assessed were; high climate match, tolerance of a wide range of environmental conditions, flexibility in utilizing food resources,

high fecundity, small size at maturity, high reproductive effort, reliable data about parasite transmission and high invasiveness potential elsewhere while factors decreasing scores were; no hybridization with native fish, and no parental care.

Discussion

When we look at the use of AS-ISK for invasive marine fishes in the Mediterranean, only three attempts (Uyan et al., 2016; Bilge et al., 2017; Filiz et al., 2017b) have been made for marine Lessepsian fish species. Bilge et al. (2017) first attempted to study of AS-ISK for Lessepsian fishes (for 45 sp.) in the eastern Mediterranean, Turkey, and calculated a basic score threshold of 18.5. According to Basic Risk Assessment (BRA) scores of the 45 species assessed, only one was ranked as 'low risk' (-20.0>BRA>0.99), 28 were categorised as 'medium risk' (1.0>BRA>18.4), and the remaining 16 as 'high risk' (BRA≥18.5). Pufferfishes are among the “high risk” invasive Indo-Pacific origin species in the Mediterranean Sea with their calculated BRA scores (Table 1).

Main factors increasing overall AS-ISK scores of pufferfishes were their tolerance of a wide range of environmental conditions, high invasiveness potential elsewhere, and high climate match. Indeed, pufferfishes quickly invaded the Mediterranean Sea. *Lagocephalus spadiceus* was first recorded from Iskenderun Bay (Turkey) by Kosswig (1950), and expanded its distribution to Sea of Marmara (Tuncer et al., 2008) in fifty-eight years. *Lagocephalus sceleratus* was first recorded from Turkish water by Filiz and Er (2004), then quickly spread north and west along the coasts of Mediterranean basin, and reached to the coasts of Spain (Izquierdo-Muñoz & Izquierdo-Gomez, 2014). *Lagocephalus suezensis* was first recorded in the Mediterranean Sea in 1977 from the Lebanon coast [where it was misidentified as *L. sceleratus* by Mouneimné (1977)], then recorded recently from Libya (Ben-Abdallah et al., 2011). Although there have been nomenclature issues with *Lagocephalus guentheri* in the Mediterranean Sea (see Farrag et al., 2016 for details), it seemed that the first confirmed record for *L. guentheri* in the Mediterranean Sea was provided by Farrag et al. (2016) from Egyptian waters and then reported from Turkey by Akyol and Aydın (2016). *Torquigener flavimaculosus* first recorded in Haifa Bay, Israel, (Golani, 1987), and subsequently was detected in Turkish waters (Bilecenoglu, 2003), and Greek waters (Corsini-Foka et al., 2006). As concerns the distinction between invader and colonizer Indo-Pacific species, it is evidenced by the ability of invader species to exploit local indigenous resources faster than native species and to form rapidly large populations which spread at the shallow coastal habitats (see the recent case *L. sceleratus*) (EastMed, 2010). Concerning their status, the majority of the assessed pufferfishes (*L. spadiceus*, *L. sceleratus*, *L. suezensis* and *T. flavimaculosus*) were classified established and abundant (Filiz et al., 2017a) on Mugla coast.

Judging from the recent increase in pufferfishes in the eastern Mediterranean Sea, its few natural predators, and its ability to adapt to a range of habitats, we faced a rapid expansion throughout the Mediterranean Sea resulted significant impacts on local ecosystems and fisheries. The invasion of pufferfishes in the Mediterranean Sea and the potential ecological and socio-economic impacts that may follow have been largely neglected by the regional scientific community, managers and other stakeholders. They have some negative effects on the fisheries since both they are poisonous and can become quite abundant. The serious damages in the fishing gears, the loss in the catch (especially by *L. sceleratus*) and the replacement of commercially important species in the catches, reducing significantly fisheries income because of pufferfishes were noted (EastMed, 2010). Indo-Pacific pufferfishes contained tetrodotoxin (TTX,) which is known a non-protein organic compound (aminoperhy-droquinazoline) and one of the strongest

marine paralytic toxins today (Halstead, 1978). TTX can be found in the liver, gonads, intestines, and skin of these fish and can cause death in approximately 60% of persons who ingest it (Ellenhorn & Barceloux, 1988). Educating the local public was also important since for example pufferfish is a toxic species and there is a high risk of death if people consume individuals of this species (EastMed, 2010). In accordance with this, the high risk ranking of pufferfishes found in BRA in AS-ISK seems appropriate due to its establishment. Additionally, given predicted climate change scenarios for the Mediterranean, establishment success of these species is expected to exacerbate according to climate change assessment (CCA) in AS-ISK. Indeed, in native ranges, pufferfishes thrive in tropical waters, so the predicted increases in water temperature of the Mediterranean resulted in positive responses to all of questions on how future climatic conditions are likely to affect the invasiveness of pufferfishes in the risk assessment area (Table 1). For example, the mean surface water temperatures of the Aegean Sea have gradually increased from 18.2 °C (1970-1979 period) to 19.0 °C (2008-2016 period) in the last 47 years (MGM, 2016). It is known that the number of introduced Lessepsian fish species is correlated significantly and positively with the Mediterranean water temperature (r= 0.77, p < 0.05; Ben Rais Lasram et al., 2010). On the contrary, recent data are suggesting that *L. sceleratus* occur also in the cold north Aegean waters (over the isotherm of 14°C) (EastMed, 2010).

The other factors increasing overall AS-ISK scores were flexibility in utilising food resources, high fecundity, small size at maturity, high reproductive effort, and reliable data about parasite transmission. Since there is no species specific diet data, except *L. sceleratus*, we used in the analysis only this species' available data. Stomach content analysis of *L. sceleratus* show that this species will share with other demersal carnivore species' food in the Mediterranean. It mainly feed on shrimps, fishes, crabs, squids and cuttlefish that densely inhibits in the habitat (Sabrah et al., 2006; EastMed, 2010; Aydın, 2011; Kalogirou, 2013). In the present study, the data about fecundity and size at maturity only obtained for *L. sceleratus* from literatures. The size at 50% maturity of females in the Mediterranean were reported as 36 cm (Kalogirou, 2013), 36.3 (Amira et al., 2015), 41 cm (Farrag, 2014), and 41.9 and 48.8 cm (Rousou et al., 2014). In the Gulf of Suez, *L. sceleratus* reached maturity during at a size of 37.1 (Amira et al., 2015) and 43.3 cm (Sabrah et al., 2006). The Mediterranean population seemed to be attain maturity earlier, except Rousou et al. (2014), than Gulf of Suez population, may reflect the reproduction adaptability when found favourable conditions as in the Mediterranean. Females *L. sceleratus* in the eastern Mediterranean released approximately 776 (Farrag, 2014), 781 (Aydın, 2011) and 854 (Amira et al., 2015) eggs g^{-1}. By comparing the relative fecundity, the results showed that the number of eggs of *L. sceleratus* were higher in eastern Mediterranean habitat than that recorded in the Gulf of Suez (619 eggs g^{-1}Amira et al., 2015). Monthly variations in GSI indicated an increase from March till June, meaning *L. sceleratus* has an extensive spawning period in early summer with a peak in June (Aydın, 2011). Overall, quantitative analyses of reproductive characteristics for pufferfishes in the Mediterranean will enhance our understanding of how reproduction supports the spread and establishment of this invader. These risks may be compensated by local mitigation factors such as predation. However, dramatic invasive success of pufferfishes result from a combination of factors such as early maturation and reproduction, anti-predatory poisonous defences and ecological versatility, coupled with native prey and the overfishing of native predators as indicated before by Côté et al., 2013 for the lionfish (*Pterois* spp.). Furthermore, the construction of a deeper and wider Suez Canal is expected to increase invasion rates (Galil et al., 2015). Recently, reliable data about parasite transmission were provided by Özak et al. (2012) and Bakopoulos et al. (2017).

The information obtained this study is expected to allow managers and agencies that are responsible for risk assessment and management of Lessepsian/invasive species to perform a better decision-making. In the case of pufferfishes, the results derived from AS-ISK suggests a more detailed (i.e. full) risk assessment is necessary (cf. NAPRA: Baker et al., 2007). Specifically, lower confidence factors observed in AS-ISK assessments on undesirable traits, resource exploitation, reproduction and dispersal mechanism of the pufferfishes call for the need for more detailed studies on these subjects.

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