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RESEARCH ARTICLE

Assessment of invasiveness potential of *Pterois miles* by the Aquatic Species Invasiveness Screening Kit

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Abstract

Risk screening tools to identify species with a high or low risk of invasiveness are being increasingly used for effective management purposes. Whilst non-native freshwater fish species have been studied more widely, there is little effort for implementing the risk-screening tools for Lessepsian or invasive marine fish species in the Mediterranean. The aim of the present study was therefore to assess the invasiveness risk of *Pterois miles* in the eastern Mediterranean using the recently-developed Aquatic Species Invasiveness Screening Kit (AS-ISK). Calculated Basic Risk Assessment (BRA= 45.5) and Climate Change Assessment (CCA= 57.5) scores indicated a high risk of invasiveness of *P. miles* for the Mediterranean. The factors increasing overall AS-ISK scores were; high climate match, tolerance of a wide range of environmental conditions, flexibility in utilising food resources, high fecundity, small size at maturity, high reproductive effort and high invasiveness potential elsewhere while factors decreasing scores were; no hybridization with native fish, no parental care, and no data about parasite transmission. This information is expected to allow managers and agencies that are responsible for risk assessment and management of *P. miles* to perform a better decision-making.

Keywords: AS-ISK, Lessepsian fish, biological invasion, Mediterranean

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Introduction

Pterois miles (Bennett, 1828) naturally occurs in the Indian Ocean from South Africa to the Red Sea and Persian Gulf, and east to Sumatra (Froese and Pauly 2016). *P. miles* is currently considered amongst the most successful marine invaders in the history of aquatic invasions (Bariche *et al.* 2013). In the Mediterranean Sea, a single specimen of *P. miles* was recorded from the Levantine coast in 1991 (Golani and Sonin 1992). In 2012, two *P. miles* individuals were captured separately off the village of Al Minie (34°29'26.15''N; 35°54'47.73''E) in the northern part of Lebanon (Bariche *et al.* 2013). Soon after, additional sightings were reported along the coasts of Cyprus (Iglésias and Frotté 2015; Oray *et al.* 2015; Kletou *et al.* 2016), Turkey (Turan *et al.* 2014; Turan and Öztürk 2015) and Rhodes (Crocetta *et al.* 2015) between 2014 and 2015. These records were reported indicating a westerly migration of the species towards the Aegean Sea. To confirm this, a specimen of *P. miles* has recently been recorded from the southern Aegean Sea (off Datça) (Bilge *et al.* 2016). Previously, it has been suggested two introduction ways for *P. miles*: (*i*) entered through the Suez Canal, like other hundreds of marine organisms (Zenetos *et al.* 2012), and (*ii*) released from captivity (Golani *et al.* 2002). Considering that *P. miles* is a common fish in the Red Sea and the proximity of the Suez Canal to the recent sightings, the Suez Canal seems to be the most likely pathway for the introduction of the species into the Mediterranean Sea (Bariche *et al.* 2013). The recent findings of *P. miles* may be an indication of a new wave of arrivals of the species in the Levant, raising justifiable concerns of a possible onset of a new invasion in the Mediterranean Sea (Bariche *et al.* 2013).

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). The characteristic features of the risk screening methods include question-answer format information scans, use of simple computer programs, high reliability estimates, and flexibility in usage for many different taxonomic groups (Pheloung *et al.* 1999; Daehler *et al.* 2004). 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 (Kolar and Lodge 2002; Daehler *et al.* 2004; Baker *et al.* 2007; Gordon *et al.* 2008). The first 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. Almeida *et al.* 2013; Copp 2013; Simonović *et al.* 2013; Vilizzi and Copp 2013; Tarkan *et al.* 2014; 2017). However, only one attempt (Uyan *et al.* 2016) has been made for marine Lessepsian fish species, *Nemipterus randalli*, in the Mediterranean Basin. It is expected that the native/endemic fish fauna of the seas can be affected to a different extent by the introduction of non-native fish species. Clearly, there is a need to assess risks posed by non-native fishes at local scales.

The aim of the present study was therefore to assess the invasive potential of Lessepsian *Pterois miles* in the eastern Mediterranean Sea (EMS) using a tool recently developed, AS-ISK, and to evaluate the applicability of AS-ISK to a large risk assessment (RA) area for a single fish species. 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 potential, existing and future undesired translocations of Lessepsian/invasive fishes in the Mediterranean Sea.

Materials and Methods

Before starting the risk screening, through literature reviews were carried out to obtain all available information on biogeographical and historical traits (AS-ISK Section 1) and biological and ecological characteristics (AS-ISK Section 2) of *P. miles*, 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) and Poursanidis (2015) 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 and Stefan (1996): $T_w = 3.47 + 0.898T_a$.

Using AS-ISK v1 (available at https://www.cefas.co.uk/nns/tools/), the eastern Mediterranean Sea was identified as the risk assessment (RA) area. Assessment was carried out by the first 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:

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

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

Overall, the results of the assessment indicated that *P. miles* under study have a high chance to survive and establish in the eastern Mediterranean. Considering the high Basic Risk Assessment (BRA) score 45.5, *P. miles* should have a high risk at different levels for almost all ecoregions of the Mediterranean. Climate Change Assessments (CCA) revealed score of 57.5 meaning maximum score (12) on top of BRA score of 45.5. This suggests that risks of entry, dispersal and establishment, impact on biodiversity, ecosystem structure and socio-economic factors of *P. miles* in the eastern Mediterranean under predicted future climatic conditions are highly likely (Table 1).

Table 1. AS-ISK (v1) scoring output for *Pterois miles* in the eastern Mediterranean

Statistics	**Scores**
BRA Score	45.5
CCA Score	57.5
Score partition	
A. Biogeography/Historical	**19.5**
1. Domestication/Cultivation	4.0
2. Climate, distribution and introduction risk	2.0
3. Invasive elsewhere	13.5
B. Biology/Ecology	**26.0**
4. Undesirable (or persistence) traits	9.0
5. Resource exploitation	7.0
6. Reproduction	3.0
7. Dispersal mechanisms	4.0
8. Tolerance attributes	3.0
C. Climate change	**12.0**
9. Climate change	12.0
Confidence factor	0.65

The factors increasing overall AS-ISK scores were; high climate match, tolerance of a wide range of environmental conditions, flexibility in utilising food resources, high fecundity, small size at maturity, high reproductive effort and high invasiveness potential elsewhere whereas factors decreasing scores were; no hybridization with native fish, no parental care, and no data about parasite transmission (Table 2). Finally, the assessor confidence in responses to questions was relatively high, with 60% of the responses ranging from 'high' to 'very high' confidence and with the most uncertain answers being related to questions regarding undesirable traits, resource exploitation and dispersal mechanism.

Discussion

We have faced a rapid invasion of *P. miles* in the Mediterranean, from a single specimen in 1991 to several sightings between 2014 and 2016 (for further information see Bilge *et al.* 2016). This increase is similar to the pattern of invasion recorded in some other areas (e.g. Albins and Hixon 2013). *P. miles* first appeared in the eastern Mediterranean at about the same time that they were first reported off Florida where it spread rapidly and colonised almost all warm parts of the east coast of United States, the Gulf of Mexico and the entire Caribbean Sea (Schofield *et al.* 2016) whereas more than two decades passed for a second sighting to occur in the Mediterranean Sea. For *P. miles* in the western Atlantic, Morris (2009) reported that females mature at approximately 175 mm total length or 1 year of age and release approximately 25000 eggs per spawning event. Based on the presence of hydrated oocytes, mature females appeared capable of spawning every 3.6-4.1 days throughout the year, although the proportion of females with ovaries in spawning condition was higher in summer (June-August). The combination of its high spawning frequency (year round, ~every 4 d) and protracted pelagic larval phase (~26 d, Ahrenholz and Morris 2010), coupled with release in a region with multiple oceanographic currents has resulted in the rapid dispersal of lionfishes into the western Atlantic Ocean (Fogg *et al.* 2013). Quantitative analyses of reproductive characteristics for *P. miles* in the Mediterranean will enhance our understanding of how reproduction supports the spread and establishment of this invader.

In 2008, *P. miles* were observed in the waters surrounding Little Cayman Island, and they were considered established there by 2009 (Schofield 2010). At this location, they have reached densities of up to 650 fish ha^{-1} (Frazer *et al.* 2012), which is far greater than the 26.3 fish ha^{-1} recorded in the native range (Kulbicki *et al.* 2012). Judging from the recent increase in *P. miles* in the eastern Mediterranean Sea, its few natural predators, the dispersal capabilities of their planktonic larvae and its ability to adapt to a range of habitats, we suspect that a rapid expansion throughout the Mediterranean Sea may soon be followed by significant impacts on local ecosystems and fisheries. In accordance with this, the high risk ranking of *P. miles* found in BRA in AS-ISK seems appropriate due to its potential for establishment, with the potential for associated adverse impacts. Additionally, given predicted climate change scenarios for the Mediterranean, potential translocation and establishment success of this species are expected to exacerbate according to climate change assessment (CCA) in AS-ISK. Indeed, in both its native and introduced ranges, *P. miles* thrives in warm 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 *P. miles* in the risk assessment area (Table 1). 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).

Controversially, lionfishes (*Pterois* spp.) can potentially spread and survive in a large part of the Mediterranean Sea because they have shown extensive dispersal capabilities and can survive to a minimum temperature of 10°C (Kimball *et al.* 2004).

These risks may be compensated by local mitigation factors such as predation. However, dramatic invasive success of *P. miles* results from a combination of factors such as early maturation and reproduction, anti-predatory venomous defences and ecological versatility, coupled with native prey and the overfishing of native predators (Côté *et al.* 2013). Furthermore, CO_2 emissions warming the Mediterranean Sea and the construction of a deeper and wider Suez Canal are expected to increase invasion rates (Galil *et al.* 2015).

The possibility of *P. miles* invasion 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. 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. In the case of *P. miles*, 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 and dispersal mechanism of the species call for the need for more detailed studies on these subjects.

Although risk identification tools such as FISK and its updated version AS-ISK have been largely used for freshwater fishes so far (Copp *et al.* 2009; Simonović *et al.* 2013; Lawson *et al.* 2013; Almedia *et al.* 2013; Tarkan *et al.* 2014), AS-ISK can be used for all relevant aquatic non-native organisms including in the marine environment as proved in the present study. Given increasing concern regarding Lessepsian fishes in the Mediterranean, this risk identification tool may be useful for identifying which non-native and translocated species in the Mediterranean are likely to become invasive. Using this tool is strongly recommended for multiple assessments of species that are likely to be invasive (e.g. Almeida *et al.* 2013). Besides, single assessor assessment should be avoided for more reliable results (e.g. Tarkan *et al.* 2014). Finally, this tool can be applicable also for small risk assessment areas such as separate ecoregions in the Mediterranean as previously done for river or lake basins (i.e. Glamuzina *et al.* 2017; Tarkan *et al.* 2017).

*Pterois miles'*in istila potansiyelinin sucul türlerde istilacılık tarama aracı ile değerlendirilmesi

Öz

Etkin yönetim için, bir türün istilacılık riskinin düşük ya da yüksek olduğunu belirlemede risk tarama araçlarının kullanımı gittikçe artmaktadır. Her ne kadar yerli-olmayan tatlısu balık türleri çok geniş olarak çalışılmışsa da, Akdeniz'de risk tarama araçlarının Lesepsiyen veya istilacı deniz balığı türleri için kullanımı bulunmamaktadır. Bu bağlamda, bu çalışmanın amacı doğu Akdeniz'deki *Pterois miles*'in istilacılık riskini son zamanlarda geliştirilmiş olan Sucul Türlerde İstilacılık Tarama Aracı (ST-ITA) kullanılarak değerlendirmektir. Hesaplanan Temel Risk Değerlendirme (TRD= 45.5) ile İklim Değişim Değerlendirme (İDD= 57.5) puanları Akdeniz için oldukça yüksek bir istilacılık riskini işaret etmiştir. Genel olarak ST-ITA puanlarını arttıran etkenler yüksek iklim benzerliği, çevre şartlarına olan yüksek tolerans, besin kaynaklarının kullanımındaki esneklik, yüksek fekondite, küçük boyda olgunluk, yüksek üreme çabası ve herhangi bir yerdeki yüksek istilacılık potansiyeli iken, puanları azaltan etkenler yerli türler ile melezleme olmaması, ailesel ilginin bulunmaması ve parazit taşınımı ile ilgili veriye rastlanmaması olarak bulunmuştur. Bu bilginin, Lesepsiyen/istilacı türlerin risk değerlendirme ve yönetiminde sorumlu olan yöneticiler ile ajanslara daha iyi bir karar vermede yardımcı olması beklenmektedir.

Anahtar kelimeler: ST-ITA, Lesepsiyen balıklar, biyolojik istilalar, Akdeniz

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Table 2. AS-ISK (v1) output report for the *Pterois miles* assessed for the eastern Mediterranean.

		Question	Response	Justification	Confidence
A. Biogeography/Historical					
1. Domestication/Cultivation					
1	1.01	Has the taxon been the subject of domestication (or cultivation) for at least 20 generations?	Y	They are thought to have been introduced to the western Atlantic in the mid 1990's (Ruttenberg *et al.* 2012).	2
2	1.02	Is the taxon harvested in the wild and likely to be sold or used in its live form?	Y	FishBase reports that it is usually used as commercially in the aquarium	3
3	1.03	Does the taxon have invasive races, varieties, sub-taxa or congeners?	Y	Few worldwide fish invasions of similar magnitude are documented; the introduction of the red lionfish *Pterois volitans* and the devil firefish *P. miles* in the western Atlantic is one of the fastest and most ecologically harmful marine fish introductions to date (Albins and Hixon 2013).	4
2. Climate, distribution and introduction risk					
4	2.01	How similar are the climatic conditions of the RA area and the taxon's native range?	2	Lionfishes can potentially spread and survive in a large part of the Mediterranean Sea because they have shown extensive dispersal capabilities and can survive to a minimum temperature of 10°C (Kimball *et al.* 2004).	3
5	2.02	What is the quality of the climate matching data?	2	New modelling approaches are needed (Hall-Spencer and Allen 2015).	2
6	2.03	Is the taxon already present outside of captivity in the RA area?	Y	The taxon is already in the risk assessment area.	3
7	2.04	How many potential pathways could the taxon use to enter in the RA area?	>1	It has been suggested that this specimen entered through the Suez Canal (Zenetos *et al.* 2012; Bariche *et al.* 2013), or released from captivity (Golani *et al.* 2002).	3
8	2.05	Is the taxon currently found in close proximity to, and likely to enter into, the RA area in the near future (e.g. unintentional and intentional introductions)?	Y	The taxon is already in the risk assessment area.	4
3. Invasive elsewhere					
9	3.01	Has the taxon become naturalised (established viable populations) outside its native range?	Y	Information from Golani and Sonin (1992), Bariche *et al.* (2013), Turan *et al.* (2014), Crocetta *et al.* (2015), Iglésias and Frotté (2015), Oray *et al.* (2015), Turan and Öztürk (2015), Kleteu *et al.* (2016), and Bilge *et al.* (2016).	4
10	3.02	In the taxon's introduced range, are there known adverse impacts to wild stocks or commercial taxa?	Y	According to Gardner *et al.* (2015), in the Caribbean and western Atlantic, the spread of lionfish exacerbates concern for the health of coral reefs already threatened by other stresses. For example, manipulative field studies in the Bahamas have shown that lionfish can reduce recruitment of native reef fish by up to 79%, with predation being the likely mechanism After lionfish invaded a mesophotic reef, Lesser and Slattery (2011) reported a shift from coral and sponge communities to algal dominated communities. Few worldwide fish invasions of similar magnitude are documented; the introduction of the red lionfish *Pterois volitans* and the devil firefish *P. miles* in the western Atlantic is one of the fastest and most ecologically harmful marine	4

		Question	Response	Justification	Confidence
				fish introductions to date (Albins and Hixon 2013).	
11	3.03	In the taxon's introduced range, are there known adverse impacts to aquaculture?	Y	It may be a negative effect on natural fish species using as feed raw material in aquaculture	1
12	3.04	In the taxon's introduced range, are there known adverse impacts to ecosystem services?	Y	According to Gardner *et al.* (2015), in the Caribbean and western Atlantic, the spread of lionfish exacerbates concern for the health of coral reefs already threatened by other stresses. After lionfish invaded a mesophotic reef, Lesser and Slattery (2011) reported a shift from coral and sponge communities to algal dominated communities. Increased predation on herbivorous fish was implicated as the cause of the shift because it was not associated with bleaching, fishing, storms and disease (Lesser and Slattery, 2011).	3
13	3.05	In the taxon's introduced range, are there known adverse socio-economic impacts?	Y	Economically, the appearance of the lionfish can cause a serious impact on the local's economy. Its presence in other areas has resulted in a reduction on the number of groupers and other commercial species, affecting the economies of the coastal fishing. The lionfish can also have an impact on artisanal fisheries and the nautical or diving tourism as it is highly toxic and its presence could inhibit tourists and divers alike. The lionfish sting should be treated as soon as possible as it can cause allergic reactions. On the other hand, some non-Mediterranean countries have begun to commercialize this fish as edible commercial species, which could be an alternative way, among other measures, to better control the populations (https://www.iucn.org/news/new-lionfish-sightings-turkey-and-cyprus-marine-protected-areas). According to Kleteu *et al.* (2016), the possibility of a lionfish invasion in the Mediterranean Sea and the potential ecological and socio-economic impacts.	4
B. Biology/Ecology					
4. Undesirable (or persistence) traits					
14	4.01	Is it likely that the taxon will be poisonous, or pose other risks to human health?	Y	The dorsal- and anal-fin spines of the lionfish contain a potent venom that can administer a painful sting. Lionfish venom has been found to cause cardiovascular, neuromuscular, and cytolytic effects ranging from mild reactions such as swelling to extreme pain and paralysis in upper and lower extremities (Kizer *et al.* 1985).	4
15	4.02	Is it likely that the taxon will smother one or more native taxa (that are not threatened or protected)?	Y	In western Atlantic, lionfish are more abundant than in their native region and have become the dominant predator on coral reefs with a great impact on native reef fishes, decreasing the abundance of >40 prey species by 65% on average, in just 2 years (Green *et al.* 2012).	3
16	4.03	Are there threatened or protected taxa that the non-native taxon would parasitise in the RA area?	N	No data	3

		Question	Response	Justification	Confidence
17	4.04	Is the taxon adaptable in terms of climatic and other environmental conditions, thus enhancing its potential persistence if it has invaded or could invade the RA area?	Y	For the Mediterranean no data is available. But, the combination of their high spawning frequency (year round, ~every 4 d, Morris 2009) and protracted pelagic larval phase (~26 d, Ahrenholz and Morris 2010), coupled with release in a region with multiple oceanographic currents (e.g., Gulf Stream, Caribbean Current, Yucatan Current and Loop Current) has resulted in the rapid dispersal of lionfishes into the western Atlantic Ocean, including the Caribbean Sea and Gulf of Mexico (Kletou *et al.* 2016, and reference therein). The recent findings of Lionfish may be an indication of a new wave of arrivals of *P. miles* in the Levant, raising justifiable concerns of a possible onset of a new invasion in the Mediterranean Sea (Bariche *et al.* 2013). Lionfishes can potentially spread and survive in a large part of the Mediterranean Sea because they have shown extensive dispersal capabilities and can survive to a minimum temperature of 10°C (Kimball *et al.* 2004).	3
18	4.05	Is the taxon likely to disrupt food-web structure/function in aquatic ecosystems it has or is likely to invade in the RA area?	Y		3
19	4.06	Is the taxon likely to exert adverse impacts on ecosystem services in the RA area?	Y	For the Mediterranean no data. But, in 2008, lionfish were observed in the waters surrounding Little Cayman Island, and they were considered established there by 2009 (Schofield 2010). At this location, they have reached densities of up to 650 fish ha^{-1} (Frazer *et al.* 2012), which is far greater than the 26.3 fish ha^{-1} recorded in the native range (Kulbicki *et al.* 2012).	3
20	4.07	Is it likely that the taxon will host, and/or act as a vector for, recognised pests and infectious agents that are endemic in the RA area?	N	There are no known OIE-reportable diseases for this species (https://www.fws.gov/fisheries/ans/erss/highrisk/Pterois-miles-WEB-7-28-2014.pdf)	1
21	4.08	Is it likely that the taxon will host, and/or act as a vector for, recognised pests and infectious agents that are absent from (novel to) the RA area?	N	There are no known OIE-reportable diseases for this species (https://www.fws.gov/fisheries/ans/erss/highrisk/Pterois-miles-WEB-7-28-2014.pdf)	1
22	4.09	Is it likely that the taxon will achieve a body size that will make it more likely to be released from captivity?	Y	Yes, it may be reach a 35.0 cm SL (Froese and Pauly 2016)	2
23	4.10	Is the taxon capable of sustaining itself in a range of water velocity conditions (e.g. versatile in habitat use)?	Y	According to Kleteu *et al.* (2016) in the western Atlantic, lionfish have been found in a wide variety of habitats including hard bottom and patchy reefs, seagrass beds and wrecks.	3
24	4.11	Is it likely that the taxon's mode of existence (e.g. excretion of by-products) or behaviours (e.g. feeding) will reduce habitat quality for native taxa?	Y	The lionfish are generalist carnivores and can feed on a large variety of fish and crustaceans although large individuals prey almost exclusively on fish (Côté *et al.* 2013).	2
25	4.12	Is the taxon likely to maintain a viable population	N	No data	1

		Question	Response	Justification	Confidence
		even when present in low densities (or persisting in adverse conditions by way of a dormant form)?			
5. Resource exploitation					
26	5.01	Is the taxon likely to consume threatened or protected native taxa in RA area?	Y	No study in the Mediterranean region but outside of the region: according to Schofield *et al.* (2016): "Research on small patch reefs in the Bahamas provided the first evidence of negative effects of lionfish on native Atlantic coral-reef fishes. The recruitment of coral-reef fishes was studied during the 2007 recruitment period (July-August) on small patch reefs in the Bahamas with and without lionfish. Over the five week period, net recruitment (i.e., accumulation of new juvenile fishes via settlement of larvae) was reduced by 79% on reefs with a single lionfish compared to reefs with no lionfish. Stomach content analyses and observations of feeding behavior showed that reductions in native fish density were almost certainly due to predation by lionfish. Prey items found in lionfish stomachs included the fairy basslet *Gramma loreto*, bridled cardinalfish *Apogon aurolineatus*, white grunt *Haemulon plumierii*, bicolor damselfish Stegastes pertitus, several wrasses *Halichoeres bivittatus*, *H. garnoti* and *Thalasoma bifasciatum*, striped parrotfish *Scarus iserti*, and dusky blenny *Malacoctenus gilli*. Initial examination of crustacean prey suggests that lionfish may also eat the juvenile spiny lobster *Panulirus argus*. The reduction in recruitment of coral-reef fishes suggests that lionfish may also compete with native piscivores by monopolizing this important food resource. In addition, lionfish have the potential to decrease the abundance of ecologically important species such as parrotfish and other herbivorous fishes that keep seaweeds and macroalgae from overgrowing corals."	1
27	5.02	Is the taxon likely to sequester food resources (including nutrients) to the detriment of native taxa in the RA area?	Y	No data	2
6. Reproduction					
28	6.01	Is the taxon likely to exhibit parental care and/or to reduce age-at-maturity in response to environmental conditions?	Y	Good adaptability in a short time	2
29	6.02	Is the taxon likely to produce viable gametes or propagules (in the RA area)?	Y	Spreading in a short time	3
30	6.03	Is the taxon likely to hybridize naturally with native taxa?	N	No data	1
31	6.04	Is the taxon likely to be hermaphroditic or to display asexual reproduction?	N	*P. miles* is gonochoristic; males and females exhibit minor sexual dimorphism only during reproduction (Fishelson 1975).	3

		Question	Response	Justification	Confidence
32	6.05	Is the taxon dependent on the presence of another taxon (or specific habitat features) to complete its life cycle?	N	No evidence	3
33	6.06	Is the taxon known (or likely) to produce a large number of propagules or offspring within a short time span (e.g. <1 year)?	Y	*P. miles* from the western Atlantic: Morris (2009) reported that females matured at approximately 175 mm total length or 1 year of age and released approximately 25000 eggs per spawning event. Based on the presence of hydrated oocytes, mature females appeared capable of spawning every 3.6–4.1 days throughout the year, although the proportion of females with ovaries in spawning condition was higher in summer (June–August).	3
34	6.07	How many time units (days, months, years) does the taxon require to reach the age-at-first-reproduction? [In the Justification field, indicate the relevant time unit being used.]	1	*P. miles* from the western Atlantic: Morris (2009) reported that females matured at approximately 175 mm total length or 1 year of age	4
7. Dispersal mechanisms					
35	7.01	How many potential internal pathways could the taxon use to disperse within the RA area (with suitable habitats nearby)?	1	Mediterranean's current system can be used. Similarly, it is reported that their eggs are planktonic and can ride the currents and cover large distances for about a month before they settle in the Atlantic (Ahrenholz and Morris 2010).	3
36	7.02	Will any of these pathways bring the taxon in close proximity to one or more protected areas (e.g. MCZ, MPA, SSSI)?	Y	The Marine Protected Areas of Kas-Kekova (Turkey) and Cape Greco (Cyprus) have been the locations where sightings have taken place for the lionfish (https://www.iucn.org/news/new-lionfish-sightings-turkey-and-cyprus-marine-protected-areas).	4
37	7.03	Does the taxon have a means of actively attaching itself to hard substrata (e.g. ship hulls, pilings, buoys) such that it enhances the likelihood of dispersal?	N	Nekton	3
38	7.04	Is natural dispersal of the taxon likely to occur as eggs (for animals) or as propagules (for plants: seeds, spores) in the RA area?	Y	The combination of their high spawning frequency (year round, ~every 4 d, Morris 2009) and protracted pelagic larval phase (~26 d, Ahrenholz and Morris 2010), coupled with release in a region with multiple oceanographic currents (e.g., Gulf Stream, Caribbean Current, Yucatan Current and Loop Current) has resulted in the rapid dispersal of lionfishes into the western Atlantic Ocean, including the Caribbean Sea and Gulf of Mexico (Kletou *et al.* 2016, and reference therein).	2
39	7.05	Is natural dispersal of the taxon likely to occur as larvae/juveniles (for animals) or as fragments/seedlings (for plants) in the RA area?	Y	Same with above answer.	2
40	7.06	Are older life stages of the taxon likely to migrate in the RA area for reproduction?	Y	No data	1

		Question	Response	Justification	Confidence
41	7.07	Are propagules or eggs of the taxon likely to be dispersed in the RA area by other animals?	N	No data	2
42	7.08	Is dispersal of the taxon along any of the pathways mentioned in the previous seven questions (7.01–7.07; i.e. both unintentional or intentional) likely to be rapid?	Y	1991- *P. miles* has been recorded from the Levantine coast (Golani and Sonin, 1992). 2012- Lebanon (Bariche *et al.* 2013). Soon after, two more were reported in a newspaper article, captured off Cyprus and in 2014, another specimen was sighted in Turkey, then in 2015 two more were captured in Cyprus and then sighted in Rhodes in Greece (Turan *et al.* 2014; Oray *et al.* 2015; Crocetta *et al.* 2015; Iglésias and Frotté 2015). Recently, several records were reported from south Turkey indicating a westerly migration of the species towards the Aegean Sea (Turan and Öztürk 2015). Hitherto, lionfish were reported only occasionally in the eastern Mediterranean Sea and their successful invasion in the Mediterranean Sea is questionable due to unfavourable oceanographic conditions that limit the wide dispersion of lionfish larvae (Johnston and Purkis 2014). The lionfish *P. miles* has spread rapidly and colonised almost the entire south eastern coast of Cyprus, from Limassol (south) to Protaras (south east) in just 1 year	3
43	7.09	Is dispersal of the taxon density dependent?	Y	Lionfishes were found abundant in some invaded areas as well as dominating reef fish communities (Kulbicki *et al.* 2012). Their dramatic invasive success results from a combination of factors such as early maturation and reproduction, anti-predatory venomous defences and ecological versatility of the lionfish coupled with native prey and the overfishing of native predators (Côté *et al.* 2013).	3
8. Tolerance attributes					
44	8.01	Is the taxon able to withstand being out of water for extended periods (e.g. minimum of one or more hours) at some stage of its life cycle?	N	Marine fish	4
45	8.02	Is the taxon tolerant of a wide range of water quality conditions relevant to that taxon? [In the Justification field, indicate the relevant water quality variable(s) being considered.]	Y	No data	1
46	8.03	Can the taxon be controlled or eradicated in the wild with chemical, biological, or other agents/means?	Y	Their highly venomous needle-sharp dorsal, anal and pectoral fin spines offer protection and signifiantly reduce predation (Bariche *et al.* 2013). Nevertheless, one potential natural predator of *P. miles* already exists in the Mediterranean. In the northern Red Sea, a juvenile Lionfish (10 cm SL) was discovered in the stomach of *Fistularia commersonii* Rüppell, 1838, the blue spotted cornetfish who has invaded the Mediterranean Sea within the last decade and established large populations in the eastern part (Azzurro *et al.* 2012) and may act as a biological control of a future possible invasion. Other possible predators could be native Mediterranean groupers, as found in the Caribbean (Mumby *et al.* 2011).	3

		Question	Response	Justification	Confidence
				Native groupers such as *Epinephelus marginatus* will learn to prey on lionfish and control their invasion. Restrictions on fishing seem sensible to help threatened IUCN Red List species such as the Mediterranean dusky grouper, as these may in turn help control the spread of invasive fish (Mumby *et al.* 2011).	
47	8.04	Is the taxon likely to tolerate or benefit from environmental/human disturbance?	Y	Judging from the recent increase in *P. miles* in the eastern Mediterranean Sea, its few natural predators, the dispersal capabilities of their planktonic larvae and its ability to adapt to a range of habitats, we suspect that a rapid expansion throughout the Mediterranean Sea may soon follow with significant impacts on local ecosystems and fisheries. Furthermore, CO_2 emissions which are warming the Mediterranean Sea and the construction of a deeper and wider Suez Canal are expected to increase invasion rates (Galil *et al.* 2015; Hall-Spencer and Allen 2015). Increased predation on herbivorous fish was implicated as the cause of the shift because it was not associated with bleaching, fishing, storms and disease (Lesser and Slattery, 2011). Their dramatic invasive success results from a combination of factors such as early maturation and reproduction, anti-predatory venomous defences and ecological versatility of the lionfish coupled with native prey and the overfishing of native predators (Côté *et al.* 2013).The warming and acidification of Mediterranean waters due to CO_2 emissions may require new modelling approaches (Hall-Spencer and Allen 2015).	2
48	8.05	Is the taxon able to tolerate salinity levels that are higher or lower than those found in its usual environment?	Y	From Froese and Pauly (2016): Tropical; 30°N - 36°S, 19°E - 112°E.	3
49	8.06	Are there effective natural enemies (predators) of the taxon present in the RA area?	Y	*Fistularia commersonii* Rüppell, 1838 (Azzurro *et al.* 2012). *Epinephelus marginatus* will learn to prey on lionfish and control their invasion (Mumby *et al.* 2011).	2
C. Climate change					
9. Climate change					
50	9.01	Under the predicted future climatic conditions, are the risks of entry into the RA area posed by the taxon likely to increase, decrease or not change?	+	Overall, the number of introduced Lessepsian fish species is correlated significantly and positively with the Mediterranean water temperature ($r = 0.77$, $p<0.05$) (Ben Raiş Lasram *et al.* 2010).	3
51	9.02	Under the predicted future climatic conditions, are the risks of establishment posed by the taxon likely to increase, decrease or not change?	+	The recent findings of Lionfish may be an indication of a new wave of arrivals of *P. miles* in the Levant, raising justifiable concerns of a possible onset of a new invasion in the Mediterranean Sea (Bariche *et al.* 2013).	3

		Question	Response	Justification	Confidence
52	9.03	Under the predicted future climatic conditions, are the risks of dispersal within the RA area posed by the taxon likely to increase, decrease or not change?	+	Since the 1980s the rate of introduction of Lessepsian species from the Red Sea to the Mediterranean has increased (Ben Rais Lasram *et al.* 2010).	3
53	9.04	Under the predicted future climatic conditions, what is the likely magnitude of future potential impacts on biodiversity and/or ecological integrity/status?	+	Professional assessment	2
54	9.05	Under the predicted future climatic conditions, what is the likely magnitude of future potential impacts on ecosystem structure and/or function?	+	Professional assessment	2
55	9.06	Under the predicted future climatic conditions, what is the likely magnitude of future potential impacts on ecosystem services/socio-economic factors?	+	Professional assessment	2