EU NON-NATIVE ORGANISM RISK ASSESSMENT SCHEME

Name of organism: ***Rugulopteryx okamurae*** (E.Y. Dawson) I.K. Hwang, W.J. Lee & H.S. Kim 2009

Author: Ministry for Ecological Transition and Demographic Challenge (MTERD)

Risk Assessment Area: Europe

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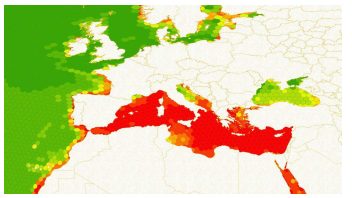
EU CHAPPEAU	
QUESTION	**RESPONSE**
1. In how many EU member states has this species been recorded? List them.	It has been recorded in 2 countries: France (Verlaque *et al*. 2009) and Spain (Altamirano *et al.* 2016).
2. In how many EU member states has this species currently established populations? List them.	It has established populations in 2 countries: France (Verlaque *et al*. 2009) and Spain (Altamirano *et al.* 2016).  Figure.1: Map showing countries in Europe with *Rugulopteryx okamurae populations.* (Altamirano *et al.* 2019)
3. In how many EU member states has this species shown signs of invasiveness? List them.	This species has only showed invasive behaviour in Spain (Altamirano *et al.* 2016, 2019).
4. In which EU Biogeographic areas could this species establish?	*R. okamurae* could proliferate within the ecological environment of the Atlantic coast of Andalusia, the Mediterranean area and the Black Sea. (Altamirano *et al.* 2016, 2019). This species could establish at least in the following regions:

	Mediterranean area and Atlantic area, and Black Sea area.
5. In how many EU Member States could this species establish in the future [given current climate] (including those where it is already established)? List them.	In the future the organism could establish in France, Spain, Portugal, Italy, Greece, Croatia, Slovenia, Cyprus and Malta.
6. In how many EU member states could this species become invasive in the future [given current climate] (where it is not already established)?	It could become invasive in all the previous countries.

SECTION A – Organism Information and Screening		
Stage 1. Organism Information	**RESPONSE** **[chose one entry, delete all others]**	**COMMENT**
1. Identify the organism. Is it clearly a single taxonomic entity and can it be adequately distinguished from other entities of the same rank?	Yes, it is cleraly a single taxonomic entity. It has been described in 2009 as *Rugulopteryx okamurae* (E.Y. Dawson) I.K. Hwang, W.J. Lee & H.S. Kim 2009. Taxonomic reappraisal of *Dilophus okamurae* (Dictyotales, Phaeophyta) from the western Pacific Ocean. *Phycologia* 48 (1): 1-12. It can be confused with other species of the genus *Dictyota*, such as *Dictyota pinnatifida, D. dichotoma, D. spiralis, D. cyanoloma or D. fasciola.*	Kingdom: Chromista; Phylum: Ochrophyta; Class: Phaeophyceae; Subclass: Dictyotophycidae; Order: Dictyotales; Family: Dictyotaceae, Tribe: Dictyoteae
2. If not a single taxonomic entity, can it be redefined? (if necessary, use the response box to re-define the organism and carry on)	NA	
3. Does a relevant earlier risk assessment exist? (give details of any previous risk assessment)	Yes. A rapid risk assessment for *Rugulopteryx okamurae* was produced by the University of Málaga (Spain) commissioned by the Ministry of Ecological Transition of Spain in October 2019 (Altamirano et al. 2019).	The mentioned risk assessment for *Rugulopteryx okamurae* describes the species as an effective invasive species, because since its detection in Ceuta (Spain, Northern Africa) in 2016, its expansion along the Andalusian coast has been exponential. This, together with the amounts of biomass recovered from invaded locations, pose a significant ecological pressure to native ecosystems and affect local economies.  Figure 2: Environmental favourability model for *Rugulopteryx*

		okamurae in the Mediterranean and western European coasts, based on the native and introduced distribution. The areas that present favourable conditions to accommodate the species are shown in warm colours.
4. If there is an earlier risk assessment is it still entirely valid, or only partly valid?	The previous mentioned risk assessment can be considered valid, although species distribution may have change locally.	
5. Where is the organism native?	*Rugulopteryx okamurae* is a common native of the warm and temperate North-western Pacific Ocean, of Korea, Japan, China, Taiwan and Philippines.	 Figure 3: Native distribution of *R. okamurae*
6. What is the global distribution of the organism (excluding Europe)?	North-western Pacific Ocean coasts (Korea, Japan, China, Taiwan and Philippines); Also in Mexico (cited as *Dilophus okamurai*) and in the Gulf of California (but it must be confirmed in this last location).	
7. What is the distribution of the organism in Europe?	*Rugulopteryx okamurae* is now established in the Thau Lagoon of France near Montpellier, and in the Atlantic and Mediterranean coasts of southern Spain: Ceuta, Cádiz, Málaga, and the Chafarinas Islands. Drifted material of the species has also been found in Huelva, Granada and Almería.	 Figure 4: Distribution of the *R. okamurae* in Europe
8. Is the organism known to be invasive (i.e. to threaten organisms, habitats or ecosystems)	Since the detection of the species in 2016 in Spain, the species is producing important ecological	<u>Ecological impacts in Southern Spain:</u> - Habitat modification due to rapid

anywhere in the world?	impacts, such as alterations of the marine habitat, loss of biodiversity, affection to species and protected areas included in the Natura 2000 Network. No invasive behaviour has been reported anywhere else.	colonization and homogenization of marine ecosystems, and accumulations of detached biomass. - Loss of marine biodiversity and alteration of the structure of the communities, causing the physical displacement of the rest of the species due to the occupation of the substrate and preventing the fixation of larvae or propagules of other species. - Affectation to habitats and species of community interest in spaces of the Natura 2000 Network. - Affected communities: Kelp forests, *Cystoseira spp* communities, *Posidonia oceanica* meadows, eulitoral and infralitoral communities of seaweeds, maerl communities, epiphytic fauna of invertebrates. - Affected species: *Laminaria ochroleuca*, *Saccorhiza polyschides*, *Cystoseira* species (eg *C. usneoides*), *Lithophyllum byssoides*, *Gymnogongrus crenulatus, Sphaerechinus granularis, Leptogorgia sarmentosa, Eunicella* spp., *Paramuricea clavata, Charonia lampas, Astroides calycularis, Corallium rubrum, Patella ferruginea.*
9. Describe any known socio-economic benefits of the organism in the risk assessment area.	There are no socio-economic benefits of *R. okamurae* although the species could have interests for the industry because of secondary metabolites.	The taxonomic family to which *R. okamurae* belongs, presents a considerable diversity of secondary metabolites with possible commercial interest, with terpenes being one of the best represented groups, but there is no evidence of practical application (Yamase et al. 1999; De Paula et al. 2011, Suzuki et al. 2002).

		Terpenoids have been shown to have various biological activities that make them very interesting as a target for exploitation by the industry. On the one hand, its compounds have been shown to possess α- inhibitory activity glucosidase, enzyme that regulates processes of synthesis and quantity of glycoproteins and glycolipids. These last two are involved in diseases such as diabetes, certain forms of hyperlipoproteinemia and obesity. Therefore, it has been proposed that glucosidase inhibitors may be useful for the treatment of this type of diseases. Other biological activities of *R. okamurae* compounds are antifungal, antibiotic, anti-inflammatory, insecticide and antiviral (De Paula et al. 2011). High efficacy against cancers such as leukemia has been demonstrated (Harada and Kamei 1997). Finally, a very interesting use of terpenes is their antifouling ability, so they are good candidates to be included as a component in the paints used for coating the ships, taking into account They are also less toxic than other synthetic compounds (Fusetani 2004).

SECTION B – Detailed assessment

PROBABILITY OF ENTRY

Important instructions:

- Entry is the introduction of an organism into Europe. Not to be confused with spread, the movement of an organism within Europe.
- For organisms which are already present in Europe, only complete the entry section for current active pathways of entry or if relevant potential future pathways. The entry section need not be completed for organisms which have entered in the past and have no current pathways of entry.

QUESTION	RESPONSE [chose one entry, delete all others]	CONFIDENCE [chose one entry, delete all others]	COMMENT
1.1. How many active pathways are relevant to the potential entry of this organism? (If there are no active pathways or potential future pathways respond N/A and move to the Establishment section)	**moderate number**	**high**	The main pathways are unintentional. Intentional ways could happen, but these are uncertain.
1.2. List relevant pathways through which the organism could enter. Where possible give detail about the specific origins and end points of the pathways. For each pathway answer questions 1.3 to 1.10 (copy and paste additional rows at the end of this section as necessary).	**-Maritime transportation (Ballast waters and hull fouling); -Marine aquaculture**	**high**	
Pathway name:	**Maritime transportation (Ballast waters and hull fouling)**		
1.3. Is entry along this pathway intentional (e.g. the organism is imported for trade) or accidental (the organism is a contaminant of imported goods)?	**accidental**	**high**	Marine transport is one of the main introduction vectors of exotic species in the marine environment, the most important being fouling (inlays in boat hulls) and ballast waters (Ruíz *et al.* 1997; Ribera 2003). Both routes could explain the introduction of *R. okamurae* in the Strait of Gibraltar from its native areas in the Pacific, taking into account the intense maritime traffic in this

			region.
1.4. How likely is it that large numbers of the organism will travel along this pathway from the point(s) of origin over the course of one year? Subnote: In your comment discuss how likely the organism is to get onto the pathway in the first place.	**very likely**	**very high**	Most macroalgae species are capable of adhering to ship hulls (Schaffelke *et al.* 2006; Hewitt *et al.* 2007). Ribera (2003) mentions 39 species of exotic macroalgae that have been introduced in new regions in this way. Recruitment can occur through macroscopic thallus, such as juvenile sporophytes of the invading brown seaweed *Undaria pinnatifida* (Hay 1990); but mainly, it is microscopic stages of the life cycle that travel by fouling (Lewis *et al.* 2004), as was the case of gametophytes of *U. pinnatifida* (Wotton *et al.* 2004) and other species of brown algae such as *Phloiocaulon* or *Punctaria* (Coutts 1999). Ballast waters has also been responsible for the introduction of many exotic marine species (Ruíz *et al.* 1997; Gollasch *et al.* 2000), including macroalgae (Carlton & Geller 1993; Smith *et al.* 1999; Gollasch *et al.* 2002; David *et al.* 2007). Up to 15 different species were identified from ballast water samples from 12 vessels that made routes through the Mediterranean, with different origins, both from within the Mediterranean and from other waters (Flagella *et al.* 2007). The importance of this vector in the transport of macroalgae propagules is not well known yet, but it is known that conditions during transport in ballast waters can induce the formation of spores and propagules (Kolwalkar *et al.* 2007), and that microscopic stages of macroalgae can withstand long periods of darkness and subsequently develop when conditions improve (Leukart & Lüning 1994; Worm *et al.* 2001; Santelices *et al.* 2002; Carney & Edwards 2006).

1.5. How likely is the organism to survive during passage along the pathway (excluding management practices that would kill the organism)? Subnote: In your comment consider whether the organism could multiply along the pathway.	**very likely**	**very high**	Given the high capacity of *R. okamurae* to produce vegetative propagules and asexual monospores and the presence of two large commercial ports in the Strait of Gibraltar (Algeciras and Tangier Med in Morocco), it is suspected that ballast waters may be an important vector of introduction of the species, especially in this region. In fact, Rosas-Guerrero *et al.* (2018) observed that portions of adult thallus of the species had survival rates between 80-100% after being grown in dark conditions for three weeks, depending on the temperature during cultivation, and that these thalli even increased their biomass during this time. Moreover, after the dark period the thallus maintained the same survival rates when they passed to lighting conditions, which simulated the release phase after dark transport. Although there is no evidence that *R. okamurae* has reached the shores of the western Mediterranean adhered to ship hulls, it has been observed that the species is capable of adhering to surfaces of very diverse nature and composition, such as glass, iron, rubber or ceramic (García-Gómez *et al.* 2018). However, the changing conditions associated with this kind of transport, especially those related to the temperature and salinity, as well as the effect of friction caused by the crossing speed and the swell, and the presence of antifouling substances, make the transport by this vector a hard test for those species that use it to enter new geographical regions (Hewitt *et al.* 2007).
1.6. How likely is the organism to survive existing management practices during passage along the pathway?	**moderately likely**	**medium**	Conditions during transport in ballast waters can induce the formation of spores and propagules (Kolwalkar et al. 2007), and microscopic stages of macroalgae can

			withstand long periods of darkness and subsequently develop when conditions improve (Leukart & Lüning 1994; Worm et al. 2001; Santelices et al. 2002; Carney & Edwards 2006). Given the high capacity of *R. okamurae* to produce vegetative propagules and asexual monospores and the presence of two large commercial ports in the Strait of Gibraltar (Algeciras and Tangier Med in Morocco), it is suspected that ballast waters may be an important vector of introduction of the species, especially in this region. However, if adequate systems for ballast water control were implemented the probability of entry of the species would be greatly reduced.
1.7. How likely is the organism to enter Europe undetected?	**very likely**	**very high**	Through the ballast waters, spores and propagules can be introduced (Kolwalkar *et al.* 2007) undetected Furthermore, due the morphological similarity with native *Dictyota* species, *R. okamurae* may be producing cryptic invasion.
1.8. How likely is the organism to arrive during the months of the year most appropriate for establishment?	**very likely**	**high**	There is no information available of the most appropriate time of the year for the establishment of *R. okamurae*. However, meanwhile there is no control on introduction vectors, new inoculums may be arriving along the whole year, increasing the probabilities of success of these inoculums. In a study about the reproductive phenology of *R. okamurae* on the coast of Cádiz carried out between January and August 2017, monospores and vegetative propagules were observed in the thallus that were collected all months of the study, although its abundance increased from May and show a maximum in August. In addition, during the months of April and

			August some thallus also presented tetrasporangia, but less abundant and frequent than the monosporangia (Pulido 2017; Pulido & Altamirano 2017).

1.9. How likely is the organism to be able to transfer from the pathway to a suitable habitat or host?	**likely**	**high**	The Spanish marine habitats are suitable for the establishment of *R. okamurae*, as evidenced by the presence of the species fixed to the substrate on the coasts of Malaga and Cádiz, and as revealed by the distribution models in other areas of the Spanish coast, mainly in its Mediterranean side. These models were developed for the previous risk analysis and show that there are other areas on the planet with potential to accommodate the species, in addition to its native area. These include the Mediterranean Sea as a whole, including the Spanish coasts and the Balearic archipelago, and the Atlantic coast of Andalusia, which have very high favourable values.
1.10. Estimate the overall likelihood of entry into Europe based on this pathway?	**very likely**	**high**	In Europe the species is present in France and Spain, not excluding other areas. But it is very likely that propagules continue to arrive from the area of origin through maritime transport. Also, short to medium dispersal through marine currents is highly likely.
1.11. Estimate the overall likelihood of entry into Europe based on all pathways (comment on the key issues that lead to this conclusion).	**very likely**	**high**	As mentioned before, the species is already established in Southern Spain coasts, spreading mainly to the Mediterranean Sea. New introductions may be still happening through maritime transport at the Strait of Gibraltar or at other ports in the Mediterranean.
End of pathway assessment, repeat as necessary.			
Pathway name:	**Marine aquaculture**		
1.3. Is entry along this pathway intentional (e.g. the organism is imported for trade) or accidental (the organism is a contaminant of imported goods)?	**accidental**	**high**	As in the case of some other non-native species, the introduction of *R. okamurae* in Thau Lagoon is associated with oyster imports (*Crassostrea gigas*) carried out later in 1977, probably in 1994, from Korea (Verlaque *et al.* 2009). It is an accidental entry.

1.4. How likely is it that large numbers of the organism will travel along this pathway from the point(s) of origin over the course of one year? Subnote: In your comment discuss how likely the organism is to get onto the pathway in the first place.	**moderately likely**	**low**	In the Alboran Sea, although it is not considered an important commercial activity, several companies are engaged in oyster farming (Robles 2010), so the presence of *R. okamurae* may be associated with this introduction vector. However, due to the high capacity of the species to produce vegetative propagules and asexual monospores, few individuals may carry big amounts of inoculum through this pathway.
1.5. How likely is the organism to survive during passage along the pathway (excluding management practices that would kill the organism)? Subnote: In your comment consider whether the organism could multiply along the pathway.	**likely**	**medium**	It is likely that microscopic stages of *R. okamurae* can survive the passage through this pathway, especially propagules and monospores.
1.6. How likely is the organism to survive existing management practices during passage along the pathway?	**unlikely**	**medium**	It will depend on the management treatment.
1.7. How likely is the organism to enter Europe undetected?	**very likely**	**very high**	Through the ballast waters, spores and propagules can be introduced (Kolwalkar *et al.* 2007) and microscopic stages can withstand long periods of darkness (Leukart & Lüning 1994; Worm *et al.* 2001; Santelices *et al.* 2002; Carney & Edwards 2006). Furthermore, due the morphological similarity with native *Dictyota* species, *R. okamurae* may be producing cryptic invasion.
1.8. How likely is the organism to arrive during the months of the year most appropriate for establishment?	**very likely**	**high**	There is no information available of the most appropriate time of the year for the establishment of *R. okamurae*. However, meanwhile there is no control on introduction vectors, new inoculums may be arriving along the whole year, increasing the probabilities os success of these inoculums.
1.9. How likely is the organism to be able to transfer from the pathway to a suitable habitat or host?	**likely**	**high**	The Spanish marine habitats are suitable for the establishment of *R. okamurae*, as evidenced by the

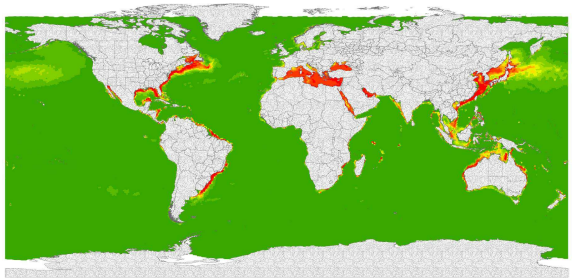
			presence of the species fixed to the substrate on the coasts of Malaga and Cádiz, and as revealed by the distribution models in other areas of the Spanish coast, mainly in its Mediterranean side. These models were developed for the previous risk analysis and show that there are other areas on the planet with potential to accommodate the species, in addition to its native area. These include the Mediterranean Sea as a whole, including the Spanish coasts and the Balearic archipelago, and the Atlantic coast of Andalusia, which have very high favourable values.
1.10. Estimate the overall likelihood of entry into Europe based on this pathway?	**very likely**	**high**	The presence of the species in France is associated to this pathway (Verlaque *et al.* 2009). Due to the commercial success of oyster farming, the Thau lagoon acts as a donor of oyster populations for its cultivation in many parts of the Mediterranean and Atlantic (Mineur *et al.* 2007; Verlaque *et al.* 2007). Therefore, this could be a potential entry pathway and the cause of unintentional or accidental dispersal of invasive macroalgae such as *R. okamurae* in the Mediterranean. In the Alboran Sea, although it is not considered an important commercial activity, several companies are engaged in oyster farming (Robles 2010), so the presence of *R. okamurae* may be associated with this activity.
1.11. Estimate the overall likelihood of entry into Europe based on all pathways (comment on the key issues that lead to this conclusion).	**very likely**	**high**	As mentioned before, the species is already established in Southern Spain coasts, spreading mainly to the Mediterranean Sea. New introductions may be still happening through maritime transport at the Strait of Gibraltar or at other ports in the Mediterranean. Ballast water may be the main vector for introduction of the species, as pieces of thalli can survive long periods of

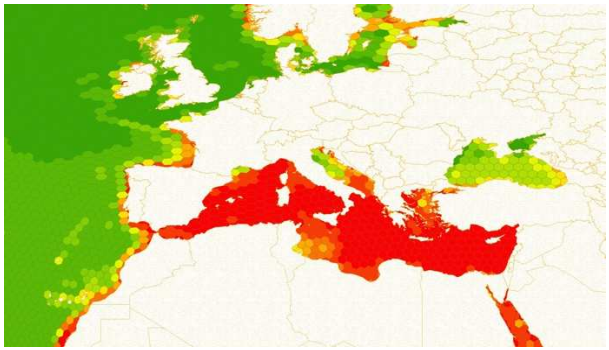
			darkness (Rosas-Guerrero *et al.* 2018).

PROBABILITY OF ESTABLISHMENT

Important instructions:

- For organisms which are already well established in Europe, only complete questions 1.15 and 1.21 then move onto the spread section. If uncertain, check with the Non-native Species Secretariat.

QUESTION	RESPONSE	CONFIDENCE	COMMENT
1.12. How likely is it that the organism will be able to establish in Europe based on the similarity between climatic conditions in Europe and the organism's current distribution?	**very likely**	**very high**	Though the distribution of this species was originally concentrated in the northwestern end of the Pacific, the model detects climatically suitable areas for *R. okamurae* in other sites of the Earth, like the northern and western coast of Australia, Persic Gulf, Red Sea, most of the American coast and, remarkably, the Mediterranean Sea.  Figure 5: Environmental suitability model for *R. okamurae* on a global scale, showing (in red) the areas that present the most similar conditions to the native distribution area of this species
1.13. How likely is it that the organism will be able to establish in Europe based on the similarity between other abiotic conditions in Europe and the organism's current distribution?	**very likely**	**very high**	Based on the global model that considers the known distribution of this species (the native distribution plus the areas where it was introduced), the obtained results show the potential distribution of the species according to the

			environmental conditions to which *R. okamurae* is exposed in all its distribution range. Therefore, the map shows the ecologically suitable places for this species. The variables that were used to create this model are obtained from the platform Bio-ORACLE (Assis et al. 2017), and these variables were water temperature, salinity, nutrients, chlorophyll, speed of the ocean currents, phytoplankton, primary production, iron, light and surface light. These variables were chosen based on their predictive potential and are expected to characterize the environmental conditions that favor the presence of *R. okamurae* in its native range. Moreover, it is intended to identify other areas out of its native range that would favour the establishment of this species if it is introduced.  Figure 6: Areas where environmental conditions could favour the establishment of *R. rugulopterix* in Europe based on native and introduction distribution.
1.14. How likely is it that the organism will become established in protected conditions (in which the environment is artificially maintained, such as wildlife	**very likely**	**high**	

parks, glasshouses, aquaculture facilities, terraria, zoological gardens) in Europe? Subnote: gardens are not considered protected conditions			
1.15. How widespread are habitats or species necessary for the survival, development and multiplication of the organism in Europe?	**widespread**	**high**	On the basis of the environmental characteristics of the native areas of distribution, it can be concluded that the Andalusian Atlantic coast, the Strait of Gibraltar region and the entire Mediterranean coast are environmentally very similar and therefore these areas could host the species. The Black Sea and south of Europe are also shown as suitable. Furthermore, it has to be considered that the host communities of *R. okamurae* in the new invaded areas (mainly *Cystoseira* species communities and photophilic ones), are well represented in the Mediterranean Sea.
1.16. If the organism requires another species for critical stages in its life cycle, then how likely is the organism to become associated with such species in Europe?	**NA**	**low**	
1.17. How likely is it that establishment will occur despite competition from existing species in Europe?	**very likely**	**high**	In the Strait of Gibraltar, *R. okamurae* causes community homogenization, just a few species remain in the ecosystem, mainly invasive macro-algae like *Asparagopsis armata*, *Asparagopsis taxiformis* and *Caulerpa cylindracea.* The host ecosystems in the early stages of invasion are Fucaidean algae bottoms with species of the genus *Cystoseira* or *Sargassum*, kelp forests and photophilic bottoms where coralline algae prevail. The invasive species competes successfully with all these species and homogenize the seabottom.
1.18. How likely is it that establishment will occur despite predators, parasites or pathogens already present in	**likely**	**high**	It is not known if there is any species that could prevent the establishment of *R. okanurae* if the

Europe?			conditions are suitable.
1.19. How likely is the organism to establish despite existing management practices in Europe?	**very likely**	**very high**	Effective control methods for *R. okamurae* that can be applied in the current stage of invasion are not known. The most important action and with more probability of success is, without any doubt, to prevent the introduction and spread of this species. Moreover, actions towards the management of established populations should be considered in order to minimize the impact they cause.
1.20. How likely are management practices in Europe to facilitate establishment?	**moderately likely**	**medium**	If the algae are collected, it is important to prevent its dispersion, having in mind the high propagation capacity of *R. okamurae.* Moreover, if it is located in the sea bottom and is taken above the photic zone it can survive and colonize new shallow areas (this species can survive up to three weeks in the darkness).
1.21. How likely is it that biological properties of the organism would allow it to survive eradication campaigns in Europe?	**very likely**	**very high**	*Rugulopteryx okamurae* shows an extraordinary competition and colonization ability, with a disproportionate and unprecedented biomass increment that is not even comparable to previous invasions in the Spanish coasts by any other macroalgae like *Asparagopsis taxiformis* (Altamirano *et al.* 2008; Zanolla *et al.* 2018a, b, c), *Lophocladia lallemandii* (Patzner 1998; Cabanelles *et al.* 2010; Deudero *et al.* 2010), *Caulerpa taxifolia* (Verlaque *et al.* 2015) or *Caulerpa cylindracea* that was considered the most severe invasive macroalgae in the Mediterranean in history, until this moment (Klein & Verlaque 2008). Part of the successful invasion performance of the species is due to its high

			propagation capacity through vegetative and asexual structures that can easily scape from management strategies such as eradications campaigns. Thalli can grow directly over the hard substrata but also on other seaweeds as epiphyte or even over animals such as sea urchins and sea cucumbers, which may become eradication campaigns with low success rate when the population is big and no monitoring is followed.
1.22. How likely are the biological characteristics of the organism to facilitate its establishment?	**very likely**	**high**	Reproduction takes place mainly by asexual and vegetative mechanisms. *R. okamurae* is a marine species that occurs from eulitoral basins to depths of more than 30 m. The invasive potential of a single thallus is substantial, taking into account that more than 100 spores and 25 propagules have been counted in 1 cm^2 of thallus (Altamirano *et al.* pers. obs.) and each one can generate a new clone individual, regardless of whether the thallus is fixed to the substrate or free in the water column. However, there is still a big lack of knowledge on the biological performance of the species in the introduction areas.
1.23. How likely is the capacity to spread of the organism to facilitate its establishment?	**likely**	**medium**	It is necessary to know which vectors allow the translocations of *R. okamurae* from the populations where the species is established to new areas in order to be able to act and stop its spread. Taking into account the high propagation ability of *R. okamurae* that was observed in the Strait of Gibraltar, it is essential to avoid its dispersal by vectors linked to human activities, because it is not possible to stop dispersal by marine currents.

			However, secondary introductions must indeed be happening, mainly due to fisheries activities (thalli entangled in the nests and released for cleaning in the sea again). Although floating material can sink to depths out of the photic zone, due to its survival capacity to darkness (Rosas-Guerrero et al. 2018), these thalli can get again the photic area thanks to sea currents and establish in new distant places, with high proliferation capacity due to monospores and propagules.
1.24. How likely is the adaptability of the organism to facilitate its establishment?	**very likely**	**high**	Ecological performance of the species at the new invaded areas at the Strait of Gibraltar, like perennial status, wide bathymetric range and host communities, presence of allelopathic substances like terpenes) leads to suggest a high adaptability of the species, although ecological performance along the year is still unknown.
1.25. How likely is it that the organism could establish despite low genetic diversity in the founder population?	**very likely**	**very high**	Up to now the presence of the gametophyte of *R. okamurae* in the invaded area has not been confirmed, as only thalli with vegetative propagules, asexual monospores or in scarce cases, tetraspores, have been found. This leads to think that invasive populations are formed and increase by clonal mechanisms with low or absent genetic variability.
1.26. Based on the history of invasion by this organism elsewhere in the world, how likely is to establish in Europe? (If possible, specify the instances in the comments box.)	**very likely**	**very high**	The species has not exhibited invasive performance elsewhere in the world besides Southern Spain and Norhtern coast of Morocco and Spanish cities, up to now.
1.27. If the organism does not establish, then how likely is	**likely**	**medium**	While maritime vectors are not under control, new

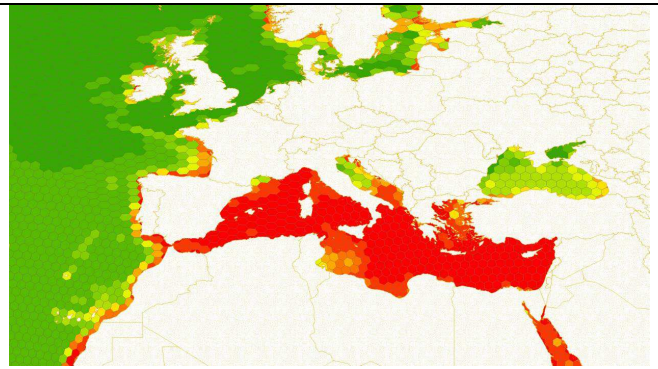
it that transient populations will continue to occur? Subnote: Red-eared Terrapin, a species which cannot re-produce in GB but is established because of continual release, is an example of a transient species.			inoculum of the species may enter in the new areas.
1.28. Estimate the overall likelihood of establishment (mention any key issues in the comment box).	**very likely**	**very high**	The species is already established and very well widespread in Southern Spain. Considering that in three years the species has widespread along the whole southern Spain coast and part of northern coast of Morocco, and that new local populations are being reported in the last months expanding its distribution, it is expected that the species may confirm the distribution model proposed.

PROBABILITY OF SPREAD

Important notes:

- Spread is defined as the expansion of the geographical distribution of a pest within an area.

QUESTION	RESPONSE	CONFIDENCE	COMMENT
2.1. How important is the expected spread of this organism in Europe by natural means? (Please list and comment on the mechanisms for natural spread.)	**massive**	**high**	A large amount of material has been observed both deposited in the bottom, occupying platforms with little slope, or occupying clearings of sand, whose thickness has reached 45-50 cm in height. In addition to this deposited material, a large amount of drifting material is also observed in the water column which can move tens of kilometres and cause the colonization of new areas mediated by sea currents or local vectors.
2.2. How important is the expected spread of this organism in Europe by human assistance? (Please list and comment on the mechanisms for human-assisted spread.)	**major**	**high**	The dispersion by ballast water can be an important vector of introduction, but also of dispersion to nearby areas, as would be the case with green algae forming massive proliferations (also known as “blooms”) *Ulva ohnoi* and *Ulva pertusa* (Zanolla et al. 2019). In addition, other potential vectors responsible for the dispersion of the species and secondary introductions must be added. Fishermen in the areas affected by the species report that they suffer decreases in their catches and deterioration of their nets because thalli of the species are trapped in them in massive quantities. These nets in many cases are dragged and / or cleaned within the sea, which favors the dispersion of the species, especially taking into account the reproductive thalli with the presence of monospores and vegetative propagules. Similarly, other fishing gear may be favouring the dispersal of the species unintentionally.

			Other possible dispersion vectors of the species may be anchors or boat anchoring systems, both fishing and recreational, diving equipment or marine litter.
2.3. Within Europe, how difficult would it be to contain the organism?	**very difficult**	**very high**	Effective control methods for *R. okamurae* that can be applied in the current state of the invasion are unknown. Only prevention of new introductions are suggested as a potential way to contain the species.
2.4. Based on the answers to questions on the potential for establishment and spread in Europe, define the area endangered by the organism.	Mediterranean, eastern and western European coasts	**high**	 Figure 7: Environmental favourability model for *Rugulopteryx okamurae* in the Mediterranean and western European coasts, based on the native and introduced distribution. The areas with favourable conditions to accommodate the species are shown in warm colours (Altamirano *et al.* 2019).
2.5. What proportion (%) of the area/habitat suitable for establishment (i.e. those parts of Europe were the species could establish), if any, has already been colonised by the organism?	**0-10**	**medium**	The presence in other areas of Europe is not disposable. However, where the species is already established in Alboran Sea, the coverage ranged from 100% to 40% of the seabottom depending on substrata and depth, occupying areas of hectares (Altamirano *et al*. 2019). Considering the predicted distribution of the species in the Mediterranean, it is suspected that the species is at the beginning of its expansion phase.

2.6. What proportion (%) of the area/habitat suitable for establishment, if any, do you expect to have been invaded by the organism five years from now (including any current presence)?	**33-67**	**medium**	New records of *R. okamurae* are available in remote areas to places where it is known that the species is currently established. The new records come from Adra, 150 km to the east, and Punta Umbría, 110 km to the west of the nearest places, from material collected by trawlers and uprights, respectively. Both localities are within the areas that the model recognizes of high favourability for the species, with values greater than 0.9, and it would be important to know if this presence of material deposited on the beaches affects subsequent establishment of the species on the coast. If the presence of fixed plants to the substrate on the coasts of Almería and Huelva are confirmed, the high dispersal capacity of the species and the high potential to continue colonization of favourable environments would be revealed. Furthermore, in-between localities in Granada are reported for new focus of introduction of the species, and new local focus are reported often during the lasts months.
2.7. What other timeframe (in years) would be appropriate to estimate any significant further spread of the organism in Europe? (Please comment on why this timeframe is chosen.)	**10**	**medium**	Considering that since its identification in 2016 and first record in Ceuta, the species has spread along Southern coast of Spain in Alboran Sea in three years, and taking into account its propagation capacity by propagules and monospores together with secondary vectors and natural sea currents, it is expected that *R. okamurae* will keep this active spread performance along its predicted suitable areas on European coasts.
2.8. In this timeframe what proportion (%) of the endangered area/habitat (including any currently occupied areas/habitats) is likely to have been invaded by this organism?	**10-33**	**medium**	The adequacy models have not considered an answer to this specific question. However, 10-33% would be a reasonable estimate in view of the models and taking into account the knowledge about the biology of this species.

2.9. Estimate the overall potential for future spread for this organism in Europe (using the comment box to indicate any key issues).	**rapidly**	**high**	Marine aquatic ecosystems are difficult to manage to prevent the dispersion of invasive species. Marine currents along with human activities can cause a difficult to predict dispersion. However, taking into consideration that the species has spread along Southern coast of Spain in Alboran Sea in three years, and on the other hand its propagation capacity by propagules and monospores together with secondary vectors and natural sea currents, it is expected that *R. okamurae* will keep this active spread performance along its predicted suitable areas on European coasts.

PROBABILITY OF IMPACT

Important instructions:

- When assessing potential future impacts, climate change should not be taken into account. This is done in later questions at the end of the assessment.
- Where one type of impact may affect another (e.g. disease may also cause economic impact) the assessor should try to separate the effects (e.g. in this case note the economic impact of disease in the response and comments of the disease question, but do not include them in the economic section).
- Note questions 2.10-2.14 relate to economic impact and 2.15-2.21 to environmental impact. Each set of questions starts with the impact elsewhere in the world, then considers impacts in Europe separating known impacts to date (i.e. past and current impacts) from potential future impacts. Key words are in bold for emphasis.

QUESTION	RESPONSE	CONFIDENCE	COMMENTS
2.10. How great is the economic loss caused by the organism within its existing geographic range, including the cost of any current management?	**massive**	**very high**	The species produced no economic loss in its native geographic range nor in Thau Lagoon in France. However, economic impact associated to fishering activities and beach management in Southern Spain, has been estimated massive in short time, rising to nearly one million and three hundred thousand euros in nine months (Altamirano et al. 2019).
2.11. How great is the economic cost of the organism currently in Europe excluding management costs (include any past costs in your response)?	**major**	**very high**	*Rugulopteryx okamurae* produces significant detrimental economic impacts since its detection in Spanish waters in 2016, with significant direct and indirect economic losses. The costs are mainly related to the fishing sector as well as to the management of massive accumulation of biomass on the beaches. Losses in ecosystems services has not been estimated. As an indicator of the economic impact on the fishering sector, it has been estimated the economic losses in captures by fisheries associations from Huelva, Cádiz and Málaga provinces. The economic lost in captures in nine months was nearly nine hundred thousand euros,

			and this amount is underestimated due to lacks information provided by fisheries associations (Altamirano *et al*. 2019). Thirteen different fish species captures have been affected by *R. okamurae* with decreases in captures ranging from 20 to 48%. Damage to fishering arts (mainly nets) should be considered as well (Altamirano *et al.* 2019). Another important economic impact in Southern Spain due to *R. okamurae* is derived from the removal of drifted material on the beaches. In a nine months period, a total of 11 thousand tons of drifted material have been removed from beaches in affected localities with expenses rising to nearly four hundred thousand euros (Altamirano *et al.* 2019). Management of this biomass means a big problem for local administrations, and a big public impact. The total economic cost of impacts on fisheries and local administration for beaches managements rises to nearly one million and three hundred thousand euros in nine months.
2.12. How great is the economic cost of the organism likely to be in the future in Europe excluding management costs?	**major**	**high**	The expansion of the organism along the Mediterranean Sea, will entail significant economic losses because of its effects on numerous species, on which the fishing sector depends. Losses in the tourism sector as well as the loss in ecosystems services are also important. The total economic cost of impacts on fisheries and local administration for beaches managements rises to nearly one million and three hundred thousand euros in nine months. If these values are put in time and geographic context in the predicted expansion of the species in the Mediterranean Sea, a bigger economic

			impact can be expected, besides the economic losses in ecosystem services.
2.13. How great are the economic costs associated with managing this organism currently in Europe (include any past costs in your response)?	**major**	**high**	Currently *R. okamurae* is producing high economic costs, associated with the management of massive accumulation of biomass on the beaches, as well as its impact on tourism when the beaches are covered by this seaweed until it can be withdrawn. Furthermore, cleaning of the fishing nets produces important economic impacts in terms of salaries and affected goods. Regional administration in Andalusia is allocating short economic and human resources in monitoring the species but none is done for managing the prevention, content or control (eradication) of the species in these moments.
2.14. How great are the economic costs associated with managing this organism likely to be in the future in Europe?	**major**	**high**	As the established area of the species is currently big and predicted to increase, it can be suggested that costs in managing the species may increase exponentially together with its expansion.
2.15. How important is environmental harm caused by the organism within its existing geographic range excluding Europe?	**minimal**	**high**	Bibliographic references indicate that *R. okamurae* in France (Thau Lagoon) does not exhibit invasive behaviour (Verlaque *et al.* 2009). In its native geographic range, the species behaves as an accompanying species of kelp forest for example, with no invasive performance (Sano *et al.* 2001).
2.16. How important is the impact of the organism on biodiversity (e.g. decline in native species, changes in native species communities, hybridisation) currently in Europe (include any past impact in your response)?	**major**	**high**	*Rugulopteryx okamurae* has shown a very explosive development, colonizing most of the hard seabed substrates in those areas where the species is present. In addition to the impacts related to the tourism and fisheries sector, the expansion of *R. okamurae* is causing a significant environmental impact on native benthic communities, immediately reflected in a very

			important loss of biodiversity as the first and most obvious consequence, and in the long term in a change in the structure and composition of species (Altamirano *et al.* 2017, 2019; Ocaña *et al.* 2016; El Aamari *et al.* 2018; García-Gómez *et al.* 2018; CAGPYDS 2018, 2019). Present coverage of the species has not been estimated, but the value will be over hundreds of hectares including marine protected areas. *Rugulopteryx okamurae* exhibits an extraordinary competitive and colonization capacity with an increase in excessive biomass, unprecedented and nothing comparable with respect to previous invasions in the Spanish coast by other macroalgae such as *Asparagopsis taxiformis* (Altamirano et al. 2008; Zanolla *et al.* 2018a, b, c), *Lophocladia lallemandii* (Patzner 1998; Cabanelles *et al.* 2010; Deudero et al. 2010), the so-called "killer algae" *Caulerpa taxifolia* (Verlaque *et al.* 2015), or the most recent *Caulerpa cylindracea* considered in its moment as the most serious in the history of invasive macroalgae species in the Mediterranean (Klein & Verlaque 2008). In relation to the affected natural spaces, it should be noted that this species is affecting very relevant spaces of great ecological value included in the Natura 2000 Network, such as the ZEC Natural Park of the Strait (ES0000337), the Natural Park Cabo de Gata, the ZEC Seabed of the Bay from Estepona (ES6170036), the ZEC El Saladillo-Punta de Baños (ES6170037), and the ZEC from Calahonda (ES6170030) on the Andalusian coast and for the Spanish African coast the ZEC from Monte Hacho (ES0000197), unique species in Europe where marine biodiversity shows the transition zone between Atlantic and Mediterranean waters, and where

			several protected and endemic specie inhabit. Although the impacts caused by *R. okamurae* on the native communities have not been quantified so far, and the ecological effects of this species remain to be determined, it has been possible to observe the important impact that the development and expansion of this species is causing during the last three years, a fact that has already been revealed (Altamirano *et al.* 2017, 2019; Ocaña *et al.* 2016; El Aamari *et al.* 2018; García-Gómez *et al.* 2018; CAGPYDS 2018, 2019). The observations carried out show a colonization of most hard substrates, with a wide bathymetric range in which *R. okamurae* develops showing large variation of the coverage with the depth. Between the surface and 20m deep there is a practically complete covering of the hard substrates by *R. okamurae*, with covers that can reach 90-100%. It produces a full homogenization of the substrata, being the only single species of macroalga happening, with few spots of another invasive species, *Asparagopsis armata*. Towards deeper levels the coverage decreases but remains at high levels, around 30-40%. In depths around 50m, the presence of the species has also been observed, but less abundant. Due to the high proliferation of this species, the benthic landscape is being deeply transformed affecting all biocenosis and marine communities. In the area of the ZEC of the Strait of Gibraltar, impacts on kelp forests have been detected, significantly affecting the most representative species of these formations such as *Laminaria ochroleuca* and *Saccorhiza polyschides*. These forests have desapeared from the zone due to *R. okamurae*. Forests of *Cystoseira usneoides* and other species of the genus have also been affected, included

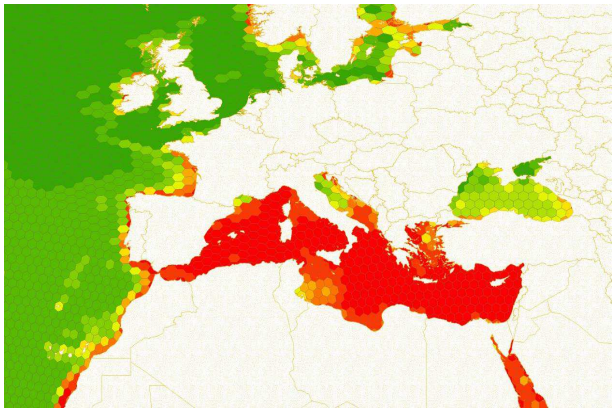
			in the List of Threatened and Endangered Species of the Protocol on Specially Protected Areas and Biological Diversity in the Mediterranean (BOE April 23, 2014); these fucales forest have also dessapeared in many places at the Strait of Gibraltar with the losses in important ecosystem services. Due to the lack of available substrate as a result of the occupation carried out by *R. okamurae*, the macroalgae communities are the first affected. Development of *R. okamurae* beds are so fast that native species cannot compete, and new recruits seem not to be able to grow under the invader canopy. Only previous invasive species like *Asparagopsis armata* or *Caulerpa cylindracea* are able to coexists with few thalli with *R. okamurae*. Other species included in the Protocol mentioned above, such as *Lithophyllum byssoides* or *Gymnogongrus crenulatus*, are also being affected by the presence of *R. okamurae*. In Malaga coast, in the El Saladillo-Punta de Baños ZEC, *Posidonia oceanica* meadows, endemic species of the Mediterranean, and also included in the Paris Protocol have also been affected (CAGPYDS, 2019). *Rugulopteryx okamurae* widely colonizes all the rhizomes of this plant occupying both the interior spaces of the meadow and the edges. Regarding wildlife, numerous invertebrates, especially those of sessile life, are being affected by the development of *R. okamurae*, such as tubes of tubular polychaetes, soft surfaces of holoturias, shellfish crustaceans, sponges, *Sphaerechinus granularis* hedgehog (García-Gómez *et al.* 2018). The coraligen communities have also been affected by the development of *R. okamurae* (Ocaña *et al.* 2016, García-Gómez *et al.* 2018; CAGPYDS 2018). Various types of gorgonians such as *Leptogorgia sarmentosa*,

			Eunicella spp and *Paramuricea clavata* are being particularly affected, in which the algae adheres and covers them almost completely. Furthermore, species of interest such as the *Charonia lampas* horn, the star coral *Astroides calycularis*, the red coral *Corallium rubrum*, or even intertidal species such as the endangered species *Patella ferruginea* are also affected (García-Gómez *et al.*, 2018; El Aamri *et al.*, 2018). Invertabrate fauna associated to *R. okamurae* differs significantly from that of the native *Dictyota dichotoma*, with unpredictable consequences (Navarro-Barranco et al. 2019). Additionally, the accumulation and decomposition of the large biomass generated by this species both in the intertidal and beaches and on the seabed could be causing indirect impacts on coastal ecosystems.
2.17. How important is the impact of the organism on biodiversity likely to be in the future in Europe?	**major**	**high**	Given the significant impact on ecosystems and marine species, which is occurring in the area where it has been detected as invasive in Spain, it is foreseeable that the future impact in the area of possible colonization, such as the Mediterranean and Black Sea, will be of the same level.
2.18. How important is alteration of ecosystem function (e.g. habitat change, nutrient cycling, trophic interactions), including losses to ecosystem services, caused by the organism currently in Europe (include any past impact in your response)?	**major**	**very high**	The significant deterioration of different marine communities in invaded areas reflects the important alteration of functions of ecosystems, so far not studied, given the recent invasion of the species *Rugulopterix okamura*. In addition, its explosive development poses a serious problem for the progressive adaptation of ecosystems to its presence, since it colonizes most of the hard seabed substrates in those areas where it is present.

			Losses in species richness and biodiversity, exhibited in sea bottoms covered only by *R. okamurae* and another invasive species, i.e. *Asparagopsis armata*, represent severe alterations of ecosystem function. The species also produces a change in species composition of invertebrate fauna associated with unpredictable effects on the biota (Navarro-Barranco *et al.* 2019). Important ecosystem services like refuge and breed zones for commercial fishes have been affected, as economic impact data shows. Recreational and social services linked to touristic activities in a touristic hot spot in Europe has also been affected, like beaches in worldwide wellknown beaches of Marbella and Tarifa.
2.19. How important is alteration of ecosystem function (e.g. habitat change, nutrient cycling, trophic interactions), including losses to ecosystem services, caused by the organism likely to be in Europe in the future?	**major**	**high**	Taking into account the effects of R. okamurae on the biocenosis invaded so far, an important alteration of the ecological function of the ecosystems is foreseen, also considering its explosive colonization in most of the hard seabed substrates where the species is present until now in Spain. Considering this in the context of predicted favourable areas in the Mediterranean, it is expected an important alteration of ecosystems function in Europe in the future. Important ecosystem services like refuge and breed zones for commercial fishes have been affected, as economic impact data shows. Recreational and social services linked to touristic activities in a touristic hot spot in Europe has also been affected, like beaches in world wide well known beaches of Marbella and Tarifa.
2.20. How important is decline in conservation status (e.g. sites of nature conservation value, WFD classification) caused by the organism currently in Europe?	**major**	**high**	The decline in conservation status caused by the organism currently in Europe is very important, because of the damage to species and habitats in several

			protected areas of the Natural Network 2000. Great environmental impact on native benthic communities, immediately reflect in an important loss of biodiversity, In the long term a change in the structure and composition of species is expected, as observed in other macroalgae invasions in the Mediterranean. Additionally, the accumulation and decomposition of the large biomass generated by this species both in the intertidal and beaches and on the seabed could be causing indirect impacts on coastal ecosystems. *Rugulopteryx okamurae* exhibits an extraordinary competitive and colonization capacity with an increase in excessive biomass, unprecedented and nothing comparable with respect to previous invasions in the Spanish coast by other macroalgae. In relation to the affected natural spaces, it should be noted that this invasion is affecting very relevant spaces of great ecological value included in the Natura 2000 Network, such as ZEC Natural Park of the Strait (ES0000337), ZEC Seabed of the Bay from Estepona (ES6170036), ZEC El Saladillo-Punta de Baños (ES6170037), and ZEC from Calahonda (ES6170030) on the Andalusian coast and for the Spanish African coast ZEC from Monte Hacho (ES0000197). Although preliminary the observations carried out so far show a colonization of most hard substrates, a wide bathymetric range in which *R. okamurae* develops with large variation of the coverage within depth. Between the surface and 20 m deep there is a practically complete covering of the hard substrates by *R. okamurae*, with covers that can reaching 90-100%. Towards deeper levels the coverage decreases but

			remains at high levels, around 30-40%.
2.21. How important is decline in conservation status (e.g. sites of nature conservation value, WFD classification) caused by the organism likely to be in the future in Europe?	**major**	**high**	It is expected an important decline in the conservation status of protected areas and of special ecological value is expected where this seaweed settles.
2.22. How important is it that genetic traits of the organism could be carried to other species, modifying their genetic nature and making their economic, environmental or social effects more serious?	**Unknown**		
2.23. How important is social, human health or other harm (not directly included in economic and environmental categories) caused by the organism within its existing geographic range?	**minimal**	**medium**	There is no evidence on risks to human health, and / or animal or plant health due to parasites or pathogens of *R. okamurae*. However, the study of substances released into the environment by decomposing biomass is recommended.
2.24. How important is the impact of the organism as food, a host, a symbiont or a vector for other damaging organisms (e.g. diseases)?	**Unknown**	**low**	
2.25. How important might other impacts not already covered by previous questions be resulting from introduction of the organism? (specify in the comment box)	**moderate**	**low**	No other effects are known. It must be studied, for example, the effect of the released that may be produced by the release to substances to the surroundings as a result of the decomposition of the biomass of *R. okamurae*, both in the water column, and in the air in gaseous form, and that could produce some kind of sanitary impact to both the native flora and fauna and the people. It is planned to conduct an analysis of the toxicity of these substances. It may produces impact on marine infrastructures, like

			waste water pipes or energy plants pipes.
2.26. How important are the expected impacts of the organism despite any natural control by other organisms, such as predators, parasites or pathogens that may already be present in Europe?	**major**	**high**	Considering that there are no known predatory organisms, parasites or pathogens that may affect it in the invaded area the impact of *R. okamurae* is of the greatest importance.
2.27. Indicate any parts of Europe where economic, environmental and social impacts are particularly likely to occur (provide as much detail as possible).	[insert text + attach map if possible]	**medium**	Given the invasive potential of the species and its environmental characteristics detected in the invaded area, it is expected that both the Mediterranean Sea and Black Sea will be affected.  Figure 8: Economic, environmental and social impacts are particularly likely to occur in the areas that present favourable conditions to accommodate the species (shown in warm colors) Specially fishing and touristic areas may suffer important impacts, similar to what already happened in Southern Spain.

RISK SUMMARIES

	RESPONSE	CONFIDENCE	COMMENT
Summarise Entry	**very likely**	**high**	Based on the introduced distribution area of *R. okamurae*, since 2002 there is evidence of its presence in France, in the Thau lagoon, on the French Mediterranean coast near Montpellier (Verlaque et al. 2009) while in Spain the species was identified for the first time in Ceuta (Altamirano *et al*. 2016). The entry routes are a priori unintentional, potentially associated with ballast waters and marine crops, without ruling out others. It is unknown if there are intentional routes of entry. The seaweed has shown survival rates between 80-100% after being grown in dark conditions for three weeks, depending on the temperature during cultivation, and that the thallus even increased their biomass during this time (Rosas-Guerrero et al, 2018), which makes the entry of organisms with ballast water very easy, which is supposed to be the main route of entry. In addition to the entire coast of the Iberian Peninsula, including the Spanish and French Atlantic coast and the Balearic archipelago, there are other areas on the planet with the potential to accommodate the species, highlighting the whole of southern Europe, which have very high probability to be colonized. It is necessary to make effective the International Convention for the control and management of ballast water and sediments of ships, made in London on February 13, 2004, and ratified by the Head of State in 2016, for its entry into force in Spain on September 8, 2017 (BOE 282, of November 22, 2016). This is especially important in regions such as the Strait of

			Gibraltar, which withstand heavy commercial maritime traffic in the form of large merchants and tourist cruises, which maintain their maneuverability thanks to ballast waters.
Summarise Establishment	**very likely**	**high**	Species distribution models highlight the high and widespread favourability for the species in the Mediterranean Sea, but also in other areas of Europe such as the Black Sea and other places on the planet, such as the northern and western coasts of Australia, Gulf Persian, much of the east coast of the Americas. Taking into account the high propagation capacity of *R. okamurae*, observed in the populations introduced in the Strait of Gibraltar, and the extraordinary competitive and colonization capacity with an increase in excessive biomass, it can be affirmed that the establishment is very probable once entered in an area that presents favorable conditions (environmental factors). However, the sources and vectors that help their establishment must still be identified.
Summarise Spread	**very rapidly**	**high**	Other potential vectors responsible for the dispersion of the species and secondary introductions are the following: - Fishing arts - Recreational boats - Diving equipment - Funding systems - Marine litter Possibly the most important vectors at the moment in the dispersion of R. okamurae are those linked to fishing activities, whose gear accidentally collects fixed

			and / or suspended algae material, sometimes in large quantities. Taking into account the high propagation capacity of R. okamurae, mainly due to asexual and vegetative mechanisms, it is essential to avoid the dispersion of thallus by vectors linked to human activities, since it is not possible to stop dispersal by sea currents.
Summarise Impact	**massive**	**very high**	*Rugulopteryx okamurae* has shown a very explosive development extending from the area of the Strait of Gibraltar to the Atlantic and Mediterranean coasts of Cadiz and Malaga, as well as to the Chafarinas Islands, Huelva and Almeria, in just over 3 years. The species exhibits an extraordinary competitive and colonization capacity with an increase in excessive biomass, unprecedented and nothing comparable with respect to previous invasions in the Spanish coast by other macroalgae. The accumulation and decomposition of the excessive biomass generated by this species, both in the intertidal and beaches, as in the seabed, could be causing indirect impacts on marine ecosystems, as well as large economic losses. **Environmental impact:** *R. okamurae* shows large variation of the coverage with the depth. Between the surface and 20m deep there is a practically complete covering of the hard substrates by *R. okamurae*, with covers that can reach 90-100%. It produces a full homogenization of the substrata, being the only single species of macroalga happening, with few spots of another invasive species, *Asparagopsis armata*. Towards deeper levels the coverage decreases

			but remains at high levels. The benthic landscape is being deeply transformed affecting all biocenosis and marine communities. With a a very important loss of biodiversity as the first and most obvious consequence, and in the long term in a change in the structure and composition of species (Altamirano *et al.* 2017, 2019; Ocaña *et al.* 2016; El Aamari *et al.* 2018; García-Gómez *et al.* 2018; CAGPYDS 2018, 2019). Its competition with native species and the alteration of the ecological balance of marine ecosystems in this area, produces a very important loss of biodiversity. Regarding wildlife, numerous invertebrates, are being affected by the development of *R. okamurae*: i.e. tubes of tubular polychaetes, soft surfaces of holoturias, shellfish crustaceans, sponges, *Sphaerechinus granularis* hedgehog (García-Gómez *et al.* 2018). The coraligen communities have also been affected by the development of *R. okamurae* (Ocaña *et al.* 2016, García-Gómez *et al.*, 2018; CAGPYDS, 2018). Various types of gorgonians such as *Leptogorgia sarmentosa*, *Eunicella* spp and *Paramuricea clavata* are being particularly affected, in which the algae adheres and covers them almost completely. Furthermore, species of interest such as the *Charonia lampas* horn, the star coral *Astroides calycularis*, the red coral *Corallium rubrum*, or even intertidal species such as the endangered species *Patella ferruginea* are also affected (García-Gómez *et al.*, 2018; El Aamri *et al.*, 2018). In relation to the affected natural spaces, it should be noted that this species is affecting very relevant spaces

			of great ecological value included in the Natura 2000 Network, such as the ZEC Natural Park of the Strait (ES0000337), the Natural Park Cabo de Gata, the ZEC Seabed of the Bay from Estepona (ES6170036), the ZEC El Saladillo-Punta de Baños (ES6170037), and the ZEC from Calahonda (ES6170030) on the Andalusian coast and for the Spanish African coast the ZEC from Monte Hacho (ES0000197), unique species in Europe where marine biodiversity shows the transition zone between Atlantic and Mediterranean waters, and where several protected and endemic specie inhabit. Additionally, the accumulation and decomposition of the large biomass generated by this species both in the intertidal and beaches and on the seabed could be causing indirect impacts on coastal ecosystems. **Economic impact:** Economic impact of the species is high associated to losses in fisheries and beach management. Total amount estimated for nine months is higher than one million euros. Tourism sector also has experimented great losses the last summers. **Social impact:** Recreational and social services linked to touristic activities in a touristic hot spot in Europe has also been affected. The large amounts of biomass accumulated on the beach creates social alarm.
Conclusion of the risk assessment	**high**	**high**	*Rugulopteryx okamurae* exhibits an extraordinary competitive and colonization capacity with an increase in excessive biomass, unprecedented and nothing

			comparable with respect to previous invasions in the Spanish coast by other macroalgae such as *Asparagopsis taxiformis* (Altamirano et al. 2008; Zanolla et al. 2018a, b, c), *Lophocladia lallemandii* (Patzner 1998; Cabanelles et al. 2010; Deudero et al. 2010), the so-called "killer algae" *Caulerpa taxifolia* (Verlaque et al. 2015), or the most recent *Caulerpa cylindracea* considered in its moment as the most serious in the history of invasive macroalgae species in the Mediterranean (Klein & Verlaque 2008). There are no previous control experiences that can serve as a reference. Among the possible actions associated with the control of the species, the inclusion in the European list is considered very positive to help identify the vectors of introduction and dispersion of human activities (ballast water, marine crops, boat hulls, as well as fishing-related activities), prevent new introductions in order to prevent the spread of the species, especially in some space or habitat of community interest. Due to the predicted high favourability for the species of Mediterranean European coasts, it is of interest to alert other exposed countries about this new invasive species, so they can implement prevention management mechanisms to avoid the invasion in their waters.

ADDITIONAL QUESTIONS - CLIMATE CHANGE			
3.1. What aspects of climate change, if any, are most likely to affect the risk assessment for this organism?	[Temperature rise]	**medium**	There is no scientific evidence that can confirm that climate change can affect this species, although it can be suspected that changes in seawater temperature could affect its vegetative and reproductive activity, not knowing the trends of these potential changes.
3.2. What is the likely timeframe for such changes?	**10 years**	**medium**	
3.3. What aspects of the risk assessment are most likely to change as a result of climate change?	[EU CHAPPEAU]	**low**	Probably the distribution of the organism in Europe would be different.
ADDITIONAL QUESTIONS - RESEARCH			
4.1. If there is any research that would significantly strengthen confidence in the risk assessment please summarise this here.	[molecular tools and genetic analysis]	**medium**	It would be necessary to identify other vectors and assess their importance, such as transport in ship hulls or marine cultures of various kinds, in order to act on them. It is also necessary to be able to know the sources of origin of the inoculums that are introduced in the Mediterranean, so that, together with the knowledge of the vectors, to be able to identify the main transport routes of *R. okamurae*, and to be able to develop management plans focused on an efficient prevention. In this sense, molecular tools and genetic analysis can be of great help, as already shown. It is also necessary to study the life cycle of the species in the new introduced areas, in order to understand its propagation systems and identify its weaknesses in biology. It is also interesting to study the ecological performance of the species in order to understand its competitive capacity with native communities. Assessing its genetic variability is important to predict

			its fate in the invaded area and resistance to changes in the environment.

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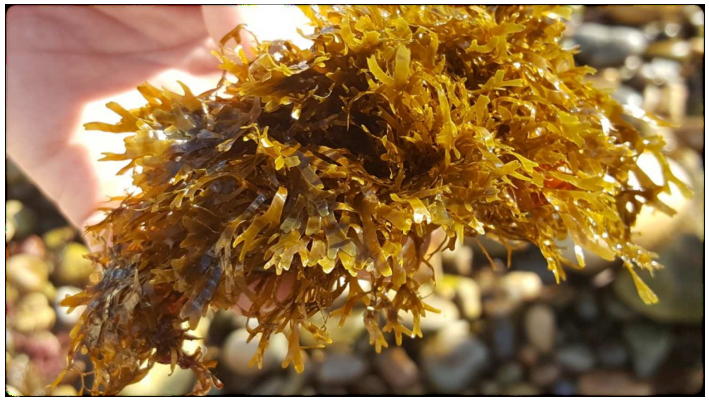
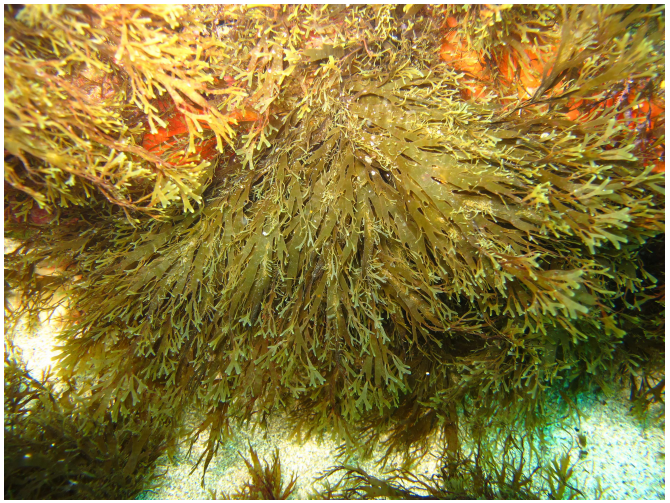
PHOTOGRAPHIC ANNEX



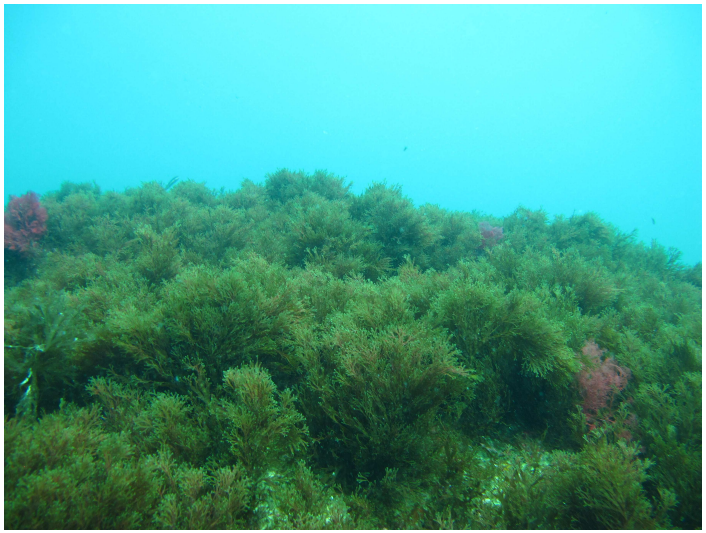
Ruguloptery okamurae. M. Altamirano



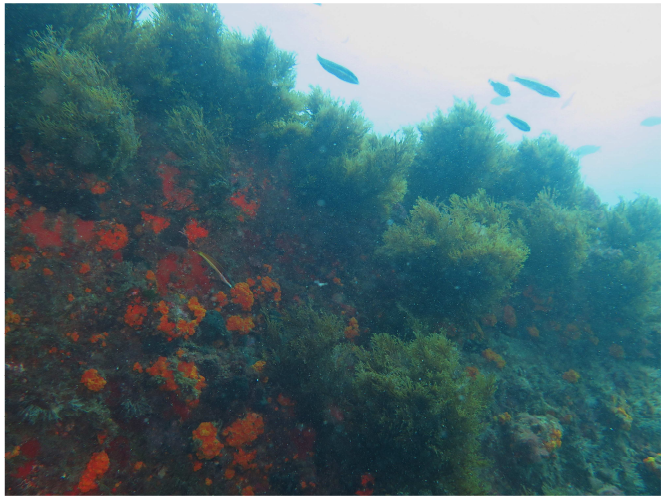
Ruguloptery okamurae. M. Altamirano

Los Lances beach, Tarifa, Cádiz, may 2019. M. Altamirano.

Benzú beach, Ceuta, july 2016. J. Martínez.



Massive development of R. okamurae on platforms shallow rockies (-20 m). J. De la Rosa



Development of R. okamurae on *Astroides calycularis*. J. De la Rosa