

EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION ORGANISATION EUROPEENNE ET MEDITERRANEENNE POUR LA PROTECTION DES PLANTES



17-23146

Pest Risk Analysis for Hygrophila polysperma



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This pest risk analysis scheme has been specifically amended from the EPPO Decision-Support Scheme for an Express Pest Risk Analysis document PM 5/5(1) to incorporate the minimum requirements for risk assessment when considering invasive alien plant species under the EU Regulation 1143/2014. Amendments and use are specific to the LIFE Project (LIFE15 PRE FR 001) 'Mitigating the threat of invasive alien plants to the EU through pest risk analysis to support the Regulation 1143/2014'.

Cite this document as:

EPPO (2017) Pest risk analysis for Hygrophila polysperma. EPPO, Paris. Available at:

Photo: Hygrophila polysperma (Andreas Hussner)

EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION

Pest risk analysis for Hygrophila polysperma

This PRA follows EPPO Standard PM5/5 Decision support scheme for an Express Pest Risk Analysis

PRA area: EPPO region

First draft prepared by: Andreas Hussner

Location and date: Paris (FR), 2016-10-03/07

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The pest risk analysis for *Hygrophila polysperma* has been performed under the LIFE funded project:



LIFE15 PRE FR 001

Mitigating the threat of invasive alien plants to the EU through pest risk analysis to support the Regulation 1143/2014

In partnership with

EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION

And

NERC CENTRE FOR ECOLOGY AND HYDROLOGY





Review Process

- This PRA on Hygrophila polysperma was first drafted by Andreas Hussner
- The PRA was evaluated under an expert working group at the EPPO headquarters between 2016-10-03/07
- Following the finalisation of the document by the expert working group the PRA was peer reviewed by the following:
 - (1) The EPPO Panel on Invasive Alien Plants (October and November 2016)
 - (2) The EPPO PRA Core members (December 2016)

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Summary¹ of the Express Pest Risk Analysis for Hygrophila polysperma

PRA area: EPPO region (see https://www.eppo.int/ABOUT_EPPO/images/clickable_map.htm.)

Describe the endangered area:

Hygrophila polysperma is not naturalised in any natural environment within the EPPO region. The species is present in thermally heated waters (which are uncharacteristic of natural conditions) in Austria, Germany, Hungary and Poland. Hygrophila polysperma is a frost sensitive species. Climate modelling indicates that under the current projections, the majority of the EPPO region is unsuitable for the establishment of the species (see Appendix 1). Under current climatic conditions very small areas of Turkey, Greece and Algeria are marginally suitable along the Mediterranean coastline (the Mediterranean biogeographical region).

Furthermore, thermally abnormal waters in other EPPO countries provide potential habitats for *H. polysperma*.

Habitats within the endangered area include slow moving rivers, canals, irrigation and drainage systems, lakes, reservoirs.

Main conclusions

The results of this PRA show that *Hygrophila polysperma* poses a low risk to the endangered area under current climatic projections (very small areas of Turkey, Greece and Algeria are marginally suitable along the Mediterranean coastline) with a moderate uncertainty. *Hygrophila polysperma* is not naturalised in any natural environment within the EPPO region.

The Expert Working Group does not recommend any phytosanitary measures for this species.

The Expert Working Group recommends that the PRA is reviewed every ten years and/or when significant new information (e.g. naturalisation in natural environment of the endangered area or ecological data) becomes available.

Entry and establishment

Hygrophila polysperma is not naturalised in any natural environment within the EPPO region. The overall likelihood of Hygrophila polysperma entering the EPPO region is high. Hygrophila polysperma is imported into the EPPO region, traded and normally established in protected conditions, for example under glass. The species can establish in artificial, especially thermally influenced water bodies.

Potential impacts in the PRA area

Note: a lot of the information on impacts for this species, i.e. in the form of factsheets available on the internet, has been disqualified in this PRA because they contain generalised, unreferenced and unsupported statements about impacts throughout its invasive range.

In thermally abnormal waters in the River Erft, Germany, *H. polysperma* has locally suppressed a native plant species (Personal Communication, A. Hussner, 2016, see Appendix 3, Fig. 5). In Poland, within a dense stand of *H. polysperma*, the oxygen concentration was found to be 3.1 mg per litre (Gabka & Owsianny, 2009), below concentrations required to support cyprinids (EEC, 1978). Negative effects on fishes and macroinvertebrates, which are reported from other countries where *H. polysperma* is invasive, can be expected if *H. polysperma* reaches similar levels of distribution. *Hygrophila polysperma* blocks the sunlight and reduces the wind induced mixing of the water column, and these effects are independent of the region in which *H. polysperma* becomes invasive.

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¹ The summary should be elaborated once the analysis is completed

Hygrophila polysperma reduces the functioning of drainage and irrigation systems and flood control canals. H. polysperma stands provide a suitable habitat for mosquitoes, which might carry diseases.

Climate modelling indicates that under the current conditions, the majority of the EPPO region is unsuitable for the establishment of the species (see Appendix 1). Very small areas of Turkey, Greece and Algeria are marginally suitable along the Mediterranean coastline. Impacts are not predicted to happen under the current climate as the species will not establish.

Habitats within the endangered area include slow moving rivers, canals, irrigation and drainage systems, lakes, reservoirs.

Hygrophila polysperma is not naturalised in any natural environment within the EPPO region.

Climate change

Under climate change scenario RCP8.5 (**Note:** RCP8.5 is the most extreme of the RCP scenarios, and may therefore represent the worst-case scenario for reasonably anticipated climate change) for 2070s, Europe and the Mediterranean are projected to remain largely unsuitable for *H. polysperma*. However, some areas projected as marginally to moderately suitable appear in northern Portugal, southwest France, Greece, Italy, the eastern Adriatic coast, southern Turkey and Georgia. Under this climate change scenario, the biogeographic regions where the species can potentially establish are the Mediterranean, Continental, Black Sea and Atlantic biogeographical regions.

Phytosanitary measures:

The major pathway being considered is:

Plants for planting

Given the low risks for establishment and impact on the natural and managed environment within the endangered area the Expert Working Group does not recommend any phytosanitary measures for this species.

National awareness raising measures:

- There are no national prevention measures for the sale of *Hygrophila polysperma* in any country within the endangered area. The Expert Working Group recommends *H. polysperma* should be monitored where it occurs in the wild.
- The Expert Working Group encourages industry to assist with public education campaigns associated with the risk of aquatic non-native plants.

For additional information see:

See Standard PM3/67 'Guidelines for the management of invasive alien plants or potentially invasive alien plants which are intended for import or have been intentionally imported' (EPPO, 2006).

See Standard PM9/19 (1) 'Invasive alien aquatic plants' (EPPO, 2014).

See Standard PP 3/74 (1) 'EPPO guidelines on the development of a code of conduct on horticulture and invasive alien plants' (EPPO, 2009).

Phytosanitary risk for the endangered area (current/future climate) Pathways for entry Plants for planting: High/High Establishment: Natural environment: Low/Moderate Managed environment: Low/Moderate Spread: High/High Impacts (in current area of distribution) Impact on biodiversity: Moderate/Moderate Impacts on ecosystem services: Moderate/Moderate Socio-economic impacts: High/High Impacts (in the PRA area) Impact on biodiversity: Low/Moderate Impacts on ecosystem services: Low/Moderate	High	Moderate	Low	x
Socio-economic impacts: Low/Moderate Level of uncertainty of assessment (current/future climate)				
Pathways for entry Pathways for entry Plants for planting: Low/Low Establishment: Natural environment: Low/High Managed environment: Moderate/High Spread: Moderate/High Impacts (in current area of distribution) Impact on biodiversity: High/High Impacts on ecosystem services: High/High Socio-economic impacts: Moderate/High Impact on biodiversity: Moderate/High Impacts on ecosystem services: Moderate/High Socio-economic impacts: Moderate/High Socio-economic impacts: Moderate/High	High	Moderate X	Low	

Other recommendations:

Inform EPPO or IPPC or EU

• The Expert Working Group recommends *H. polysperma* should be monitored where it occurs in the wild within the endangered area.

Inform industry, other stakeholders

• Encourage industry to assist with public education campaigns associated with the risk of aquatic non-native plants.

Express Pest Risk Analysis: *Hygrophila polysperma* (Roxb.) T. Anderson

First draft prepared by: Dr. Andreas Hussner, Jackels Umweltdienste GmbH, Siemensring 9, 41334 Schwalmtal

Date: 2016-08-03

Stage 1. Initiation

Reason for performing the PRA:

Hygrophila polysperma (Roxb.) T. Anderson has a strong negative impact in other regions of the world, which warrants an evaluation of its potential impacts in the EPPO region. The high phenotypic plasticity allows the species to grow in variable habitats, and the predicted climate change will result in increasing suitable habitat in the EPPO region. Overall, species biology, its impacts and the predicted spread potential make a PRA for the EPPO region essential. H. polysperma currently has a limited distribution in the EPPO region. The species is present in thermally heated waters in Austria, Germany, Hungary and Poland. H. polysperma was added to the EPPO Alert List in 2010 and transferred to the EPPO List of Alien Invasive Plants in 2012. In 2016, the species was prioritized (along with 36 additional species from the EPPO List of Invasive Alien Plants and a recent horizon scanning study²) for PRA within the LIFE funded project "Mitigating the threat of invasive alien plants to the EU through pest risk analysis to support the Regulation 1143/2014'. H. polysperma scored a high priority for PRA and was thus included in the list of 16 species to undergo risk analysis as part of the LIFE project.

PRA area:

The EPPO region (see https://www.eppo.int/ABOUT_EPPO/images/clickable_map.htm.)

²

 $http://ec.europa.eu/environment/nature/invasivealien/docs/Prioritising \%\,20 prevention \%\,20 efforts \%\,20 through \%\,20 horizon \%\,20 scanning.pdf$

Stage 2. Pest risk assessment

1. Taxonomy: *Hygrophila polysperma* (Roxb.) T. Anderson (Kingdom Plantae; Phylum Spermatophyta; Class Dicotyledonae; Order: Lamiales; Family Acanthaceae; Genus *Hygrophila*) (according to CABI)

EPPO Code: HYGPO

Synonyms:

Hemiadelphis polysperma (Roxb.) Nees, Justicia polysperma Roxb. (ThePlantList)

Common names: Indian swampweed, East Indian hygrophila, Miramar weed, Dwarf Hygrophila, Green hygro, **German name**: Indischer Wasserfreund, **Dutch**: Belgisch groen

Plant type: Rooted amphibious perennial herb

Related species in the EPPO region:

Native: none

Non-native: Hygrophila corymbosa (Blume) Lindau, Hygrophila difformis Blume

Additional species used within the aquatic plant trade are: Hygrophila costata Nees (Syn. H. guianensis Nees, Hygrophila lacustris (Cham. & Schltdl.) Nees), Hygrophila odora (Nees) T. Anderson, Hygrophila surinamensis Bremek.; Hygrophila angustifolia R. Br. (Syn. of Hygrophila ringens (L.) R. Br. ex Spreng.), Hygrophila corymbosa (Blume) Lindau; Hygrophila salicifolia (Vahl) Nees (Syn. of Hygrophila ringens (L.) R. Br. ex Spreng.), Hygrophila stricta (Vahl.) Lindau; Unresolved names: Hygrophila balsamica Raf., Hygrophila difformis Blume, Hygrophila pinnatifida (Dalzell) Sreem. (Hussner et al. 2014).

2. Pest overview

Introduction

Hygrophila polysperma is a submerged or emerged growing, rooted aquatic plant. H. polysperma grows in stagnant and running water, marshes and rice fields (Thayer et al., 2016). It is a native to Asia (Bangladesh, Bhutan, Cambodia, China, India, Laos, Malaysia, Myanmar, Nepal, Pakistan, Taiwan, Thailand, Vietnam) (Angerstein & Lemke, 1994) and was introduced into the US in the 1950s (Nault & Mikulyuk, 2009) and Mexico (Mora-Olivo et al., 2008). In the EPPO region, H. polysperma was first reported from the thermally heated River Erft, Germany (Hussner et al., 2007). Gabka & Owsianny (2009) found H. polysperma in a reservoir which is used as a cooling pond for nearby power plants in Poland, and Lukács et al. (2016) from thermally heated channels in Hungary. In addition, the species has been recorded from thermal Villacher Warmbad waters in Austria (Bundesministerium für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft, 2013).

Environmental requirements

Hygrophila polysperma grows best at temperatures between 22 – 28 °C, with a minimum temperature of 4 °C (Spencer & Bowes, 1985; Kasselmann, 1995). In Virginia, *H. polysperma* was documented to tolerate freezing temperatures for brief periods (Cuda & Sutton, 2000 citing Reams, 1953), while a study in New Zealand found that emergent plants did not survive during the winter even without freezing water temperatures (Burnett, 2008). Emerged plants show generally increased vegetative growth than submerged plants (Botts *et al.*, 1990), and the growth rates were highest when emerged plants root in 5 cm water depth (Fast *et al.*, 2008). The growth rate of *H. polysperma* is highly related to the availability of ammonia-nitrogen in the sediment (Sutton & Dingler, 2000).

A temperature decrease from 30 °C to 10 °C reduced net photosynthesis only by about 25 % (Spencer & Bowes, 1985). The light saturation of photosynthesis of submerged and emerged shoots is at 400 μE and 600 μE respectively. Submerged plants usually grow in waters with a pH <7.8 (Spencer & Bowes, 1995), but in calcareous waters it has been found to grow at pH up to 8.5 (Personal Communication, W. Haller, 2016), indicating a limited ability to use HCO₃. At pH 9 a biomass loss was documented (Spencer & Bowes, 1995). This might explain why *H. polysperma* grows best in flowing waters (Van Dijk *et al.*, 1986) as the flow reduces boundary layer effects during photosynthetic carbon uptake. This has been also been demonstrated where plants have a higher growth rate when flushed twice per week compared to when grown under stagnant conditions (Fast *et al.*, 2008). Field observations of *H. polysperma* growing in a fast flowing spring outlet were made but the species was not observed in the downstream adjacent lake (Walk in Water Lake) (Personal Communication, W. Haller, 2016).

H. polysperma predominantly spreads via plant fragments, as high regeneration rates were found for small stem fragments with nodes (Spencer & Bowes, 1985). Apical shoot fragments of 3cm show high regeneration rates, but stem fragments of 2cm with one node are also able to regenerate, even though in a lesser extent (Personal communication, A. Hussner, 2017).

Plant fragments showed regrowth capacities of 100 % for shoot fragments with three or more nodes per fragment (Spencer & Bowes, 1985). Even single detached leaves are able to regrowth into new plants (Sutton, 1995). However, the number of produced plant fragments was documented as low compared to other invasive aquatic plants (like *Egeria densa* or *Vallisneria spiralis*) in a study in the River Erft, Germany (Heidbüchel *et al.* 2016).

Submerged plants withstand environments with freezing air temperatures, as long as the water temperature does not drop below 9 °C, as found in the River Erft, Germany (Hussner, 2014). But even single detached leaves are able to regrowth into new plants (Sutton, 1995). In the invasive range in the USA, no seed production was found (Spencer & Bowes, 1985).

Habitats

In both the native and introduced range, *Hygrophila polysperma* grows in both aquatic and riparian habitats, and particularly in shallow slow flowing waters high biomass densities were reported (Van Dijk *et al.*, 1986; Cuda & Sutton, 2000). *H. polysperma* prefers flowing rivers but can be found as well in stagnant waters like canals, ditches, irrigation ditches and lakes and also grows in marshes, swamps and wetlands (Nault & Mikulyuk, 2009, Thayer *et al.*, 2016). *H. polysperma has* been observed to grow on damp soils in seasonally flooded areas.

Identification

Hygrophila polysperma is a rhizomatous perennial aquatic plant with stems four angled and opposite leaves. The plants predominantly grow submerged, but shoots can reach the water surface and become floating and emergent (see Appendix 3, Fig. 1 & 2). The stems reach lengths of up to 2 m (CABI, 2016). The roots are either rooted in the sediment or float freely in the water column from floating shoots. The leaves are oblong to elliptic, sparsely hairy and broader to the tips (See Appendix 3, Fig. 3). Even though *H. polysperma* does not show heterophyllous leaves (Sutton, 1985), the submerged leaves tend to be larger than emerged leaves (Cuda & Sutton, 2000). Stems are often prostrate, 4-angled, slightly swollen above nodes. Leaves are hairless, opposite, petiole to 5 mm; leaf blade oblong-lanceolate to ovate, $2-3.5 \times 0.6-1.3$ cm. Flowers in terminal spikes, white with a blue tinge, around 5 mm in length produced from September to November. Fruit a capsule linear-oblong, 5.5-8 mm, 20-30-seeded. Seeds ca. 1×0.5 mm.

Symptoms

In the invaded region of North America, *H. polysperma* builds up high biomass densities which occupies the whole water column and can outcompete and shade out both native and alien invasive plant species (e.g. *Hydrilla verticillata*, Van Dijk *et al.*, 1986) in shallow water and river ecosystems (Spencer & Bowes, 1985; Angerstein & Lemke, 1994; Cuda & Sutton, 2000; Ramey, 2001; Doyle *et al.*, 2003). The species has also been found to be a weed in rice fields in Asia (Krombholz, 1996). Dense stands can clog waterways and interfere with irrigation and flood control systems (Schmitz & Nall, 1984; Sutton, 1995). Navigation and the recreational use of water bodies (for fishing, diving, swimming and boating) can be limited (Cuda & Sutton, 2000).

Shading of the water column by dense floating mats can cause oxygen depletion due to reduced water circulation and light limitation for photosynthesis of primary producers accompanied with their increased die off and decomposition (Cuda & Sutton, 2000), which is the case for several floating aquatic plants and thus must be considered as highly likely also for *H. polysperma* mats. Similar to other aquatic plants with a similar growth form, dense mats of *H. polysperma* can provide habitat for mosquitoes, and the mosquito *Coquillettida perturbans* (a vector for encephalomyelitis) was found attached to submerged roots of *H. polysperma* (Cuda & Sutton, 2000).

Existing PRAs for Hygrophila polysperma

Europe:

In Europe *H. polysperma* was added to the EPPO Alert List in 2010 and transferred to the List of Invasive Alien Plants in 2012 (EPPO, 2012).

USA:

In the USA, weed risk assessments classified *H. polysperma* as a species of high risk (USDA, 2015). *H. polysperma* is a State Noxious Weed in eight States, and is considered as a U.S. Federal Noxious Weed (USDA, 2015). *H. polysperma* was evaluated for Florida using a modified version of the AWRA (Pheloung *et al.*, 1999). Under this assessment *H. polysperma* scored 25, indicating a high probability of invasion (Invasive Plant Working Group, 2016).

New Zealand:

In New Zealand, *H. polysperma* scored 44 out of 100 points in the Aquatic Weed Risk Assessment (AWRAM), indicating a moderate weed risk (Champion & Clayton, 2001).

Australia:

In Australia *H. polysperma* scores 53 out of 130 points using the aquatic Australian version of the Aquatic Weed Risk Assessment Model (Champion *et al.* 2008), indicating some weed risk. The study recommended that further evaluation was required to properly assess its weed potential.

Pacific Islands:

For the Pacific Islands, a weed risk assessment based on the New Zealand and Australian method and adapted to the Pacific Islands identified *H. polysperma* as a species of high risk (PIER, 2016; http://www.hear.org).

Socio-economic benefit

Hygrophila polysperma is a high value species to the aquatic trade. One aquarium supplier in Australia advised that prior to its declaration as a noxious weed in New South Wales it was their third highest species traded in that State (Personal Communication, Andrew Petroeschevsky, 2016). In aquarium environments its attractiveness and easiness to grow and hardiness make it a popular plant particularly amongst beginners.

In the EPPO region, the plant is sold in large quantities (Brunel, 2009) and is available from numerous online suppliers (Hussner *et al.* 2014). The Ornamental Aquatic Trade Association (UK based) carried out a survey with its members in August 2016 requesting advise on the number of plants and value that they had sold in the calendar year for 2015. Thirty-three members responded to this survey and detailed that in total 478 459 *H. polysperma* plants were sold in the UK in 2015 with a value of GBP 559 677.

3. Is the pest a vector?

No

Although not a direct vector of organisms, indirectly *H. polysperma* can create suitable habitats for the mosquito species *Coquillettida perturbans*, a vector for encephalomyelitis (Cuda & Sutton, 2000).

4. Is a vector needed for pest entry or spread? No

No. A vector is not needed for the entry of this weed species into the PRA area.

5. Regulatory status of the pest

EPPO region:

There are no regulations for *H. polysperma* in the EPPO region.

USA:

In the USA, *H. polysperma* has varying classifications at a federal, government or state level. In Alabama: Class A – noxious weed; California: Quarantine; Florida: Prohibited aquatic plant, Class 2; Massachusetts: Prohibited; North Carolina: Class A – noxious weed; Oregon: Quarantine; South Carolina: Invasive aquatic plant, plant pest; Vermont: Class A – noxious weed (USDA 2016; http://plants.usda.gov/core/profile?symbol=HYPO3).

Australia:

H. polysperma is declared as a noxious weed in New South Wales (http://weeds.dpi.nsw.gov.au/Weeds/Details/154).

6. Distribution

Continent	Distribution (list countries, or provide a general indication, e.g. present in West Africa)	Provide comments on the pest status in the different countries where it occurs (e.g. widespread, native, introduced)	Reference
Asia	Bangladesh, Bhutan, Cambodia, China, India, Laos, Malaysia, Myanmar, Nepal, Pakistan, Taiwan, Thailand, Vietnam	Widespread and native throughout tropical Asia	CABI (2016)
North America	(1) Present in the USA: Florida and Texas (naturalized), Virginia (current status unknown), Kentucky (established) (2) Present in Mexico	(1) North America (restricted Southern distribution, introduced) (2) Mexico (locally established populations, introduced)	GBIF.org (2017), USDA (2016); Angerstein and Lemke (1994); USDA NRCS, (2016).
Europe	Present in Austria, Germany, Poland, Hungary	Local occurrences in thermally heated waters, introduced	Hussner et al. (2007), Gabka & Owsianny, (2009), Lukács et al. (2014), Bundesministerium für Land- und Forstwirtschaft, Umwelt und

Continent	Distribution (list countries, or provide a general indication, e.g. present in West Africa)	Provide comments on the pest status in the different countries where it occurs (e.g. widespread, native, introduced)	Reference
			Wasserwirtschaft (Ed) (2013).
Oceania	Australia	Restricted eastern distribution in Australia, introduced	www.weeds.dpi.ns w.gov.au/Weeds/D etails/154

Introduction

H. polysperma is found in Asia, Australia, Europe, the USA and Mexico. The centre of origin of *H. polysperma* is reported to be in Asia (Nault & Mikulyuk, 2009) (See Appendix 4, Fig.1).

Asia

H. polysperma is native and widespread in Bangladesh, Bhutan, Cambodia, China, India, Laos, Malaysia, Myanmar, Nepal, Pakistan, Taiwan, Thailand and Vietnam (See Appendix 4, Fig.2).

America

H. polysperma was introduced into the USA in 1945 (Innes, 1947). In Virginia, *H. polysperma* was reported in the 1950s for the first time in the wild and became established for 15-20 years, until extreme cold winters in the 1970s killed the populations. Established in Kentucky in 2009 (USGS, 2016). In Florida, *H. polysperma* was found in the wild in 1965 (Les & Wunderlin, 1981) and became established and spread into rivers, canals, lakes and ditches. In Texas, the species was reported for the first time in 1969 and has become established (Angerstein & Lemke, 1994) (See Appendix 4, Fig.3).

In Mexico, *H. polysperma* was found in 1985 in a lagoon (laguna del Chairel) (Mora-Oliva *et al.* 2008).

Australia

The species is reported from New South Wales and Queensland (http://weeds.dpi.nsw.gov.au/Weeds/Details/154). It was first discovered growing in the

Caboolture River in South East Queensland in 2005. In 2006 further occurrences were discovered in New South Wales.

Europe

In Europe, *H. polysperma* was found 2005 in the Kasterer Mühlenerft, a side branch of the River Erft, Germany (Hussner *et al.* 2007). The species spread within this thermally abnormal river and occurred within a >30 km river stretch with small populations (Hussner, 2014).

In Austria the species has been recorded from thermal waters in Villacher Warmbad though there are no further details on timing of occurance or population size (Bundesministerium für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft (Ed) 2013).

In Poland, *H. polysperma* was found in 2008 in a cooling reservoir of power stations (Gabka & Owsianny, 2009).

In Hungary, a population of *H. polysperma* was reported from a thermally heated water system though no further details on the population size are detailed (Lukács *et al.*, 2014) (See Appendix 4, Fig.4).

7. Habitats and their distribution in the PRA area

Habitats	EUNIS habitat types	Present in PRA area (Yes/No)	Comments (e.g. major/minor habitats in the PRA area)	Reference
Freshwater bodies including canals, rivers (slow moving), ponds, irrigation channels, estuaries and lakes	C1 : Surface standing waters C2 : Surface running waters	Yes	Major habitat(s) within the PRA area and the habitat(s) at the highest risk of invasion	Hussner (2014)
Riverbanks	C3: Littoral zone of inland surface waterbodies	Yes	Major habitat within the PRA area.	Personal Communication Petroeschevsky (2016) (see Appendix 3, Fig.4).
Wetlands	C3: Littoral zone of inland surface waterbodies	Yes	Major habitats within the PRA area.	Personal Communication Petroeschevsky (2016)

In both the native and introduced range, *Hygrophila polysperma* grows in both aquatic and riparian habitats, and particularly in shallow slow flowing waters high biomass densities were reported (Van Dijk *et al.*, 1986; Cuda & Sutton, 2000). *H. polysperma* prefers flowing rivers but can be found as well in stagnant waters like canals, ditches, irrigation ditches and lakes and also grows in marshes, swamps and wetlands (Nault & Mikulyuk, 2009, Thayer *et al.*, 2016). *H. polysperma has* been observed to grow on damp soils in seasonally flooded areas.

Many freshwater bodies and wetland sites are protected within the EPPO region. Freshwater habitats are detailed within the Habitats Directive 1992 and the Water Framework Directive 2000. Such habitats often harbour rare or endangered species.

8. Pathways for entry

Possible pathways	Pathway: Plants for planting
Short description explaining why it is considered as a pathway	The species is widely sold in aquarium and garden shops with 450, 000 units imported into the PRA region (from Asia) in a given year (Brunel, 2009) and is very popular because of its attractive growth form (Hussner <i>et al.</i> 2014). Plants are released intentionally (for 'wild harvesting' purposes) or unintentionally (unintentional disposal of plant material where <i>H. polysperma</i> is a contaminant) into the field (Brunel, 2009; Hussner <i>et al.</i> 2014).
Is the pathway prohibited in the PRA area?	No. There are no restrictions for the trade of <i>H. polysperma</i> . Currently the species is traded within the EPPO region as an ornamental plant for aquaria.
Has the pest already been intercepted on the pathway?	Yes
What is the most likely stage associated with the pathway?	Live plants would be associated with this pathway.
What are the important factors for association with the pathway?	There are no current import restrictions in the EPPO region. <i>H. polysperma</i> was found to be widely sold in shops in Germany (Hussner <i>et al.</i> 2014), and additionally it is frequently sold in online marketplaces such as ebay.
Is the pest likely to survival transport and storage in this pathway?	Yes. As an import for ornamental purposes, care would be taken to ensure plants survive during transportation.
Can the pest transfer from this pathway to a suitable habitat?	Only through human agency (i.e. intentional introductions or the unintentional disposal of plants into wild habitats). The species could be misused and introduced directly into freshwater bodies and ecosystems (e.g. streams, lakes, dams). The unintended habitats are freshwater bodies and ecosystems (semi-natural and natural waterbodies). Plants used in confined waterbodies could spread to unintended habitats very easily through human activities as well as through natural spread by floods downstream. Releases of aquarium contents have been a source of introduction of aquatic plants in some countries, even if it is considered as an accidental pathway of introduction (e.g. <i>Cabomba caroliniana</i> in the Netherlands, see the EPPO PRA on the species; <i>Hydrilla verticillata</i> in the USA, Langeland, 1996. See Petroeschevsky & Champion (2008) for reference to wild harvesting operations.
Will the volume of movement along the pathway support entry?	Yes. <i>H. polysperma</i> is listed and can be purchased by a number of internet suppliers (worldwide) and is available throughout the EPPO region (www.ppp-index.de).
Will the frequency of movement along the pathway support entry?	Yes, the frequency of supply is related to the demand of the species.

Likelihood of entry	Low	Moderate	High x
Uncertainty	Low x	Moderate	High

9. Likelihood of establishment in the natural environment in the PRA area

Climate modelling indicates that under the current projections, the majority of the EPPO region is unsuitable for the establishment of the species (see Appendix 1). The establishment of the plant is limited by temperature and although ssubmerged plants withstand environments with freezing air temperatures, as long as the water temperature does not drop below 9 °C, as found in the River Erft, Germany (Hussner, 2014).

Hygrophila polysperma grows best at temperatures between 22 – 28 °C, with a minimum temperature of 4 °C (Spencer & Bowes, 1985; Kasselmann, 1995). In Virginia, *H. polysperma* was documented to tolerate freezing temperatures for brief periods (Cuda & Sutton, 2000 citing Reams, 1953), while a study in New Zealand found that emergent plants did not survive during the winter even without freezing water temperatures (Burnett, 2008).

Very small areas of Turkey, Greece and Algeria are marginally suitable along the Mediterranean coastline.

In the EPPO region, *H. polysperma* was first reported from the thermally heated River Erft, Germany (Hussner *et al.*, 2007). Gabka & Owsianny (2009) found *H. polysperma* in a reservoir which is used as a cooling pond for nearby power plants in Poland, and Lukács *et al.* (2016) from thermally heated channels in Hungary. In addition, the species has been recorded from thermal Villacher Warmbad waters in Austria (Bundesministerium für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft, 2013).

Habitats within the endangered area include slow moving rivers, canals, irrigation and drainage systems, lakes, reservoirs.

Hygrophila polysperma is not naturalised in any natural environment within the EPPO region.

Rating of the likelihood of establishment in the natural environment	Low X	Moderate	High
Rating of uncertainty	Low X	Moderate	High

10. Likelihood of establishment in managed environment in the PRA area

Hygrophila polysperma is traded and normally established in protected conditions, for example under glass. The species can establish in artificial, especially thermally influenced water bodies (irrigation channels, reservoirs, drainage ditches etc.). For example, submerged plants withstand environments with freezing air temperatures, as long as the water temperature does not drop below 9 °C, as found in the River Erft, Germany (Hussner, 2014).

Plants are tolerant of mechanical damage, such as mowing and cutting, which may enhance spread through production of viable fragments spread by water movement or contaminated machinery (Nault and Mikulyuk, 2009).

Rating of the likelihood of establishment in the managed	Low X	Moderate	High
environment			
Rating of uncertainty	Low	Moderate X	High

11. Spread in the PRA area

Natural spread

H. polysperma predominantly spreads via plant fragments, as high regeneration rates were found for small stem fragments with nodes (Spencer & Bowes, 1985). Apical shoot fragments of 3cm show high regeneration rates, but stem fragments of 2cm with one node are also able to regenerate, even though in a lesser extent (Personal communication, A. Hussner, 2017).

Plant fragments showed regrowth capacities of 100 % for shoot fragments with three or more nodes per fragment (Spencer & Bowes, 1985). Even single detached leaves are able to regrowth into new plants (Sutton, 1995). However, the number of produced plant fragments was documented as low compared to other invasive aquatic plants (like *Egeria densa* or *Vallisneria spiralis*) in a study in the River Erft, Germany (Heidbüchel *et al.* 2016).

In the invasive range in the USA, no seed production was found (Spencer & Bowes, 1985). Due to the absence of viable seed production in the invasive range, the likelihood of long-distance dispersal of seeds via waterfowls, which has been reported as likely for other invasive aquatic plants (Garcia-Alvarez *et al.* 2015) is low. The natural spread of *H. polysperma* via whole plant fragments is documented only within connected water bodies.

The species can spread rapidly to form dense monoculture stands; in the USA it has been shown to expand from 0.04 ha to over 0.41 ha in one year (Vandiver 1980). Other examples again highlight the rapid spread of the species in Texas (where it spread rapidly to occupy over 20 % of the Comal River, but no time factor was included (Doyle et al., 2003).

Human assisted spread

Intended and/or unintended release of *H. polysperma* plants by humans is the most significant pathway of human mediated spread in the EPPO region. The species is widely sold in aquarium and garden shops with 450, 000 units imported into the region in a given year (Brunel, 2009) and is very popular because of its attractive growth form (Hussner *et al.* 2014). Similar to other aquatic plants, recreational equipment and boating equipment can act as a vector into new unconnected water bodies, however, this has not been demonstrated in the EPPO region to date. The likelihood of a species to spread by transported plant fragments largely depends on its resistance to desiccation (Barnes *et al.* 2013).

Rating of the magnitude of spread	Low	Moderate	High X
Rating of uncertainty	Low	Moderate X	High □

12.01 Impact in the current area of distribution

Note: a lot of the information on impacts for this species, i.e. in the form of factsheets available on the internet, has been disqualified because they contain generalised, unreferenced and unsupported statements about impacts throughout its invasive range.

Impacts on biodiversity and the environment

Florida

Similar to other invasive aquatic plants with a similar growth form, dense stands of *H. polysperma* can block sunlight (which causes the death and decomposition of other vegetation) and reduce

wind induced mixing of the water column, resulting in decreased oxygen levels in the water column (Nault & Mikulyuk, 2009).

Hygrophila polysperma is reported as a strong competitor (Doyle *et al.* 2003, Van Dijk *et al.* 1986) and can displace native vegetation. Decomposing plants and oxygen depletion can cause fish and macroinvertebrates kill (Nault & Mikulyuk, 2009).

Australia

In Australia, although established the species is not showing the strong invasive attributes as reported in Florida. *H. polysperma* at naturalised sites has not been observed to be outcompeting native vegetation or smothering waterways (Personal Communication, A. Petroeschevsky, 2016).

EPPO region

In the River Erft, Germany, *H. polysperma* has locally suppressed the submerged form of the native *Sparganium emersum* (Personal Communication, A. Hussner, 2016). In Poland, within a dense stand of *H. polysperma*, the oxygen concentration was found to be 3.1 mg per litre (Gabka & Owsianny, 2009), below concentrations required to support cyprinids (EEC, 1978).

To-date there are no impacts recorded on red list species and species listed in the Birds and Habitats Directives.

Control methods

Manual and physical control

Reports detail the control of *H. polysperma* has had a limited efficacy due to its ability to propagate vegetatively through fragments (Nault and Mikulyuk, 2009). Attempts to mechanically harvest may only serve as means of creating and introducing more plant fragments, and potentially aiding in dispersal to new locations (Ramey, 2001).

As for all aquatic plants, removal by hand is recommended for early infestations and small areas only. Weed harvesters can be used for the biomass reduction of large infestations, but eradication is only achievable in combination with other control options (e.g. hand removal, chemical control).

Chemical control

Fast *et al.* (2009) tested various herbicides for the control of *H. polysperma*. Triclopyr showed the highest efficiency to control *H. polysperma*, and in combination with other herbicides (2,4-D and / or glyphosate) the efficiency for the control is higher than for the application of Triclopyr alone.

Biological control

Even though *H. polysperma* is considered as a good candidate for biological control (Cuda & Sutton, 2000), there is no biological control agent which is used for the control of *H. polysperma* so far. Grass carp, the most widely used biological control agents for submerged aquatic plants, do not control *H. polysperma*, as *H. polysperma* is unpalatable to these fish (Cuda & Sutton 2000). Several insects have been found in the native range feeding on *H. polysperma*, including caterpillars (*Precis alamana* L. and an unidentified noctuid moth) defoliating emerged shoots (Mukherjee *et al.* 2012). In addition, a *Puccinia* species has been found infecting the plant in the native Indian range (Mukherjee *et al.* 2012).

In the introduced range, in Florida, an aquatic caterpillar (*Parapoynx bilinealis* Snellen) and a leafmining beetle (*Trachys* sp.) have been observed feeding on submerged leaves (Mukherjee *et al.* 2011). Additionally, Habeck & Cuda (2014) reported the waterlily leafcutter (*Elophila obliteralis* Walker) feeding on *H. polysperma*. Some phytoparasitic nematodes are associated with the rhizosphere of *H. polysperma* both in the native and introduced ranges (Mukherjee *et al.* 2012).

The rating of magnitude is moderate due to inconsistent reported impacts within different parts of its introduced range. The uncertainty rating is therefore assessed as high.

Rating of the magnitude of impact in the current area of distribution	Low	Moderate X	High
Rating of uncertainty	Low	Moderate	High X

12.01 Impacts on ecosystem services

Ecosystem service	Does the IAS impact on this Ecosystem service?	Short description of impact	Reference
Provisioning	Yes	Limits water availability in arid zones.	Nault & Mikulyuk (2009)
Regulating	Yes	Increases mortality of fish species and macroinvertebrates, displaces submerged plants.	Nault & Mikulyuk (2009)
Supporting	Yes	Alters the chemical composition of the water column.	Nault & Mikulyuk (2009)
Cultural	Yes	Restrict access for recreation and tourism. May provide breeding habitat for mosquitoes.	Nault & Mikulyuk (2009) Cuda and Sutton (2000)

H. polysperma can form dense mats that impede recreational activities such as boating, fishing, swimming, water skiing, canoeing, and kayaking (Nault and Mikulyuk, 2009). In addition, unsightly mats of vegetation decrease aesthetic values. These declines in recreational and aesthetic values decrease tourism, which can be a major source of livelihood within the community (Nault and Mikulyuk, 2009).

The rating for high uncertainty is given due to the limited number of publications and lack of specifics.

Rating of the magnitude of impact in the current area of distribution	Low	Moderate X	High
Rating of uncertainty	Low	Moderate	High X

12.02. Describe the adverse socio-economic impact of the species in the current area of distribution

Dense stands of *Hygrophila polysperma* limit water flow and thus limit the functioning of irrigation and drainage systems (Nault & Mikulyuk, 2009). The species is reported as a weed in rice fields but there is no information on yield reduction. Dense mats of *H. polysperma* provide a suitable habitat for disease-carrying mosquitoes such as *Coquillettida perturbans* (a vector for encephalomyelitis). The covering of water surfaces interact with recreational water sports activities, like boating, fishing and swimming (Nault & Mikulyuk, 2009).

Herbicides typically used in controlling *H. polysperma* are estimated at costing between US\$988 to US\$1482 per hectare (US\$400 - 600 per acre), and total costs are even higher when labour and equipment are included (Cuda and Sutton, 2000). In an extreme case involving the use of fluridone in flowing water, control was achieved for a period of 20 months at a cost of US\$34,580 per hectare (US\$14,000 per acre) (Sutton, 1996).

Rating of the magnitude of impact in the current area of distribution	Low	Moderate	High X
Rating of uncertainty	Low	Moderate X	High

13. Potential impact in the PRA area

In the River Erft, Germany, *H. polysperma* has locally suppressed a native plant species (Personal Communication, A. Hussner, 2016, see Appendix 3, Fig. 5). In Poland, within a dense stand of *H. polysperma*, the oxygen concentration was found to be 3.1 mg per litre (Gabka & Owsianny, 2009), below concentrations required to support cyprinids (EEC, 1978). Negative effects on fishes and macroinvertebrates, which are reported from other countries where *H. polysperma* is invasive, can be expected if *H. polysperma* reaches similar levels of distribution. *Hygrophila polysperma* blocks the sunlight and reduces the wind induced mixing of the water column, and these effects are independent of the region in which *H. polysperma* becomes invasive.

Hygrophila polysperma reduces the functioning of drainage and irrigation systems and flood control canals. *H. polysperma* stands provide a suitable habitat for mosquitoes, which might carry diseases.

Climate modelling indicates that under the current conditions, the majority of the EPPO region is unsuitable for the establishment of the species (see Appendix 1). Very small areas of Turkey, Greece and Algeria are marginally suitable along the Mediterranean coastline. Impacts are not predicted to happen under the current climate as the species will not establish. With this in mind, there are no impacts envisaged on red list species and species listed in the Birds and Habitats Directives in the near future though this could potentially change if the species establishes under future climate conditions.

Habitats within the endangered area include slow moving rivers, canals, irrigation and drainage systems, lakes, reservoirs.

Hygrophila polysperma is not naturalised in any natural environment within the EPPO region.

13.01. Negative environmental impacts with respect to biodiversity and ecosystem patterns and processes

Rating of the magnitude of impact in the potential area of distribution	Low X	Moderate	High
Rating of uncertainty	Low	Moderate X	High

13.02. Negative impact the pest may have on categories of ecosystem services

Rating of the magnitude of impact in the potential area of distribution	Low X	Moderate	High
Rating of uncertainty	Low	Moderate X	High

13.03 Socio-economic impact of the species

Rating of the magnitude of impact in the potential area of distribution	Low X	Moderate	High
Rating of uncertainty	Low	Moderate X	High

14. Identification of the endangered area

Hygrophila polysperma is not naturalised in any natural environment within the EPPO region. Hygrophila polysperma is a frost sensitive species. Climate modelling indicates that under the current projections, the majority of the EPPO region is unsuitable for the establishment of the species (see Appendix 1, Fig. 5). Very small areas of Turkey, Greece and Algeria are marginally suitable along the Mediterranean coastline. Furthermore, thermally abnormal waters in other EPPO countries provide potential habitats for Hygrophila polysperma. Habitats within the endangered area include slow moving rivers, canals, irrigation and drainage systems, lakes, reservoirs.

15. Climate change

15.01. Define which climate projection you are using from 2050 to 2100*

Climate projection RCP 8.5 (2070)

Note: RCP8.5 is the most extreme of the RCP scenarios, and may therefore represent the worst-case scenario for reasonably anticipated climate change.

15.02 Which component of climate change do you think is most relevant for this organism? Delete (yes/no) as appropriate

Temperature (yes) Precipitation (no) C0₂ levels (no)

Sea level rise (no) Salinity (no) Nitrogen deposition (no)
Acidification (no) Land use change (no) Other (please specify)

Are the introduction pathways likely to change due to climate change?	
(If yes, provide a new risk and uncertainty score)	Reference
The introduction pathways are unlikely to change as a result of climatic change as the species enters the EPPO region as a result of	Days of (2000). Hosping at al.
the horticultural trade. The overall rating for introduction will not	Brunel (2009), Hussner <i>et al.</i> (2014)
change.	(2014)
Is the risk of establishment likely to change due to climate change? (If	
yes, provide a new risk and uncertainty score)	Reference
The risk of establishment will increase with increasing temperature in some countries, in which frost events currently hinders <i>H. polysperma</i> becoming established. Under climate change scenario RCP8.5 (Note: RCP8.5 is the most extreme of the RCP scenarios, and may therefore represent the worst-case scenario for reasonably anticipated climate change) for 2070s, Europe and the Mediterranean are projected to remain largely unsuitable for <i>H. polysperma</i> . However, some areas projected as marginally to moderately suitable appear in northern Portugal, southwest France, Greece, Italy, the eastern Adriatic coast, southern Turkey and Georgia. Under this climate change scenario, the biogeographic regions where the species can potential establish are the Mediterranean, Continental, Black Sea and Atlantic biogeographical regions. The risk of establishment in the natural environment will increase and the rating would change to moderate with a high uncertainty	Hussner <i>et al.</i> (2007); Gabka & Owsianny (2009) (see appendix 1, Figure 6).
and the rating would change to moderate with a high uncertainty	
Is the risk of spread likely to change due to climate change? (If yes, provide a new risk and uncertainty score)	Reference
The risk of spread into countries, in which frost events currently hinder <i>H. polysperma</i> becoming established will increase with increasing temperature. The risk of spread will remain high with a high uncertainty.	Hussner <i>et al.</i> (2007); Gabka & Owsianny (2009) (see appendix 1, Figure 6).
Will impacts change due to climate change? (If yes, provide a new risk and uncertainty score)	Reference

With increasing temperature, the effects of <i>H. polysperma</i> will be more profound than under current climatic conditions. With increasing temperature, the establishment and spread of the species is likely to increase. <i>H. polysperma</i> will potentially have a high negative impact on plant species and the associated fauna in the EPPO region.	
The EWG consider that all impacts in the PRA area will increase from low to moderate with a high uncertainty.	

16. Overall assessment of risk

The overall likelihood of *Hygrophila polysperma* entering into the EPPO region is high. The plant is imported into the EPPO region under its proper name and its synonyms and sold for aquarium. *Hygrophila polysperma* was already found in thermally abnormal waters in Austria, Germany, Poland and Hungary. The risk of the species spreading within the EPPO region is low. The risk of the species establishing in the EPPO region is low. The potential impact of the species within the EPPO region is low with moderate uncertainty.

Pathways for entry:

Plants for planting

Likelihood of entry	Low	Moderate	High x
Likelihood of uncertainty	Low x	Moderate	High

Likelihood of establishment in the natural environment in the PRA area

Rating of the likelihood of establishment in the natural	Low X	Moderate	High
environment			
Rating of uncertainty	Low X	Moderate	High

Likelihood of establishment in managed environment in the PRA area

Rating of the likelihood of establishment in the managed environment	Low X	Moderate	High
Rating of uncertainty	Low	Moderate X	High

Spread in the PRA area

Rating of the magnitude of spread	Low	Moderate	High X
Rating of uncertainty	Low	Moderate X	High

Impacts in the current area of distribution

Impacts on biodiversity and the environment

Rating of the magnitude of impact in the current area of distribution	Low	Moderate X	High
Rating of uncertainty	Low	Moderate	High X

Rating of the magnitude of impact in the current area of distribution	Low	Moderate X	High
Rating of uncertainty	Low	Moderate	High X

Socio-economic impacts

Rating of the magnitude of impact in the current area of distribution	Low	Moderate	High X
Rating of uncertainty	Low	Moderate X	High

Impacts in the PRA area

Will impacts be largely the same as in the current area of distribution? No

Rating of the magnitude of impact in the potential area of distribution	Low X	Moderate	High
Rating of uncertainty	Low	Moderate X	High

Negative impact the pest may have on categories of ecosystem services

Rating of the magnitude of impact in the potential area of distribution	Low X	Moderate	High
Rating of uncertainty	Low	Moderate X	High

Socio-economic impact of the species

Rating of the magnitude of impact in the potential area of distribution	Low X	Moderate	High
Rating of uncertainty	Low	Moderate X	High

Stage 3. Pest risk management

17. Phytosanitary measures

The results of this PRA show that *Hygrophila polysperma* poses a low risk to the endangered area under current climatic projections (very small areas of Turkey, Greece and Algeria are marginally suitable along the Mediterranean coastline) with a moderate uncertainty. *Hygrophila polysperma* is not naturalised in any natural environment within the EPPO region.

The Expert Working Group recommends that the PRA is reviewed every ten years and/or when significant new information (e.g. naturalisation in natural environment of the endangered area or ecological data) becomes available.

The major pathway being considered is:

Plants for planting

Given the low risks for establishment and impact on the natural and managed environment within the endangered area the Expert Working Group does not recommend any phytosanitary measures for this species.

National awareness raising measures:

- There are no national prevention measures for the sale of *Hygrophila polysperma* in any country within the endangered area. The Expert Working Group recommends *H. polysperma* should be monitored where it occurs in the wild.
- The Expert Working Group encourages industry to assist with public education campaigns associated with the risk of aquatic non-native plants.

For additional information see:

See Standard PM3/67 'Guidelines for the management of invasive alien plants or potentially invasive alien plants which are intended for import or have been intentionally imported' (EPPO, 2006).

See Standard PM9/19 (1) 'Invasive alien aquatic plants' (EPPO, 2014).

See Standard PP 3/74(1) 'EPPO guidelines on the development of a code of conduct on horticulture and invasive alien plants' (EPPO, 2009).

18. Uncertainty

An overall moderate uncertainty rating has been given due to the lack of ecological studies. Uncertainty should also be considered in the context of species distribution modelling (SDM). Here records for *H. polysperma* and synonyms were retrieved from GBIF and other online sources, and were also digitised from occurrences that were either mapped or clearly georeferenced in published sources. This may mean that the realised climatic niche of *H. polysperma* is under-characterised.

19. Remarks

Inform EPPO or IPPC or EU

• The Expert Working Group recommends *H. polysperma* should be monitored where it occurs in the wild within the endangered area.

Inform industry, other stakeholders

• Encourage industry to assist with public education campaigns associated with the risk of aquatic non-native plants.

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Projection of climatic suitability for Hygrophila polysperma establishment

Aim

To project the suitability for potential establishment of *Hygrophila polysperma* in the EPPO region, under current and predicted future climatic conditions.

Data for modelling

Climate data were taken from 'Bioclim' variables contained within the WorldClim database (Hijmans *et al.*, 2005) originally at 5 arcminute resolution (0.083 x 0.083 degrees of longitude/latitude) but bilinearly interpolated to a 0.1 x 0.1 degree grid for use in the model. Based on the biology of the focal species, the following climate variables were used in the modelling:

- Mean temperature of the warmest quarter (Bio10 °C) reflecting the growing season thermal regime. USDA APHIS (2015) mentions 4 °C as a minimum growth temperature, so low temperatures should limit growth.
- Mean minimum temperature of the coldest month (Bio6 °C) reflecting exposure to frost. CABI (2015) suggests that *H. polysperma* requires coldest month temperatures above 0°C.
- <u>Mean annual precipitation</u> (Bio12 ln+1 transformed mm). Although the species is aquatic and will therefore have limited direct dependence on precipitation, sufficient precipitation for the presence of wetland habitat may be required.

To estimate the effect of climate change on the potential distribution, equivalent modelled future climate conditions for the 2070s under the Representative Concentration Pathway (RCP) 8.5 were also obtained. This assumes an increase in atmospheric CO₂ concentrations to approximately 850 ppm by the 2070s. Climate models suggest this would result in an increase in global mean temperatures of 3.7 °C by the end of the 21st century. The above variables were obtained as averages of outputs of eight Global Climate Models (BCC-CSM1-1, CCSM4, GISS-E2-R, HadGEM2-AO, IPSL-CM5A-LR, MIROC-ESM, MRI-CGCM3, NorESM1-M), downscaled and calibrated against the WorldClim baseline (see http://www.worldclim.org/cmip5_5m). RCP8.5 is the most extreme of the RCP scenarios, and may therefore represent the worst case scenario for reasonably anticipated climate change.

In the models we also included two measures of habitat availability:

- <u>Cover of inland waterbodies</u> was estimated from the Global Inland Water database (Feng *et al.*, 2016). The original database is a remote sensed estimate at a 30 x 30 m resolution of the presence of inland surface water bodies, including fresh and saline lakes, rivers, and reservoirs. For the PRA, this was supplied as a 0.1 x 0.1 degree raster indicating the proportion of the constituent 30 x 30 m grid cells classified as inland waters.
- <u>Density of permanent rivers</u> was estimated from the Vector Map VMAP0 (United States National Imagery Mapping Agency, 1997). River vectors were rasterised at 0.02 x 0.02 degree resolution. Then, we calculated the proportion of these grid cells containing rivers within each of the 0.1 x 0.1 degree cells used in the model.

Species occurrences were obtained from the Global Biodiversity Information Facility (www.gbif.org), supplemented with records from the USGS Nonindigenous Aquatic Species Database (https://nas.er.usgs.gov/Default.aspx), the scientific literature and the Expert Working Group. Occurrence records with insufficient spatial precision, potential errors or that were outside of the coverage of the predictor layers (e.g. small island or coastal occurrences) were excluded. The remaining records were gridded at a 0.1 x 0.1 degree resolution (Figure 1).

A small number were either examples of casual occurrences introduced to climatically unsuitable regions (for example, where severe winter frosts are known to kill all individuals) or records of persistent populations known to occupy climatically anomalous micro-habitats such as thermal streams or warmed industrial outflows. These were removed from the occurrence data as they will impede the model's ability to characterise climatic suitability. Specifically these records were from thermally abnormal stretches of the River Erft in Germany, a power station outflow in Poland, Stockholm botanic garden in Sweden and two records from aquaria in New Zealand. This represented all the records from Europe.

In total, there were 144 grid cells with recorded occurrence of *H. polysperma* available for the modelling (Figure 1), which is a low sample size for trying to model a species' climatic and other environmental requirements.

Figure 1. Occurrence records obtained for *Hygrophila polysperma* used in the model, after exclusion of casual and thermally-anomalous records.



Species distribution model

A presence-background (presence-only) ensemble modelling strategy was employed using the BIOMOD2 R package v3.3-7 (Thuiller *et al.*, 2009, Thuiller *et al.*, 2014). These models contrast the environment at the species' occurrence locations against a random sample of the global background environmental conditions (often termed 'pseudo-absences') in order to characterise and project suitability for occurrence. This approach has been developed for distributions that are in equilibrium with the environment. Because invasive species' distributions are not at equilibrium and subject to dispersal constraints at a global scale, we took care to minimise the inclusion of locations suitable for the species but where it has not been able to disperse to. Therefore the background sampling region included:

- The native continent of *H. polysperma*, Asia, for which the species is likely to have had sufficient time to cross all biogeographical barriers; AND
- A relatively small 50 km buffer around all non-native occurrences, encompassing regions likely to have had high propagule pressure for introduction by humans and/or dispersal of the species; AND
- Regions where we have an *a priori* expectation of high unsuitability for the species (see Fig. 2). The following rules were applied to define the region expected to be highly unsuitable for *H. polysperma*:
 - Mean minimum temperature of the coldest month (Bio6) < -10 °C. There is little information on frost tolerance of *H. polysperma*, but as the species can exist as a submerged plant of flowing water it is likely to exhibit some frost tolerance. The USDA APHIS risk assessment suggests the species can tolerate USDA Plant Hardiness Zone 7, where the average annual extreme low temperature can be as low as -17.8 °C (USDA)

APHIS, 2015). However, this was based on a single occurrence in Richmond, while the heavily invaded parts of the USA are substantially further south and warmer than this. Weather records for the coldest known location in Australia show an average July minimum temperature of 5 °C with a lowest recorded of -3.4 °C (A. Petroeschevsky, personal comment). The coldest location with a presence in our dataset has Bio6 = 0.7 °C.

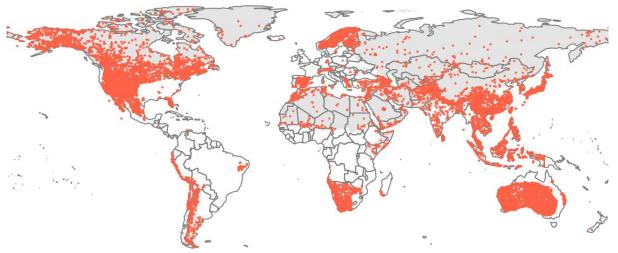
o Annual precipitation (Bio12) < 500 mm. There is little information on precipitation requirements and the USDA APHIS risk assessment does not assume a lower limit on annual precipitation. The driest occurrence has 842 mm of precipitation.

We did not specify a limitation by growing season temperatures because minimum growing temperatures of 4 °C are reported (Nault & Mikulyuk, 2009). Locations with growing seasons as cold as this will likely be included in the unsuitable region as they should also have very cold winter temperatures.

Within this sampling region there will be substantial spatial biases in recording effort, which may interfere with the characterisation of habitat suitability. Specifically, areas with a large amount of recording effort will appear more suitable than those without much recording, regardless of the underlying suitability for occurrence. Therefore, a measure of vascular plant recording effort was made by querying the Global Biodiversity Information Facility application programming interface (API) for the number of phylum Tracheophyta records in each 0.1 x 0.1 degree grid cell. The sampling of background grid cells was then weighted in proportion to the Tracheophyte recording density. Assuming Tracheophyte recording density is proportional to recording effort for the focal species, this is an appropriate null model for the species' occurrence.

To sample as much of the background environment as possible, without overloading the models with too many pseudo-absences, five background samples of 10,000 randomly chosen grid cells were obtained (Figure 2).

Figure 2. Randomly selected background grid cells used in the modelling of *Hygrophila polysperma*, mapped as red points. Points are sampled from the native continent (Asia), a small buffer around non-native occurrences and from areas expected to be highly unsuitable for the species (grey background region), and weighted by a proxy for plant recording effort.



Each dataset (i.e. combination of the presences and the individual background samples) was randomly split into 80% for model training and 20% for model evaluation. With each training dataset, ten statistical algorithms were fitted with the default BIOMOD2 settings (Thuiller *et al.*, 2009, Thuiller *et al.*, 2014), except where specified below:

- Generalised linear model (GLM)
- Generalised boosting model (GBM)

- Generalised additive model (GAM) with a maximum of four degrees of freedom per smoothing spline.
- Classification tree algorithm (CTA)
- Artificial neural network (ANN)
- Flexible discriminant analysis (FDA)
- Multivariate adaptive regression splines (MARS)
- Random forest (RF)
- MaxEnt
- Maximum entropy multinomial logistic regression (MEMLR)

Since the background sample was much larger than the number of occurrences, prevalence fitting weights were applied to give equal overall importance to the occurrences and the background. Variable importances were assessed and variable response functions were produced using BIOMOD2's default procedure. Model predictive performance was assessed by calculating the Area Under the Receiver-Operator Curve (AUC) for model predictions on the evaluation data, that were reserved from model fitting. AUC can be interpreted as the probability that a randomly selected presence has a higher model-predicted suitability than a randomly selected absence. This information was used to combine the predictions of the different algorithms to produce ensemble projections of the model. For this, the three algorithms with the lowest AUC were first rejected and then predictions of the remaining seven algorithms were averaged, weighted by their AUC. Ensemble projections were made for each dataset and then averaged to give an overall suitability.

Results

The ensemble model had a better predictive ability (AUC) than any individual algorithm and suggested that suitability for *H. polysperma* was most strongly determined by the annual precipitation, mean temperature of the warmest quarter and minimum temperature of the coldest month (Table 1). Inland water cover and river density had very little effect on model fit (Table 1, Fig. 3). From Fig. 3, the ensemble model estimated the optimum conditions for occurrence with approximately:

- Annual precipitation = 1289 mm (≥50% suitability with >823 mm)
- Mean temperature of warmest quarter = $27.5 \,^{\circ}\text{C} \ (\geq 50\% \text{ suitability with } > 23.9 \,^{\circ}\text{C})$
- Mean minimum temperature of the coldest month = 9.3 °C ($\geq 50\%$ suitability for -9.8 to 17.0°C)

These optima and ranges of high suitability described above are conditional on the other predictors being at their median value in the data used in model fitting.

There was substantial variation among modelling algorithms in the partial response plots (Fig. 3). In part this will reflect their different treatment of interactions among variables. Since partial plots are made with other variables held at their median, there may be values of a particular variable at which this does not provide a realistic combination of variables to predict from. It also demonstrates the value of an ensemble modelling approach in averaging out the uncertainty between algorithms.

Global projection of the model (Fig. 4) indicates that most of the native and known invaded records all fell within regions predicted to be suitable for the species. Florida was highlighted as an invaded region with especially high suitability for establishment. The model also predicts large regions of suitability in Northern Australia, South America and Africa where the species has not been recorded as invasive.

In Europe and the Mediterranean region, the model predicts very limited opportunity for establishment (Fig. 5). Areas predicted to have marginal suitability can be found in isolated locations around the Mediterranean coast, especially in western Greece and southern Turkey.

Under climate change scenario RCP8.5 for the 2070s, Europe and the Mediterranean are projected to remain largely unsuitable for *H. polysperma* (Fig. 6). However, some areas projected as marginally to moderately suitable appear in northern Portugal, southwest France, Italy, the eastern Adriatic coast, southern Turkey, Georgia and a few other places.

Table 1. Summary of the cross-validation predictive performance (AUC) and variable importances of the fitted model algorithms and the ensemble (AUC-weighted average of the best performing seven algorithms). Results are the average from models fitted to five different background samples of the data.

Algorithm	Predictive	Variable importance				
	AUC	Minimum temperature of coldest month	Mean temperature of warmest quarter	Annual precipitation	Inland water cover	River density
GBM	0.9894	11.3%	38.2%	50.1%	0.3%	0.1%
MaxEnt	0.9888	15.3%	36.2%	42.0%	4.4%	2.2%
GAM	0.9878	10.4%	37.4%	51.9%	0.2%	0.1%
MARS	0.9874	20.4%	26.4%	50.8%	2.2%	0.2%
GLM	0.9840	18.7%	29.1%	51.7%	0.3%	0.1%
ANN	0.9832	15.2%	36.6%	40.6%	6.5%	1.0%
RF	0.9828	13.9%	37.8%	42.2%	4.9%	1.2%
FDA	0.9680	16.1%	32.9%	51.0%	0.0%	0.0%
CTA	0.9368	11.7%	38.3%	44.8%	5.3%	0.0%
MEMLR	0.8318	55.0%	10.7%	10.3%	22.5%	1.5%
Ensemble	0.9914	15.0%	34.5%	47.0%	2.7%	0.7%

Figure 3. Partial response plots from the fitted models, ordered from most to least important. Thin coloured lines show responses from the seven algorithms, while the thick black line is their ensemble. In each plot, other model variables are held at their median value in the training data. Some of the divergence among algorithms is because of their different treatment of interactions among variables.

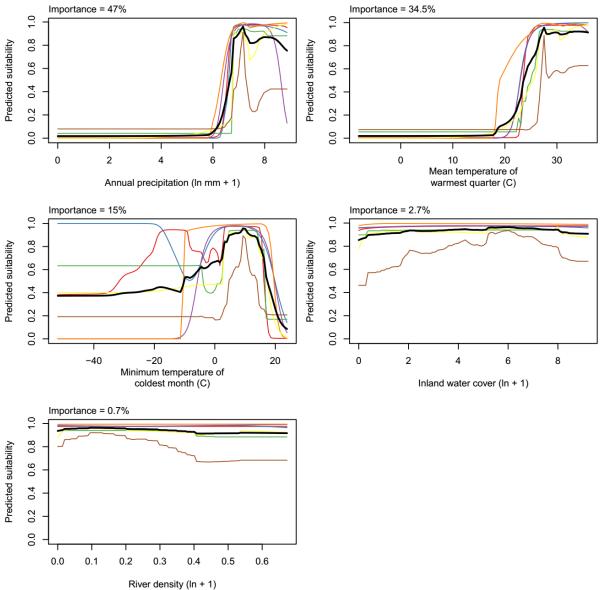


Figure 4. Projected global suitability for *Hygrophila polysperma* establishment in the current climate. For visualisation, the projection has been aggregated to a 0.5×0.5 degree resolution, by taking the maximum suitability of constituent higher resolution grid cells. Values > 0.5 may be suitable for the species. The white areas have climatic conditions outside the range of the training data so were excluded from the projection.

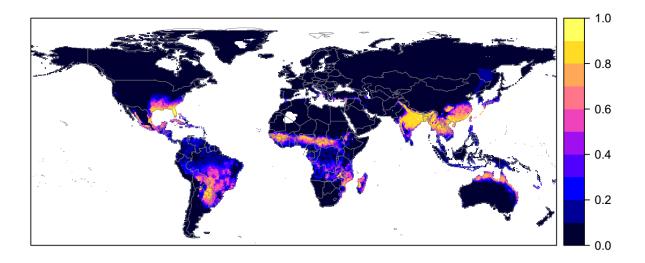


Figure 5. Projected current suitability for *Hygrophila polysperma* establishment in Europe and the Mediterranean region. For visualisation, the projected suitability has been smoothed with a Gaussian filter with standard deviation of 0.1 degrees longitude/latitude. The white areas have climatic conditions outside the range of the training data so were excluded from the projection.

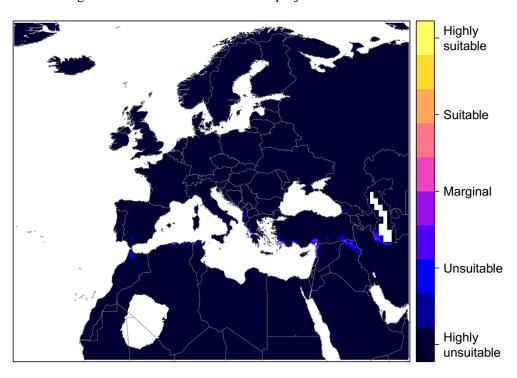
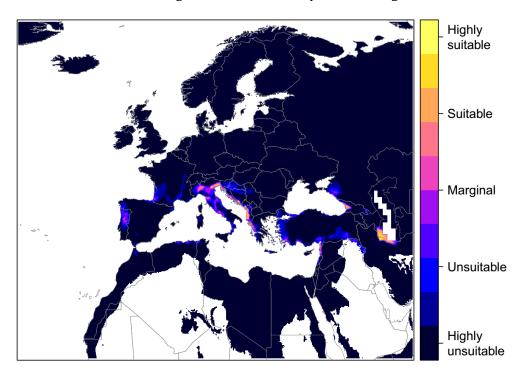


Figure 6. Projected suitability for *Hygrophila polysperma* establishment in Europe and the Mediterranean region in the 2070s under climate change scenario RCP8.5, equivalent to Fig. 5.



Caveats to the modelling

The sample size of 144 grid cells with occurrences is low and adds considerable uncertainty to the modelling.

To remove spatial recording biases, the selection of the background sample was weighted by the density of Tracheophyte records on the Global Biodiversity Information Facility (GBIF). While

this is preferable to not accounting for recording bias at all, a number of factors mean this may not be the perfect null model for species occurrence:

- The GBIF API query used to did not appear to give completely accurate results. For example, in a small number of cases, GBIF indicated no Tracheophyte records in grid cells in which it also yielded records of the focal species.
- We located additional data sources to GBIF, which may have been from regions without GBIF records.
- Levels of Tracheophyte recording may not be a consistent indicator of the recording of aquatic plants. There is a suggestion that aquatic plants may be disproportionately under-recorded in tropical regions (Jonathan Newman, *pers. comm*), which could have been responsible for an under-prediction of suitability in tropical regions.

Air temperatures were used in the model, while water temperatures may be more appropriate for an aquatic plant. In some cases air and water temperatures can markedly diverge, for example warming associated with industrial outflows. Wherever the water temperature is warm enough, the species is likely to be able to persist, regardless of the model's estimate of suitability.

Water chemistry and quality may have a large effect on the ability of the species to persist but were not used in the model. Factors such as dissolved inorganic carbon, pH and nutrient concentration are likely to be important modifiers of habitat suitability.

The climate change scenario used is the most extreme of the four RCPs. However, it is also the most consistent with recent emissions trends and could be seen as worst case scenario for informing risk assessment.

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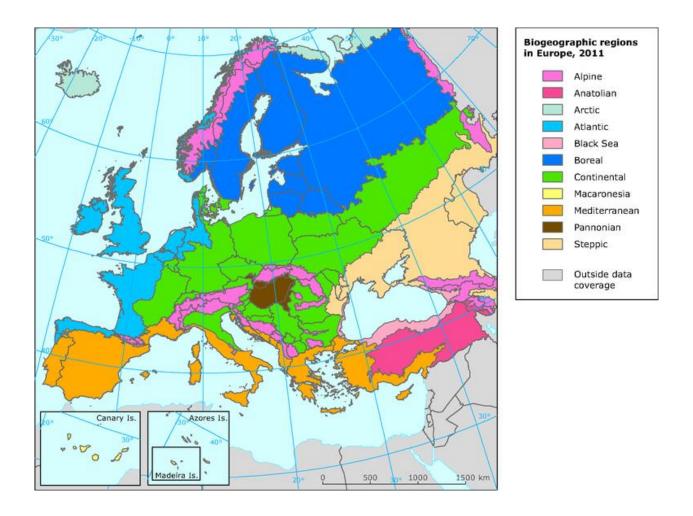
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Appendix 2 Biogeographical regions in Europe



Appendix 3. Relevant illustrative pictures (for information)

Figure 1. *Hygrophila polysperma* (River Erft, Germany).



Figure 1. Hygrophila polysperma (River Erft, Germany).



 $\label{eq:Figure 3.} \textit{Hygrophila polysperma} - \text{emergent leaves}$



Figure 4. *Hygrophila polysperma* growing up a river bank (Australia).

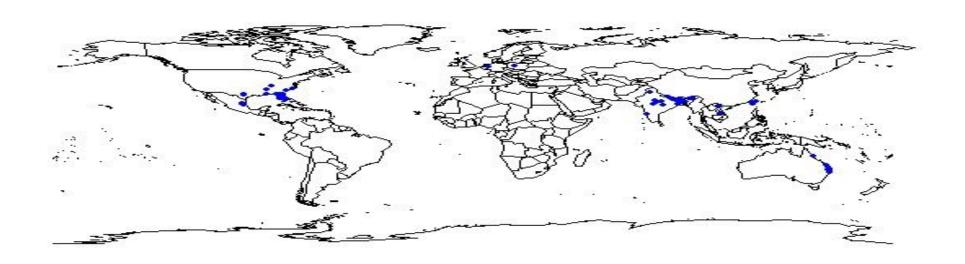


Figure 5. Hygrophila polysperma suppressing native plants (Germany)



Appendix 4. Distribution maps of *Hygrophila polysperma*

Figure 1. Global occurrence of *Hygrophila polysperma*



Russia 60 Kazakhstan Mongolia Turkmenistan, China South Korea Afghanistan Iran East China Sea Pakistan i Arabia Myanmar (Burma) 20-Thailand Yemen Bay of Gengal ulf of Aden Vietnam Philippines Arabian Sea Laccadive Sea Malaysia nalia Indonesia Papua New Guinea Map data ©2016 Google

100

X

125

150

Figure 2. Occurrence of Hygrophila polysperma in Asia

75

Figure 3. Occurrence of Hygrophila polysperma in USA

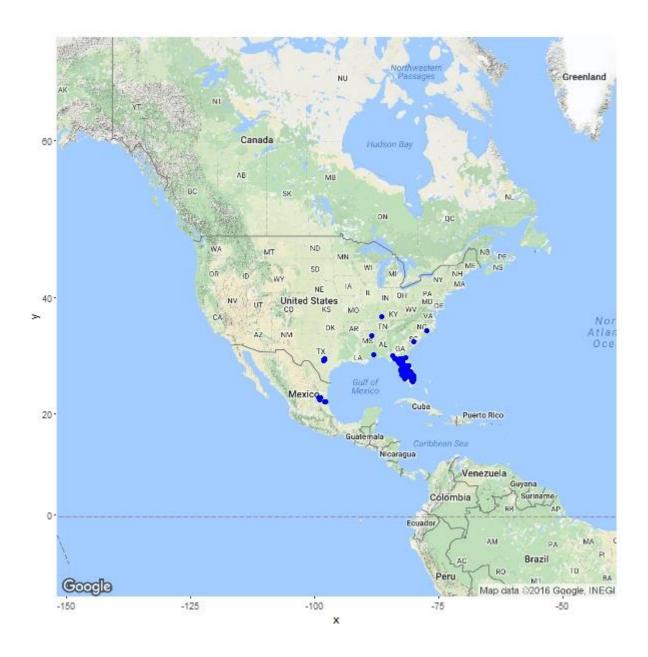


Figure 4. Occurrence of *Hygrophila polysperma* in Europe

