

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

21-26956 (19-24688)

This PRA document was modified in 2021 to clarify the phytosanitary measures recommended

#### **Pest Risk Analysis for**

#### Ambrosia trifida L. (Asteraceae)



Ambrosia trifida in agricultural fields in the EPPO region. Swen Follak (EPPO Global Database)

September 2019

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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.

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Based on this PRA, *Ambrosia trifida* was added to the EPPO A2 List of pests recommended for regulation as quarantine pests in 2019. Measures for *A. trifida* are recommended.

#### Pest Risk Analysis for Ambrosia trifida L. (Asteraceae)

PRA area: EPPO region

Prepared by: EWG on Ambrosia trifida

Date: 19-21 February 2019. Further reviewed and amended by EPPO core members and Panel on

Invasive Alien Plants.

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This Pest Risk Analysis (PRA) is based on a national French PRA on *Ambrosia trifida* (Anses, 2017) and expanded during the EPPO Expert Working Group to encompass the whole EPPO region. Additional information and data have been added where appropriate.

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. For the determination of ratings of likelihoods and uncertainties, experts were asked to provide a rating and level of uncertainty individually during the meeting, based on the evidence provided in the PRA and on the discussions in the group. Each EWG member provided anonymously a rating and level of uncertainty, and proposals were then discussed together in order to reach a final decision.

Following the EWG, the PRA was further reviewed by the following core members: Alan MacLeod and Muriel Suffert.

The Panel on Invasive Alien Plants considered the management options in 2019-06. The Working Party on Phytosanitary Regulations agreed that *Ambrosia trifida* should be added to the A2 Lists of pests recommended for regulation as a quarantine pest in 2019.

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#### Summary of the Express Pest Risk Analysis for Ambrosia trifida

PRA area: The EPPO region

#### **Describe the endangered area:**

Ambrosia trifida is best suited to a continental climate and parts of the Pannonian biogeographical region, but able also to establish in part of the Mediterranean, Steppic and Anatolian biogeographical regions<sup>1</sup>. All EPPO countries south of 55° latitude have potential for establishment.

Habitats most at risk in the endangered area include ruderal disturbed habitats (including transportation networks), riparian systems, field crops, (annual summer crops, particularly maize, soybean and sunflower) and open habitats. Within the climatically suitable area, the distribution of the preferred crop types of *A. trifida* (maize, soybean and sunflower) are mostly restricted to the warmer Continental, Pannonian and Steppic parts of the EPPO region. The present occurrences of *A. trifida* in the eastern part of Germany and Russia coincide with wheat cropping.

Based on the experience of the occurrence of *A. trifida* in North America, the species is likely to occur more in moist habitats (including artificial irrigation) than drier areas in the EPPO region.

The Expert Working Group (EWG) considers the species distribution modelling conducted as part of this PRA (see Appendix 3) to be a realistic projection of the potential occurrence of *A. trifida* in the EPPO region. However, the model may over predict the potential occurrence in the warmer and drier Mediterranean area due to the inclusion of the layer for crop land cover in the model. Actual suitability of these areas may be restricted to irrigated fields.

#### **Main conclusions**

#### Ambrosia trifida presents a high phytosanitary risk for the endangered area with low uncertainty.

The likelihood of new introductions to the EPPO region occurring via contamination of seed (maize seed, soybean seed, spring crops (sunflower and sorghum) and contamination of grain (e.g. soybean and maize) is moderate and high respectively. Within the EPPO region, *A. trifida* seems to preferentially become established in crops and ruderal environments. It is found on riverbanks within the EPPO region and the likelihood of further establishment in natural habitats is considered high with a low uncertainty. It is also found in arable land and fallows, road networks, rail networks and domestic and non-domestic gardens. The likelihood of further establishment in the managed environment is considered very high with a low uncertainty. The potential magnitude of spread within the EPPO region is high with moderate uncertainty. *A. trifida* has both short and long-distance natural dispersal pathways. Human assisted spread facilitated by agriculture machinery and movement within the EPPO region as a contaminant of seed or grain can act to move seeds over long distances.

The main impacts of the species at a global level (North America and the EPPO region) are the reduction of crop yields and human health impacts which are translated in the PRA into socio-economic impacts. The EWG consider the potential socio-economic impacts in the EPPO region very high with a moderate uncertainty. Potential impacts on biodiversity and ecosystem services are moderate with high uncertainty. The species has the potential to establish along rivers where it could compete with native vegetation and reduce access to water bodies for recreation. The high uncertainty reflects the lack of quantitative studies on the impact of the species in natural habitats.

<sup>&</sup>lt;sup>1</sup> Geographical regions are defined in Appendix 5

The species is particularly difficult to manage due to early and p growth. An established population is very difficult to control as soil for 4 to 21 years, depending on burial depth. Within the E economical control options, and chemical control options are countries in the EPPO region.	s the seeds of EPPO region, t	A. trifida can rem here is a lack of e	ain viab	ole in and
Phytosanitary risk for the <u>endangered area</u>	High X	Moderate □	Low	

High

П

Moderate

Low

Χ

#### Other recommendations:

Level of uncertainty of assessment

The EWG recommends that further studies be conducted on the effect of ecotypes on establishment in the PRA area. In North America, there are ecotypes which are adapted to varying climatic conditions but knowledge of the presence of ecotypes in the EPPO region is currently unknown. In addition, the EWG recommends that surveys be conducted to determine the current establishment and spread of the species in the EPPO region along with surveys and inspection of the contamination of imported grain and seed from North America. It would also be of value, and could inform phytosanitary decision making, to evaluate the potential spread of the species through the contamination of seed and grain within the EPPO region and thus surveys and inspections could be carried out to assess this. The EWG considers that studies should be conducted on the emergence period of the species in the EPPO region along with studies on the cultural, mechanical, chemical and biological control of the species

#### **EPPO Pest Risk Analysis:**

## Ambrosia trifida L

Prepared by: ANSES and the EPPO Expert Working Group

Date: 2019-02-19/21

**Note:** This Pest Risk Analysis (PRA) is based on a national French PRA on *Ambrosia trifida* (Anses, 2017) and expanded during the EPPO Expert Working Group to encompass the whole EPPO region. Additional information and data have been added where appropriate.

#### Stage 1. Initiation

#### **Reason for performing the PRA:**

This PRA was conducted to determine the likelihood and extent of entry into the EPPO region and spread of A. trifida in the EPPO region, along with the magnitude of impacts. Ambrosia trifida presents a phytosanitary risk to the PRA area (EPPO region). Ambrosia trifida presents risks to the economy, agricultural production and human health. The species is very harmful to crops due to its competitive ability (Regnier et al., 2016). This species has a very allergenic pollen that contributes to the pollinosis observed in summer in genetically predisposed people. As the species was introduced into the EPPO region as a contaminant of imports of seed (for planting: e.g. maize, wheat, soybeans, barley and clover), and grain for animal feed and products intended for use in the food industry (e.g. soybeans, maize) (Shamonin and Smetnik, 1986; Stoyanov et al., 2014), it could negatively affect international trade and exchanges (Karnkowski, 2001). The species has been found in a number of EPPO countries where both established and casual populations occur. More recent occurrences in spring and summer crops which impact on yield in Southern France (Chauvel et al., 2015) and Slovenia highlights its potential harmful impacts on agriculture (pers. comm. Marisavljevic, 2019).

**PRA area:** EPPO region (see https://www.eppo.int/ABOUT\_EPPO/images/clickable\_map.htm)

#### Stage 2. Pest risk assessment

#### 1. Taxonomy

**Taxonomy:** Kingdom: *Plantae*, Division, *Spermatophyta*, Sub-Division, *Angiospermae*, Class *Dicotyledonae*, Order *Asterales*, Family *Asteraceae*, Genus *Ambrosia*, Species *Ambrosia trifida* L., according to Linnaeus (1753).

EPPO code: AMBTR

#### **Synonyms**

Ambrosia aptera DC., Ambrosia integrifolia Mulh. ex Willd., Ambrosia trifida var. aptera (DC.) Kuntze, Ambrosia trifida var. heterophylla Kuntze, Ambrosia trifida var. integrifolia (Mulh ex. Willd) Torr. & A.Gray, Ambrosia trifida f. integrifolia (Mulh ex. Willd) Fernald, Ambrosia trifida var. polyploidea J.Rousseau, Ambrosia trifida var. texana Scheele, Ambrosia trifida subsp. trifida, Ambrosia trifida var. trifida

Ref: The Plant List (http://www.theplantlist.org/tpl1.1/gcc-7636) and EWG.

#### **Common names**

English: Great ragweed, Blood ragweed, Buffalo-weed, Crownweed, Horseweed, Giant ragweed, Bitterweed, Buffalo weed, Crown-weed, Horse-cane, Horse-weed, Kinghead, Tall ambrosia, Tall ragweed Wild hemp. Chinese: san lie ye tun cao, Czech: Ambrozie trojklaná, Dutch: Driedelige Ambrosia, Estonian: Kolmehõlmane ambroosia, French: Ambroisie trifide, Grande herbe à poux (Québec), Finish: Sormituoksukki, German: Dreilappige Ambrosie; Dreispaltige Ambrosie; Dreilappiges Traubenkraut Italian: Ambrosia trifida, Japanese: Kuwamodoki; Oobutakusa, Latvian: Trisdaivu ambrozija, Lithuanian: Triskiaute ambrozija, Norwegian: Hesteambrosia, Polish: Ambrozja trójdzielna, (Russian).Амброзия трехраздельная, Slovakian: Ambrózia trojzárezová, Slovenian: Trikrpata žvrklja, Swedish: Hästambrosia

**Plant type:** Annual herbaceous

#### **Related species in the EPPO region:**

**-Non-native species:** *Ambrosia artemisiifolia* (AMBEL), *Ambrosia confertiflora* (FRSCO), *Ambrosia psilostachya* (AMBPS), *Ambrosia tenuifolia* (AMBTE), *Ambrosia tomentosa* (FRSTO). **-Native species:** *Ambrosia maritima* (AMBMA).

Appendix 1 provide a comparison of *Ambrosia* species present within Europe which can all be confused with *A. trifida*. Careful identification is needed to tell the species apart as many of the species traits overlap (see figures 1 and 2 and section 2 pest overview: identification).

#### 2. Pest overview

#### Introduction

Ambrosia trifida is a summer annual plant that can measure from 1 to 5 m in height (Basset and Crompton, 1982) (see figures 1, 2 and 3 in Appendix 2). The species is native to North America where it is expanding as a crop weed and has impacts in agricultural areas (see figures 4 and 5 in Appendix 2). (Regnier *et al.*, 2016). Ambrosia trifida was introduced into the EPPO region at the end of the 19th century most likely with contaminated animal feed and seeds for planting (Follak et al., 2013, Chauvel et al., 2015)."

#### **Identification**

Ambrosia trifida has large leaves (4-15 cm long). They are oppositely arranged, simple, and palmately lobed, generally with three lobes (they may also have five lobes or be unlobed). Alone, the upper leaves can be alternate. They are borne on a long petiole (3-12 cm). Male and female flowers are separated on the same individual (monoecious plant – Payne, 1964). The inflorescences are long terminal clusters (30 cm) consisting of florets of male flowers. The female flowers are grouped into florets at the base of the male clusters and sometimes in the axils of the upper leaves. The fruit is a cup-shaped cypsela, tipped with a long central beak surrounded by a crown of approximately 5 or more shorter tips (see figure 6, Appendix 2). It measures from 0.5 to 1.2 cm long and from 0.3 to 0.5 cm wide. Ambrosia trifida is characterised by enormous variability in the size and shape of its seeds, which may correspond to an ability to germinate in a variety of conditions (Harrison et al. 2007; Hovick et al. 2018; Sako et al. 2001; Schutte et al. 2008).

Ambrosia trifida is a diploid species (2n = 24; Payne, 1964) that essentially reproduces through cross-pollination. Within the genus Ambrosia, A. trifida can hybridise with A. artemisiifolia (Vincent et al., 1987; Vincent et al., 1988) to give a new hybrid taxon, A. x helenae Rouleau 1944, but this taxon is described as sterile (Vincent et al., 1988). Such hybrids were observed in the 1940s in France in the Bordeaux botanical garden (Chauvel et al., 2015).

#### Life cycle

Ambrosia trifida has a comparatively low fecundity (compared to other Ambrosia species), transient seedbank characteristics and a high percentage of non-viable or low-survivorship seeds (Harrison *et al.* 2001; Harrison *et al.* 2007). Golpen *et al.*, 2016 reports that plants produced an average of 1,818 seeds per plant in soybean and field margins, with 66% being potentially viable. The majority (90% or more) of *A. trifida* seeds buried 10 cm or less lost viability after 4 years (Harrison *et al.* 2007; Stoller and Wax 1974), however some seeds remained viable for 9 to 21 years when buried 20 cm or deeper (Harrison *et al.* 2007; Toole and Brown 1946). Due to their high nutritional value the seed are often eaten by animals (e.g. birds and rodents) causing high losses (Harrison *et al.* 2003). It should be noted that *A. trifida* only reproduces by seed and not vegetatively.

Within the EPPO region and the native range, seedlings typically emerge early in the growing season (e.g. March) and over a prolonged period (March until the end of July) (Regnier *et al.*, 2016). Flowering occurs in response to shortening day length and begins in the male inflorescences (Allard, 1945). In the native range (North America), it flowers from mid-June to the end of August, or even early September (Bassett and Crompton, 1982). The species can flower two to three weeks earlier than *A. artemisiifolia*. In southwest France, the flowering dates observed are similar to those in its area of origin (pers. comm. B. Chauvel, 2019).

#### **Habitats**

In its area of origin, A. trifida primarily grows in east-central USA (see Appendix 4, Fig. 2), on the shores of lakes and the banks of the Ohio and Mississippi rivers, as well as in southern Canada (Bassett and Crompton, 1982). In the native range (North America), A. trifida has historically been recorded in naturally

disturbed habitats (for example the banks of water courses), (Bassett and Crompton, 1982). Today it is regarded as a major weed in the native range (Ganie *et al.*, 2017; Regnier *et al.*, 2016). It has also become established in gardens, ditches, abandoned industrial sites and disturbed habitats (roadsides and near fences). It is a meso-hygrophilic species, preferring wet meadows to drier areas (Bassett and Crompton, 1982; Uva *et al.*, 1997). Within the EPPO region, the species occupies crop fields, ruderal habitats, including railway tracks and naturally disturbed habitats like riparian systems (Chauvel, 2015; Follak *et al.*, 2013).

#### **Environmental requirements**

Ambrosia trifida is not well adapted to drought and it is not recorded in areas with a long summer drought unless there is irrigation (Allard, 1945; Regnier <u>et al.</u> 2016). Establishment is favoured by moist environments (Korres et al., 2015). Ambrosia trifida can tolerate a wide variety of soil types (Regnier et al., 2016).

Seeds germinate under a wide range of temperatures with an optimum germination between  $10 - 24^{\circ}$ C (Abul-Fatih and Bazzaz, 1979). The seedlings are able to develop very quickly within four to 13 days (Abul-Fatih and Bazzaz, 1979). A. trifida can emerge over a long period of time (March to June/July). In France, it emerges together with spring crops or a few days after crop emergence. Soybean is seeded in May in France. In south-west France, germination and emergence can begin as early as in the end of March and continue later until the end of summer, especially in irrigated fields (Mamarot and Rordiguez, 2014). A. trifida has a high photosynthetic ability compared to most annual species (Barnett, 2012). A trifida is damaged (i.e. damage to the foliage), but not killed by moderate frost (Stevens, 1924).

In North America, there is variation in *A. trifida* plant traits at both large and small geographic scales. Populations in the western USA corn belt had nearly four times greater fecundity and a 50% greater allocation to reproduction than populations in the eastern USA corn belt (Hovick *et al.* 2018). In addition, seedling emergence patterns differ among populations in agricultural fields (Schutte *et al.*, 2006; Schutte *et al.* 2008; Sprague *et al.*, 2004). For example, the latter author showed that seeds which were from Iowa (western USA corn belt) produced seedlings in a rapid flush during early April, whereas seeds from Illinois and Ohio (eastern USA corn belt) produced seedlings in a more gradual flush that extended into late July. Seedling emergence patterns also differ between agricultural and non-agricultural environments. Populations from agricultural habitats exhibited a more prolonged emergence pattern than those from riparian, early successional, railroad siding, or forest border habitats (Hartnett *et al.* 1987; Hovick *et al.* 2018; Schutte *et al.* 2012). A variant, *A. trifida* L. var. *texana* Scheele - is recognized in the southwestern USA (USDA, https://plants.usda.gov/core/profile?symbol=amtrt). This variant has small fruits (0.4 mm long) (http://artemis.austincollege.edu/acad/bio/gdiggs/NCTXpdf.htm). At present no different phenotypes are known in the EPPO region.

#### **Existing PRAs:**

The following PRAs are available:

- PRA area Poland: A PRA was conducted by Poland in 2001 on *Ambrosia* spp.: "Pest Risk Analysis and Pest Risk Assessment for the territory of the Republic of Poland (as PRA area) on *Ambrosia* spp." by Karnkowski (2000). *Ambrosia* spp. (A. artemisiifolia, A. trifida and A. psilostachya) have been categorised as quarantine organisms.
- PRA area Lithuania: A PRA was conducted for Lithuania: "Pest risk analysis and pest risk assessment for the territory of Lithuania on *Ambrosia* spp. (A. artemisiifolia, A. trifida and A. psilostachya)" (Anonymous, 2003). Species of the genus *Ambrosia* have been described as quarantine organisms that require the use of phytosanitary measures.

EFSA (2007a, b) highlighted that both PRAs were not up to standards in particular with regard to detailed information on *A. trifida* and the fact that the PRAs assessed three species within one PRA document and as a result, *Ambrosia* species were not listed as quarantine pests for the EU.

• PRA area EU: A PRA was conducted by ANSES (2017) on *Ambrosia trifida* 'Risk analysis relating to the giant ragweed (*Ambrosia trifida* L.) in order to formulate management recommendations' for the EU'. The PRA concluded that the regular entry of *A. trifida* into the PRA area, and the establishment of the species in all regions with warm summers is likely. Economic impact (effect on crop yield) can be high and will depend on the species becoming established within the region.

The PRA conducted by ANSES recommended the following measures:

- Monitoring the absence of A. trifida seeds in seed imports on entering the PRA area
- Monitoring the emergence and development of new populations of A. trifida
- Implementing early eradication measures for newly-reported populations
- Implementing a containment or eradication plan for already-established populations

A PRA has been conducted for the COSAVE region (Argentina, Bolivia, Brazil, Chile, Paraguay, Peru and Uruguay) (COSAVE, 2018). The outcome of the PRA reports that the probability of entry on the pathway contamination of seed imported for sowing as medium with a medium uncertainty. For the contaminant of imported grain, the rating is low with a medium uncertainty. Contamination of imported wool was assessed and rated as negligible for risk and uncertainty. The overall probability of establishment and spread was medium with medium uncertainty. The potential economic and environmental consequences was rated as high with a low uncertainty. The PRA recommends that *A. trifida* is included in the list of quarantine pests for the COSAVE region and requirements for imported seed for sowing should include:

- The place of production/ site of production is inspected during the growing season and found to be free from *Ambrosia trifida*,
- The shipment is free of Ambrosia trifida, according to the result of an official laboratory analysis.

Wilson *et al.* (2016) provides a pathway risk analysis of weed seed in imported grain into Canada and provides information on A. trifida seeds intercepted in maize, soybean, cereals and pulses from the USA between 2007 and 2015. Wilson et al. (2016) highlights that grain cleaning and processing methods differ for human use and animal consumption, with grain for human use presenting a lower risk of introducing weed seed into new environments compared to grain that undergo minimal processing for livestock feed. These factors have been taken into consideration in this PRA.

#### Other documents considered:

EFSA (2010) 'Scientific Opinion on the effect on public or animal health or on the environment on the presence of seeds of *Ambrosia* spp. in animal feed' detail that animal feed material is extensively processed and processing destroys *Ambrosia* seeds suggesting that compounded feed has a negligible effect on dispersing *Ambrosia* seed. However, bird seed can remain unprocessed and may play a role in dispersing *Ambrosia* seed. These factors have been taken into consideration in this PRA.

3. Is the pest a vector?	Yes	No	X
4. Is a vector needed for pest entry or spread?	Yes	No	Х

#### 5. Regulatory status of the pest

All *Ambrosia* species are regulated in Directive 2002/32/EC as undesirable substances in animal feed. In the EU, grain intended for bird feed is subject to regulations that severely restrict the presence of seeds of species of the genus *Ambrosia* (50 mg/kg of grain, Regulation (EU) 2015/186 of 6 February 2015).

In the USA, *A. trifida* has the status of "restricted noxious weed" in 4 states (California, Delaware, New Jersey, Wisconsin) under the Federal Seed Act (USDA, 2018; https://www.ams.usda.gov/rules-regulations/fsa) and the status of "noxious weed" in 4 states (California, Delaware, Illinois, Minnesota (in two counties, only)) under the Federal Noxious Weed Act and Minnesota Noxious Weed Law (USDA - https://plants.usda.gov/java/noxComposite?stateRpt=yes, Minnesota Department of agriculture - https://www.mda.state.mn.us/plants/pestmanagement/weedcontrol/noxiouslist/countynoxiousweeds).

In Canada, it is listed as a "primary noxious weed" under the Weed Seeds Order of the Seeds Act (http://www.gazette.gc.ca/rp-pr/p2/2016/2016-05-18/html/sor-dors93-eng.html) and a noxious weed under the noxious weed laws in the provinces of Ontario, Quebec, and Manitoba (Ontario Ministry of Agriculture, Food and Rural Affairs - http://www.omafra.gov.on.ca/english/crops/facts/info\_ragweed.htm).

In the EPPO region the species is regulated (or listed) at least in the following countries:

- Asia: Kazakhstan and Uzbekistan: A1 List,
- **Europe**: Azerbaijan, France (regulated by Ministerial Order (26 April 2017)), Serbia, Ukraine: A1 List, Belarus: Quarantine pest, Moldova and Russia: A2 List.

It is also listed by the Eurasian Economic Union (A2 List) and EPPO (List of Invasive Alien Plants)

In 2016, China put in place a new grain import law to keep invasive weeds and other plant pests from entering their country. *Ambrosia trifida* is included in the list of invasive weeds (USDA, 2018).

In Peru it is classified as a quarantine pest not present (SENASA-PERÚ, 2017). *Ambrosia trifida* appears on the list of regulated pests for Argentina as a quarantine pest, not present (IPPC, 2017).

## **6.** Distribution

Continent	Distribution	Comments	Reference
Africa	Absent		
North America	Canada <sup>n</sup> (Alberta, British Colombia, Manitoba*, New Brunswick, Nova Scotia, Ontario***e, Prince Edward Island, Quebec*, Saskatchewan*), Mexico (Chihuahua), USA (Alabama, Arizona, Arkansas **e, California**, Colorado*e, Connecticut, Delaware**n, Florida, Georgia, Idaho, Illinois***en, Indiana***, Iowa***e, Kansas **e, Kentucky*, Louisiana, Maine, Maryland*, Massachusetts, Michigan, Minnesota **e, Mississippi, Missouri **e, Montana, Nebraska **e, New Hampshire, New Jersey**n, New Mexico, New York, North Carolina, North Dakota, Ohio***, Oklahoma, Oregon, Pennsylvania**e, Rhode Island, South Carolina, South Dakota, Tennessee***e*e, Texas, Utah, Vermont, Virginia*, Washington, West Virginia, Wisconsin***e*e, Wyoming)	Native to North America * reported as having invasive tendencies  w reported as being one of the most competitive, abundant, problematic, or difficult to control (often due to resistance) weeds in agricultural crops  e reported as expanding, i.e. becoming a more important weed compared to earlier, or as having appeared as a crop weed only within the last 15 years  n identified as a noxious weed in this state or country through the state or country noxious weed or seed laws	EPPO (2019); USDA-NRCS (2012) Regnier et al. (2016); See additional references under 5. regulatory status of the pest, above.
Asia	Mongolia*	Non-native to all countries in Asia.  * Casual	EPPO (2019); Gagnidze, (2005); Qin et al. (2014); Wan et al. (2012); Yan et al. (2001).
	China**, Japan**, South Korea**	**Established	Kee Dae (2017)
Europe	Albania*, Austria*, Belarus*, Belgium*, Denmark*, Estonia*, Georgia*, Ireland*, Latvia*, Lithuania*, Luxembourg*, Moldova*, Netherlands*, Norway*, Poland*, Portugal*, Romania*, Slovakia*, Slovenia*, Spain*, Switzerland*, United Kingdom*, Ukraine*.	Non-native to all countries in Europe *Casual	(pers. Obs. Follak), (Fedorov 1994), (Verloove 2006), (EPPO, 2019), (BSBI Atlas), (BSBI Atlas), pers. Obs. Johan van Valkenburg, 2019), (Gudzinskas 1993), (Karnkowski 2001),

Continent	Distribution	Comments	Reference
	Bulgaria**, Czech Republic**, France**, Germany**, Italy**, Russia**, Serbia**.	**Established	(Tabaka <i>et al.</i> 1988), (Anthos 2019), (EPPO 2014), (Stoyanov <i>et al.</i> 2014), (Sirbu & Opera 2008),
			(Follak et al. 2013), (Chauvel et al. 2015), (Buttler et al. 2016), (Celesti-Grapow et al., 2009)
Oceania	Absent		

#### Introduction

Ambrosia trifida is native to North America where the species is recorded as being weedy in many States (USDA, 2012). The species has been introduced to Asia and the EPPO region (see Appendix 4, Fig. 1).

#### **North America**

In North America, *A. trifida* seems to prefer establishment at latitudes between 45° and 30° North, because of fairly strict photoperiodic constraints for flowering, which may maximise its reproduction (Allard, 1943).

#### **EPPO** region

Ambrosia trifida was introduced into the EPPO region at the end of the 19<sup>th</sup> century, and it has expanded its range since the Second World War (Follak *et al.*, 2013, Chauvel *et al.*, 2015).

Many of the occurrences of *A. trifida* in the EPPO region are considered casual populations (for example see table above and Appendix 4, Fig. 3). Of 324 observations of *A. trifida* in Central Europe, only 27% were considered as established by Follak *et al.* (2013). There are, however, well-established populations in Western Europe with high densities in south-west France (Chauvel *et al.*, 2015). It is also considered established in a large part of Italy (<a href="http://dryades.units.it/floritaly/index.php?procedure=taxon\_page&tipo=all&id=5573).">http://dryades.units.it/floritaly/index.php?procedure=taxon\_page&tipo=all&id=5573).</a>

#### Specific details about the distribution in selected EPPO countries

Austria: A. trifida was found for the first time in 1948 in Graz, in south-east Austria. The species is thought to be no longer present within Austria (pers comm, Follak, 2019), however, it cannot be certain that casual populations do not occur.

Belarus: Fedorov (1994) detailed A. trifida to be casual in Belarus.

Belgium: A. trifida was mentioned for the first time in 1894 in Heverlee by Suttor (Lawalrée, 1947). The species may have been introduced by imports of contaminated wool in the valley of the Vesdre.

Bulgaria: A. trifida has been repeatedly introduced to Bulgaria. It has been recently found in the Varna District (NE-Bulgaria) as well as in Kostinbrod town (Sofia region) in 2014. The introduction of the species in the area of the Kostinbrod dates back to 1993 (Stoyanov *et al.*, 2014).

Czech Republic: The first *A. trifida* plant was reported in 1960 in Brno. Since then, the species has spread to different points of the Czech Republic (Follak *et al.*, 2013). There are currently a few established populations in the Kolín district (Rydlo *et al.* 2011).

Estonia: A. trifida has been collected from five localities in 1989-1990, four of these were railway stations in North and Central-Estonia and one exact locality is unknown (Kapp, 2016).

France: A. trifida has been mentioned in French botanical gardens since 1765 (Paris). The first observations of A. trifida were made in Alsace between 1901 and 1904 (under German occupation and related to imports into Germany carried out at this time). Other observations were made during the First World War concerning populations introduced with forage from the USA. Establishment of the species in France seems to be recent and unrelated to the first introductions at the beginning of the 20th century; it may be linked to more recent and more southerly introductions, probably with seed imports of soybeans (Chauvel et al., 2015).

Germany: Confirmed to be present by Buttler *et al.* (2016). Mentioned for the first time in 1877 in Hamburg with early reports established populations (e.g. one population occurring continuously from1889-1904, Poppendieck 2007), *A. trifida* may have been introduced with foreign wheat seed (Follak *et al.*, 2013). Several populations in Berlin were found from 1930s to 1990 but have disappeared since then (Senatsverwaltung für Umwelt, Verkehr und Klimaschutz 2019).

Ireland: *A. trifida* is rare or casual in Ireland. No recent observations have been made (Bruce Osborn, pers. comm., 2019, University College Dublin, Ireland). It would seem that the species was introduced by imports of contaminated seed (EPPO, 2014).

Italy: *A. trifida* is mainly found in northern Italy (Atzori *et al.*, 2009; Ardenghi, 2010) almost always on riverbanks but also in cultivated fields (Acta Plantarum - Flora delle Regioni italiane, 2019). It is reported in the whole of northern Italy (Piedmont, Lombardy, Veneto, Tuscany, etc.; <a href="http://luirig.altervista.org/flora/taxa/index2.php?scientific-name=ambrosia+trifida">http://luirig.altervista.org/flora/taxa/index2.php?scientific-name=ambrosia+trifida</a>), especially in the plain of the Po (Atzori *et al.*, 2009; Ardenghi, 2010).

Israel: The plant was found in the summer of 2001 and it has since been eradicated (EPPO, 2019 citing T. Yaacoby, Israeli Plant Protection Organization, pers. comm., 2014). Other individual plants have been found since but immediately eradicated. The species is officially reported as absent, pest eradicated in Israel (EPPO, 2019).

Lithuania: The first observation of the species was in 1947 in Vilnius. New observations of *A. trifida* were then made 40 years later (1987). Its introduction may be linked to imports of North American seed (Gudzinskas, 1993).

Luxembourg: The species was observed at Neudorf in 1950 on ruins, in conjunction with *A. artemisiifolia* (Beck *et al.*, 1951).

Netherlands: The species is traditionally found at ports (grain imports), and in cereal processing companies (EPPO, 2019). As a casual it is found on riverbanks of major rivers suggesting the species is colonising from populations upstream.

Poland: During the period from 1900 to 1997, 20 outbreaks of *A. trifida* were identified in Poland (Karnkowski, 2001).

Romania: *A. trifida* has been mentioned in South-west Romania since 1970-1980 (Culita and Opera, 2011) as well as 2005 in south-eastern Romania (Ialomița county) (Sirbu and Oprea 2008).

Russia: *Ambrosia trifida* was first reported in the beginning of XX<sup>th</sup> century in Abkhazia (Flora of the USSR. 1959). The species is recorded as having limited distribution in Russia by Vinogradova *et al.*, (2010). The species spread after the Second World War to some areas of the European part of Russian Federation (especially in South Ural (Republic of Bashkortostan and some places in Volga region). Its introduction may be linked to feed imports from Ukraine (Terekhina, 2015 citing Abramova, 1997). In the eastern part of Russia, *A. trifida* has been found in the Rostov region, St. Petersburg, Leningrad, Samara region, Udmurtia and Bashkiria (Terekhina, 2015).

Spain: There are a few occurrences known in the Basque Country (Anthos, 2019). The plant has been collected along the highway in Santurtzi and Portugalete, near Bilbao but the population disappeared (M. Herrera, 2019, pers. comm.). It has also been collected in the vicinity of the oil factory of Marina de Cudeyo, in Cantabria (pers. comm. M. Herrera, 2019).

Serbia: Ambrosia trifida was recorded in Serbia for the first time in 1981 in the town of Čoka but in the following period it disappeared from this regularly visited locality (Koljadžinski et al., 1982). Ambrosia trifida was recorded again in Serbia in 1995, at the locality Despotovo, about 150 km away. At this locality the species has formed a stable population and had started to spread very quickly. Presently it may be recorded in a wider area in a phase of active spreading. This species and other species of genus Ambrosia were introduced accidentally, with seed material and mineral matter.

Slovakia: It was found for the first time in 1980 (EPPO, 2019). The introduction of *A. trifida* was due to imports of North American maize grain via the USSR (Jehlik and Dostalek, 2008).

Slovenia: A. trifida was observed for the first time in 1980 (EPPO, 2014).

United Kingdom: First tangible records of *A. trifida* in the wild were from 1897, however the species might have already escaped from cultivation before (Murray 1808, EPPO, 2019). Since 1970, the frequency of recordings has decreased (BRC, 2019). The species is recorded as very scattered in GB (Stace, 2019).

#### Asia

In Japan (see Appendix 4, Fig. 4), the first record was in 1952 from the Shizuoka Prefecture (Honshu) and currently occurs in almost entire Japan (https://www.nies.go.jp/biodiversity/invasive/DB/detail/80410e.html).

In South Korea, A. trifida was first recorded in Seoul metropolitan area during the 1970s and now it is widely naturalized in the country (Jung et al. 2017).

CABI (2019) citing Qin *et al.* (2014) detail that *A. trifida* was introduced into China in 1935 from North America. For China, the literature shows differences in the number of provinces where *A. trifida* occurs. For example, Xu *et al.* (2012) detailing 5 and Fang Hao (2011) detailing 12.

## 7. Habitats and where they occur in the PRA area (Habitat classification based on **EUNIS** habitat types)

Habitat (main)	Classification	Status of habitat	Is the pest present in the habitat in the PRA area (Yes/No)	Comments (e.g. major/minor habitats in the PRA area)	Reference
C: Inland surface waters	Banks of major waterways, littoral zone of inland surface waterbodies	Protected in part	Yes	Major	Jehlik and Hejny, 1974
E: Grasslands and lands dominated by forbs, mosses or lichens;	Ruderal environments	Protected in part	Yes	Major	Follak <i>et al.</i> , 2013;
I: Regularly or recently cultivated agricultural, horticultural and domestic habitats,	Cultivated fields, bare tilled, fallow or recently abandoned arable land (I1-5),	None	Yes	Major	Rydlo <i>et al.</i> , 2011; Chauvel <i>et al</i> , 2015
X: Habitat complexes	Domestic and non- domestic gardens	None	Yes	Major	Follak <i>et al.</i> , 2013
J: Constructed, industrial and other artificial habitats	Road networks (J4-2), rail networks (J4-3)	None	Yes	Major	Follak et al., 2013

In North America, *A. trifida* grows in different types of herbaceous communities, including ruderal habitats such as railroad sidings and roadsides, and cultivated fields, on rather rich and moist soil (Bassett and Crompton, 1982; Hartnett *et al.*, 1987; Krippel and Colling, 2006; Regnier *et al.* 2016). It is also found in damp natural environments, particularly on river banks and floodplains as well as managed moist environments such as ditchbanks, irrigation ditches, and waterways (Regnier *et al.* 2016; Sickels and Simpson, 1985).

In Japan, *A. trifida* can be found preferably along riverbanks, mostly in disturbed locations (artificial banks, bridges and quarries) but also in the riverine vegetation (Ishikawa *et al.*, 2003; Miyawaki & Washitani, 2004; Shimizu *et al.*, 2007). In South Korea it occurs in the riparian systems of streams and rivers and around agricultural fields, on road edges, landfill sites and recently, it has also invaded forest edges and interiors (Lee *et al.*, 2010; Kim, 2017). In Japan and Korea, *A. trifida* grows also in semi-natural areas (Miyawaki, 2004; Lee *et al.*, 2010).

Suitable habitats occur for the establishment of *A. trifida* in the PRA area. It occupies different environments: agricultural land (Rydlo *et al.*, 2011), the banks of major water courses such as the Rhine and the Elbe, the banks of streams or canals (Jehlik and Hejny, 1974), road networks, and other disturbed environments (e.g. abandoned industrial sites), as well as green urban areas (gardens) (Follak *et al.*, 2013).

For *A. trifida* most natural habitats of high conservation value are unsuitable, and thus negative effects of this plant on biodiversity are considered to be of low importance. Nevertheless, some data is available on *A. trifida* showing that it is able to invade natural riverside vegetation. There is no data for negative impacts of the species on rivers, especially for where it occurs in the Po Valley (IT) in the EPPO region. In central and eastern Europe, *A. trifida* mainly occupies ruderal habitats including railway tracks. and cultivated fields (Rydlo *et al.*, 2011). According to Stoyanov *et al.* (2014), *A. trifida* may be established around *Robinia pseudoacacia* bushes close to the railway at the exit of the town of Dalgopol (Bulgaria). In western Europe (France), the species only occupies cultivated fields.

#### 8. Pathways for entry

Seed and grain should be understood in this PRA as defined in ISPM 5:

Seeds: cypsela/ seeds (in the botanical sense) for planting

Grain: Seeds (in the botanical sense) for processing or consumption, but not for planting.

Globally, there have been numerous interceptions of *A. trifida* as contaminant of seed for planting or as a contaminant of grain for human or animal consumption. COSAVE (2018) highlights a number of examples detailing the primary literature:

- Egypt: between 2009 and 2010 grain of wheat, corn and sorghum had contamination of Ambrosia seed (including *A. trifida*). Shipments originating from Ukraine, USA and Russia had 3.7 %, 1 % and 2.3 % contamination, respectively.
- Australia: between 1994 and 1995 seeds of *A. trifida* were detected in imports of corn and sorghum from the USA and from soybean (origin not included),
- Peru: in 2017 and 2018, *A. trifida* was detected on 43 occasions in shipments of corn and soybean grain imported from the USA.

A. trifida has been introduced in Europe with imports of animal feed and seed. Other pathways of entry have been described, such as imports of grain for the agri-food industry (Verloove, 2006). There are documented cases of introduction of A. trifida into the EPPO region (Europe) via seed from crops imported from North America (Follak et al., 2013; Chauvel et al., 2015). This includes contaminant of spring wheat seed for planting (Follak et al., 2013), soybean seed (Chauvel et al., 2015), maize seed (Chauvel et al., 2015; Stoyanov et al., 2014) and seed of other spring crops (sunflower, sorghum) (pers. comm. G. Fried, 2019).

Contaminant of spring wheat seed is considered a historical pathway and the EWG considers that it remains very unlikely that the species will enter via this commodity due to certification and regulations on seed as well as the lifecycle of *A. trifida* versus spring wheat.

Many of the occurrences in the EPPO region are historic casual occurrences. This may be related to the pathway of entry and over recent time the movement of seed and grain into the EPPO region from areas where the species occurs has increased.

Possible pathway (in order of importance)	Pathway: Contamination of seed (for planting) of cereals, soybean, sunflower from areas where <i>A. trifida</i> is established	
Short description explaining why it is considered as a pathway	Ambrosia trifida has been recorded as growing in crop fields following the sowing of maize seed, soybean seed, spring crops (sunflower and sorghum) from USA within the EPPO region.	
	Contaminant of soybean, maize seed and seed from other spring crops (sunflower, sorghum) are considered current pathways.	
	There will be less risk of contamination in seed which has been certified.	
Is the pathway prohibited in the PRA area?	No, this pathway is not prohibited in the PRA area.	
	There are some requirements at EU level in marketing Directives for seed <a href="https://ec.europa.eu/food/plant/plant_propagation_material/legislation/eu_marketing_requirements_en">https://ec.europa.eu/food/plant/plant_propagation_material/legislation/eu_marketing_requirements_en</a>	
	Ambrosia trifida seeds in seed materials is controlled by phytosanitary requirements on the territory of the EEU countries (Armenia, Belarus, Russia, Kazakhstan, Kyrgyzstan) (Decision of the Council of the Eurasian Economic Commission of November 30, 2016 №157)	
Has the pest already been intercepted on the pathway?	Yes. There are documented cases of introduction of <i>A. trifida</i> into the EPPO region (Europe) via seed from crops imported from North America (Follak <i>et al.</i> , 2013; Chauvel <i>et al.</i> , 2015). This includes contaminant of soybean seed (Chauvel <i>et al.</i> , 2015), maize seed (Chauvel <i>et al.</i> , 2015; Stoyanov <i>et al.</i> , 2014) and seed of other spring crops (sunflower, sorghum) (pers. comm. G. Fried, 2019).	
	A. trifida has also been intercepted in seeds of grasses from the Netherlands (most likely the seed was imported into the Netherlands from other regions) (Karnkowski, 2000) and in Russia in imported flax seed (see https://vniikr.ru/main/events/ob-obnaruzhenii-voronezhskim-filialom-fgbu-«vniikr»-zarazhennoj-partii-semyan-lna)	
What is the most likely stage associated with the pathway?	Seeds of <i>A. trifida</i> may become associated with seeds of spring crops at harvest.	

What are the important factors for association with the pathway?	The probability that seeds of <i>A. trifida</i> are associated with the pathway at origin depends mainly on the crop species concerned (spring crops are more likely to be contaminated), on the exact origin of the imported product and the degree of infestation of this region by <i>A. trifida</i> . The timing or harvest can influence if <i>A. trifida</i> contaminates the commodity.  The likelihood that <i>A. trifida</i> seeds are associated with the pathway at the point of origin greatly depends on the effectiveness of the management measures implemented during cultivation, the degree of resistance of local populations to glyphosate or to ALS inhibitors, and the cleaning procedures that can be implemented at the origin before export.  Seeds are sorted after harvest, and submitted to quality requirements in particular when they are certified, which will reduce the probability of association (EU marketing directive, OECD Standards)
Is the pest likely to survive transport and storage along this pathway?	The seeds of <i>A. trifida</i> can remain viable in dry storage for four years (Stoller and Wax 1974) enabling their survival along the pathway.
Can the pest transfer from this pathway to a suitable habitat?	Seed for sowing contaminated by <i>A. trifida</i> is directly sown in agricultural fields, which is an optimal habitat for this species. This is particularly the case with soybean or maize fields when irrigated.
Will the volume of movement along the pathway support entry?	Yes, <i>A. trifida</i> has repeatedly been intercepted as a contaminant in seeds for planting and seed imports into the EPPO region occur in large volumes. As an example, Appendix 6 provides figures on the quantities of maize, sorghum, soybean and sunflower imported into the EPPO region from the USA from 2015-2018. Although there is variation year on year, there are significant volumes of the aforementioned seed entering the EPPO region. It is likely that the volume of <i>A. trifida</i> as a contaminant along this pathway will proportionate to imports into the PRA area.
Will the frequency of movement along the pathway support entry?	As mentioned, although the frequency of movement of maize, sorghum, soybean and sunflower imported into the EPPO region from the USA, varies year on year, the frequency of seed imports is regular, with equivalent volumes each year (an increase for maize, a decrease for soybeans).
	The frequency of movements along the pathway has no impact on the viability of the seeds introduced or on their quantity. Only the volumes imported can have an impact on the likelihood of introduction.
Rating of the likelihood of entry	Moderate X
Rating of uncertainty	Low X

Possible pathway	Pathway: Contaminant of grain e.g. used for animal feed mixture and human consumption		
Short description explaining why it is considered as a pathway	Seed of <i>A. trifida</i> maybe a contaminant in grain imported for (1) animal feed mixture and (2) human consumption. The likelihood of contamination in grain for animal feed is higher than in seed. There are some regulations for the latter but these are less restrictive. The grain important for human consumption is likely to be less contaminated than for animal consumption as regulation are stricter.		
	Grain for human consumption is cleaned to a very high standard to ensure quality and consistency for the end product. In addition, the processing of grain for human consumption may be partially or totally destructive. This is different for the processing of grain for animal feed where the standards are less restrictive, and grain may be cleaned and processed to a lesser degree. In addition, grain may be used whole for animal feed. Bird seed can remain unprocessed.		
	Therefore, although the entry into the EPPO region would be the same for both human consumption and animal feed, differences in processing should be taken into account.		
	Both commodities would be transferred to a processing facility and then separated for the two different uses.		
Is the pathway prohibited in the PRA area?	No. The EU Directive 2002/32/EC has requirements on the purity of the grain for animal feed.		
	Ambrosia trifida seeds in food is controlled by phytosanitary requirements on the territory of the EEU countries (Armenia, Belarus, Russia, Kazakhstan, Kyrgyzstan) (Decision of the Council of the Eurasian Economic Commission of November 30, 2016 №157)		

Has the pest already been intercepted on the pathway?	Yes, <i>A. trifida</i> has been intercepted along this pathway both globally and cominto the EPPO region. The species has been intercepted in animal feed from No America into Russia nine times in a period of nine months between 2009-20 (Petson <i>et al.</i> , 2011). <i>Ambrosia trifida</i> has also been intercepted in Netherlands in animal feed (soybean) from North America (van Denderen <i>et a</i> 2010).  In Egypt, between 2009 and 2010 grain of wheat, corn and sorghum has contamination of <i>Ambrosia</i> seed (including <i>A. trifida</i> ). Shipments originating from Ukraine, USA and Russia had 3.7 %, 1 % and 2.3 % contamination		
	from Ukraine, USA and Russia had 3.7 %, 1 % and 2.3 % contamination respectively (COSAVE, 2018 and references within).  In Peru, in 2017 and 2018, <i>A. trifida</i> was detected on 43 occasions in shipments of corn and soybean grain imported from the USA (COSAVE, 2018 and references within).		
	In Canada, <i>A. trifida</i> has been intercepted in grain of maize, soybean, cereals and pulses from the USA between 2007 and 2015 (Wilson <i>et al.</i> , 2016).		
What is the most likely stage associated with the pathway?	Seeds of <i>A. trifida</i> may become associated with seeds of spring crops at harvest where the species occurs.		
What are the important factors for association with the pathway?	The probability that seeds of <i>A. trifida</i> are associated with the pathway at the point of origin depends mainly on the crop species concerned (spring crops are more likely to be contaminated), on the exact origin of the imported product and the degree of infestation of this region by <i>A. trifida</i> . The timing or harvest calcainfluence if <i>A. trifida</i> contaminates the commodity. The likelihood that <i>A. trifida</i> seeds are associated with the pathway at origin greatly depends on the effectiveness of the management measures implemented during cultivation, the degree of resistance of local populations to glyphosate or to ALS inhibitors, and the cleaning procedures that can be implemented at origin before export.		
T 4 411 1 4	Grain can become contaminated at harvest in the area of origin.		
Is the pest likely to survive transport and storage along this pathway?	The seeds of <i>A. trifida</i> can remain viable in dry storage at room temperature for four years (Harrison <i>et al.</i> , 2007; Stoller and Wax 1974; Toole and Brown 1946 enabling their survival along the pathway.		
	However, the processing procedure for human consumption may act to crush and destroy the seeds. For animal feed, the seeds may stay whole and survive.		

Can the pest transfer from this pathway to a suitable habitat?	In the areas of introduction such as ports, airports or freight stations where cargos of seed for sowing or grain for industry or livestock pass through, any seeds falling to the ground would have more difficulty becoming established as shown by the species' historical decline in such sites (Chauvel <i>et al.</i> , 2015), with a disappearance from all the historical stations.		
	Grain lots may be sorted before processing to remove external matters such as weed seeds. If the waste from the sorting is put in fields, they may become infested		
	There may also be deviation froused as seed).	om the intended use (i.e. imported as grain, and	
Will the volume of movement along the pathway support entry?	It is likely that movement with volumes of the commodity will support entry. Appendix 6 shows volumes of grain (soybean and maize) entering the EPPO region from USA. In addition, the <i>A. trifida</i> has been intercepted as a contaminant of grain numerous times within the EPPO region.		
	Potentially, these figures may contain volumes for various uses (including potential industrial use), but the main volume would be for animal feed or human consumption. The figures for soybean and maize grain imports show a high volume and reasonably consistent volume of import from the USA into the EPPO region.		
Will the frequency of movement along the pathway support entry?	The frequency of movement along the pathway is likely to support entry. Although there are no figures to highlight the frequency of movement of <i>A. trifida</i> seeds as a contaminant of grain it is likely that movement with volumes of the commodity will support entry. Grain is frequently imported into the EPPO region from North America (see Appendix 6).		
Rating of the likelihood	Grain for animal feed mixture	High	
of entry	Grain for bird seed mixture	High	
	Grain for human consumption	Moderate	
Rating of uncertainty	Grain for animal feed mixture	Low	
	Grain for bird seed mixture	Low	
	Grain for human consumption	Moderate	

### Historical pathways considered by the EWG as unlikely for the entry of the species

- Contaminant in fodder for horses. This has been historically recorded as fodder was imported into the American camps in France in the First World War (Brandicourt, 1918).
- Contaminant of straw introduced into Poland in 1903 (Lackowitz, 1903),
- Contaminant of cotton fibres introduced for the textile industry and found in a field fertilised with cotton compost in Issenheim (France) in 1971 (Herbier G.),
- Wool contaminants (Verloove, 2006).

#### Other pathways where the species could travel along but the EWG consider are unlikely:

- Contaminant of used machinery and equipment (See ISPM 41). However, this may play a role for local spread.
- Contamination of processed grain (e.g. sunflower or soybean meal) seeds are unlikely to survive the digestive system of animals.
- Contaminant of growing media (See ISPM 40). Import as such is prohibited in most EPPO countries, not likely to be associated with growing media attached to plants for planting
- Import as plants for planting. The species was imported into botanical gardens in the 18<sup>th</sup> century (within Europe). The EWG considers that the trade of *A. trifida* is unlikely as it is not usually used or traded as an ornamental species.

#### No other pathways need to be considered.

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

Ambrosia trifida is already established in Bulgaria, Czech Republic, France, Germany, Italy, Russia and Serbia.

Follak *et al.* (2013) modelled the climatic zones favourable to the species, in central and eastern Europe. According to these authors, because the species is constrained by temperature and precipitation, only 16% of the territory considered (central Europe) would be climatically favourable to the species. The species currently occupies less than 1.5% of the climatically favourable areas in central and eastern Europe.

Based on modelling conducted in the national French PRA (used as the basis for this PRA), using distribution data (covering the area of origin and the area of introduction GBIF and additional data) an ecological niche model with the MaxEnt algorithm (Elith *et al.*, 2011) was produced using Worldclim climate data. European populations regarded as casual (historical observations not confirmed recently) were not taken into account. Three Bioclim variables (Hijmans *et al.*, 2005) were selected on the basis of the biology of the species (the species is annual with spring germination and a summer growing period: mean diurnal temperature range (monthly mean of the difference between max. and min. temperature, Bio2), mean temperature of the warmest quarter (Bio10), and precipitation of the warmest quarter (Bio18). The model's results suggest that *A. trifida* encounters favourable climatic conditions in France (in the southwest, the valleys of the Rhône, the Saône and the Rhine), Spain (north-east), northern Italy (mainly the plain of the Po), very locally in Switzerland, in southern Germany, southern Poland, the southern Czech Republic and Slovakia, eastern Austria, Hungary, northern Slovenia, Croatia, Bulgaria and Romania. These results suggest that a large part of the PRA area can still be colonised by *A. trifida*.

Based on species distribution modelling conducted during the development of this PRA (Appendix 3), and evaluated by the EWG, the model suggests that suitability for *A. trifida* at the global scale and resolution of the model was most strongly determined by winter temperature (Bio6), winter precipitation (Bio19), energy availability (PET) and moisture availability (CMI) (see Appendix 3, Table 1, Figure 3). There were also appreciable effects of human modification and the distribution of preferred crop types. Across Europe and the Mediterranean region, the model predicts a large climatically suitable range spanning most of Europe below ~55 °N, excluding the Mediterranean coastline, and generally increasing in suitability towards the more continental east. *Ambrosia trifida* is currently absent, recorded sporadically or unrecorded from the central part of this distribution (see Figure 1), suggesting a potential for much wider establishment.

The model suggests that establishment in northern Europe and the Ural Mountains of Russia will be limited by low energy availability (low PET), while warm winters mainly limit establishment around the Mediterranean and Black Sea coastlines (Figure 6). Drought stress (low CMI) was suggested to be the most important limiting factor in the driest parts of southern Russia and in Turkey. The EWG considers the modelling conducted within this PRA to be a realistic projection of the potential occurrence of *A. trifida* in the EPPO region. The model may overpredict the potential occurrence of the species in the warmer and drier Mediterranean area due to the inclusion of the layer for crop land cover in the model. Actual suitability of these areas may be restricted to irrigated fields.

Within the EPPO region, the species mostly grows in disturbed habitats. Based on data from the native range, in the PRA area the species might be capable of establishing in flood plains, grassland and forest borders. Actual occurrences are mostly along riverbanks in Italy (Follak *et al.*, 2013) and the Netherlands, flood plains and moist meadows in Russia (Abramova, 2011). In stable natural environments interspecies competition may limit population sizes.

A. trifida is not well adapted to drought and it is not recorded in areas with a long summer drought unless there is irrigation (Allard, 1945). Establishment is favoured by moist environments (Korres et al., 2015). A. trifida can tolerate a wide variety of soil types though it prefers moist fertile soils. Seeds germinate under a wide range of temperatures with an optimum germination between 10 - 24°C (Abul-Fatih and Bazzaz, 1979). There is no information on the effect of day length on germination.

Like A. artemisiifolia (Kiss, 2007), A. trifida has few natural enemies in the PRA area likely to reduce its establishment potential.

Rating of the likelihood of establishment in the natural environment	Very low □	Low 🗆	Moderate □	High <b>X</b>	Very high □
Rating of uncertainty			Low X	Moderate □	High □

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

In ruderal environments contiguous to agricultural areas, due to preferential colonisation of bare soil and a competitive advantage linked to rapid germination and development (Bassett and Crompton, 1982), it is unlikely that the potential area of establishment of *A. trifida* would be reduced by competition with other ruderal species. In the agricultural environment, it is also unlikely that competition with cultivated plants would prevent the establishment of the species.

The high frequency of spring and summer crops such as maize, soybean and sunflower in the crop rotation system (including tillage and irrigation) is a factor that strongly promotes the establishment of *A. trifida* once the field has become contaminated. Monocultures of spring and summer crops as well as reduced tillage are likely to promote *A. trifida* (Regnier *et al.*, 2016). Irrigation could also favour this mesohygrophilic species, particularly beyond its climate envelope in areas where the limiting factor is the level of summer precipitation (areas of southern Europe, North Africa, Israel and Jordan).

When A. trifida is present in crop fields, common weed control methods for soybean and sunflower are not sufficient to limit the development of the plant, due to its prolonged emergence and rapid growth. In addition, further complications may arise from the reduction in the number of herbicide compounds, the

glyphosate resistance in this species (Norsworthy *et al.*, 2011) and the decrease in the number of herbicides treatments associated with the reduction in the use of plant protection products. All of the aforementioned factors can potentially promote the establishment of *A. trifida*. In contrast, in crop systems where different weed control practices are used, in maize crops, conventional broad-leaf pre-emergent (mesotrione, thiencarbazone-methyl) and/or post-emergent (e.g. dicamba and 2,4-D) herbicides if applied in a timely way should be able to effectively prevent the establishment of this species.

Rating of the likelihood of establishment in the managed environment	Very low □	Low 🗆	Moderate □	High □	Very high X
Rating of uncertainty		Low X	Moderate □	High	

#### 11. Spread in the PRA area

#### Natural spread

The seeds of *A. trifida* are relatively large in size (cypsela 0.5 to 1.2 cm long) and naturally spread mainly by barochory (falling from the parent plant) and hydrochory (dispersal via water). In the case of barochory, dispersal takes place over very limited distances (a few metres around the mother plant). In contrast, for populations growing near rivers or on sloping land, dispersal by hydrochory may carry the seeds over several kilometres. The speed and distance of dispersal can therefore vary greatly according to the situation of the contaminated area in the toposequence and depending on the presence of a water course in the immediate vicinity. The seeds can be displaced from a few centimetres (earthworms) to a few metres or more (Goplen *et al.*, 2016; Harrison *et al.*, 2003; Payne, 1962; Regnier *et al.*, 2008) by animal species in agro- ecosystem communities (rodents, birds).

Ambrosia trifida has been present within the EPPO region (often as casual populations) since the end of the 19<sup>th</sup> century and the species has not been shown to spread through natural means significantly throughout the region. The reasons for this could be due to the species not predominantly growing in more natural habitats within the region and therefore spread has not been facilitated by natural pathways such as rivers. The seed is also relatively large and thus not spread by wind. Additionally, the species may be in a lag phase within the EPPO region and further spread may be realised in the future.

#### **Human assisted spread**

The rate of spread by human assistance can be very high, either by contamination and subsequent spillage from transporting containers of crops intended for seed or grain for processing or feed for livestock or wild animals, or dispersal of seeds by agricultural machinery. Its introduction may be linked to feed imports from Ukraine (Abramova, 1997). This is particularly the case with harvesters operating in contaminated fields of soybean, maize or sunflower seed. Because some seeds are still attached to the plant at the time of harvest (Goplen *et al.*, 2016), *A. trifida* can be dispersed by combine harvesters, which may then transfer the seeds to any other fields they subsequently visit. Tillage can also act to spread seeds within and between fields.

The areas favourable to the development of *A. trifida* are currently isolated from each other. Contamination of the entirety of a favourable area may be fairly rapid (a few years) once the species establishes a presence. However, contamination from one area to another will be much slower if the ecologically favourable areas are far away from each other and the harvesting machines do not circulate from one area to another.

Jehlik and Dostalek (2008) highlights that *A. trifida* has been spread through the transportation of grain from the former USSR to Slovakia. Trade of seed and grain within the EPPO region is a major pathway for long distance spread of the species.

Rating of the magnitude of spread in the PRA area	Very low □	Low 🗆	Moderate □	High X	Very high □
Rating of uncertainty		Low □	Moderate <b>X</b>	High □	

#### 12. Impact in the current area of distribution

#### 12.01 Impacts on biodiversity

A. trifida can be a dominant species within the current area of distribution. The species is highly competitive and can form annual monospecific stands in ruderal, forest border, and grassland habitats, and on riparian levees (Regnier *et al.*, 2016; Sickels and Simpson 1985).

For *A. trifida* most natural habitats of high conservation value have a low potential to be invaded as they have low levels of disturbance, and thus negative effects of this plant on biodiversity are considered to be of low importance. Nevertheless, some data is available on *A. trifida* showing that it is able to invade natural riverside vegetation. There is no data for negative impacts of the species on rivers, especially for where it occurs in the Po Valley (IT) in the EPPO region. However, there is some anecdotal evidence that the species may have impacts on biodiversity from online forums (e.g. Acta Plantarum, an Italian forum for botanists: <a href="https://www.floraitaliae.actaplantarum.org">https://www.floraitaliae.actaplantarum.org</a>) where comments include that the species has increased in a small area from 1 to 100 plants in one year (which can act to displace native plants), and the stream near to a house has been invaded by *A. trifida*.

In Japan, a study on the floral diversity of infested river banks highlighted a decrease in diversity as a function of the density of *A. trifida* (Washitani, 2001). Miyawaki and Washitani (1996) found that plant species diversity was negatively correlated with the abundance of *A. trifida* in a nature reserve of moist tall grasslands along the Arakawa River, near Tokyo/Japan. Lee *et al.* (2010) demonstrated that the vegetation dominated by *A. trifida* in Central-Western Korea differed with regard to the composition and diversity of the species to that of the uninvaded riparian vegetation

There is little data on the impact of the species on habitats, except those on the problems of rehabilitation of fragile grassland environments in the USA (Megyeri, 2011). There are very few data in the invasion area on the environmental impact of infestations of *A. trifida*.

Compared to the low or absence of impact of *A. artemisiifolia* on biodiversity (Fried *et al.*, 2014), the EWG expect a higher effect of *A. trifida* due to its high stature and high leaf area, two traits known to be related to competition for light.

A moderate rating of magnitude of impact has been given with a moderate level of uncertainty. The moderate uncertainty score reflects the low number of scientific studies conducted on the impacts of the species in its invaded range. The EWG consider the impact could be either lower or higher of the moderate rating.

Rating of the magnitude of impact on biodiversity in the current area of distribution	Very low □	Low 🗆	Moderate X	High □	Very high □
Rating of uncertainty		Low □	Moderate <b>X</b>	High	

#### 12.02. Impact on ecosystem services

Ambrosia trifida can impact on ecosystem services within the current area of distribution (see table below).

Ecosystem service	Does the pest impact on this Ecosystem service? Yes/No	Short description of impact	Reference
Provisioning	Yes	Reduces yields in agricultural cropping systems	Barnet and Steckel, 2012; Harrison <i>et al.</i> , 2001; Webster <i>et al.</i> , 1994
Regulating	Yes	Can impact on native plant species and can impact on air quality regulation (due to the release of pollen).	Golstein <i>et al.</i> , 1994; Washitani, 2001
Supporting	NA	NA	NA
Cultural	Yes	The species may have impacts on recreation where the species occurs along riverbanks. The presence of the species may act to reduce access to waterbodies for recreational purposes. In addition, pollen can cause allergic reaction in people that are susceptible, and this may limit outdoor activities where the species is present.	EWG opinion

A moderate rating of magnitude of impact has been given with a high level of uncertainty. The direction of uncertainty lies with the fact that there are few studies on the negative impact of the species on regulating, supporting and cultural ecosystem services and thus the EWG consider that the impact could be lower.

Rating of the magnitude of impact on ecosystem services in the current area of distribution	Very low □	Low □	Moderate X	High □	Very high □
Rating of uncertainty			Low □	Moderate □	High <b>X</b>

#### 12.03. Socio-economic impact

Detailed information on socio-economic impacts is available from USA and China. There is some evidence from the EPPO region, but further studies need to be conducted.

All socio-economic impacts within agricultural systems are expressed as crop yield reductions. The EWG did not find any monetary figures within the literature for this species.

#### In North America:

In its area of origin, the economic consequences associated with the presence of *A. trifida* are considered to be major from an agricultural point of view and a public health point of view.

In agricultural environments, the plant's significant and rapid development gives it a strong ability to enter into competition with different summer crops: soybean, cotton, maize. Even at very low densities (one plant per 25 m²), loss of crop yield (of around 5%) has been shown, a phenomenon rarely observed for other weeds (Harrison *et al.*, 2001). Yield reductions of 13 to 50% have been observed in crop situations, with the losses being greatest when the crop and the weed grow simultaneously (Barnett and Steckel, 2012; Harrison *et al.*, 2001). In North America, complete crop losses have been reported due to the presence of *A. trifida* (pers. comm E. Regnier, 2019).

COSAVE (2019) citing the references within, state 'dense *A. trifida* populations reduced soybean seed yields by approximately 50% (Baysinger and Sims, 1991). There was also a 55% reduction in corn yield in Michigan (Michigan State University, 2018). A density of 1 plant of *A. trifida* per m2 reduced the yield of sweet corn by approximately 40% and affected several parameters of crop quality (Williams and Masiunas, 2006). In Tennessee a density of 0.26 plants of A. trifida per metre of row reduced the yield of cotton by 50% (Barnett and Steckel, 2013)'.

In 1994, Webster *et al.* (1994) estimated the loss of yield in the USA associated with *A. trifida* in soybeans to be 5 to 7% of the yield of the crop. A recent study (Regnier *et al.*, 2016) among farmers in the USA showed that *A. trifida* was the most difficult weed to manage for 45% of them, while 57% also reported a problem of herbicide resistance, either to acetolactate synthase (ALS) inhibitors or glyphosate (or resistance to both).

In Northeast China A. trifida is considered one of the weeds that causes the most economic damage to wheat and other annual crops. It was found that the plant and its residues have allelopathic effects that reduce wheat growth (Kong et al., 2007).

From a public health point of view, in the USA, *A. trifida* has been identified as a problem since the 1930s, due to its allergenic pollen and its presence in urban areas. Historically, Gahn (1933) had already indicated that hundreds of thousands of people were affected by allergy problems without any quantified costs being mentioned. The allergens are well-known (Golstein *et al.*, 1994). Today, *A. trifida* (and its congener *A. artemisiifolia*) are the main cause of seasonal allergic rhinitis in eastern and middle

USA. The *Ambrosia* pollen also contributes to the exacerbation of asthma and allergenic conjunctivitis (Oh, 2018). It is recommended, that individuals allergic to *Ambrosia* pollen may adjust their outdoor activities to avoid contact with the allergen (e.g., https://www.aafa.org/ragweed-pollen/). The health effect remains significant to such a point that visitor numbers at certain tourist sites are affected according to the presence of species of the genus *Ambrosia*. Consequently, tourism can be impaired if visitors avoid areas with high *Ambrosia* occurrence (Durham, 1949; also see <a href="http://ragweed.digitalbishop.com/">http://ragweed.digitalbishop.com/</a>).

#### In the invaded areas in the EPPO region

Yield losses have been assessed at between a few hundred euros and a few thousand euros in the region of Toulouse (FR) in soybean plots infested by *A. trifida* (pers comm, A. Rodriguez, 2019). However, no specific study has yet been conducted in this region that can be examined scientifically. Farmers in this region report additional operating costs associated with hand weeding, and even the destruction of plots before harvesting due to very high densities of *A. trifida* (pers. comm., A. Rodriguez).

Allergy impacts have not been recorded for A. trifida in the EPPO region to date.

Rating of the magnitude of impact on the socio-economy in the current area of distribution	Very low □	Low □	Moderate □	High □	Very high <b>X</b>
Rating of uncertainty		Low X	Moderate □	High □	

#### 13. Potential impact in the PRA area

#### 13.01. Potential impacts on biodiversity in the PRA area

As mentioned in 12.01, there is no data for negative impacts of the species on native biodiversity in the EPPO region, even from where it occurs along rivers (Po Valley (IT)). Currently in France, it has only been observed in cultivated environments, with the exception of reports by the Botanical Conservatory of Midi-Pyrénées of populations present in a gravel pit in the region (pers. comm. J. Dao). This is similar for most of the EPPO region (e.g. Serbia), where the species is recorded for agricultural habitats.

There is the potential for impacts on biodiversity within the EPPO region, but the habitat preferences of the species should be taken into account when evaluating this. Within the EPPO region, the species mostly grows in disturbed habitats. The environmental impact of this species in meso-hygroscopic environments (river banks, wet grasslands, gravel pits and ditches) could damage local biodiversity.

Abramova (1997) highlights that in the future in Southern Siberia, with further spread, ecosystems of the steppe zone and southern forest-steppe, will lead to significant changes in synanthropic vegetation. However, there are no known studies that have followed up and assessed the damage in Southern Siberia.

There is no evidence that A. trifida invades protected areas within the EPPO region or areas with a high conservation status. There are no reports of impacts on specific plant or animal species within the EPPO region.

A moderate rating of magnitude of impact has been given with a high level of uncertainty. The direction of uncertainty reflects that there are no scientific studies conducted on the impacts of the species in its invaded range. The EWG consider the impact could be higher of the low rating.

Rating of the magnitude of impact on biodiversity in the potential area of distribution	Very low □	Low	Moderate X	High □	Very high □
Rating of uncertainty		Low □	Moderate □	High <b>X</b>	

#### 13.02. Potential impact on ecosystem services in the PRA area

The growth of the species along riverbanks can restrict access to the waterbody for recreation activities, however, there is currently no data for the EPPO region on such impacts. If the population increases, pollen can cause allergic reaction in people that are susceptible, and this may limit outdoor activities where the species is present. In addition, the species can reduce yields in agricultural cropping systems.

A moderate rating of magnitude of impact has been given with a high level of uncertainty. The direction of uncertainty lies with the fact that there are few studies on the negative impact of the species on ecosystem services and thus the EWG consider that the impact could be lower.

Rating of the magnitude of impact on ecosystem services in the potential area of distribution	Very low □	Low □	Moderate X	High □	Very high □
Rating of uncertainty		Low □	Moderate □	High <b>X</b>	

#### 13.03 Potential socio-economic impact in the PRA area

In Europe, it is not currently possible to quantify the economic impacts of this species. In France, in the region of Toulouse, farmers report additional costs associated with hand weeding, and even the destruction of plots before harvesting due to very high densities of plants, meaning the total loss of the crop (pers. comm. A. Rodriguez, 2017). These costs (from a few hundred euros to a few thousand euros per ha) have not yet been studied to a precise enough degree. At the national level, given the limited distribution of the species and the highly localised nature of the existing populations in the PRA area (Chauvel *et al.*, 2015; Follak *et al.*, 2013), the costs in terms of health or losses of agricultural yields attributable to this species are negligible so far.

Any action targeting control of this species will generate additional production costs (cost of weeding practices, establishment of less profitable crops or fallow). In the absence of plant health regulations relating to the control of introduction into the PRA area of seed lots of maize, soybeans, sorghum and sunflower, the risk of introduction of herbicide-resistant genotypes of *A. trifida* appears high and such an introduction would result in a very high increase in control costs, based on the studies carried out in the USA (Ganie *et al.*, 2017).

In annual summer crops where it is present, *A. trifida* is managed like other weeds without it being subject to additional control measures. Note, however, the arrival on the European market of sunflower varieties tolerant to herbicides intended to control species of the genus *Ambrosia* (and Asteraceae more generally).

These varieties, through their tolerance to two herbicides from the class of ALS inhibitors, enable weed control in a post-emergence situation; they were placed on sale in 2010 to improve the post-emergence weed control of sunflower crops in general, and more specifically against *A. artemisiifolia*. These new varieties make it easier to manage the recent problems with *A. trifida*. However, the repeated use of such varieties and the associated herbicides risks causing the significant and rapid selection of populations of *A. trifida* resistant to these active ingredients in the PRA area, as is currently occurring with *A. artemisiifolia* (Chauvel and Gard, 2010). An additional problem is the emerging resistance of *A. trifida* to glyphosate and ALS-inhibiting herbicides (Norsworthy *et al.*, 2011; Regnier *et al.* 2016), thus further decreasing the possible avenues for its control, both in agriculture (both conventional and GMO crops) and ruderal areas, such as railways, roadsides etc.

Based on the results of studies conducted in the USA (Ganie *et al.*, 2017) in 2013 and 2014, the absence of management measures against this species resulted in a total loss of maize yield, even at low weed densities. These results suggest the same level of impact in the PRA area if no control measures are implemented against *A. trifida*.

Without the implementation of integrated control against this species – effective chemical weed control, rotation including winter crops and appropriate tillage – the negative effects of *A. trifida* will probably increase, as suggested by the situation with certain plots in south-west France. However, until now, no published information is available to quantify the negative effects of *A. trifida* in the PRA area.

Some countries such as Russia, Israel, and Egypt refuse imports of cereals contaminated by species of the genus *Ambrosia*. *Ambrosia trifida* is not mature when winter cereals are harvested in Europe and will not directly contaminate these crops. On the other hand, it is mature at the time of harvesting summer crops (maize, soybean, sunflower and sorghum). Contamination of these crops could prevent their export. As an example, in 2015 the maize export sector from the EU accounted for more than 63 million tonnes (Eurostat, 2019). There is a great risk of the additional costs of weed control and/or post-harvest sorting being reflected in market losses due to a higher production cost compared with situations free from *A. trifida*.

If significant *A. trifida* populations become established in the PRA area in either cultivated or uncultivated areas, the substantial pollen production will contribute to seasonal allergic rhinitis caused by *Ambrosia* pollen, already a major health concern in the PRA, where sensitization rates can be as high as 70% (compared to 26% in the USA, Chen *et al.* 2018). Further, recent research suggests that immunotherapy practices for *A. trifida* differ from those for *A. artemisiifolia* (Asero *et al.* 2001), which could necessitate additional plant monitoring and therapeutic costs.

Rating of the magnitude of impact on the socio-economy in the potential area of distribution	Very low □	Low 🗆	Moderate □	High □	Very high <b>X</b>
Rating of uncertainty			Low □	Moderate <b>X</b>	High □

#### 14. Identification of the endangered area

Ambrosia trifida is capable of establishing in predominately a continental climate. The species is most suited to continental and Pannonian biogeographical region and in addition, part of the Mediterranean, Steppic and parts of Anatolian biogeographical regions are also suitable. All North African EPPO member countries have a potential for establishment in the northern Mediterranean areas. All EPPO countries south of 55° latitude have potential for establishment.

Habitats most at risk in the endangered area include ruderal disturbed habitats (including transportation networks), riparian systems, field crops, (annual summer crops, particularly maize, soybean and sunflower) and open habitats. Within the climatically suitable area, the distribution of the preferred crop types (maize, soybean and sunflower) is mostly restricted to the warmer continental, Pannonian and Steppic parts of the EPPO region. The present occurrences in the eastern part of Germany and Russia coincide with wheat cropping.

Based on the experience in North America, the species is likely to occur more in moist habitats (including artificial irrigation) than drier areas.

The EWG considers the modelling in Appendix 3 to be a realistic projection of the potential occurrence of *A. trifida* in the EPPO region. The model may overpredict the potential occurrence of the species in the warmer and drier Mediterranean area due to the inclusion of the layer for crop land cover in the model. Actual suitability of these areas may be restricted to irrigated fields.

#### 15. Climate change

Consider the influence of projected climate change scenarios on the pest.

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

Climate projection RCP: 4.5 and RCP 8.5 (see Appendix 3)

## 15.02. Which component of climate change do you think is the most relevant for this organism?

Temperature (yes)	Precipitation ( <b>yes</b> )	$CO_2$ levels (yes)
Sea level rise ( <b>no</b> )	Salinity ( <b>no</b> )	Nitrogen deposition ( <b>no</b> )
Acidification (no)	Land use change ( <b>yes</b> )	

## 15.03. Consider the influence of projected climate change scenarios on the pest.

Are the pathways likely to change due to climate change? (If yes, provide a new rating for likelihood and uncertainty)	Reference
The pathways are unlikely to change as a result of climate change.  For contamination of seed and the contamination of grain for animal feed mixture and human consumption there may be fluctuations in the import of the commodity.	EWG opinion
However, the EWG do not consider the scores will change as a result.	
Is the <b>likelihood of establishment</b> likely to change due to climate change? ( <b>If yes, provide a new rating for likelihood and uncertainty</b> )	Reference
The potential for establishment may change as a result of climate change. Changes in land use which could be both favorable and unfavorable (e.g. an increase or decrease in ruderal habitats) to the establishment of the species may occur within the EPPO region  On the other hand, it has been detailed that the species is not well adapted to drought and thus areas that are currently suitable for the species may become unsuitable especially in the Mediterranean region in the absence of irrigation. More extreme weather events are likely, including flooding, which will act to increase the establishment of the species.  The model shows there will be little change in the climate envelope from the current predictions to the future predictions.  However, the EWG do not consider the scores will change as a result.	EWG opinion
Is the magnitude of <b>spread</b> likely to change due to climate change? ( <b>If yes</b> , <b>provide</b> a <b>new rating for the magnitude of spread and uncertainty</b> )	Reference
The spread of the species may change with climate change. More extreme weather events are likely, including flooding, which will act to increase the spread capacity of the species.  However, the EWG do not consider the scores will change as a result.	EWG opinion
Will impacts in the PRA area change due to climate change? (If yes, provide a new rating of magnitude of impact and uncertainty for biodiversity, ecosystem services and socio-economic impacts separately)	Reference
Ambrosia trifida may become more competitive due to increased biomass production and increased seed production with increased atmospheric CO <sub>2</sub> (Ziska and Beggs 2012). In addition, increased pollen production and longer duration of pollen production period may be realised with climate change which may increase its allergenicity (Ziska and Beggs 2012).	EWG opinion

#### 16. Overall assessment of risk

	Likelihood	Uncertainty
Entry		
Contamination of seed (for planting) of cereals, soybean,	Moderate	Moderate
sunflower from areas where A. trifida is established		
Contaminant of grain e.g. used for animal feed mixture		
and human consumption		
Grain for animal feed mixture	High	Low
Grain for bird seed mixture	High	Low
Grain for human consumption	Moderate	Moderate
Likelihood of establishment in the natural environment	High	Low
in the PRA area		
Establishment in the managed environment in the PRA	Very high	Low
area		
Spread	High	Moderate
Impact in the current area of distribution		
Impacts on biodiversity	Moderate	Moderate
Impact on ecosystem services	Moderate	High
Socio-economic impact	Very high	Low
Potential impact in the PRA area		
Impacts on biodiversity	Moderate	High
Impact on ecosystem services	Moderate	High
Socio-economic impact	Very High	Moderate

#### Ambrosia trifida presents a high phytosanitary risk for the endangered area with low uncertainty.

The likelihood of new introductions occurring via contamination of seed and contamination of grain is moderate and high respectively. Within the EPPO region, *A. trifida* seems to preferentially become established in crops and ruderal environments. It is found in the littoral zone of inland surface waterbodies, bare tilled, fallow or recently abandoned arable land, road networks, rail networks and domestic and non-domestic gardens. The likelihood of further establishment in natural habitats is considered high with a low uncertainty. The likelihood of further establishment in the managed environment habitats is considered very high with a low uncertainty. The potential magnitude of spread within the EPPO region is high with moderate uncertainty. *Ambrosia trifida* has both short and long-distance natural dispersal pathways. Human assisted spread facilitated by agriculture machinery and movement within the EPPO region as a contaminant of seed or grain can act to move seeds over long distances.

The main impacts of the species at a global level are on the reduction of crop yields and human health impacts which are translated in the PRA into socio-economic impacts. In the area of distribution, the species has a dual economic cost, it can impact on yield production and it has a human health impact. The EWG consider the potential socio-economic impacts in the EPPO region are very high with a moderate uncertainty. Impacts on biodiversity and ecosystem services are moderate with high uncertainty. The high uncertainty reflects the lack of quantitative studies on impacts.

The species is particularly difficult to manage due to early and prolonged emergence and very rapid biomass growth. An established population is very difficult to control as the seeds of *A. trifida* can remain viable in soil for 4 to 21 years, depending on burial depth (Harrison *et al.*, 2007; Stoller and Wax 1974, Toole and Brown 1946). Within the EPPO region, there are a lack of effective and economical control options, and chemical control options are becoming increasing restricted in Member countries in the EPPO region.

#### Stage 3. Pest risk management

#### 17. Phytosanitary measures

#### 17.01 Management measures to prevent further introduction and spread within the EPPO region

The results of this PRA show that *Ambrosia trifida* poses a high risk to the endangered area with a low uncertainty.

The major pathway(s) being considered are:

- Contaminant of seeds (maize seed, soybean seed spring crops (sunflower and sorghum))
- Contaminant of grain (soybean and maize)

Possible pathways (in	Measures identified						
order of importance)							
Contaminant of seeds	Seed has been produced in a pest-free area (PFA)						
· · · · · · · · · · · · · · · · · · ·	Or						
spring crops (sunflower	Pest-free place of production/production site consist in the following						
and sorghum))	combination of measures: visual inspection at the place of production,						
	specified treatment of the crop, inspection of the commodity.						
	Or						
	Certification scheme for seeds: seeds certified free of <i>Ambrosia trifida</i>						
	seeds						
Contaminant of grain	Grain has been produced in a pest-free area (PRA)						
(soybean and maize)	Or						
	Pest-free place of production/production site consist in the following						
	combination of measures: visual inspection at the place of production,						
	specified treatment of the crop, inspection of the commodity.						
	Or						
	Cleaning and treatment of grain lot to remove <i>Ambrosia trifida</i> seeds,						
	AND						
	Cleaning and treatment of ships and containers.						
Used agricultural	ISPM 41 'International movement of used vehicles, machinery and						
_	equipment' should be implemented						
equipment							
	ISPM 40 'International movement of growing media in association with plants for						
media	planting'						

#### 17.02 Management measures for eradication, containment and control

#### **National measures**

Early detection is important to identify new occurrences of the species. *Ambrosia trifida* should be monitored and eradicated, contained or controlled where it occurs in the endangered area. In addition, public awareness campaigns to prevent spread from existing populations or from botanic gardens in countries at high risk are necessary. If these measures are not implemented by all countries, they will not be effective since the species could spread from one country to another. National measures should be combined with international measures, and international coordination of management of the species between countries is

recommended.

Agricultural machinery should be cleaned and decontaminated before movement.

The EWG recommends the prohibition of selling and movement of the plant. These measures, in combination with management plans for early warning; obligation to report findings, eradication and containment plans, and public awareness campaigns should be implemented.

#### Containment and control of the species in the PRA area

Eradication measures should be promoted where feasible with a planned strategy to include surveillance, containment, treatment and follow-up measures to assess the success of such actions. As highlighted by EPPO (2012), regional cooperation is essential to promote phytosanitary measures and information exchange in identification and management methods. Eradication may only be feasible in the initial stages of infestation, and this should be a priority.

General considerations should be taken into account for all potential pathways, where, as detailed in EPPO (2014), these measures should involve awareness raising, monitoring, containment and eradication measures. NPPOs should facilitate collaboration with all sectors to enable early identification including education measures to promote citizen science and linking with universities, land managers and government departments.

#### **Natural spread** (method of spread within the EPPO region):

Increase surveillance in areas where there is a high risk the species may invade. NPPO's should provide land managers and stakeholders with identification guides and facilitate regional cooperation, including information on site specific studies of the plant, control techniques and management.

At the plot scale, it is technically possible to achieve total control of *A. trifida* by a combination of chemical and mechanical weed control and agronomic practices. Currently, the development of resistance to herbicides, particularly to ALS-inhibitors and glyphosate, is reducing the effectiveness of control (Heap, 2017). Moreover, supplementary mechanical management is not really feasible on a large scale. At the regional scale, it is likely that the spread cannot be reliably prevented, as shown by the progression of *A. trifida* on the North American continent (Royer and Dickinson 1999).

There is not presently any programme for eradicating *A. trifida* on the scale of the PRA area. *Ambrosia trifida* is present in various environments, including natural moist environments (riverbanks) where implementation of an eradication programme is very difficult. The large size and morphological characteristics of individuals make identification of this species very easy, allowing early detection of any new incursion, which can help the rapid implementation of local eradication schemes.

As a problematic weed of crops, control measures for *A. trifida* seem very likely. While the use of preemergence (e.g. imazaquin) and/or post-emergence (e.g. dicamba and 2,4-D) herbicides allows effective control of the species on the scale of the agricultural plot (Soltani *et al.*, 2011; Vink *et al.*, 2012), this is rarely total (Soltani *et al.*, 2011). Moreover, many cases of resistance to herbicides have been reported in the area of origin (Heap, 2017; Regnier *et al.* 2016; Vink *et al.*, 2012). Bearing in mind the substantial trade of GMO soybean seeds from the USA to EPPO countries, although no glyphosate-resistant populations of *A. trifida* have been registered so far, it can be expected that glyphosate resistance will also become a problem in the EPPO region. Control then becomes more difficult to implement and requires a combination of tillage and pre- and post-emergence herbicide treatments to reduce the density of *A. trifida* at the beginning of the season, which would seem to provide an integrated approach for effective management of the species (Ganie *et al.*, 2017). Tillage helps reduce the development of a population in an agricultural plot for a given year, but is not intended to eradicate the species. Furthermore, as *A. trifida* is capable of establishing a soil seedbank for 4 to 21 years, depending on seed burial depth (Harrison *et al.*, 2007; Stoller and Wax 1974, Toole and Brown 1946) with germination spread over time (Abul Fatih and Bazzaz, 1979), and of occupying non-agricultural habitats, it is likely, with a low uncertainty, that the conventional control measures considered will not be able to completely eradicate the species. Adaptation of the crop rotation system is recommended e.g. inclusion of winter cereals, autumn seeded cover crops and perennial pasture and hay crops, avoidance of sunflower and soybean (Regnier *et al.*,2016).

While eradication seems difficult, containment measures could help curb the invasion of the species within the PRA area. Limiting the local development of a population can be achieved by the use of herbicides (Soltani *et al.*, 2011), at least where this is possible, or by grubbing-up. Nevertheless, effective containment requires rapid detection and measures to prevent the dispersal of the species. There is no coherent surveillance system enabling early detection of invasion outbreaks on the scale of the PRA area. On cultivated land, early detection followed by rapid reasoned intervention can effectively contain a new outbreak. In addition, the species can be spread by land transport and agricultural machinery, via contaminated seed lots, or by flooding along water courses. These pathways are difficult to control: it therefore appears only moderately likely that the pest can be contained if an outbreak occurs in the PRA area.

#### 18. Uncertainty

Overall, the level of uncertainty associated with the PRA is low. The species is present within the PRA area, it has established in a number of EPPO countries and the species has a negative impact on agriculture. There is a high level of uncertainty whether traded crop seed can be contaminated with *A. trifida* as it is not clear how the species can become incorporated into seed lots given the low amount of contamination that is authorized for exporting such commodities. There is a low level of uncertainty associated with contamination of grain for animal feed and human consumption. There is a low level of uncertainty associated with likelihood of establishment in the natural and managed environments, and a moderate level of uncertainty for spread in the EPPO region. There is a moderate and high level of uncertainty for impacts on biodiversity and ecosystem services in the current area of distribution as there are few studies that have evaluated such impacts. This is the same for the potential impact of the species on biodiversity and ecosystem services in the PRA area.

There is uncertainty associated with the species distribution modelling because the potential distribution of range-expanding species is always difficult and uncertain. In this case study, uncertainty arises because:

- The models were constructed using convenient climate and habitat layers, which may not be the most appropriate for *A. trifida*. Specific predictors layers capturing requirements for different stages of the life cycle (e.g. for germination in spring or seed ripening in late summer) may have improved the predictions. Additionally, there was little ecophysiological information available to define the unsuitable background region.
- The selection of the background sample was weighted by the density of vascular plant records on the Global Biodiversity Information Facility (GBIF) to reduce spatial recording biases. 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 recording, especially because additional data sources to GBIF were used.
- The distribution databases included many casual occurrences from central Europe and western Russia, in places predicted to be climatically suitable. Therefore, the species may not always establish when introduced in apparently climatically suitable conditions. Local habitat, management and biotic factors not included in the model may play a role in this.

• A. trifida has a very large native range, across which locally adapted ecotypes are thought to occur. The model attempts map the suitability of the whole species, but locally adapted populations are likely to have a narrower niche. Additionally, Ambrosia species are known to be adaptable and may be able to expand their niche during invasion.

#### 19. Remarks

The EWG recommends that further studies be conducted on the effect of ecotypes on establishment in the PRA area. In the USA, there are ecotypes which are adapted to varying climatic conditions but information regarding the presence of ecotypes in the EPPO region is currently unavailable. In addition, the EWG recommend that surveys be conducted to determine the current establishment and spread of the species in the EPPO region along with surveys and inspection on the contamination of imported grain and seed from North America. It would also be of interest to evaluate the potential spread of the species through the contamination of seed and grain within the EPPO region and thus surveys and inspections could be carried out to assess this. The EWG consider that studies should be conducted on the emergence period of the species in the EPPO region along with studies on the cultural, mechanical and chemical control of the species.

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## Appendix 1 Comparison of Ambrosia species present within Europe

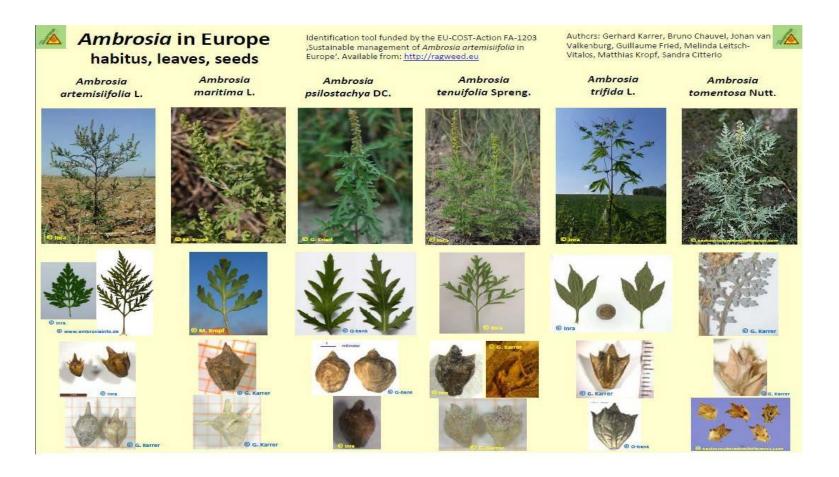


Figure 2. Comparison of six *Ambrosia* species traits in Europe (Taken from <a href="http://internationalragweedsociety.org/docs/Ambroisia">http://internationalragweedsociety.org/docs/Ambroisia</a> Europe GB.pdf).





# Six Ambrosia species in Europe comparison of traits

Species	A. artemisiifolia L.	A. maritima L.	A. psilostachya DC.	A. tenuifolia Spreng.	A. trifida L.	A. tomentosa Nutt.
Life form	Annual	Biennial, perennial	Perennial	Perennial	Annual	Perennial
Plant size (cm)	10 to 250	20-80	10 to 90	20-100	40 to 400	15-60
Belowground	Taproot	Taproot	Root sprouter	Root sprouter	Taproot	Root sprouter
Stem	+/- intensively branched, branches with wide angles	Intensively branched, stems lignified towards the base, sprouts from lower aerial stem buds	Few branches, with narrow angles	Few branches, with narrow angles	+/- intensively branched	Few subordinated branches
Leaves	Pinnatifid to bipinnate, rarely entire; leaf segments broadened and separated, rarely narrow; lower leaves with distinct narrow petiole; upper leaves alternate; long and short hairs mixed		Pinnatifid, rarely entire; leaf segments lineal and connected, often sharped towards the tip; +/- sessile; upper leaves alternate; dense short hairs	Bipinnate to pinnatifid; leaf segments as narrow as the rachis, lineal, connected; lower leaves with distinct narrow petiole; upper leaves alternate; dense short hairs		Pinnatifid, leaf segments narrow and rounded; middle and upper leaves alternate; extremely dense white hairs below; above greenish-grey
Diaspore (mm)	2-4; 1-seeded	3.5-5; 1-seeded	2-3; 1-seeded	2-3.5; 1-seeded	>6; 1-seeded	4-6; 2-seeded
Diaspore coat	Few hairs and glands; 2-5 short lateral spines with sharpened tips; dark brown	4-6 distinct lateral spines	Few glands and short hairs; blunt, short lateral spines few or none; dark brown	Short hairs and glands, 2-5 lateral short blunt spines; olive to dark brown	Glabrous or few hairs; 2-4 indistinct lateral spines; dark brown to black	
Reproductive mode	Sexual (seeds)	Sexual (seeds)	Mostly vegetative, rarely by seeds	Vegetative; very rarely by seeds	Sexual (seeds)	Vegetative, and sexual (seeds)
Smell (leaves)	None	Distinct, aromatic	Distinct	None	None	None
Origin	N-America	Europe, Africa	N-America	S-America	N-America	N-America
Habitat types (in Europe)	Field, riverbank, ruderal, roadside, orchard, pasture	Coastal dune	Sandy coast, riverbank, field, dry grassland, roadside, ruderal, orchard, vineyard		Field, sandy riverbank	Abandoned field, pasture
Distribution in Europe	Widespread; all Europe except Mediterranean and far North	Critically endangered, in many places extinct; formerly cultivated	Scattered; European lowlands and coastal areas, more frequent to the South	Scattered; Southern Europe	Scattered; Central and Southern Europe	Very rare; Spain
Allergenicity	High	Unknown	Medium	Unknown	High	Medium

# **Appendix 2. Relevant illustrative pictures (for information)**



Fig. 1. Ambrosia trifida (EPPO Global Database)



Fig 2. Ambrosia trifida invading a wheat crop in Russia (EPPO Global Database).



Fig 3. Ambrosia trifida invading a soybean field in the south west of France.



Fig 4. Ambrosia trifida flower (EPPO Global Database).



Fig 5. *Ambrosia trifida* flower (EPPO Global Database)



Fig 6. Ambrosia trifida burs

# Appendix 3: Projection of climatic suitability for *Ambrosia trifida* establishment in the EPPO region

Daniel Chapman, 25<sup>th</sup> February 2019

#### Aim

To project the climatic suitability for potential establishment of *Ambrosia trifida* in Europe and the Mediterranean region, under current and predicted future climatic conditions.

#### Data for modelling

Species occurrence data were obtained from the Global Biodiversity Information Facility (GBIF), EcoEngine, iNaturalist, the AMBROTRIF database (Follak, Dullinger, Kleinbauer, Moser, & Essl, 2013), published sources (Abramova, 2018; Stoyanov, Vladimirov, & Milanova, 2014) and databases of the Expert Working Group (EWG) performing the Pest Risk Assessment. With the EWG, the records were scrutinised to remove any considered too old (<1980), from regions where the species is not known to be established, that appeared to be dubious, where the georeferencing was too imprecise (e.g. records referenced to a country or island centroid) or outside of the coverage of the predictor layers (e.g. small island or coastal occurrences). This included removing records from the European countries classified as casual in the Pest Risk Assessment, records from the Moscow region of Russia and replacing records from France with known naturalised occurrences from the EWG's databases.

The remaining records were gridded at a 0.25 x 0.25 degree resolution for modelling (Figure 1a). This resulted in 766 grid cells containing records of *A. trifida* for the modelling (Figure 1a), which is a sufficient number for distribution modelling.

Based on the life history requirements of *A. trifida* and likely limiting factors for establishment in Europe, the following predictor variables were assembled on the same grid:

- Minimum temperature of the coldest month (Bio6 °C) from WorldClim v1 (Hijmans *et al.*, 2005), reflecting exposure to winter cold necessary for seed stratification (ref).
- <u>Precipitation of the coldest quarter (Bio19, mm, ln+1 transformed)</u> from WorldClim v1 (Hijmans *et al.*, 2005), reflecting moisture requirements during stratification.
- <u>Annual potential evapotranspitation (PET mm)</u> indicating the radiation and heat energy available for plant growth. PET was estimated following Zomer *et al* (2008).
- <u>Climatic moisture index (CMI, ln+1 transformed)</u> calculated as annual precipitation (Bio12 from Worldclim v1; Hijmans *et al.*, 2005) divided by PET and reflecting drought stress.
- <u>Human modification gradient (ln+1 transformed)</u> combining human settlement, agriculture, transportation, mining and energy production and electrical infrastructure (Kennedy, Oakleaf, Theobald, Baruch-Mordo, & Kiesecker, 2019). This was chosen to capture an association of *A. trifida* with anthropogenic habitats.
- <u>Urban cover (ln+1 transformed)</u> derived from GlobCover 2009 v2.3 urban class ("Artificial surfaces and associated areas (Urban areas >50%)") (Bontemps *et al.*, 2011).
- <u>Cropland cover (ln+1 transformed)</u> derived from GlobCover 2009 v2.3 cropland classes ("Post-flooding or irrigated croplands (or aquatic)", "Rainfed crops", "Mosaic cropland (50-70%) / vegetation (grassland/shrubland/forest) (20-50%)" and "Mosaic vegetation (grassland/shrubland/forest) (50-70%) / cropland (20-50%)") (Bontemps *et al.*, 2011).

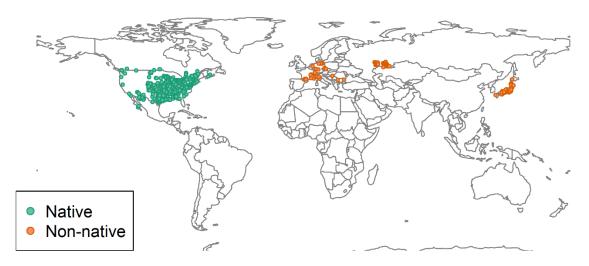
- <u>Preferred crop cover</u> (ln+1 transformed) derived from global maps of maize, soybean and sunflower harvested areas (Monfreda, Ramankutty, & Foley, 2008). Spring wheat could not also be included as the crop cover maps do not differentiate spring and winter wheat.
- <u>Maximum flow accumulation</u> (ln+1 transformed) (Domisch, Amatulli, & Jetz, 2015) as an indicator of the presence of wetlands and major river systems, which may provide suitable habitat for *A. trifida*.

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) 4.5 and 8.5 were also obtained. For both scenarios, 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 v1 baseline (see <a href="http://www.worldclim.org/cmip5.5m">http://www.worldclim.org/cmip5.5m</a>).

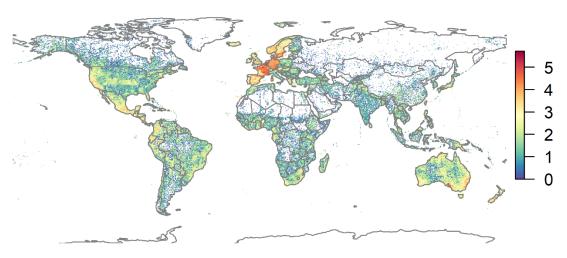
RCP 4.5 is a moderate climate change scenario in which CO<sub>2</sub> concentrations increase to approximately 575 ppm by the 2070s and then stabilise, resulting in a modelled global temperature rise of 1.8 °C by 2100. RCP8.5 is the most extreme of the RCP scenarios, and may therefore represent the worst case scenario for reasonably anticipated climate change. In RCP8.5 atmospheric CO<sub>2</sub> concentrations increase to approximately 850 ppm by the 2070s, resulting in a modelled global mean temperature rise of 3.7 °C by 2100.

Finally, the recording density of vascular plants (phylum Tracheophyta) on GBIF was obtained as a proxy for spatial recording effort bias (Figure 1b).

### (a) Occurrence records used in the modelling



# (b) Log10 Tracheophyta records from GBIF



**Figure 1.** (a) Occurrence records obtained for *Ambrosia trifida* and used in the modelling, showing the native range and (b) a proxy for recording effort – the number of vascular plant records held by the Global Biodiversity Information Facility, displayed on a log<sub>10</sub> scale.

#### **Species distribution model**

The modelling was based on a recent adaptation of standard presence-background (presence-only) ensemble distribution nmodelling approaches for emerging invasive non-native species (Chapman, Pescott, Roy, & Tanner, 2019). This attmepts to account for dispersal constraints on non-equilibrium invasive species' distributions (Elith, Kearney, & Phillips, 2010) by minimising the inclusion of locations suitable for the species but where it has not been able to disperse to. To do this background samples (pseudo-absences) were sampled from two distinct background regions:

A presence-background (presence-only) ensemble modelling strategy was employed using the BIOMOD2 R package v3.3-7 (Thuiller, Georges, Engler, & Breiner, 2016; Thuiller, Lafourcade, Engler, & Araújo, 2009). Because invasive species' distributions are not at equilibrium and subject to dispersal constraints at

a global scale (Elith *et al.*, 2010), we took care to minimise the inclusion of locations suitable for the species but where it has not been able to disperse to. Therefore background samples (pseudo-absences) were sampled from two distinct regions (Figure 2):

- An accessible background includes places close to A. trifida populations, in which the species is likely to have had sufficient time to disperse and sample the range of environments. The accessible background was defined as a 400 km buffer around the native range (minimum convex polygon bounding native occurrences) and a 30 km buffer around non-native occurrences (capturing a 4-cell neighbourhood of the non-native occurrences). Sampling was more restrictive from the invaded range to account for stronger dispersal constraint over a shorter residence time.
- An <u>unsuitable background</u> includes places expected to be unsuitable for the species and in which absence is irrespective of dispersal constraints. No specific ecophysiological information was available to define the unsuitable region, but based on the likely limits on *A. trifida* occurrence in Europe and the extreme values of the predictors at the species occurrences, unsuitability was defined as:
  - o Minimum temperature of the coldest month (Bio6) > 7 °C (presumed too warm for seed stratification), OR
  - Precipitation of the coldest quarter (Bio19) < 25 mm (presumed too dry for seed stratification),</li>
     OR
  - Potential evapotranspiration (PET) < 660 mm (presumed too cold for growth and seed maturation), OR
  - o Climatic moisture index < 0.2 (presumed too dry for growth), OR
  - Human modification gradient < 5% (presumed to undisturbed for occurrence)

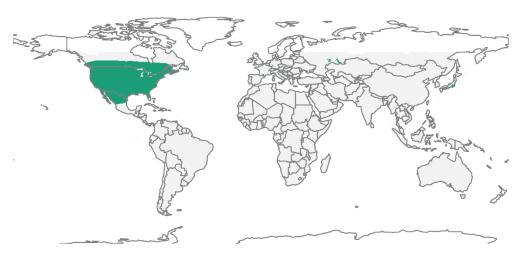
Twenty three of the occurrences (3%) fell in the unsuitable background.

For modelling, 10 random background samples were obtained:

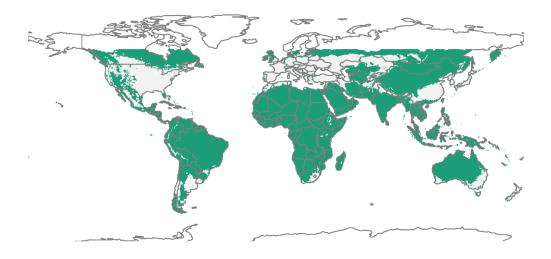
- From the accessible background 766 samples were drawn, which is the same number as the occurrences. Sampling was performed with similar recording bias as the distribution data using the target group approach (S. J. Phillips, 2009). In this, sampling of background grid cells was weighted in proportion to GBIF recording density (Figure 1b). Taking the same number of background samples as occurrences ensured the background sample had the same level of bias as the data.
- From the unsuitable background 5000 simple random samples were taken. Sampling was not adjusted for recording biases as we are confident of absence from these regions.

Model testing on other datasets has shown that this method is not overly sensitive to the choice of buffer radius for the accessible background or the number of unsuitable background samples.

### (a) Accessible background



# (b) Unsuitable background



**Figure 2.** The background regions from which 'pseudo-absences' were sampled for modelling. (a) The accessible background is assumed to represent the range of environments the species has had chance to sample. (b) The unsuitable background is assumed to be environmentally unsuitable for the species. Note that predictor coverage did not extend to beyond 60° N, so this region is excluded from the modelling.

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, seven statistical algorithms were fitted with the default BIOMOD2 settings (except where specified below) and rescaled using logistic regression:

- Generalised linear model (GLM)
- Generalised boosting model (GBM)
- Generalised additive model (GAM) with a maximum of four degrees of freedom per effect.
- Artificial neural network (ANN)

- Multivariate adaptive regression splines (MARS)
- Random forest (RF)
- Maxent (Steven J Phillips, Dudík, Dudík, & Phillips, 2008)

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. Normalised variable importance was 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, which were reserved from model fitting. AUC is the probability that a randomly selected presence has a higher model-predicted suitability than a randomly selected pseudo-absence.

An ensemble model was created by first rejecting poorly performing algorithms with relatively extreme low AUC values and then averaging the predictions of the remaining algorithms, weighted by their AUC. To identify poorly performing algorithms, AUC values were converted into modified z-scores based on their difference to the median and the median absolute deviation across all algorithms (Iglewicz & Hoaglin, 1993). Algorithms with z < -2 were rejected. In this way, ensemble projections were made for each dataset and then averaged to give an overall suitability.

Global model projections were made for the current climate and for the two climate change scenarios, avoiding model extrapolation beyond the ranges of the input variables. The optimal threshold for partitioning the ensemble predictions into suitable and unsuitable regions was determined as that equalising sensitivity and specificity.

Limiting factor maps were produced following Elith *et al.* (2010). Projections were made separately with each individual variable fixed at a near-optimal value. These were chosen as the median values at the occurrence grid cells. Then, the most strongly limiting factors were identified as the one resulting in the highest increase in suitability in each grid cell. Partial response plots were also produced by predicting suitability across the range of each predictor, with other variables held at near-optimal values.

#### **Results**

The ensemble model suggested that suitability for *A. trifida* at the global scale and resolution of the model was most strongly determined by winter temperature (Bio6), winter precipitation (Bio19), energy availability (PET) and moisture availability (CMI) (Table 1, Figure 3). There were also appreciable effects of human modification and the distribution of preferred crop types.

Global projection of the ensemble model in current climatic conditions indicates that the native and known invaded records in Europe and Asia generally fell within regions predicted to have high suitability (Figure 4). The main exception was non-native occurrences in northeast Germany, which were predicted unsuitable for *A. trifida* establishment and about which there is some uncertainty over their established status.

Across Europe and the Mediterranean region, the model predicts a large climatically suitable range spanning most of Europe below ~55 °N, excluding the Mediterranean coastline, and generally increasing in suitability towards the more continental east. *Ambrosia trifida* is currently absent, recorded sproadically or unrecorded from the central part of this distribution (see Figure 1), suggesting a potential for much wider establishment.

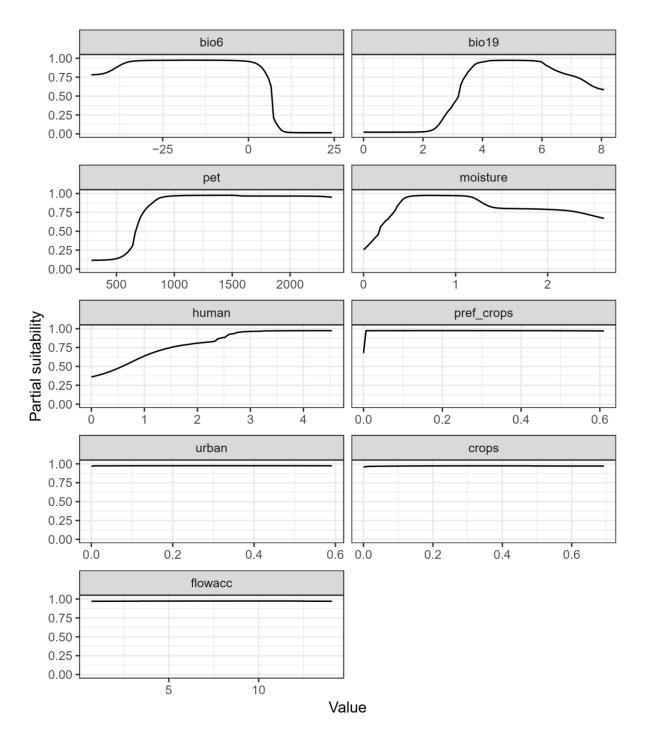
The model suggests that establishment in northern Europe will be limited by low energy availability (low PET), while warm winters mainly limit establishment around the Mediterranean and Black Sea coastlines (Figure 6). Drought stress (low CMI) was suggested to be the most important limiting factor in the driest parts of southern Russia and in Turkey.

Predictions of the model for the 2070s, under the moderate RCP4.5 and extreme RCP8.5 climate change scenarios, suggest relatively small changes in suitability (Figure 7-8). A small northwards range expansion is predicted in Russia, presumably driven by warmer summer temperatures. A small reduction in suitability in Mediterranean regions is also predicted for the more extreme climate scenario, presumably reflecting drying out and warmer winters.

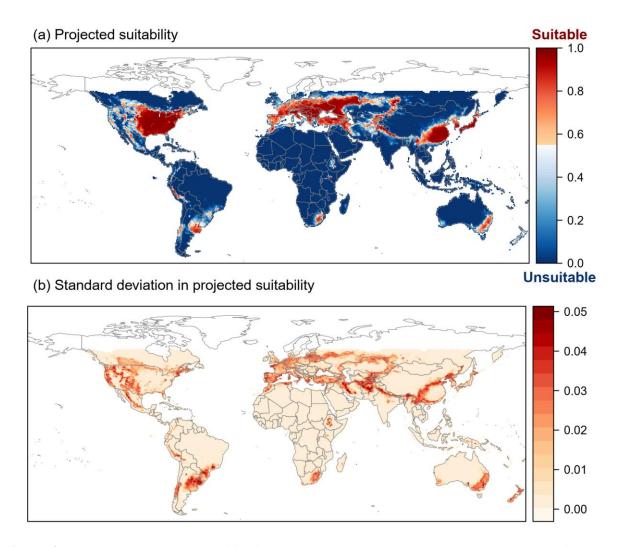
These results are reflected in the suitability of different European Biogeographical Regions (Bundesamt fur Naturschutz (BfN), 2003) (Figure 9). The most suitable regions under current and future climate scenarios are the Pannonian, Anatolian, Continental, Steppic and Black Sea and Alpine.

**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 algorithms). Results are the average from models fitted to ten different background samples of the data.

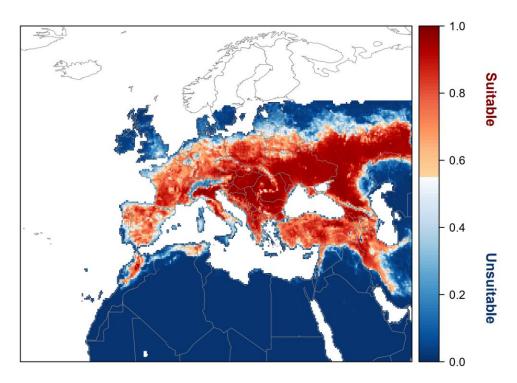
			Variable importance								
Algorithm	AUC	In the ensemble	Minimum temperature of coldest month	Precipitation of coldest quarter	Potential evapotranspiration	Climatic moisture index	Human modification	Cropland cover	Preferred crop cover	Urban cover	Flow accumulation
GLM	0.969	yes	43%	15%	18%	12%	9%	2%	0%	0%	0%
GAM	0.971	yes	41%	13%	19%	14%	9%	2%	0%	0%	0%
ANN	0.972	yes	35%	18%	21%	16%	6%	2%	0%	0%	0%
GBM	0.972	yes	41%	20%	7%	6%	5%	1%	15%	5%	0%
MARS	0.966	no	45%	17%	22%	6%	4%	2%	4%	0%	0%
RF	0.973	yes	37%	18%	7%	7%	6%	1%	19%	5%	1%
Maxent	0.961	yes	37%	16%	14%	10%	5%	3%	10%	3%	1%
Ensemble	0.975		39%	17%	15%	11%	7%	2%	7%	2%	0%



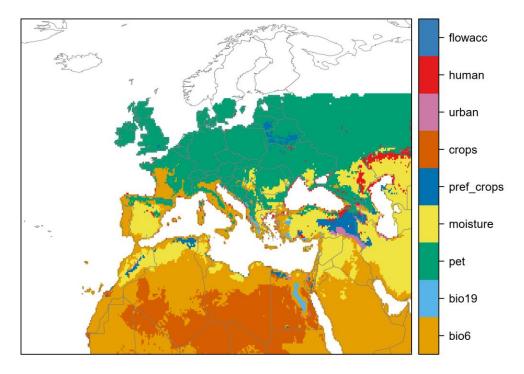
**Figure 3.** Partial response plots from the ensemble model, ordered from most to least important. In each plot, other model variables are held at their median value in the training data. Variable codes: bio6 = minimum temperature of coldest month; bio19 = precipitation of coldest quarter (ln+1); pet = potential evapotranspiration; moisture = climatic moisture index (ln+1); human = human modification gradient (ln+1);  $pref\_crops = cover$  of preferred  $pref\_crops = cover$  of  $pref\_crops = cover$  of



**Figure 4.** (a) Projected global suitability for *Ambrosia trifida* establishment in the current climate. For visualisation, the projection has been aggregated to a 0.5 x 0.5 degree resolution, by taking the maximum suitability of constituent higher resolution grid cells. Red shading indicates suitability, according to the selected threshold. White areas have climatic conditions outside the range of the training data so were excluded from the projection. (b) Uncertainty in the suitability projections, expressed as the standard deviation of projections from different algorithms in the ensemble model.

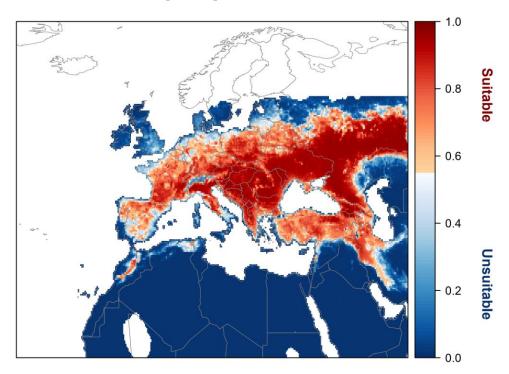


**Figure 5.** Projected current suitability for *Ambrosia trifida* establishment in Europe and the Mediterranean region. The white areas have climatic conditions outside the range of the training data so were excluded from the projection.

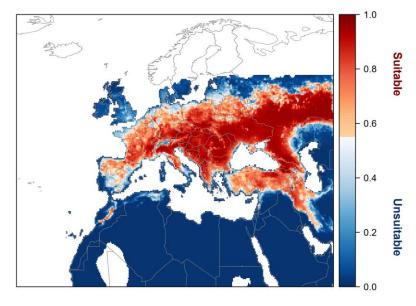


**Figure 6.** Limiting factor map for *Ambrosia trifida* establishment in Europe and the Mediterranean region in the current climate. Shading shows the predictor variable most strongly limiting projected suitability.

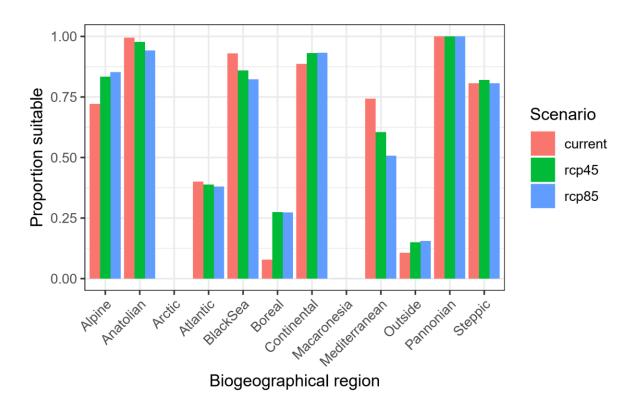
Axis bio19 = precipitation of coldest quarter (ln+1); pet = potential evapotranspiration; moisture = climatic moisture index (ln+1); human = human modification gradient (ln+1); pref\_crops = cover of preferred crops (ln+1); urban = urban cover (ln+1); crops = cropland cover (ln+1); flowacc = flow accumulation (ln+1).

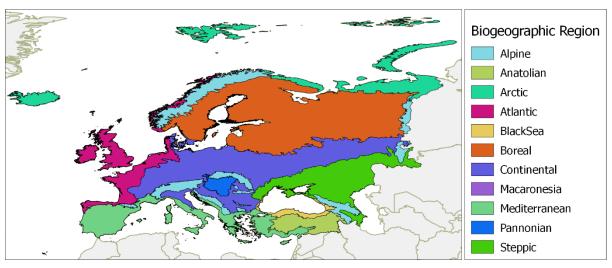


**Figure 7.** Projected suitability for *Ambrosia trifida* establishment in Europe and the Mediterranean region in the 2070s under climate change scenario RCP4.5, equivalent to Figure 5.



**Figure 8.** Projected suitability for *Ambrosia trifida* establishment in Europe and the Mediterranean region in the 2070s under climate change scenario RCP8.5, equivalent to Figure 5.





**Figure 9.** Variation in projected suitability among Biogeographical regions of Europe (Bundesamt fur Naturschutz (BfN), 2003). The bar plots show the proportion of grid cells in each region classified as suitable in the current climate and projected climate for the 2070s under emissions scenarios RCP4.5 and RCP8.5. The coverage of each region is shown in the map below.

#### **Caveats to the modelling**

Modelling the potential distributions of range-expanding species is always difficult and uncertain. In this case study, uncertainty arises because:

- The models were constructed using convenient climate and habitat layers, which may not be the most appropriate for *A. trifida*. Specific predictors layers capturing requirements for different stages of the life cycle (e.g. for germination in spring or seed ripening in late summer) may have improved the predictions. Additionally, there was little ecophysiological information available to define the unsuitable background region.
- The selection of the background sample was weighted by the density of vascular plant records on the Global Biodiversity Information Facility (GBIF) to reduce spatial recording biases. 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 recording, especially because additional data sources to GBIF were used.
- The distribution databases included many casual occurrences from central Europe and western Russia, in places predicted to be climatically suitable. Therefore, the species may not always establish when introduced in apparently climatically suitable conditions. Local habitat, management and biotic factors not included in the model may play a role in this.
- A. trifida has a very large native range, across which locally adapted ecotypes are thought to occur. The model attempts map the suitability of the whole species, but locally adapted populations are likely to have a narrower niche. Additionally, Ambrosia species are known to be adaptable and may be able to expand their niche during invasion.

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# Appendix 4 Distribution of Ambrosia trifida data used for the modelling



Figure 1. Global distribution data for Ambrosia trifida used in the modelling

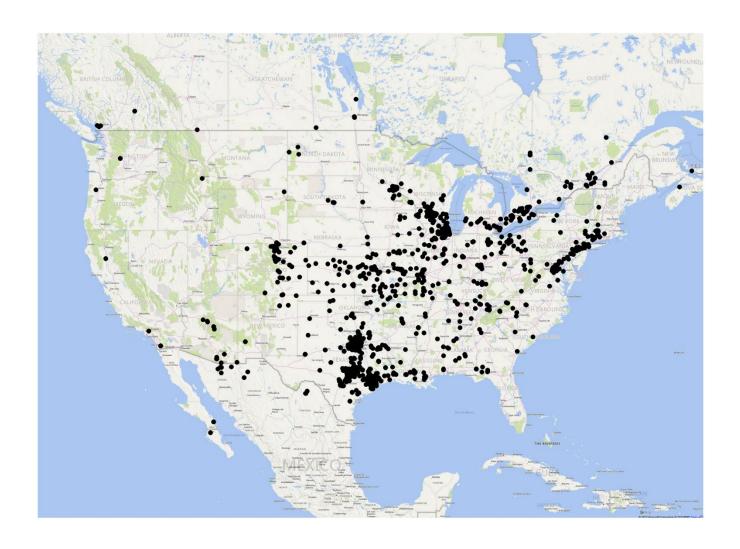


Figure 2. Distribution of Ambrosia trifida data for North America used in the modelling.

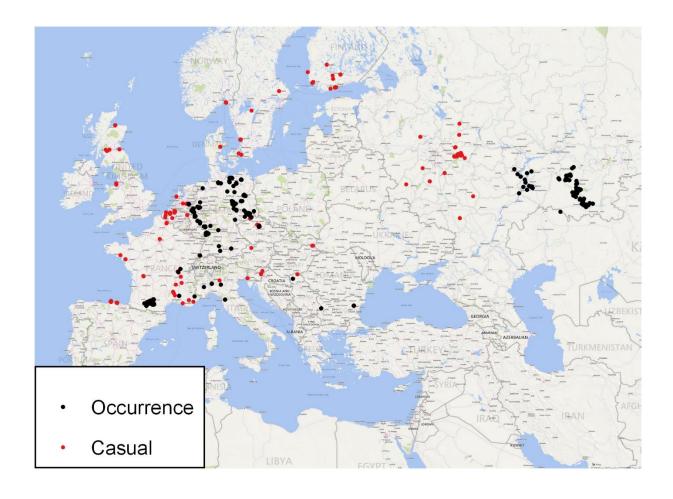


Figure 3. Distribution of *Ambrosia trifida* in the EPPO region showing casual and established populations. Established data were used for the modelling of the species.

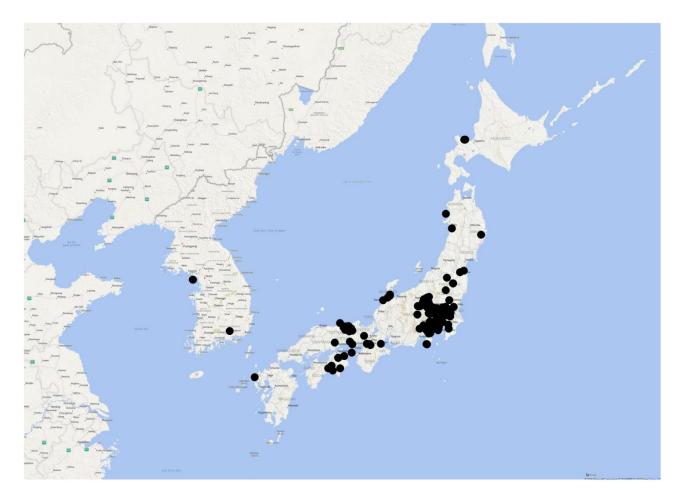
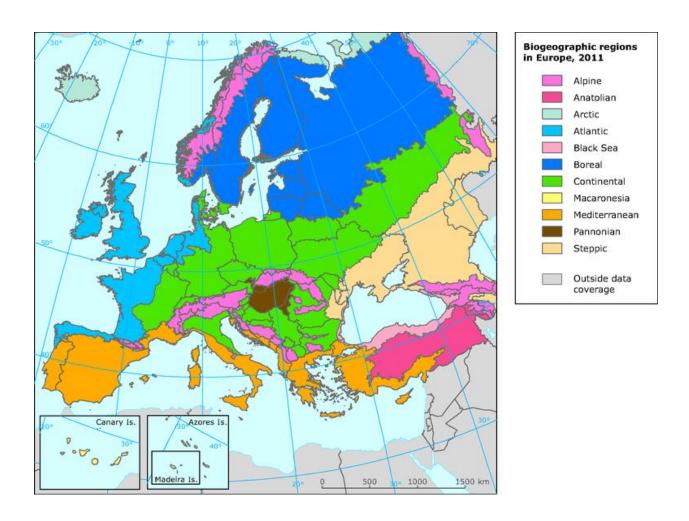


Figure 4. Distribution of *Ambrosia trifida* in Asia used in the modelling..

# Appendix 5 Biogeographical regions in Europe



# Appendix 6: Imports of seed of crops that may be contaminated by A. trifida from USA into the EPPO region (Data from FAO Stats)

**Table 1. Maize seed for planting imports into EPPO countries from the USA from 2015-2018.** The following commodities have been combined (Corn SD Other (HS code: 1005100090), Corn SD Yellow (HS code 1005100010), Sweet Corn SD (HS code: 712908550)). The data for 2018 is from Jan-Nov. Figures detail in metric tonnes per year.

Country	2015	2016	2017	2018
Albania	0	40.2	0	18.6
Algeria	0	5.9	119.9	0
Austria	52.6	67	0	221
Belgium	0.1	19.6	105.5	111.8
Croatia	2.4	3	0	0.2
Cyprus	0	0	4.5	54.3
Denmark	0	0.2	0	0.7
Finland	0.9	0	0	0
France	2848.4	2586.5	3269.5	2028.7
Germany	77	109.7	126.7	139.4
Greece	44.1	164.3	22.8	99.1
Hungary	155.2	103.4	86.6	84.5
Ireland	4.6	0	0	0
Israel	35	52.3	87.4	66.5
Italy	674.1	1123.1	693.3	485.5
Jordan	91.2	18.9	26.8	24.4
Kazakhstan	0	0.9	7	102
Kyrgyzstan	0.4	1.1	0.4	0.1
Morocco	0	0	0	2.5
Netherlands	844.2	372.5	232	308.5
Poland	0	0	40	0
Portugal	0	15	11.4	1.1
Romania	5.4	0.7	0	2.1
Russia	0	0	0	5.8
Serbia	1.6	1.2	2.2	4.2
Spain	2059.5	407	132.6	62.1
Switzerland	1.8	9.1	0	0
Turkey	236.2	133.9	103.2	72.2
Ukraine	18.3	14.3	29.2	152.2
United Kingdom	294.2	216.1	354.2	380
Uzbekistan, Republic of	3.6	5.8	6.9	1.3

**Table 2. Sorghum seed for planting imports into EPPO countries from the USA from 2015-2018.** The following commodities have been combined (Sorghum seed (HS code: 1007100000) and Sorghum/Sudan SD (HS code: 1209299150)). The data for 2018 is from Jan-Nov. Figures detail in metric tonnes per year.

Country	2015	2016	2017	2018
Algeria	641.9	1092.1	599.3	293.6
Austria	4.7	0	0.8	0
Cyprus	14	0	0	0
France	1785.3	215.1	329.2	377.2
Germany	279.9	536.9	102.3	47.5
Greece	119	118	118	72
Hungary	236.6	555.2	287.4	0
Israel	38.8	0.8	0	0
Italy	1513.4	417.7	1021.2	1379
Jordan	0	0	3	0
Kazakhstan	0	0	25.2	0
Morocco	79.1	239.9	197.5	38.8
Netherlands	0	4.1	359.5	60
Poland	0	0	20	32.5
Portugal	10	134	115	130
Romania	0	0	17	39.2
Russia	79.3	327.6	390	589
Slovenia	0	0	20	0
Spain	640.8	267.7	202.2	281.1
Tunisia	551	357	408.5	95
Turkey	434	299.2	237.5	356
Ukraine	101.5	667.5	733	334.7
United Kingdom	36	24	24	0

Table 3. Soybean seed (HS code: 1201100000) for planting imports into EPPO countries from the USA from 2015-2018. The data for 2018 is from Jan-Nov. Figures detail in metric tonnes per year.

Country	2015	2016	2017	2018
Austria	0	2.8	268.8	232
Finland	5.3	0	0	0
France	0	13.2	183.5	196.4
Germany	435.4	450.9	20.7	15.6
Israel	0	0	14	0
Italy	11261.5	12476.4	12868.4	10109.1
Malta	0	0	5.8	0
Netherlands	10.6	0	9.7	155
Poland	29.2	0	0	0
Portugal	49.1	0	0	0
Romania	1269.4	6572.5	1761.3	161.5
Spain	0	0	0	37
Switzerland	0	89	110.3	0
Ukraine	40	0	0	0
<b>United Kingdom</b>	0	41.9	11.7	15.8

Table 4. Sunflower seed (HS code: 1206000031) for planting imports into EPPO countries from the USA from 2015-2018. The data for 2018 is from Jan-Nov. Figures detail in metric tonnes per year.

Country	2015	2016	2017	2018
Austria	663.6	4165.7	7045.1	5948
Belgium	3209.3	5399.1	4286.3	2990.6
Denmark	9.5	0	14.7	1.2
France	12477.4	3425	1864.4	7570.4
Germany	774.6	127.6	215.6	282.2
Hungary	1072.2	194.7	255.4	692
Italy	1526.5	1057.4	67.7	98.7
Jordan	19.4	0	0	0
Kazakhstan	44.3	0	0	0
Netherlands	12	20.7	0	0.4
Romania	1433.3	1062.9	777.1	487.8
Russia	1882.7	206.6	657.2	1394.7
Serbia	3.1	0	0	0
Spain	485.4	14.3	0	0
Turkey	87.2	69.6	84	71.3
Ukraine	860.6	132.9	2260.8	5720.7
United Kingdom	0.3	0	1.6	1.7

# Appendix 7 Grain imports from USA into the EPPO region

**Table 1. Imports of soybean grain into EPPO countries from the USA from 2015-2018.** The following commodities have been combined (Soybean (other) HS code: 1201900095), Soybean seeds of a kind used as oil stock HS code: 1201900005). The data for 2018 is from Jan-Nov. Figures detail in metric tonnes per year.

Country	2015	2016	2017	2018
Azerbaijan	0	0	0	10493
Finland	333	234	273	272
France	104165	272466	64900	182732
Germany	2191796	1308642.3	1314686	901860
Greece	0	17000	14114	57038
Ireland	0	2600	4637	0
Israel	73	74141	79454	119956.1
Italy(	50089.7	201452	75523	881304
Lithuania	0	0	0	2.9
Morocco	109222	66092	55722	39785
Netherlands	1119010	1909165	2045877	3784707.2
Poland	1453	0	105	30000
Portugal	197565	57812	123156	472551
Romania	67822	0	0	113477
Russia	510507	155547	0	0
Spain	1041898	895232	607995	1812908.1
Tunisia	152036	362771	221094	448182
Turkey	509695.8	157369	368627	240078
Ukraine	20	232	120	47
United Kingdom	200185	229897	100	326894.5

**Table 2.** Imports of maize grain into EPPO countries from the USA from 2015-2018. The following commodities have been combined (HS Code: 1005902045 No. 4 corn X SD, HS code: 1005904055 corn white EX SD, HS code: 1005904065 corn NES, 1005902020 No. 1 Corn EX SD, HS Code: 1005902035, No. 3 corn, EX SD). The data for 2018 is from Jan-Nov. Figures detail in metric tonnes per year.

Country	2015	2016	2017	2018
Algeria	238846	678575	75373	47627
Austria	0	3396	0	0
France	0	799	19	0
Germany	0	743	343	0
Greece	0	0	0	81
Ireland	61322	280515	140149	111
Israel	16180	387811	107459	814810
Italy	0	19	27816	29502
Jordan	80441	61778	155984	38
Lithuania	0	0	0	42
Morocco	268286	772927	575272	822679
Netherlands	0	84457	210197	439800
Norway	0	0	0	47
Poland	0	0	0	51
Portugal	152089	109026	118335	227473
Romania	0	0	0	0
Russia	1313	0	0	0
Spain	66299	85079	185613	1167083
Tunisia	38189	177691	20000	451707
Turkey	13199	2679	80	585
Ukraine	0	0	42	0
United Kingdom	293	43851	434	19888