SCIENTIFIC OPINION

Risk to plant health in the EU territory of the intentional release of the bud-galling wasp *Trichilogaster acaciaelongifoliae* for the control of the invasive alien plant *Acacia longifolia*[1]

EFSA Panel on Plant Health (PLH)[2,3]

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ABSTRACT

The EFSA Panel on Plant Health was requested by the European Commission to assess the risk to plant health in the European Union if the Australian bud-galling wasp *Trichilogaster acaciaelongifoliae* was released for the control of the invasive alien plant *Acacia longifolia* in Portugal. *T. acaciaelongifoliae* feeds on *A. longifolia* and *A. floribunda*. In South Africa, following its intentional introduction in 1982 and 1983, the wasp is now present throughout the range of *A. longifolia* in that country, with most plants showing galls and seed set reductions of, initially, up to 95 %. Climatic conditions in the EU are largely suitable for establishment wherever *A. longifolia* and *A. floribunda* are present. *T. acaciaelongifoliae* is moderately likely to establish and spread in the EU, by natural means, but particularly if it is intentionally moved to control populations of *A. longifolia* other than those present in Portugal. The effects on native biodiversity and ecosystems resulting from invasive populations of *A. longifolia* are likely to be reduced by the wasp. *A. longifolia* is grown as an ornamental plant in some EU countries. *A. floribunda* is not an invasive plant in the EU and is cultivated as an ornamental plant on a small scale in France, Greece and Italy. Any effects on cultivated ornamental *A. longifolia* and *A. floribunda* are rated as moderate, although likely to be transient, as the industry could switch to the cultivation of other *Acacia* spp. For plant species other than *A. longifolia* and *A. floribunda*, consequences are expected to be minor, with low uncertainty except for *A. retinodes* and *Cytisus striatus*, where further investigation is required. No risk-reducing options in the plant health context are considered necessary, except for monitoring, sentinel planting, and care with regard to quarantine facilities and release protocols to prevent accidental release in situations and locations other than those intended.

KEY WORDS

beneficial organisms, biological control, bud-galling wasps, invasive *Acacia* spp., ornamental industry, plant health, *Trichilogaster acaciaelongifoliae*

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SUMMARY

Portugal recently informed the Commission that it is investigating the possibility of using the alien bud-galling wasp *Trichilogaster acaciaelongifoliae* Froggatt to control *Acacia longifolia* (Andrews) Willd. in its territory, since this plant is negatively affecting local biodiversity in coastal sand dunes and a variety of other habitats. *T. acaciaelongifoliae* is currently not a regulated harmful organism in the European Union and it is also not known to occur in the EU. However, it is an organism likely to be injurious to plants in the EU and is therefore subject to plant health regulation. Therefore, following discussion at the Standing Committee on Plant Health, the Member States and the Commission agreed to seek an advice from the European Food Safety Authority (EFSA) on the risks to plant health in the EU that such a release could pose.

Accordingly, this opinion presents an assessment of the risk to plant health in the EU territory posed by the intentional release of *T. acaciaelongifoliae* for the biological control of the invasive alien plant *A. longifolia*. The assessment specifically excludes the probability of entry and systematic evaluation of risk reduction options, and focuses on the probability of establishment, spread and impact in the EU territory.

This categorisation of a biological control agent (BCA) assesses all those characteristics of the organism observed outside the risk assessment area and useful to the completion of the BCA risk assessment. Essentially, this BCA risk assessment follows the pest risk assessment process outlined by the EFSA Panel on Plant Health (PLH) in 2010, with the substitution of the term “BCA” for “pest”. All consequences of release are evaluated, but no attempt is made to balance the potential positive and negative impacts.

An extensive literature search on *T. acaciaelongifoliae* and invasive alien *Acacia* spp. was conducted at the beginning of the mandate using CAB Abstracts, AGRIS, Scopus and Zoological Records, as well as a wide variety of websites, databases, Google, Google Scholar and other sources of information. The keywords used were “*Trichilogaster acaciaelongifoliae*”, “invasive *Acacia*” and numerous variants of these basic search terms. Further references and information were obtained from experts and from citations within the references.

Data from host range tests for *T. acaciaelongifoliae* were provided, upon request, by Helia Marchante, University of Coimbra, Portugal, the author of the application for release made to the Portuguese authorities. Data on the presence in the MSs of *Acacia* spp., both in the wild and cultivated as ornamental plants, were obtained through procurement from Stichting Dienst Landbouwkundig Onderzoek.

A technical hearing was held in September 2014 with three external experts: Helia Marchante, Richard Shaw (CABI, UK) and Andrea Allavena (Unità di Ricerca per la Floricoltura e le Specie Ornamentali, Consiglio per la Ricerca e la Sperimentazione in Agricoltura, Sanremo, Liguria, Italy). These experts covered the following areas of expertise, respectively: the proposed release of *T. acaciaelongifoliae* in Portugal, international regulations concerning the release of BCAs and the cultivation of ornamental *Acacia* spp. in Italy.

T. acaciaelongifoliae is native to Australia where it is restricted to the hosts *A. longifolia* and *Acacia floribunda* (Vent.) Willd. It was released intentionally in South Africa in 1982 and 1983 as a BCA for *A. longifolia* and has successfully established and spread there, with the majority of plants showing galls. Seed set on affected hosts is reduced by up to 95 %. In South Africa, spill-over to two other hosts (*Acacia melanoxylon* R. Br. and *Paraserianthes lophantha* (Willd.) I. C. Nielsen) was observed, but both are sub-optimal hosts and galls now only form sporadically with negligible effects on these hosts. The climate in the target area is likely to be largely suitable for the BCA.

The probability of establishment in the target area, after a release programme in Portugal, was rated as moderately likely because of the following:

- In the release area, by definition, host plants are present.
- The organism has been successfully established in South Africa, outside of its native range (Australia).
- In the proposed release area, the environmental conditions are similar to those in the native area of the wasp.
- There is the intention to make the release programme succeed, including the possibility of multiple releases. If release attempts are repeated often enough, the likelihood of establishment would increase to the level of likely.
- From the initial release sites in the *A. longifolia* infested dune areas in Portugal, the organism is likely to colonise the whole of the target dune area (based upon the previous experience in South Africa).
- The probability of establishment of a founder population depends on the ability to match the wasp's life history with the host's phenology in the northern hemisphere; specifically, the host must have suitably sized buds in the three days available for the wasps to find them.

Uncertainty was rated as medium as, generally, there is excellent information on all relevant aspects of the biology of the agent and its establishment in South Africa. However, there is no prior experience with its establishment in the northern hemisphere.

The probability of natural spread and subsequent establishment outside of the target area was rated as moderately likely as:

- Active dispersal is only possible over short distances; beyond a certain distance, dispersal can only be wind assisted.
- For effective dispersal, a suitable host must be found within three days of the emergence of the adults from the galls.
- Host populations are often fragmented, requiring long-distance dispersal (jumps).
- The probability of natural spread is a function of the source population size.

However, where there is close proximity of hosts in, for example, northern Portugal and Galicia, then spread is rated as likely.

Uncertainty was rated as medium as there is little information on wind-assisted dispersal.

The probability of human-assisted spread and subsequent establishment outside of the target area was rated as moderately likely as:

- Experience in South Africa has shown that intentional redistribution of the galls at the right time in a release programme is a very effective mode of spread of the organism.
- It cannot be ruled out that people would want to spread the BCA without due authorisation. If those persons were aware of the constraints imposed by the biology of the organism and were sufficiently expert, then such spread would likely be successful.
- Inadvertent spread associated with human movement is possible but is less likely to happen than with other organisms because of the constraints imposed by the biology of this organism.

- The trade in ornamental *A. longifolia* could enable the spread of the BCA.

Though not currently anticipated, authorised intentional movement outside of the target area would result in spread being likely with low uncertainty. If movement is intentional but not authorised, then the uncertainty is also low. With inadvertent movement, the likelihood of spread is low, and uncertainty is high because of the unpredictability of the process.

In conclusion, the risk assessment area is the area occupied or potentially occupied by wild or planted *A. longifolia* and *A. floribunda* in the EU territories. The probability of establishment in the target area is assessed as moderately likely (based on the previous experience in South Africa), with medium uncertainty (because of the switch between hemispheres). The probability of spread to the non-target area is assessed as:

- moderately likely for natural spread (because of fragmented host populations), with medium uncertainty because of little information on wind-assisted dispersal;

- moderately likely for human-assisted, intentional spread (based on the experience in South Africa), with low uncertainty, but unlikely for inadvertent movement (with high uncertainty).

The consequences of the release of the wasp on the invasive alien plant *A. longifolia* were rated as massive, as:

- the reproductive potential, vegetative growth and ultimately the population density of invasive alien *A. longifolia* would be reduced substantially;

- negative impacts of invasive alien *A. longifolia* on biodiversity and ecosystems would be reduced to a very significant extent;

- negative impacts of current control measures for invasive alien *A. longifolia* would be reduced substantially.

Uncertainty was rated as medium because of the unclear suitability of the climate to support high population densities of the BCA.

The consequences for commercial trade of cultivated *A. longifolia* and *A. floribunda* were rated as moderate, as:

- any use of these species would come under pressure from the BCA if it spreads to the areas of production;

- there is a trade in ornamental *A. longifolia* and *A. floribunda*, but the scale is limited compared with many other ornamental species (including other *Acacia* spp. that are not hosts of the BCA); these other ornamental species could be substituted for *A. longifolia*;

- amenity plantings are more likely to be affected than ornamentals in a dynamic production chain and trade.

Uncertainty was rated as medium as information on trade volumes and routes, and pest control in nurseries is missing.

For plant species other than *A. longifolia* and *A. floribunda*, consequences are expected to be minor, with low uncertainty except for *Acacia retinodes* Schltdl. and *Cytisus striatus* (Hill) Rothm., where further investigation is required.

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INTRODUCTION

1. Background and Terms of Reference

1.1. Background

The current European Union plant health regime is established by Council Directive 2000/29/EC on protective measures against the introduction into the Community of organisms harmful to plants or plant products and against their spread within the Community (OJ L 169, 10.7.2000, p.1).

The Directive lays down, amongst others, the technical phytosanitary provisions to be met by plants and plant products and the control checks to be carried out at the place of origin on plants and plant products destined for the Union or to be moved within the Union, the list of harmful organisms whose introduction into or spread within the Union is prohibited and the control measures to be carried out at the outer border of the Union on arrival of plants and plant products.

The long-leaved wattle *Acacia longifolia* (Andrews) Willd. is a leguminous shrub native to south-eastern Australia. It is a fast growing plant with a prolific production of seeds, which has been introduced in several parts of the world to curb erosion along costal dunes as well as an ornamental plant. It is described as an invasive species in several regions, including in the Union (Portugal). In South Africa, the Australian bud-galling wasp *Trichilogaster acaciaelongifoliae* Frogatt has been used (in combination with the seed-feeding weevil *Melanterius ventralis* Lea) with reported success as a biological control agent of *Acacia longifolia*.

Portugal recently informed the Commission that it is investigating the possibility of using *Trichilogaster acaciaelongifoliae* to control *Acacia longifolia* in its territory, since this plant is posing a substantial threat to local biodiversity in costal sand dunes and a variety of other habitats. *Trichilogaster acaciaelongifoliae* is currently not a regulated harmful organism in the Union and it is also not known to occur in there. However, this organism could be classified as a plant harmful organism that could be potentially listed in Directive 2000/29/EC if it would pose a threat to plants other than the target species *Acacia longifolia*, in particular native plants. Therefore, when the possibility of a voluntary release of *Trichilogaster acaciaelongifoliae* in Portugal was discussed at the Standing Committee on Plant Health, the Member States and the Commission agreed to seek an advice from the European Food Safety Authority (EFSA) on the risks to plant health in the Union that such a release could pose.

Portugal indicated that the two following scientific publications could be useful for EFSA's work:

- Assessing the suitability and safety of a well-known bud-galling wasp, *Trichilogaster acaciaelongifoliae*, for biological control of *Acacia longifolia* in Portugal. H. Marchante, H. Freitas and J.H. Hoffmann, Biological control 56 (2011) 193–201.

- Invasion of Portuguese dunes by *Acacia longifolia*: present status and perspectives for the future. Hélia Marchante, Doctoral Thesis, University of Coimbra, February 2011 (https://eg.sib.uc.pt/bitstream/10316/18181/1/HeliaMarchante %20PhD %20thesis.pdf)

1.2. Terms of reference

EFSA is requested, pursuant to Article 29(1) and Article 22(5) of Regulation (EC) No 178/2002, to assess the risk to plant health that would pose a voluntary release of the bud-galling wasp *Trichilogaster acaciaelongifoliae* Frogatt in the Union territory for the biological control of the invasive alien plant *Acacia longifolia* (Andrews) Willd. This pest risk assessment is to be conducted under the scenario assumption of a voluntary release of *Trichilogaster acaciaelongifoliae*. Therefore, it should focus on the risk of establishment, spread and impact for the EU territory, excluding the assessment of the probability of entry and a systematic evaluation of risk reduction options.

2. Interpretation of the terms of reference

The objectives of the risk assessment are to meet the terms of reference, as provided by the European Commission and stated in Section 1.2, concerning the release of the bud-galling wasp *Trichilogaster acaciaelongifoliae* for the biological control of the invasive alien host plant *Acacia longifolia*. The target plant population is considered to be *A. longifolia* in those regions of the European Union territory where it is invasive; however, to assess the risk to plant health more generally, the risks to this and other *Acacia* species grown and traded as ornamentals, or used in amenity plantings, were also considered. The environmental consequences of release were assessed in terms of the effects of biological control on ecosystem service provision and, particularly, in contributing to the restoration of native plant communities.

The approach taken is to characterise the biological control agent (BCA), as would normally be done in a pest categorisation, but focusing on the potential for establishment, spread and impact based on the experience gained in South Africa where the wasp was released at the beginning of the 1980s. The risk assessment part of the opinion then assesses the likelihood of establishment, spread and impact on target and non-target *Acacia* populations in the EU territory. We recognise the differing approaches used by Member States (MSs) and the EU with regard to invasive alien species, environmental health, BCA release and plant health regulations. As a consequence, recommendations will be made in a separate EFSA PLH Statement on future EU procedures for the evaluation of BCA releases.

2.1. Purpose

This opinion presents an assessment of the risk to plant health in the EU territory posed by the intentional release of the bud-galling wasp *T. acaciaelongifoliae* Froggatt for the biological control of the invasive alien plant *A. longifolia* (Andrews) Willd.

2.2. Scope

The assessment specifically excludes the probability of entry and systematic evaluation of risk reduction options and focuses on the probability of establishment, spread and impact in the EU territory.

3. Additional information

Submission of an application for a permit for the release of *T. acaciaelongifoliae* (Australian gall wasp) for the biological control of *A. longifolia* (long-leaved wattle) was made by the Centro de EcologiaFuncional/Departamento de Ciências da Vida, Universidade de Coimbra, Portugal. The application was made in accordance with the recommendations of the European and Mediterranean Plant Protection Organization (EPPO) standard PM 6/2.

4. Methodologies

The methodologies used in this opinion are taken from the EFSA Panel on Plant Health's (PLH Panel's) usual practice for pest risk assessment (EFSA PLH Panel, 2010), but adapted for the evaluation of the release of BCAs. In addition, expertise in risk assessment and specific expertise in weed biological control, plant ecology and horticulture were included.

4.1. The guidance documents

The risk assessment was conducted in line with the principles described in the documents "Guidance of the Scientific Committee on transparency in the scientific aspects of risk assessment carried out by EFSA" (EFSA Scientific Committee, 2009) and "Guidance on a harmonised framework for pest risk assessment and the identification and evaluation of pest risk management options" (EFSA PLH Panel, 2010).

The detailed questions in the EFSA-adapted EPPO pest risk assessment scheme, presented in the above-mentioned guidance document, were used as a checklist to ensure that all relevant elements were included; however, the terms of reference require that the opinion excludes the assessment of entry and a systematic evaluation of risk reduction options. The establishment section focuses on determining the area of potential establishment and spread beyond the immediate area of release, including those host plants used for ornamental trade.

The terms of reference exclude a systematic evaluation of risk reduction options. However, a restricted evaluation was made in line with the principles described in the above-mentioned guidance document (EFSA PLH Panel, 2010), as well as with those in the "Guidance on methodology for evaluation of the effectiveness of options to reduce the risk of introduction and spread of organisms harmful to plant health in the EU territory" (EFSA, 2012).

The PLH Panel developed rating descriptors to provide clear justification when a rating was given, which are presented in Appendix A of this opinion. This was done in order to follow the principle of transparency, i.e. that "…Transparency requires that the scoring system to be used is described in advance. This includes the number of ratings, the description of each rating … the Panel recognises the need for further development…", as described in Section 3.1 of the guidance document on the harmonised framework for risk assessment (EFSA PLH Panel, 2010).

Furthermore, this opinion considers the principles outlined in the International Standards for Phytosanitary Measures (ISPM) No 3 on the import and release of non-indigenous BCAs[4], as well as the related guidance on the safe use of BCAs, published by EPPO[5].

4.2. Methods used for conducting the risk assessment

The categorisation of the BCA assessed all those characteristics of the organism observed outside the risk assessment area, and useful to the completion of the BCA risk assessment. Essentially, this BCA risk assessment followed the pest risk assessment process outlined by the EFSA PLH Panel (2010), with the substitution of the term "BCA" for "pest". The level of detail provided is therefore in accordance with the relevance of the information to assessing the risk of establishment, spread and impact of the BCA in the risk assessment area. All consequences of release are evaluated, but no attempt is made to balance the potential positive and negative impacts. The consequence ratings are based on pest impacts on crops, but in this assessment they have been interpreted more widely in terms of BCA impacts on plants used in trade and in the natural environment.

The conclusions for establishment, spread and impact are presented separately and the descriptors used to assign qualitative ratings are provided in Appendix A.

4.3. Methods used for evaluating the risk reduction options

The assessment excludes the systematic evaluation of risk reduction options, i.e. ratings, but provides a description of the effectiveness and feasibility of measures post-release if required.

4.4. Level of uncertainty

For the risk assessment, conclusions on establishment, spread and impact, and the levels of uncertainty, are rated separately.

The descriptors used to assign qualitative ratings to the levels of uncertainty are shown in Appendix A.

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[4] FAO, 2005. International Standards for Phytosanitary Measures (ISPM) No 3. Guidelines for the export, shipment, import and release of biological control agents and other beneficial organisms. ftp://ftp.fao.org/docrep/fao/009/a0450e/a0450e.pdf

[5] Anonymous, 2014. PM 6/2 (3) Import and release of non-indigenous biological control agents. EPPO Bulletin 44, 320–329, doi: 10.1111/epp.12153

5. Data

5.1. Literature search

An extensive literature search on *T. acaciaelongifoliae* and invasive alien *Acacia* spp. was conducted at the beginning of the mandate. The databases used were CAB Abstracts, AGRIS, Scopus and Zoological Records, as well as a wide variety of websites, databases, Google, Google Scholar, and other sources of information (see Derkx et al., 2015). Keywords used were "*Trichilogaster acaciaelongifoliae*", "invasive *Acacia*" and numerous variants of these basic search terms. Further references and information were obtained from experts and from citations within the references.

5.2. Data collection

Data from host range tests for *T. acaciaelongifoliae* were provided, upon request, by Helia Marchante, University of Coimbra, Portugal. Data on the presence in the MSs of *Acacia* spp., both in the wild and cultivated as ornamental plants, were obtained through procurement from Stichting Dienst Landbouwkundig Onderzoek (Derkx et al., 2015).

5.3. Technical hearing

During the September 2014 PLH Panel plenary meeting[6], a technical hearing was held with three external experts: Helia Marchante (University of Coimbra, Portugal, the author of the application for release of *T. acaciaelongifoliae* referred to in Section 3), Richard Shaw (CABI, UK) and Andrea Allavena (Unità di Ricerca per la Floricoltura e le Specie Ornamentali, Consiglio per la Ricerca e la Sperimentazione in Agricoltura, Sanremo, Liguria, Italy). These experts covered the following areas of expertise, respectively: the proposed release of *T. acacialongifoliae* in Portugal, the international regulations concerning the release of BCAs and the cultivation of ornamental *Acacia* spp. in Italy.

ASSESSMENT

6. Risk assessment

6.1. Categorisation of the biological control agent

6.1.1. Identity and biology of *Trichilogaster acaciaelongifoliae*

6.1.1.1. Taxonomy

The organism under assessment is a clear, single taxonomic entity.

<u>Name</u>:

Trichilogaster acaciaelongifoliae Froggatt is currently the valid scientific name for the organism.

<u>Related species:</u>

The related species, *Trichilogaster signiventris* Girault, has been released in South Africa to control *Acacia pycnantha* Benth. (Dennill and Gordon, 1991; Prinsloo and Neser, 2007; Ndlovu et al., 2013).

<u>Taxonomic position:</u>

Class: Insecta; order: Hymenoptera; superfamily: Chalcidoidea; family: Pteromalidae; subfamily: Brachyscelidiphaginae.

[6] The minutes of the meeting are available at http://www.efsa.europa.eu/en/events/event/140924.htm

6.1.1.2. Identification

Identification (the correct nomenclature based on taxonomic criteria) is made on the basis of morphological characteristics (Prinsloo and Neser, 2007). Diagnosis (field recognition based, for example, on gall examination) is based on gall induction (details are in the submission made by Helia Marchante to the Portuguese Authorities, Section 2.2).

6.1.1.3. Organism biology

The genus *Trichilogaster* Mayr is associated with *Acacia* (Austin et al., 2004). *T. acaciaelongifoliae* forms galls (Figure 1) and maintains populations on the two closely related species *A. longifolia* and *A. floribunda* (Neser, 1982; Marchante et al., 2011a). In South Africa, this wasp has also formed galls on *A. melanoxylon* (see Section 6.1.4.3), which is an invasive alien species in South Africa but is also cultivated (for furniture), and *Paraserianthes lophanta,* which is also an invasive species in South Africa (Dennill et al., 1993).

For details on the life cycle and key aspects of the life-history strategy (development, survival, reproduction, feeding and dispersal) and ecological requirements, the reader is referred to Marchante et al. (2011a).

Figure 1: Galls of *Trichilogaster acaciaelongifoliae* on *Acacia longifolia* (photo courtesy of Jon Richfield, Wikimedia Commons)

6.1.2. Current distribution

6.1.2.1. Global distribution

The areas of origin of *T. acaciaelongifoliae* are the coastal regions of New South Wales and Victoria, in continental Australia, and Tasmania (Austin et al., 2004). *T. acaciaelongifoliae* was collected from these areas of origin (on the two closely related hosts *A. longifolia* and *A. floribunda*) and released in South Africa in 1982 and 1983 by G.B. Dennill and A.J. Gordon (Dennill, 1985, 1987). The wasp is not currently known to be present elsewhere.

6.1.2.2. Occurrence in the risk assessment area

The organism is not present in the risk assessment area, except under controlled experimental conditions for research purposes (Marchante et al., 2011a).

6.1.3. Regulatory status

The regulation of the introduction of alien BCAs (not listed as plant pests) is the responsibility of MSs. The Plant Health Directive provides protective measures against the introduction to MSs of organisms harmful to plants. Therefore, the risks associated with the release of a BCA against plants should be assessed with regard to this Directive.

In terms of EU regulations other than the Plant Health Regulation, the Invasive Alien Species (IAS) directive, Article 10, states that: "where a MS has evidence concerning the presence in, or imminent risk of introduction into its territory of an invasive alien species, which is not included on the Union list but which the competent authorities have found, on the basis of preliminary scientific evidence, to be likely to meet the criteria set out in Article 4(3), it may immediately take emergency measures, consisting of any of the restrictions set out in Article 7(1)" [Article 4 gives the characteristics of a listed IAS].

6.1.4. Potential for establishment and spread in the risk assessment area

The intentional release of *T. acaciaelongifoliae* is intended to lead to its permanent establishment as a BCA for *A. longifolia*. The constraints to such an effort are multiple and are addressed in the sections below.

6.1.4.1. Propagule pressure

It is anticipated that efforts will be made to ensure that adequate numbers of adult females (males are not needed to complete the life cycle) are released on multiple occasions to maximise the chance of establishment and this will lead to a high propagule pressure.

6.1.4.2. Health of the founding population

Given that the Portuguese researchers were successful in establishing galls from eggs from newly emerged adult females of *T. acaciaelongifoliae* under containment conditions, it can be concluded that the founding population is likely to be healthy enough to establish a colony. That said, there is always a risk that cryptic microbial pathogens may be present in the emerging adults that may affect their performance.

6.1.4.3. Host range

T. acaciaelongifoliae uses *A. longifolia* (subsp. *longifolia* and subsp. *sophorae*) as its main host (Figure 2) (Dennill and Donnelly, 1991). In Australia, it has been found on the related species *A. floribunda* (previously *A. longifolia* subsp. *floribunda*), but not on other *Acacia* species.

In South Africa, after intentional introduction, galls have been found on two other species in the Mimosoidae subfamily, namely *A. melanoxylon* and *P. lophantha* (Dennill et al., 1993). This was associated with unusually high densities of the BCA, after its first introduction on a previously unexposed, and therefore very abundant, host population of *A. longifolia*. After the initial population peak, galling on *A. melanoxylon* and *P. lophantha* has not been re-observed (Fiona Impson, October 2014, Plant Protection Research Institute, South Africa, personal communication). This type of attack has been termed "spill-over" (Taylor et al., 2007).

In Portugal, the host *A. floribunda* has not been reported (but various other *Acacia* spp. are present), while *A. melanoxylon* has been decreed an invasive alien species. *P. lophantha* (which is also a species native to Australia) has shown features of invasiveness in Portugal (Freitas and de Almeida, 2006).

Figure 2: Branches and flowers of *Acacia longifolia* (photo courtesy of Josh Jackson, Wikimedia Commons)

The original area of distribution of the host species *A. longifolia* (both subspecies) is south-eastern Australia (Figure 3).

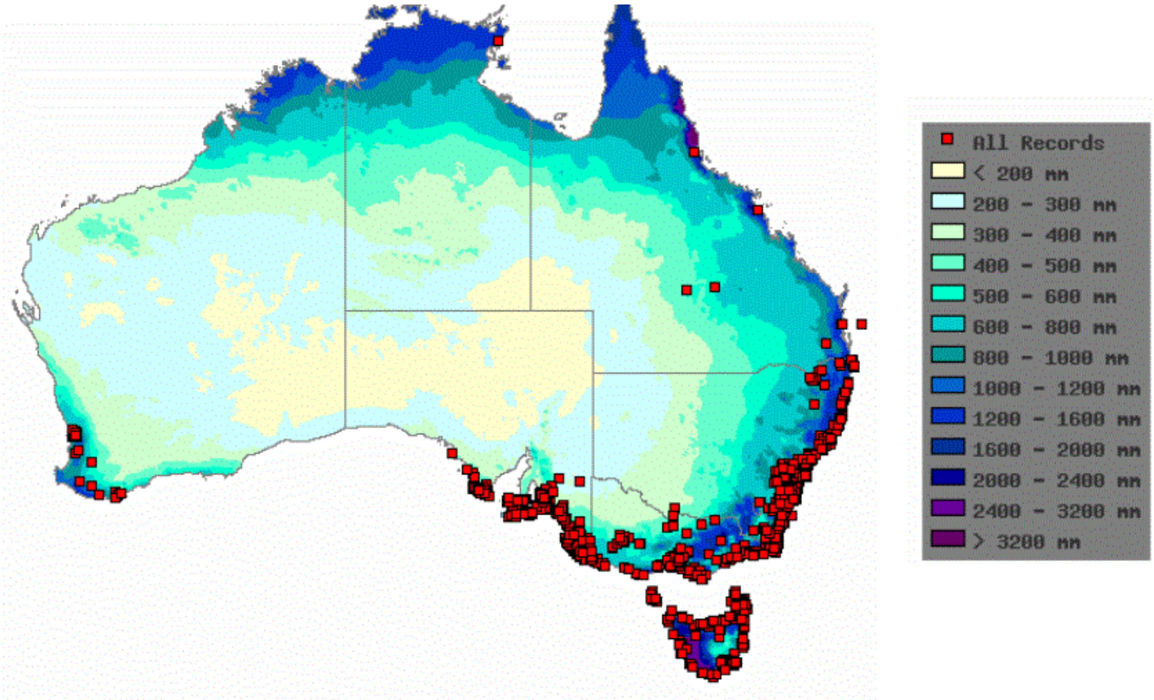

Figure 3: Distribution of *Acacia longifolia* in Australia (1 756 records), plotted on annual rainfall distribution (unknown period)—note the disjoint populations in the native range (from Hill, 2005)

In South Africa, *A. longifolia* was introduced as early as 1827 (Hill, 2005). The distribution and range expansion in South Africa of *A. longifolia* were mapped by Veldtman et al. (2010), who also reported a complete overlap in the occurrence of *A. longifolia* and *T. acaciaelongifoliae* in South Africa. Based on expert opinion, *A. longifolia* has a current estimated range in South Africa of 1 500 km^2 (as at

2000), which is much less than its potential estimated range (78 000 km^2) (Van Wilgen et al., 2004). *A. longifolia* is not just present in the Western Cape province, but is also found along the coasts of the Eastern Cape region up to Kwa-Zulu Natal, as well as in some inland locations in the Mpumalanga region (Veldtman et al., 2010).

In Portugal, *A. longifolia* was first introduced in the late 19^{th} century. It has been invasive in the dune system habitats of the Atlantic coast, but is increasingly invasive inland too. *A. longifolia* is a widespread invader, particularly in Portugal where extreme efforts have been under way to establish it for sand dune stabilisation since the beginning of the 20^{th} century (Rodríguez-Echeverría et al., 2009). *A. longifolia* is now found throughout much of Portugal (Figure 4).

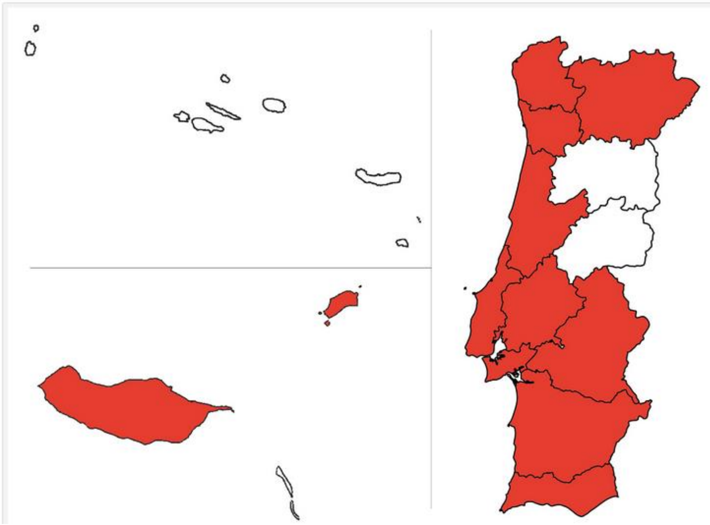

Figure 4: Distribution of *Acacia longifolia* in Portugal (provinces with presence: Trás-os-Montes, Minho, Douro Litoral, Beira Litoral, Estremadura, Ribatejo, Alto Alentejo, Baixo Alentejo, Algarve and Madeira; from http://www.invasoras.pt/gallery/acacia-longifolia/)

There is ornamental cultivation of *A. longifolia* in various MSs (e.g. in Galicia in Spain, in south-western and south-eastern France, and in some regions of Italy; see also Table 1 and Appendix C).

Table 1: Summary of the cultivation of major ornamental *Acacia* spp. in EU countries (from Derkx et al., 2015)

Acacia spp.	Host presence	Countries with cultivation	Scale	Number of nurseries
Acacia dealbata Link	No	Croatia, France, Germany, Greece, Italy, Spain, UK	About 400 ha in Liguria (Italy), 18 million stalks (France), common as a street tree in Spanish cities (some of the *A. dealbata* production in Italy and France is actually of *A. retinodes)*	11 (Germany), 5 (Greece), 7 (Italy), 36 (UK)
Acacia floribunda	Yes	France, Greece, Italy	–	3 (France), 4 (Greece), 2 (Italy)
Acacia longifolia	Yes	France, Germany, Greece, Ireland, Italy, the Netherlands, Spain, UK	Grown in many gardens in Cornwall, UK	9 (France), 6 (Germany), 3 (Greece), 1 (Ireland), 10 (Italy), 5 (the Netherlands), 7 (Spain), 3 (UK)
Acacia melanoxylon	Spill-over	Germany, Greece, UK	Popular in gardens in coastal areas of the UK	4 (Germany), 1 (Greece), 6 (UK)
Acacia saligna (Labill.) Wendl.	No	Germany, Greece, the Netherlands, UK	Commonly found in gardens and as a street tree in the UK	4 (Germany), 2 (Greece), 2 (the Netherlands)
Acacia retinodes	Under testing, but unlikely	France, Germany, Italy, UK	See above	9 (Germany), 9 (UK)

Acacia is a pan-tropical genus, with no native species in Europe (Figure 5). There are about 1 350 described spp. of *Acacia*, with about three-quarters of them originating from the Australia-Pacific region, and the rest from Asia, Africa, and Central and South America. Many *Acacia* spp. have been introduced worldwide for a variety of purposes, such as reforestation, dune stabilisation, animal fodder, tannin production, windbreaks and fuel wood, as well as for ornamental use (Kull et al., 2011). Many of these species have become invasive causing environmental consequences by outcompeting native vegetation. In Europe, *Acacia* spp. are currently cultivated as ornamentals or for perfume, but there is only sporadic cultivation of the two hosts of *T. acaciaelongifoliae* (*A. longifolia* and *A. floribunda*), whilst other *Acacia* spp. (e.g. *A. dealbata*) are intensively cultivated and traded (Table 1; Appendix C). The species named *A. floribunda* in the horticultural trade is named incorrectly; it is actually *A. retinodes*, which is not expected to be a host given its morphology (flowers in capitulate, whereas *A. longifolia* has flowers in spikes) and phylogeny. With regard to its phylogeny, *A. longifolia* does not belong to the same phylogenetic section as *A. retinodes*, not even to the same subgenus; while *A. longifolia* is from the subgenus Juliflorae, *A. retinodes* is from subgenus Phyllodineae (Helia Marchante, 10 March 2015, University of Coimbra, Portugal, personal communication). However, the ability of *A. retinodes* to act as a host is currently being tested by Helia Marchante.

Figure 5: Global native distribution of *Acacia* spp. (from World Wide Wattle, http://www.worldwidewattle.com/infogallery/distribution.php)

The phylogenetic relationships of acacias have recently been clarified in the context of biological control (Figure 6) (Kleinjan and Hoffman, 2013). *A. pycnantha*, which is also an invasive alien in South Africa, Portugal and other European countries (Hoffmann et al., 2002; Dorchin et al., 2006; Ndlovu et al., 2013), is not in the same clade as *A. longifolia*. The same applies to *A. saligna*, which is invasive in Portugal and in Italy (where it has recently been shown to be a host for the emerging bacterium *Xylella fastidiosa*). *A. pycnantha* has been the subject of biological control efforts in South Africa using a different *Trichilogaster* species (Hoffmann et al., 2002).

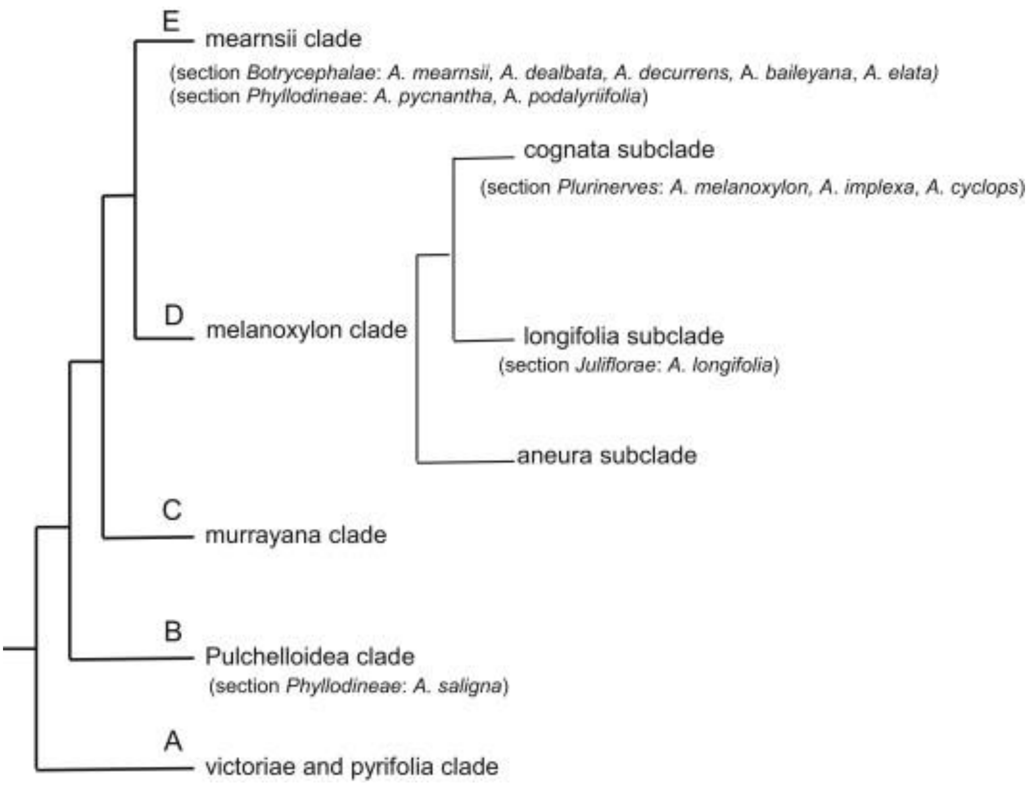

Figure 6: Phylogeny of *Acacia* spp. *Acacia longifolia* is more closely related to *A. melanoxylon* than to *A. pycnantha* or *A. saligna* (from Kleinjan and Hoffmann, 2013)

A summary of the results of host range tests for *T. acaciaelongifoliae* in South African and Portuguese experiments is provided in Table 2 (and in the Table provided by H. Marchante, Appendix B).

Table 2: Host tests of *Trichilogaster acaciaelongifoliae* and related *Trichilogaster* species (from Kleinjan and Hoffmann, 2013)

	T. arabica	***T. acacia-longifoliae***	***T. maidenii***	***T. esculenta***	***T. pendulae***	***T. flavivena***	***T. stefani***	***T. signiventris***
Vachellia	++	O						O
Senegalia		O						O
Paraserianthes lophantha (Ingeae)		+						O
Acacia s.s.								
clade A								
clade B		O				++		
clade C								
clade D								
aneura subclade				++			++	
longifolia subclade		++	++					
cognata subclade		+	++					O
unknown subclade					++			
clade E		O						+ +

++: Standard host.
+: Non-standard host (gall symptoms occurred in host-specificity tests or occurrence in the field is rare).
O: No gall symptoms developed in host-specificity tests.

6.1.4.4. Climatic conditions

T. acaciaelongifoliae prefers a warm temperate, fully humid, warm summer-type climate (classified as Cfb by the Köppen–Geiger climate classification; Kottek et al., 2006) and was collected from areas with such a climate in New South Wales and Tasmania in Australia for release in South Africa. In South Africa, the wasp has developed best under both Mediterranean, mild with dry, warm summer climates (Csb) and Cfb climates, with an average temperature of the hottest month of less than 22 °C (Dennill, 1987) and, although it occurs throughout the host range, was less likely to develop in Mediterranean, mild with dry, hot summer climate (Csa) areas, with average temperatures of more than 22 °C and a marked contrast between summer and winter temperatures (Dennill and Gordon, 1990). In Portugal, there is a Csb climate along the Atlantic coast and inland in the north of the country (Figure 7).

This suggests a slight mismatch between the most suitable climates for the wasp (Cfb and Csb) and the areas where *Acacia* species are most often found (areas with Csa climates along the Mediterranean coast). However, the presence of *A. longifolia* and *T. acaciaelongifoliae* in the sub-tropical regions of Australia (Queensland and near Perth; see above) suggests that the wasp would be able to establish under Mediterranean conditions.

According to the hearing expert Helia Marchante (25 September 2014): "In South Africa, the organism now occurs throughout the area of distribution of *A. longifolia* (as it does in Australia). However, as the host plant is more successful and is a more vigorous invader in regions without an extremely arid period, or with only a short arid period in the middle of summer, the insect is also more frequently observed in these regions."

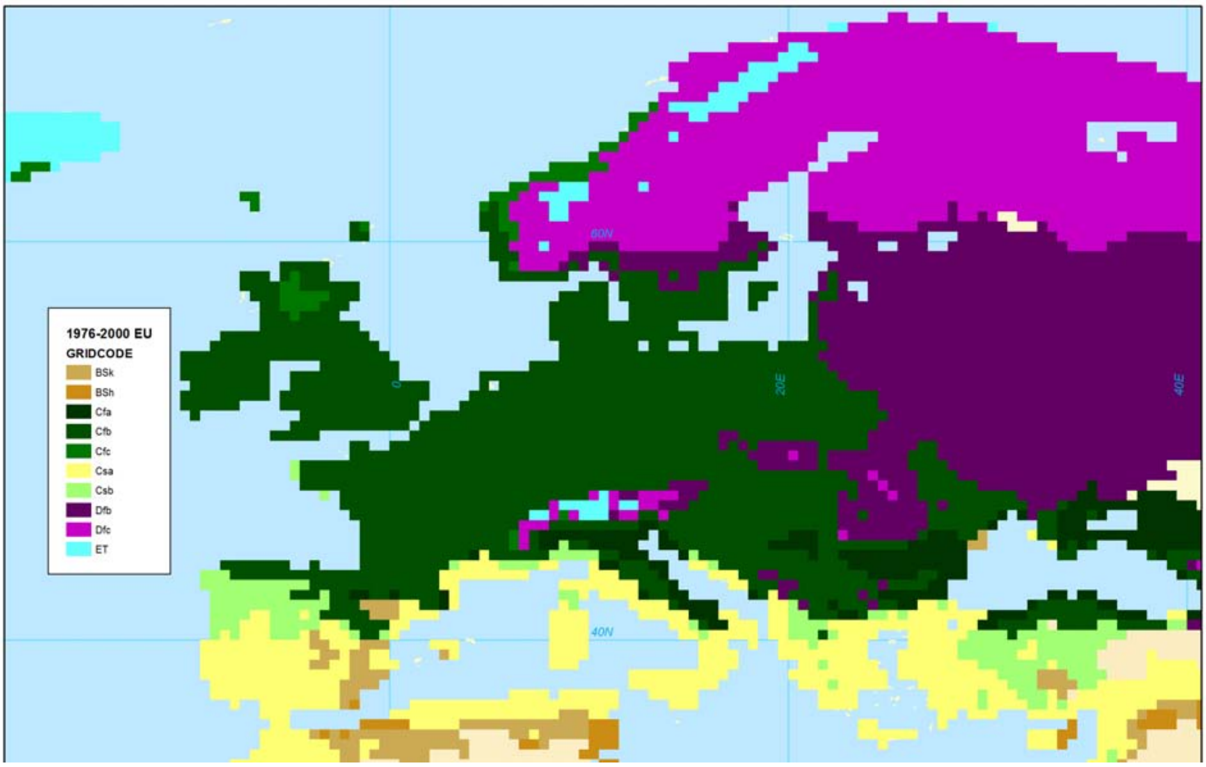

Figure 7: Köppen–Geiger climate distribution map for Europe (1976–2000) (modified from Rubel and Kottek, 2010)

Those studying the wasp in South Africa discovered that the insect performance was better in areas with Mediterranean-type IV climates (as per Walter and Leith, 1960), with winter rains and short arid spells in summer, than in hotter inland valleys with a Type III climate (Dennill and Gordon, 1990).

6.1.4.5. Current establishment in the risk assessment area

The wasp has never been introduced into the EU territory and is not established therein.

6.1.4.6. Predation and parasitism

When the wasp is released in the target area, all arthropod parasites of the wasp will be removed and destroyed before release so that no alien natural enemies of the wasp from the received shipments will be present. However, the question remains regarding whether or not parasites and/or predators already present in the EU could include *T. acaciaelongifoliae* in their diet and, therefore, have an impact on establishment, performance and spread. The South African experience provides information regarding this issue, as they found *T. acaciaelongifoliae* to have various native parasites and hyperparasitoids as well as symbionts but, in the main, these had no adverse effects on the BCA's success (Hill and Hulley, 1995; Manongi and Hoffmann, 1995; Seymour and Veldtman, 2010). However, there was reference in this work to the rates of parasitism by native parasitoid *Pseudotorymus* spp. wasps of about 21 % in the Western Cape province and 60–80 % in the Eastern Cape province. There has not been a review of likely parasitoids in the EU but native *Pseudotorymus* spp. do exist in the EU according to the Universal *Chalcidoidea* Database, although they are not listed as parasitoids of *Trichilogaster*. The following Chalcidoid (Hymenoptera) species are parasitoids of *T. acaciaelongifoliae* (main host): *Eurytoma gahani* (Eurytomidae, Australia), *Coelocyba nigrocincta* (Pteromalidae, Australia) and *Megastigmus darlingi* (Torymidae, Australia).

6.1.4.7. Allee effects

The BCA is parthenogenetic so there should be no issues relating to mate acquisition; however, the importance of males in the long-term persistence of this species is not clear and a decision needs to be made regarding whether or not males should also be collected and released (Marchante et al., 2011a).

6.1.4.8. Spread capacity

In South Africa, the wasp has dispersed effectively in *A. longifolia* stands in both coastal and inland regions. Information on natural dispersal ability is largely missing. The experience gained from the early days of South African releases shows that the excellent host-seeking ability, coupled with wind dispersal and directed flight, enabled establishment at sites 20 km from release points after two years (Dennill, 1987), most likely as a result of wind-assisted dispersal. Within two restricted sites of 1 ha each, the wasp filled the extent of the study area within two generations, because of its rapid reproduction and host-finding ability (two years). However, as female wasps that do not find a host die within three days, this may limit the extent of dispersal in the absence of contiguous host populations.

6.1.4.9. Conclusion on the potential establishment and spread in the risk assessment area

The wasp is likely to be able to establish where hosts are present in Europe. Spread over limited distances of up to 20 km has been described (Dennill, 1987), but there is no evidence for dispersal beyond this distance. In South Africa, the successful establishment of the wasp throughout the range of *A. longifolia* of 1 500 km^2 was accomplished through a release program with over 60 release sites (Neser, 1985).

6.1.5. Potential for consequences in the risk assessment area

The proposed consequences of the intentional release of the wasp as a BCA are the control of invasive alien plant species, contributing to fulfilling the objectives of nature restoration and conservation. In South Africa, no negative environmental consequences of the release of the wasp in the early 1980s have been reported. In Europe, the main (direct) plant health consequence would be on established invasive alien *A. longifolia* populations. However, consequences for cultivated *A. longifolia* need to be considered. Other potential consequences could be the reduction in the stability of sand dunes, where *A. longifolia* has successfully fulfilled a stabilisation purpose, the reduction in the use of *A. longifolia* for food and shelter by native species or as a source of pollen and nectar for bees, the modification of forest fire regimes, and social impacts (e.g. the reduction in the use of *A. longifolia* as a source of firewood and ornamental flowers).

6.1.6. Conclusion on the categorisation of the biological control agent

T. acacialongifoliae is a gall wasp native to Australia, where it is restricted to the hosts *A. longifolia* and *A. floribunda* (Marchante et al., 2011a). It was released intentionally in South Africa in 1982 and 1983 as a BCA for *A. longifolia* and has successfully established and spread there, with the majority of plants showing galls (Dennill, 1987). Seed set on affected hosts was reduced by between 73 and 95 % within three generations of release (Dennill, 1987). In South Africa, spill-over to two other hosts (*A. melanoxylon* and *P. lophantha*) was observed, but both are sub-optimal hosts and galls form only sporadically with negligible effects. The climate in the target area is likely to be largely suitable for the BCA. The present BCA categorisation shows the need for an assessment of the risks to plant health posed by its intentional release.

6.2. Probability of entry

The probability of entry is excluded from this risk assessment because there is a plan for an intentional release of the wasp as a BCA of invasive *A. longifolia*.

6.3. Probability of establishment in the risk assessment area

6.3.1. Availability of suitable hosts and alternate hosts in the risk assessment area

The genus *Trichilogaster* Mayr is associated with *Acacia* (Austin et al., 2004). Although *T. acaciaelongifoliae* builds galls (Figure 1) on the two closely related species *A. longifolia* and *A. floribunda* (Marchante et al., 2011a), in South Africa this wasp has also attacked *A. melanoxylon*, which is an invasive alien species in South Africa but is also cultivated (for furniture), as well as *P. lophantha* (Dennill et al., 1993), both of which are present in Portugal, Spain, France and Italy (Derkx et al., 2015).

These non-target attacks on *A. melanoxylon* and *P. lophantha* were unexpected; however, further communications with South African researchers (Fiona Impson, October 2014, Plant Protection Research Institute, South Africa, personal communication) revealed that at sites where galling on *A. melanoxylon* was initially observed as relatively common, galls are now rare and hard to find or, in one case, completely absent, even though galls persist on local *A. longifolia*. This is most likely to be what practitioners of biological control refer to as a “spill-over effect” (Taylor et al., 2007). As such, a plant that is within the physiological host range of the potential BCA is attacked but only under conditions where the agents are present at a very high population during the initial outbreak period. After some time, perhaps years, the BCA population declines as do the non-target attacks. In South Africa, the damage to *A. melanoxylon*, and another closely related invasive alien species *P. lophantha*, was found to be largely cosmetic. The low incidence of galling on *A. melanoxylon* and the low gall to pod dry mass ratio is expected to prevent the wasp from adversely affecting the growth, since dry gall mass should exceed that of the normal reproductive structures in order to act as significant metabolic sinks (Dennill, 1988, 1990). Although the percentages of trees infested and branches galled on *P. lophantha* in the Dennill et al. (1993) study were 95 % and 33 %, respectively, the mean dry gall mass was only 25 % of that of the pods, and there was no difference between the number of pods on galled and non-galled branches. Indeed, the South African researchers expressed disappointment that the damage to the invasive species *P. lopantha* may not reduce growth or reproduction significantly (Dennill et al., 1993).

The experience gained in South Africa indicates that *A. melanoxylon* and *P. lopantha* are likely to be attacked if *Trichilogaster* populations build up to high levels in their proximity. These two potential hosts are often present along Portuguese coasts where *A. longifolia* grows. However, this is unlikely to have any significant impact on the growth of these two non-target species and it is likely that the levels of attack will decline over time, along with the decline of populations of *A. longifolia* and the BCA. Information provided by the South African team (Johnny Hoffmann, July 2014, Zoology Department, University of Cape Town, personal communication) revealed that they had not seen any galls on *A. melanoxylon* or *P. lopantha* “for years”, and that this is certainly considered a “rare occurrence” and “the wasps certainly cannot sustain themselves permanently on either of these hosts”. However, should *P. lopantha* be present in the absence of *A. longifolia*, it is possible that, if *T. acaciaelongifoliae* arrives in a new area, the presence of *P. lopantha* would facilitate the wasp’s establishment. Other *Acacia* spp. present in those regions have been tested and shown not to be hosts of the BCA.

In host range testing, there are two types of experiments commonly used: choice and no-choice tests (Schaffner, 2001). In no-choice tests, the species is tested on its own, whereas in choice tests, it is paired with a known host. The no-choice test will show overall acceptability but can produce false positives, indicating hosts which would never act as hosts in the field. Choice tests, on the other hand, come closer to reality in providing the organism with a choice (Hinz et al., 2014).

Marchante et al. (2011a) carried out a series of no-choice oviposition and development tests under quarantine conditions to complement the data generated in advance of the release of *T. acaciaelongifoliae* in South Africa and those generated from field observations. In these tests, limited oviposition was observed on *Cytisus striatus*, a native leguminous shrub in Europe, and *Vitis* *vinifera* L., an important crop (grapes). In the latter case, only 4.3 % of buds received eggs versus 31.8 % of the buds on *A. longifolia*. More importantly, eggs were laid on the outer pubescent sheath of the developing buds and not within the bud tissues of the plant. None of these eggs were able to develop and it can probably be assumed that they were laid "by accident" and, therefore, this may be considered a laboratory artefact. This is further supported by the fact that the massive wine-producing regions in South Africa and Australia have not reported any galling caused by this insect. Subsequent choice tests on excised plant material revealed that no eggs were laid on *Vitis vinifera* in the presence of its host *A. longifolia*.

In the case of *C. striatus*, the results are less convincing since, in the no-choice tests, eggs were laid in the tissues of the non-target plant with some preference for buds of around 1 mm and with little difference in the number of eggs per branch on this non-target plant compared with the control target plant, *A. longifolia*. As far as subsequent development is concerned, the number of galls found in the target control replicates was very low, with one plant supporting three galls and two plants with one gall each, i.e. only 50 % of the six replicates developed galls. Whilst there was no development of the wasp in *C. striatus* in the weeks before they died, the results do not convincingly demonstrate a non-host status for this plant because of the poor performance of the wasp on the preferred host; therefore, a repeat of the study with more replicates would be useful. It should be noted, however, that these are no-choice tests and, therefore, the most extreme in the suite available to inform safety studies. Choice tests would be a better indicator of the oviposition that might occur in the field should *T. acaciaelongifoliae* be presented with a choice between suitable buds of *A. longifolia* and *C. striatus*. The no-choice test mimics what might happen should adult gravid females of *T. acaciaelongifoliae* be seeking hosts when only *C. striatus* is present or when only *C. striatus* is at a suitable stage for oviposition in the presence of *A. longifolia* and is therefore quite precautionary in its approach. Subsequent choice tests (see Figure 8) revealed a preference for *A. longifolia* versus *C. striatus* but not exclusivity. No egg laying was observed on *C. striatus* in additional choice tests using potted plants (Marchante submission to the Portuguese Authorities).

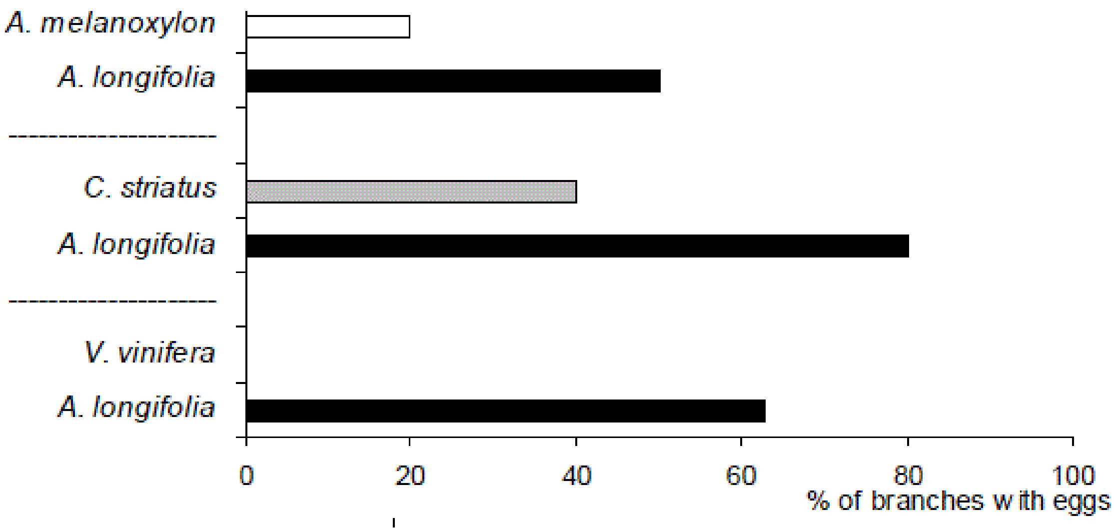

Figure 8: Percentage of branches of the target and non-target species on paired-choice tests where *Trichilogaster acaciaelongifoliae* laid eggs. The white, grey and black colours are used only to distinguish between the tested plant species (from Marchante, 2011; with kind permission of Helia Marchante)

Follow-up surveys of the closely related Spanish broom species *Spartium junceum* L. and *Teline monspessulana* (L.) K. Koch (formerly *Cytisus monspessulanus*) in South Africa and Australia revealed no galling (Marchante et al., 2011a). Both of these species produce flower buds that are very similar to those of the non-target *C. striatus*. Field observations were carried out to determine whether or not South African brooms were attacked by the wasp if present in proximity to *A. longifolia*. No attack was observed (Marchante, 2011). These findings indicate that these plants, which are very closely related to *C. striatus*, are not hosts and suggest that the observation of egg laying on *C. striatus* should be regarded with caution and not as conclusive evidence of its status as a host.

Marchante conducted further experiments on *C. striatus* and *A. retinodes* and, in both cases, these were no choice tests with limited replicates. No egg laying was observed on *C. striatus*. Egg laying but not gall formation was observed on *A. retinodes* (Helia Marchante, 10 March 2015, University of Coimbra, Portugal, personal communication).

Given the conflicting nature of the available evidence, there is uncertainty about whether or not *C. striatus* and *A. retinodes* might be hosts of the BCA.

6.3.2. Suitability of the environment

The climate is expected to be largely suitable for the establishment of *T. acaciaelongifoliae* wherever its hosts (*A. longifolia* and *A. floribunda*) are present (see Section 6.1.4.4). However, there is uncertainty about whether or not the climate in the more arid regions where *A. longifolia* is invasive in Portugal is likely to support high population densities of the wasp.

6.3.3. Other characteristics of the organism affecting the probability of establishment

6.3.3.1. Reproduction and development

Adults emerge in late spring and lay eggs on young *A. longifolia* buds (Figure 9). Through parthenogenesis, each female lays around 400 eggs in its brief three-day life, so no males are required. Eggs are often laid on living plant material close to the gall from which the female emerged. After oviposition upon buds, the hatched egg produces juveniles which, in turn, produce a substance that causes the buds to form galls within which the larval wasp spends the remainder of their pre-adult life. The galls reach their maximum size in mid-summer, and the juvenile wasps enter pupation prior to emergence of the adult females which eat their way out of the gall and seek suitably sized buds on which to lay eggs and continue the cycle. Galls can be single- or multi-chambered, but the presence of either type of gall prevents successful flowering. Most chambers contain females, but occasionally males develop in smaller chambers on the periphery of the gall (Marchante et al., 2011a).

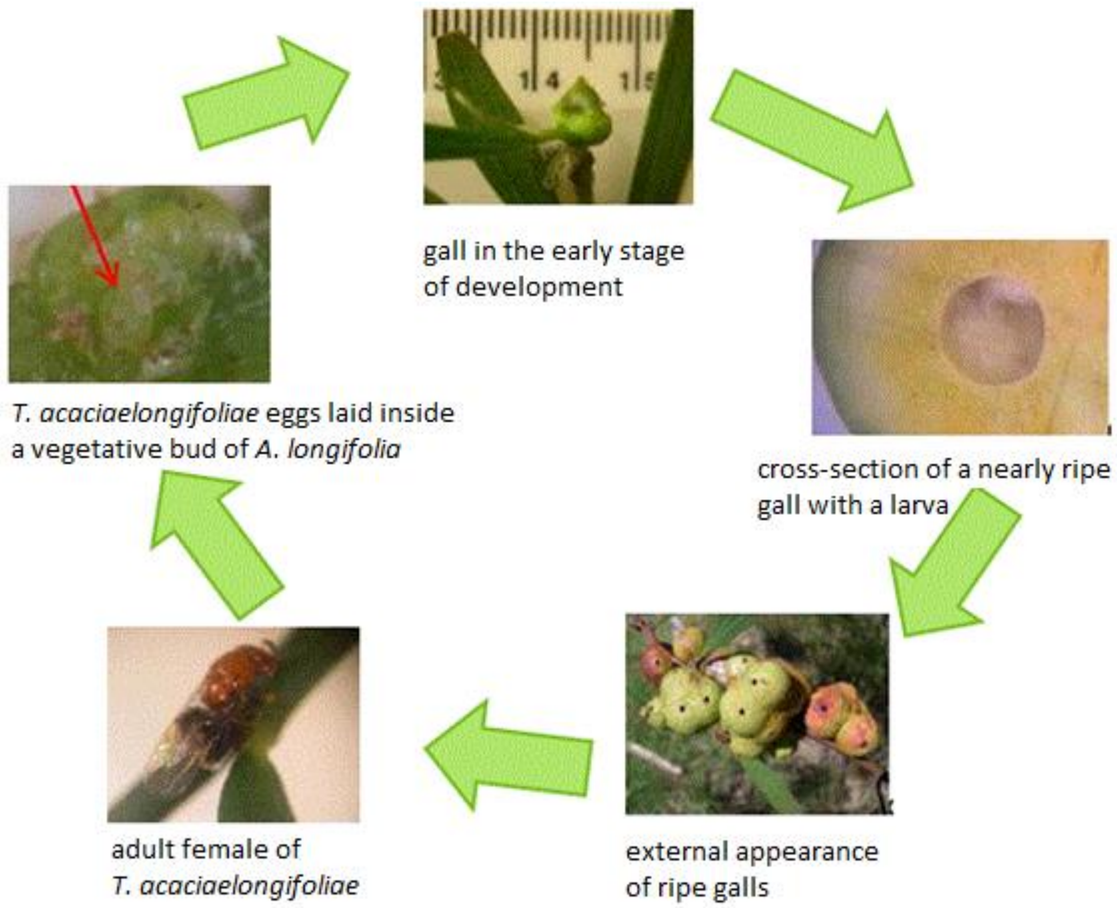

Figure 9: A schematic representation of the life cycle of *Trichilogaster acaciaelongifoliae* (modified from Marchante, 2011; with kind permission of Helia Marchante)

6.3.3.2. Survival

T. acacialongifoliae is dependent on its host plant for survival and development, as the adult females do not feed during the few days of oviposition. The gall in which the larvae develop may provide some protection against biotic and abiotic variables; however, the extremes that larvae can tolerate are untested as the areas of previous introduction and the native range do not show large climatic variations.

6.3.3.3. Dispersal

Natural dispersal of the wasp is achieved solely during the adult stage. These insects are good at finding their host plants and were observed to disperse up to 20 km in South Africa within two seasons, thanks to a combination of prevailing winds and direct flight (Dennill, 1987). However, observations in the lab and glasshouse in South Africa, prior to field release, suggest that they are likely to disperse quite well and may be capable of strong and even directed flight in the search for hosts after the females have lightened their egg load during the initial oviposition (Neser, 1985). Neser (1985) speculated that older females may fly, or be carried, long distances on windy days, and that, with high population densities, dispersal may be very effective. Since its release in South Africa in the early 1980s, *T. acaciaelongifoliae* has been found in the regions of South Africa that support the host plant. However, this cannot be presumed to be as a result of the capabilities of the wasp, since there was an active redistribution programme once galls became common; for example, the wasp was released at 64 additional sites in 1983 (Neser, 1985). Furthermore, there was an extensive programme designed to spread the wasp beyond these initial 64 sites over the entire territory infested by *A. longifolia* (Veldtman et al., 2010).

6.3.3.4. Synchrony in relation to the likelihood of establishment

In order to establish, *T. acaciaelongifoliae* adults must find buds in a suitable condition in the three days in which they are able to survive after emergence. These buds are only present on the plant in large numbers for around one month per year. The challenge facing biocontrol practitioners is to synchronise the emergence of adults with the presence of optimal bud sizes of the target plant in the field. Achieving such synchrony may be a challenge because Europe is half a year out of phase with the southern hemisphere from which the galls must be obtained, as there is no ongoing culture of the wasp in Europe. Thus, successful establishment cannot be taken for granted, even though the climatic requirements are met at least in most of the invasive range of the plant in Europe. Seasonal synchrony will also play a part in the season after release if buds are indeed galled in the first season, as it is not certain that the life cycle of the wasp is solely linked to plant developmental stage, although this is likely to have the greatest influence. An inability to resynchronise weed biocontrol agents to a new seasonal cycle has been blamed for previous failures, such as the release of the flea beetle (*Longitarsus aeneus* Kutschera) to control Paterson’s curse (*Echium plantagineum* L.) in Australia (Swiperick and Smyth, 2002).

In the early days of the South African release experience, it was clear that galls did not last long in the laboratory after being removed from their host plant and over 1 000 galls that were shipped from Australia to South Africa in 1980 yielded only four weak adult female wasps and numerous parasitoids (Neser, 1985). The South African researchers concluded that the best solution was to collect galls from which the adults were only 7–10 days from expected emergence. The Portuguese researchers propose receiving mature galls from the field in South Africa and to release emerging adults into the field where a few suitably sized buds are expected to be present. There is no proposal to release wasps from a maintained culture. This selection of an atypical founder population may not be ideal; careful consideration needs to be given to the release strategy to overcome the asynchronous phenology of the host plant and the wasp. This issue has implications for the likelihood of establishment.

6.3.4. Conclusions on the probability of establishment in the risk assessment area

	Descriptors
Rating: *moderately likely*	• In the release area, by definition, host plants are present • The organism has been successfully established in South Africa outside its native range (Australia) • In the proposed release area, the environmental conditions are similar to those in the native area of the wasp • There is the intention to make the release programme succeed, including the possibility of multiple releases over time and space. If release attempts are repeated often enough, the likelihood of establishment would increase to the level of ***likely*** • From the initial release sites in the *A. longifolia* infested dune areas in Portugal, the organism is likely to colonise the whole of the target dune area (based upon the previous experience in South Africa) • The probability of establishment of a founder population depends on the ability to match the wasp life history with the host phenology in the northern hemisphere; specifically, having suitably sized buds in the three days available for the wasps to find them
Uncertainty: *medium*	Broadly, there is excellent information on all relevant aspects of the biology of the agent and its establishment in South Africa. However, there is no prior experience with its establishment in the northern hemisphere

6.4. Probability of spread and establishment outside of the target area

The target area is defined as the regions of Portugal where invasive *A. longifolia* is present. All release sites will be located within the target area.

6.4.1. Spread and establishment by natural means

6.4.1.1. Adults

Flight

Adults of *T. acaciaelongifoliae* are relatively weak flyers, especially when carrying a high egg load. Flight distances in such conditions are tens of metres rather than kilometres. Adults are short lived and thus need to find host plants in suitable conditions (small buds) within three days (probably less in the field situation).

Passive

Wind-assisted dispersal is by far the most likely natural means of spread, as the adults are very small and were found to spread 20 km in two seasons in South Africa (Dennill, 1987). This could be much more in gale conditions. The Portuguese trade winds start in about April and last until September. On the Algarve coast, the summer winds are mostly northerly and most gales occur in winter, when the prevailing winds are westerly but the wasp would be in the larval stage.

6.4.1.2. Larvae

There is no mechanism of spread of the BCA larvae by natural means.

6.4.2. Spread and establishment by human assistance

Larvae

The dispersal of larvae is dependent on the transport of healthy galls. These decompose or dry out very rapidly in sub-optimal conditions. More likely to succeed is the transport of galls on a living plant, but this is more unlikely to occur than transport of galls on cut branches.

Adults

In all of the following cases, the most limiting step for dispersal is the ability of the adult to find its host at the destination, which would probably need to be within a few metres of the point of release.

Clothing

Adults could settle on the clothes of walkers but are unlikely to stay on them for any distance; therefore, this would only add limited dispersal capabilities.

Cars

If adult populations are high, it is quite possible that adults could find their way into cars, either by flying in or being carried on the clothes or belongings of passengers, and travel hundreds of kilometres before being able to escape.

Aeroplanes

It is quite possible that adults could find their way onto flights from Lisbon, Santarém, Porto and Faro and travel considerable distances but, again, they would immediately need to find receptive host trees at their destination.

Intentional redistribution

Because *A. longifolia* is a recognised serious invader in France, Italy and Spain, as well as in the intended region of release (Portugal), it is quite possible that concerned citizens, and even the conservation community, may be interested in receiving the BCA, especially if it is advertised as a solution to *A. longifolia* invasions. It would take some skill and knowledge to achieve this in the short-term, but these communities contain expert entomologists. Repeated intentional release of mature galls over years at multiple sites would increase the propagule pressure and therefore the likelihood of spread to non-target areas.

Curiosity

The galls are likely to be of interest to those with an interest in nature and may be picked and taken home by such people. This would increase the likelihood of movement but the chance of establishment would remain low unless this human transport was for intentional redistribution (see above).

Trade

It is recognised that there is some trade of planting material of *A. longifolia* in the Mediterranean, but there is no evidence of such trade from the target area to other regions where the host is present (Appendix C).

6.4.3. Conclusions on the probability of spread and establishment outside of the target area

6.4.3.1. Natural spread

	Descriptors
Rating: *Moderately likely*	• Active dispersal is only possible over short distances; beyond a certain distance, dispersal can only be wind-assisted • For effective dispersal, a suitable host must be found within three days of the emergence of the adult • Host populations are often fragmented, requiring long-distance dispersal (jumps) • The probability of natural spread over considerable distances increases with the size of the source population • Where there is close proximity in presence of hosts in, for example, northern Portugal and Galicia in Spain (see Figure 5 in Derkx et al., 2015), then spread is ***likely***
Uncertainty: *Medium*	There is little information on dispersal by wind (although there is experience from South Africa on the successful wind dispersal there)

6.4.3.2. Human-assisted spread

	Descriptors
Rating: *Moderately likely*	• Experience in South Africa has shown that intentional redistribution of the galls at the right time in a release programme is a very effective mode of spread of the organism • It cannot be ruled out that people would want to spread the BCA without due authorisation. If those persons were aware of the constraints imposed by the biology of the organism and were sufficiently expert, then such spread would ***likely*** be successful • Inadvertent spread associated with human movement is possible but is less likely to happen than with other organisms because of the constraints imposed by the biology of the organism • There is the potential of future trade in ornamental *A. longifolia* to enable spread of the BCA

Though not currently anticipated, authorised intentional movement outside of the target area would result in **likely** spread with **low** uncertainty.

If movement is intentional but not authorised, then the uncertainty is also **low**.

With inadvertent movement, the likelihood of spread is low, and uncertainty is **high** because of the unpredictability of the process.

6.5. Conclusion of the risk of establishment and spread in the risk assessment area

The risk assessment area is that occupied or potentially occupied by wild or planted *A. longifolia* and *A. floribunda* in the EU territory.

The probability of establishment is assessed as **moderately likely** (based on the previous experience in South Africa), with **medium** uncertainty (because of the switch between hemispheres).

The probability of spread to non-target-areas outside of Portugal is assessed as:

- **moderately likely** for natural spread (because of the fragmented host populations), with **medium** uncertainty, because of little information on wind-assisted dispersal;

- **moderately likely** for human-assisted, intentional spread (based on the experience in South Africa), with **low** uncertainty, but **unlikely** for inadvertent movement (with **high** uncertainty).

6.6. Assessment of consequences

6.6.1. Impacts of *Acacia longifolia* and invasive alien acacias more generally

Significant environmental impacts of the *A. longifolia* invasion in Portugal have been quantified by the Portuguese research team at Coimbra, and others, over the last 10 years. They include a reduction in plant diversity and species richness (the average number of plant species per plot was less than half in *A. longifolia*-covered areas compared with areas without *A. longifolia*) (Marchante et al., 2003; Marchante, 2011), alterations to the chemical and microbiological composition of the soil (Marchante et al., 2008a, b; Marchante, 2008; Rascher et al., 2012) and to the seed stock (Marchante et al., 2010), and a reduction in the resilience of the invaded ecosystems (Marchante et al., 2009, 2011b; Le Maitre et al., 2011).

Acacia species have been shown to induce simultaneous changes in above- and below-ground communities, microclimates, soil moisture regimes and soil nutrient levels (Le Maitre et al., 2011). The general flow of impacts and interactions due to invasive alien *Acacia* spp. is presented in Figure 10 (from Le Maitre et al., 2011) and Table 3.

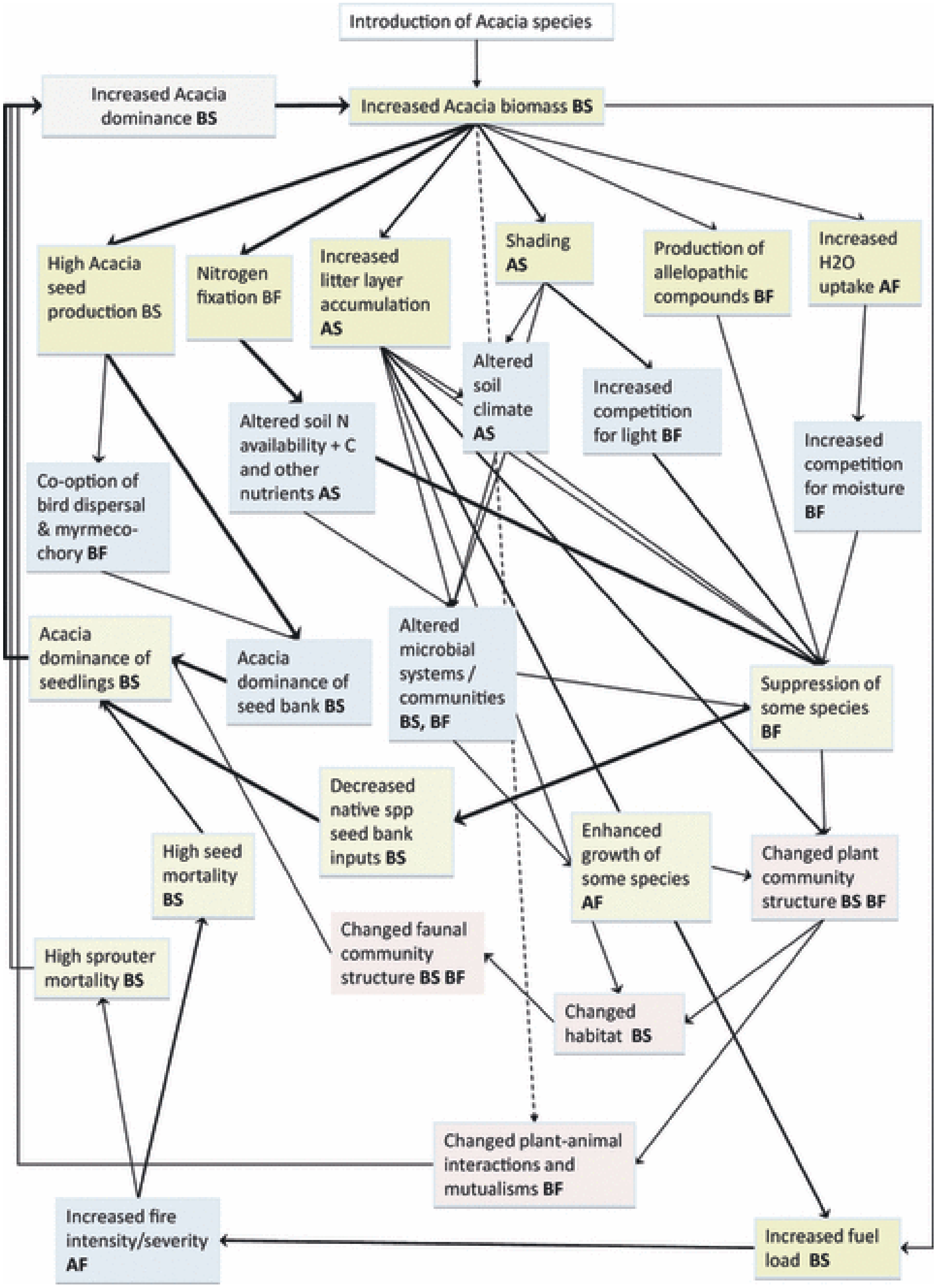

Figure 10: The general flow of impacts due to invasive alien *Acacia* spp. The width of the arrows indicates the relative importance of the pathways based on the literature; the dotted arrow indicates a probable link. B = biotic, A = abiotic, S = Structure and F = function (from Le Maitre et al., 2011)

Table 3: Summary of the effects on ecosystems of the invasion by *Acacia longifolia* and other invasive alien *Acacia* spp.

Category of transformation	From	To	Timescale	Reference
Transformation of floral structure	Herbs, few shrubs and trees	Continuous stands of *A. longifolia*	A few years	Marchante, 2011
		Accumulation of a deep *Acacia* litter layer	A few years	
Changed nutrient regime	Nutrient-poor sand dune ecosystem	Increased soil carbon and nutrients, especially total nitrogen	Decades	
Soil microbial processes	Low	More than double	10 years	
Litter biomass	0.6 km/m^2	2.05 kg/m^2	Decades	Marchante et al., 2008a
Nitrification		48- to 285-times higher NO_3/g dry soil	Decades	Marchante et al., 2008a
Soil water content	Low	Higher because of build-up of organic material		
Depletion of native seed banks	Range of native seeds	Massive seedbank of up to 1 500 seeds/m^2, exclusively *Acacia*	Decades	
Nitrogen uptake by other plant species	No discernible increase in uptake by native plants from a native N_2 fixer	Significant increase in foliar nitrogen content		Hellmann et al., 2011
Altered forest fire regimes		Increased flammability	A few years	Wilson et al., 2011

The physical removal of *A. longifolia* from the dune ecosystems in Portugal produced only partial recovery after six years (Marchante et al., 2009, 2011b), but that recovery was associated with the arrival of generalist plant species and subsequently some replacement by characteristic dune species.

6.6.2. Potential effects of *Trichilogaster acaciaelongifoliae* on invasive alien *Acacia longifolia*

T. acaciaelongifoliae inhibits seed production (up to 95 %) by galling the reproductive buds of *A. longifolia* and, by so doing, reduces reproductive potential (Dennill, 1990). In heavily galled trees, this reduction in seed production can be even higher (Dennill, 1985). The wasp can be introduced along with a seed-feeding weevil (*Melanterius ventralis* Lea), with the intention of destroying any residual seeds (Donnelly and Hoffman, 2004). In addition, the stress imposed on the plant reduces vegetative growth and competitive ability (Dennill, 1985). At 32–38 % of sites in South Africa where the wasp is present, tree mortality was observed (Dennill, 1990).

Galling increases flower abscission of unaffected inflorescences through indirect effects throughout the tree (Dennill and Donnelly, 1991). Gall formation replaces reproductive bud formation and is far more energy-demanding than normal reproduction (Dennill, 1988). This aspect of a powerful metabolic sink has been noted for other similar gall-forming systems (Goolsby et al., 2000). A detailed account of the physiological processes involved are given for the paired congeneric system *Acacia pycnantha*/*Trichilogaster signiventris* (Dorchin et al., 2006), which provides another example of biological control by a gall wasp (Hoffman et al., 2002).

Release of *T. acaciaelongifoliae* has been considered in New Zealand where similar detrimental effects on invasive alien *A. longifolia* are expected (Hill, 2005).

The production of galls has been observed in South Africa on *A. melanoxylon* and *P. lophantha*, a spill-over effect, but detrimental effects on these hosts are minimal (Dennill et al., 1993).

6.6.3. Other environmental consequences

6.6.3.1. Occurrence of the organism in natural habitats

In Australia, *T. acaciaelongifoliae* has been recorded on only two closely related species, *A. longifolia* (including both subspecies *A. longifolia* (Andr.) Willd. var. *longifolia* and *A. longifolia* (Andr.) Willd. var. *sophorae* (R. Br.) F.J. Muell.) and *A. floribunda* Sieber (Noble, 1940). The host specificity of *T. acaciaelongifoliae* has been confirmed by comprehensive tests in both South Africa and Portugal.

The gall wasp's performance on *A. longifolia*, *A. melanoxylon* and *P. lophantha*, all of which are invasive alien species in South Africa, was also studied by Dennill et al. (1993). This revealed a very high incidence of galling on the target weed *A. longifolia*, as well as on *P. lophanta*, but a low incidence of galling on *A. melanoxylon*. However, this seems to have been a transient phenomenon and not persistent.

6.6.3.2. Occurrence of the organism in private gardens, plantations or amenity land

T. acaciaelongifoliae is known to attack garden specimens of *A. floribunda* in South Africa as severely as it attacks *A. longifolia*. *A. floribunda* is not invasive in South Africa (or in Portugal). Similar effects could be expected on *A. longifolia* and *A. floribunda* cultivated as ornamentals in Europe, although the effects may be held in check by insecticides already in use to protect such cultivation from other insects.

The wasps are reported to have spread to plantations of the commercially important tree species *A. melanoxylon* (Dennill et al., 1993). However, the further away from stands of *A. longifolia*, the lower the levels of non-target infestation compared with *A. longifolia*.

Dennill et al. (1993) concluded that the chance of negative impacts on what is a valuable timber tree (*A. melanoxylon*) in South Africa is very low, as the incidence of galling and the gall mass is too low. This is also the case for *P. lophanta*.

In summary, the non-target effects in South Africa have been shown to be negligible and temporary (Dennill et al., 1999).

6.6.3.3. Other potential plant health effects of *Trichilogaster acaciaelongifoliae*

Trade consequences

Of the ornamental *Acacia* species in the trade, the main ones are *A. dealbata* and *A. retinodes* (Table 1; Derkx et al., 2015). *A. dealbata* has been tested and is not a host of *T. acaciaelongifoliae* (Appendix B). To date, limited testing has been done on *A. retinodes* (Section 6.3.1; Helia Marchante, 10 March 2015, University of Coimbra, Portugal, personal communication). According to Derkx et al. (2015), in Australia, *A. retinodes* is not considered to be a host of *T. acaciaelongifoliae*. *A. retinodes* (flowers in capitulae) is morphologically very distinct from *A. longifolia* (flowers in spikes) (Derkx et al., 2015). Moreover, *A. longifolia* does not belong to the same phylogenetic section of *A. retinodes*: while *A. longifolia* is from the subgenus Juliflorae, *A. retinodes* is from the subgenus Phyllodineae (Helia Marchante, 10 March 2015, University of Coimbra, Portugal, personal communication).

There are potential consequences to the commercial trade of cultivated *A. longifolia* and *A. floribunda* (Table 1). However, these species are not native to the risk assessment area. Moreover, the ornamental use of these species would come under pressure from the BCA only if it spreads to the areas of cultivation. There is a trade within the EU in ornamental *A. longifolia* and *A. floribunda*, which could help spread the BCA and magnify its impact for ornamental traders, but the scale is limited (see Appendix C) compared with many other ornamental species (including other *Acacia* spp. that are not hosts of the BCA; see Table 1). Given their perennial nature, amenity plantings are more likely to be affected than ornamentals traded each year, because of the dynamic nature of this market. There are other ornamental acacias that could be used instead of *A. longifolia* and that are not affected by the

BCA. It is therefore considered that the flower production chain and trade in *Acacia* planting material has alternatives if the BCA did spread outside of the release area.

Unintended ecological consequences

It is unlikely that *T. acaciaelongifoliae* will have any significant direct effect on any plant species other than the target weed *A. longifolia* in Europe, based on the findings of host range testing and the experience from the native range of Australia and the introduced range of South Africa. A possible exception is *C. striatus* based on no-choice tests, which revealed significant oviposition, but no subsequent gall development. Moreover, choice tests using potted plants did not result in any egg laying (Marchante, submission to Portuguese authorities).

The unintended ecological consequences of species introductions are extremely difficult to predict or quantify. Most species live in a complex web of interactions, making it difficult to predict the response of even well-understood systems. Some ecologists even despair of finding general patterns (Holt and Hochberg, 2001). Even host-specific natural enemies have been implicated in negative environmental impacts, via mechanisms such as ecological replacement, compensatory responses and food web interactions (Pearson and Callaway, 2003).

In the case of weed biocontrol, the most commonly perceived potential problem is that of apparent competition (Holt, 1977). In this case, the apparent competition would be due to the presence of a generalist predator of *T. acaciaelongifoliae* whose population and behaviour would change as a result of this new resource. This could happen in two ways:

1. the generalist predator could build an artificially high population and return to its normal hosts, thereby reducing the population of the host ,which may already be rare;

2. the generalist predator could leave its usual host arthropod in favour of the new prey, allowing higher populations of the original arthropod host to build up, which may negatively impact on the host plant(s).

The consequences of apparent competition may be transient or permanent. Permanent effects are only likely to occur if the biocontrol agent is able to build up to high numbers without having an ultimate impact on the host plant population, which in turn would limit the biocontrol agent’s population. In the case of *T. acaciaelongifoliae*, evidence from South Africa suggests that persistent apparent competition is unlikely. However, it is possible that there may be some transient effects in the early stages of the programme when wasp populations may boom, but only if natural enemies are able to exploit this new food source.

Socio-economic consequences

Indirect economic, environmental and social effects include the reduction in dune stability (where *A. longifolia* has successfully fulfilled the role of dune stabiliser), the loss of shade/cover for animals or flower resources for pollinators (however, this might be recovered by restoring native communities), increased numbers of fires as a result of additional dead wood (likely to be a transient effect) and the reduction in the availability of *A. longifolia* branches for flower displays and firewood (although *A. longifolia* is reported to reduce the productivity and increase the management costs of forest plantations in Portugal).

These indirect consequences will depend on the magnitude of the direct consequences of the wasp on *A. longifolia* populations, and will not exceed the importance of direct effects, because of the transience of these indirect effects and the possibility for substitution (e.g. of flower resources provided by *A. longifolia*). Given that the wasp will reduce seed production but will not result in widespread *A. longifolia* mortality, indirect effects are expected to be minor.

6.6.4. Conclusion on the assessment of consequences

6.6.4.1. Consequences of *Trichilogaster acaciaelongifoliae* on invasive alien *Acacia longifolia*

	Descriptors
Rating: *massive*	• Reproductive potential, vegetative growth and ultimately population density of invasive alien *A. longifolia* are reduced substantially • Negative impacts of invasive alien *A. longifolia* on biodiversity, ecosystem functioning and services are reduced substantially • Negative impacts of current control measures of invasive alien *A. longifolia* are reduced substantially
Uncertainty: *medium*	Because of the unclear suitability of the climate to support high population densities of the BCA

6.6.4.2. Consequences to commercial trade of cultivated *Acacia longifolia* and *Acacia floribunda*

	Descriptors
Rating: *moderate*	• Any use of cultivated *A. longifolia* and *A. floribunda* would be affected by the BCA if it spreads to the areas of production • There is a trade in ornamental *A. longifolia* and *A. floribunda*, but the scale is limited (see Derkx et al., 2015) compared with many other ornamental species (including other *Acacia* spp.) that are not hosts of the BCA • Amenity plantings are more likely to be affected than ornamentals in a dynamic production chain and trade • Other ornamental *Acacia* species can be substituted for *A. longifolia*
Uncertainty: *medium*	Information on trade and control measures is missing

6.6.4.3. Consequences for other plant species

	Descriptors
Rating: *minor*	• Within the Mimosoidae subfamily, there has been extensive testing of host range, with the status of *A. retinodes* unclear, whereas *A. melanoxylon* and *P. lophanta* are identified as a spill-over hosts (Section 6.3.1) • In other subfamilies of the Fabaceae family, only *Cytisus striatus*, *Teline monspessulana* and *Spartium junceum* have been tested. For the last two plant species, there is no evidence that they are hosts • For *C. striatus* (see Section 6.3.1), because of the lack of robust information, there is uncertainty over its host status • *Vitis vinifera*, because of its importance, has been tested and found not to be a host
Uncertainty: *Low*	For species other than *A. retinodes* and *C. striatus* that have been tested
Medium to high	For *A. retinodes* and *C. striatus*

7. Identification and evaluation of risk reduction options

7.1. Options after entry

Currently, the only MS considering the release of *T. acaciaelongifoliae* into the natural environment is Portugal. The organism will be released at multiple sites, spread over the sandy coastal regions of central and northern Portugal, where *A. longifolia* is widespread and its invasive behaviour is most vigorous. It is envisaged that the wasp will disperse widely in the natural environment after release at a site. In this respect, no risk-reducing options in the plant health context are envisaged or proposed, except with regard to the care required in quarantine facilities and release protocols to prevent accidental release in situations and locations other than those intended. However, non-target plants in the vicinity of *A. longifolia* in the release area and other known areas should be monitored to detect any unexpected gall formation. In the area of release, (female) galls should be sampled at regular intervals as part of general monitoring for biocontrol effectiveness to determine whether or not other native organisms in the environment (e.g. symbionts, predators or parasitoids) are associated with the galls. Sentinel plants not normally present in the vicinity of *A. longifolia* can be deliberately planted for further monitoring of gall formation, subject to the usual risk assessment and local/national regulations. With appropriate cooperation among MSs, such sentinel plantings could represent non-target species present in other MSs, but not in the country where release is proposed. However, this is not really a risk-reduction option as it would be too late to do anything by the time any impacts in distant countries were observed.

As mentioned throughout this opinion, the evidence indicates that *T. acaciaelongifoliae* is mono-specific, surviving and reproducing only in *A. longifolia* and the closely-related *A. floribunda*. Female wasps that hatch and do not find host plants within three days will die without laying eggs. This greatly reduces the opportunities for uncontrolled dispersal of the organism, whether by natural or accidental human intervention. The deliberate collection of galls and their transfer to other locations (not as part of the approved release programme) by third parties might lead to an unplanned range expansion. *A. longifolia* is available from a very small number of nurseries in Europe, which would facilitate targeting control measures to reduce the risk that the trade in ornamental *A. longifolia* would lead to further spread of the organism.

As with the release of other (classical) BCAs, once it has established, there is no way in which dispersal to other sites, which are contiguous or close by the release site, can be prevented. Thus, in the case of release in Portugal, because of the close proximity of invasive alien *A. longifolia* in north-western Spain, the spread of the wasp to these areas is likely to occur. However, the geographical, topographical and habitat separation between pockets of invasive alien *A. longifolia* in the Iberian peninsula and the rest of southern Europe, even though climatic conditions may be suitable for the wasp, would make the dispersal of the wasp unlikely unless intentional. If this occurred, then the recipient country with known populations of *A. longifolia* would need to decide whether or not the plant is invasive in the locations where present and whether or not there are any reasons to prevent the establishment of the wasp as a BCA. The only option for this would be to monitor for galls and remove these before the emergence of the next generation of female adults. Insecticides would not be effective and BCAs would require a period of time before they would effectively control the wasp population. Treating the *A. longifolia* plants with herbicides would defeat the objective of trying to maintain their populations (the reason for not wanting the wasp to be established). If nurseries with *A. longifolia* were invaded, a grower could either start a control program based on pesticides at the time of female egg laying, or switch to species other than *A. longifolia*.

Biological control might be an option to reduce the unwanted impacts of *T. acaciaelongifoliae* on the ornamental production of *A. longifolia* in a sustainable way. The Natural History Museum, UK (Universal Chalcidoidea Database), reports that *T. acaciaelongifoliae* has parasitoids in the order Hymenoptera, family Eupelmidae (*Eupelmus* spp.) and family Torymidae (*Antistrophoplex* spp.). It remains to be studied whether or not the intentional release of this parasitoid could effectively control the bud-galling wasp under European conditions.

7.2. Conclusions

The PLH Panel has made a pest risk assessment for the intentional release of the bud-galling wasp *T. acacialongifoliae* for the biological control of the invasive alien plant *A. longifolia* (Andrews) Willd., specifically in coastal sand dune ecosystems of Portugal. The assessment excludes the assessment of the probability of entry and focuses on the risk of establishment and spread and the consequences for the EU territory. No systematic evaluation of risk reduction options was made.

The likelihood of establishment in the target area of release is rated as moderately likely, given the experience in South Africa, with the major constraint being the need to match the wasp's biological cycle with the plant's phenology in the northern hemisphere. The likelihood of spread and further establishment in non-target areas outside of Portugal, through either natural or intentional non-authorised human-assisted spread, was rated as moderately likely. The risk of inadvertent human-assisted spread was rated as low, but with high uncertainty.

The consequences of establishment of the wasp on invasive *A. longifolia* were rated as massive with medium uncertainty, whether in the target area of release or where the wasp spreads and establishes outside of this area. There would be minor consequences on populations of other invasive or ornamental *Acacia* spp. because of the wasp's high degree of specificity, although transient spill-over effects may occur. The one native wild species that needs further investigation, because of the current inconclusive nature of the data, is the broom, *C. striatus*.

The consequences for ornamental *Acacia* spp. are limited because only *A. longifolia* and *A. floribunda* are host species, with little cultivated production in Europe, compared with the main ornamental spp. *A. dealbata* and *A. saligna*. The species often named *A. floribunda* in Europe is actually the unrelated species *A. retinodes*, which has a different floral morphology. Further investigation is required for *A. retinodes* because of the inconclusive nature of the current data.

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APPENDICES

Appendix A. Ratings and descriptors

In order to follow the principle of transparency, as described in Paragraph 3.1 of the Guidance document on the harmonised framework for risk assessment (EFSA PLH Panel, 2010)—"… Transparency requires that the scoring system to be used is described in advance. This includes the number of ratings, the description of each rating … the Panel recognises the need for further development …"—the PLH Panel has developed specific rating descriptors for this opinion to provide clear justification when a rating is given.

A1. Ratings used in the conclusion of the risk assessment

In this opinion of the EFSA PLH Panel, a rating system of five levels, with corresponding descriptors, has been used to separately formulate conclusions on establishment, spread and impact, as described in Tables 4, 5 and 6.

Table 4: Ratings of the probability of establishment

Rating	Descriptors
Very unlikely	The likelihood of establishment would be very low because, even though the host plants are present in the risk assessment area, the environmental conditions are unsuitable and/or the host is susceptible for a very short time during the year; other considerable obstacles to establishment occur
Unlikely	The likelihood of establishment would be low because, even though the host plants are present in the risk assessment area, the environmental conditions are mostly unsuitable and/or the host is susceptible for a very short time during the year; other obstacles to establishment occur
Moderately likely	The likelihood of establishment would be moderate because, even though the host plants are present in the risk assessment area, the environmental conditions are frequently unsuitable and/or the host is susceptible for a short time; other obstacles to establishment may occur
Likely	The likelihood of establishment would be high because the host plants are present in the risk assessment area, they are susceptible for a long time during the year and the environmental conditions are frequently suitable; no other obstacles to establishment occur
Very likely	The likelihood of establishment would be very high because the host plants are present in the risk assessment area, they are susceptible for a long time during the year and the environmental conditions are suitable for most of the host growing season; no other obstacles to establishment occur. Alternatively, the pest has already been established in the risk assessment area

Table 5: Ratings of the probability of spread

Rating	Descriptors
Very unlikely	The likelihood of spread would be very low because the pest: • has only one specific way to spread which is not available/possible in the risk assessment area; and/or • highly effective barriers to spread exist; and/or • the host is not or is only occasionally present in the area of possible spread; and/or • the environmental conditions for infestation are unsuitable in the area of possible spread
Unlikely	The likelihood of spread would be low because the pest: • has one or only a few specific ways to spread and its occurrence in the risk assessment area is occasional; and/or • effective barriers to spread exist; and/or • the host is not frequently present in the area of possible spread; and/or • the environmental conditions for infestation are mostly unsuitable in the area of possible spread
Moderately likely	The likelihood of spread would be moderate because the pest: • has few specific ways to spread and its occurrence in the risk assessment area is limited; and/or • effective barriers to spread exist; and/or • the host is moderately present in the area of possible spread; and/or • the environmental conditions for infestation are frequently unsuitable in the area of possible spread
Likely	The likelihood of spread would be high because the pest: • has some unspecific ways to spread, which occur in the risk assessment area; and/or • no effective barriers to spread exist; and/or • the host is usually present in the area of possible spread; and/or • the environmental conditions for infestation are frequently suitable in the area of possible spread
Very likely	The likelihood of spread would be very high because the pest: • has multiple unspecific ways to spread, all of which occur in the risk assessment area; and/or • no effective barriers to spread exist; and/or • the host is widely present in the area of possible spread; and/or • the environmental conditions for infestation are mostly suitable in the area of possible spread

Table 6: Ratings of the magnitude of the potential consequences

Rating	Descriptors
Minimal	Differences in crop production (saleable fruits and leaves, cut branches with foliage, plants for planting) are within normal day-to-day variation; no additional control measures are required
Minor	Crop production (saleable fruits and leaves, cut branches with foliage, plants for planting) is rarely reduced or at a limited level; additional control measures are rarely necessary
Moderate	Crop production (saleable fruits and leaves, cut branches with foliage, plants for planting) is occasionally reduced to a limited extent; additional control measures are occasionally necessary
Major	Crop production (saleable fruits and leaves, cut branches with foliage, plants for planting) is frequently reduced to a significant extent; additional control measures are frequently necessary
Massive	Crop production (saleable fruits and leaves, cut branches with foliage, plants for planting) is always or almost always reduced to a very significant extent (severe crop losses that compromise the harvest); additional control measures are always necessary

A2. Ratings used for describing the level of uncertainty

For the risk assessment section—establishment, spread and impact—the level of uncertainty has been rated separately in coherence with the descriptors that have been defined specifically by the Panel in this opinion.

Table 7: Ratings used for describing the level of uncertainty

Rating	Descriptors
Low	No or little information or no or few data are missing, incomplete, inconsistent or conflicting. No subjective judgement is introduced. No unpublished data are used
Medium	Some information is missing or some data are missing, incomplete, inconsistent or conflicting. Subjective judgement is introduced with supporting evidence. Unpublished data are sometimes used
High	Most information is missing or most data are missing, incomplete, inconsistent or conflicting. Subjective judgement may be introduced without supporting evidence. Unpublished data are frequently used

Appendix B. Summary of tested hosts of *Trichilogaster acaciaelongifoliae*

Table 8 was compiled by the hearing expert, Helia Marchante, in July 2014.

Table 8: Summary of tested hosts of *Trichilogaster acaciaelongifoliae*

***Acacia*/*Mimosa* spp.**	**Tested by (P/SA)**	**Type of test**	**Number of replicates**	**Result (preferred host, host, non-host)**	**Reference**
A. longifolia (both subsp. *longifolia* and *sophorae*) [(a)]	Native range	–	–	Preferred host	Neser, 1982
A. floribunda (*A. longifolia* subsp. *floribunda*) [(a)]	Native range	–	–	Host	Neser, 1982
Tests in Portugal					
A. longifolia A	P	No-choice; paired-choice	Nine; nine	Preferred host; eggs detected on buds	Marchante et al., 2011
A. melanoxylon A	P	No-choice; paired-choice	Nine; nine	Host; eggs detected on buds	Marchante et al., 2011
Tests in South Africa [(b)]					Neser, 1982
1st experiment					
A. longifolia A	SA	Quarantine: females, ready to lay eggs, confined to potted plants (30–60 cm tall) observed for egg laying and 1 year for signs of gall development	Three replicates are likely to have been done—but we cannot say with certainty	Preferred host; galls	
A. melanoxylon A	SA			Non-host; probe observed	
A. baileyana A	SA			Non-host	
A. cyclops A	SA			Non-host	
A. dealbata A	SA			Non-host	
A. decurrens A	SA			Non-host	
A. elata A	SA			Non-host	
A. floribunda A	SA			Non-host	
A. implexa A	SA			Non-host	
A. mearnsii A	SA			Non-host	
A. neriifolia A	SA			Non-host	
A. podalyriifolia A	SA			Non-host	
A. saligna A	SA			Non-host	
A. davyi SA	SA			Non-host	
A. erubescens SA	SA			Non-host; probe observed	
A. exuvialis SA	SA			Non-host	
A. kirkii SA	SA			Non-host	
A. nigrescens SA	SA			Non-host	
A. schweinfurthii SA	SA			Non-host	
A. xanthophloea SA	SA			Non-host	

***Acacia*/*Mimosa* spp.**	**Tested by (P/SA)**	**Type of test**	**Number of replicates**	**Result (preferred host, host, non-host)**	**Reference**
2nd experiment					
A. longifolia A [(c)]	SA	Natural environment: females, ready to lay eggs, confined in double sleeve cages on living *Acacia* branches on mature trees; non-choice; branches observed for one year for gall development	Two replicates; 10 females/branch	Preferred host; galls	
A. caffra SA	SA			Non-host	
A. gerrardii SA	SA			Non-host	
A. hebeclada SA	SA			Non-host	
A. karroo SA	SA			Non-host	
A. nilotica SA	SA			Non-host	
A. reficiens SA	SA			Non-host	
A. robusta SA	SA			Non-host	
A. mellifera SA	SA			Non-host	
A. nigrescens SA	SA			Non-host	
A. permixta SA	SA			Non-host	
A. senegal SA	SA			Non-host	
A. tortilis SA	SA			Non-host	
3rd experiment					
A. longifolia A	SA	Plants including young growth exposed to the wasp in presence or absence of the host; plants with ca. one year included in pots with four species (multiple choice); 60 females in each cage until dead		Preferred host, galls	
A. albida SA	SA			Non-host	
A. schweinfurthii SA	SA			Non-host	
A. brevispica SA	SA			Non-host	
A. ataxancantha SA	SA			Non-host	
A. polyacantha SA	SA			Non-host	
A. hereroensis SA	SA			Non-host	
A. senegal SA	SA			Non-host	
A. montis-usti SA	SA			Non-host	
A. erubescens SA	SA			Non-host	
A. galpinii SA	SA			Non-host	
A. nigrescens SA	SA			Non-host	
A. burkei SA	SA			Non-host	
A. mellifera SA	SA			Non-host	
A. xanthophloea SA	SA			Non-host	
A. tortilis SA	SA			Non-host	
A. hebeclada SA	SA			Non-host	
A. stuhlmannii SA	SA			Non-host	
A. robusta SA	SA			Non-host	
A. haematoxylon SA	SA			Non-host	
A. erioloba SA	SA			Non-host	

Acacia/*Mimosa* spp.	Tested by (P/SA)	Type of test	Number of replicates	Result (preferred host, host, non-host)	Reference
A. nilotica SA	SA			Non-host	
A. karroo SA	SA			Non-host	
A. davyi SA	SA			Non-host	
A. exuvialis SA	SA			Non-host	
A. grandicornuta SA	SA			Non-host	
A. gerrardii SA	SA			Non-host	
A. sienerana var. *woodii*	SA			Non-host	
A. melanoxylon A	SA			Non-host	
A. baileyana A	SA			Non-host	
A. cyclops A	SA			Non-host	
A. dealbata A	SA			Non-host	
A. decurrens A	SA			Non-host	
A. elata A	SA			Non-host	
A. floribunda A	SA			Non-host	
A. implexa A	SA			Non-host	
A. mearnsii A	SA			Non-host	
A. neriifolia A	SA			Non-host	
A. podalyriifolia A	SA			Non-host	
A. saligna A	SA			Non-host	
South Africa—Field					
Paraserianthes lophanta (Mimosoidea)	SA	Sporadic galls—field		Non-suitable host	Dennil et al., 1993
A. melanoxylon	SA	Sporadic galls—field		Non-suitable host	Dennil et al., 1993

(a): Section Juliflorae, subgenus Heterophyllum—closely related.
(b): Specificity tests were conducted in South Africa using three different, complementary experimental procedures between 1977 and 1980.
(c): Potted, with similar sleeves, amongst test plants.
A, Australian Species; P, Portugal; *SA*, South African species; SA, South Africa.

Appendix C. Tables of occurrence of *Acacia floribunda* and *A. longifolia*

Tables 9 and 10 provide an overview of the occurrence in the wild and in the trade of *A. floribunda* and *A. longifolia* (Derkx et al., 2015). The details of the references are available in Derkx et al. (2015).

Table 9: Occurrence in the wild and in the trade of *Acacia floribunda*

EU MS	Occurrence in wild	Reference	Area of cultivation	Reference
Austria				
Belgium				
Bulgaria				
Croatia				
Cyprus				
Czech Republic				
Denmark				
Estonia				
Finland				
France			The true to name *A. floribunda* (Vent.) Willd. is offered by three nurseries	Florama, Pépinieres Cavatore, Pépinieres Saint Georges
France			Cultivated in Cels' garden in France	http://www.worldwidewattle.com/speciesgallery/floribunda.php?id=18286
Germany				
Greece			Four nurseries in Greece offer *A. floribunda*. In two cases, it certainly is *A. retinodes*; in the other two cases it cannot be determined, but most likely it is not *A. floribunda*	Fytopromitheytiki, Ergotech, Papaniki Nurseries, Delta-trees
Hungary				
Irish Republic				
Italy	Not recorded, either as casual or naturalised	G. Brundu, personal communication	Not recorded, but it cannot be excluded that it is kept somewhere as cultivated species	Giuseppe Brundu, University of Sassari, Italy, personal communication, Nov 2014
Italy			The true to name *A. floribunda* (Vent.) Willd. is offered by two nurseries	Viveros del Sueve, Arboles Ornamentales
Latvia				
Lithuania				
Luxembourg				
Malta				

EU MS	Occurrence in wild	Reference	Area of cultivation	Reference
Netherlands				
Poland				
Portugal	None	H. Marchante, personal communication	None	Helia Marchante, University of Coimbra, Portugal, personal communication, Nov 2014
Romania				
Slovakia				
Slovenia				
Spain				
Sweden				
UK				

Table 10: Occurrence in the wild and in the trade of *Acacia longifolia*

EU MS	Occurrence in wild	Reference	Area of cultivation	Reference
Austria				
Belgium				
Bulgaria				
Croatia				
Cyprus				
Czech Republic				
Denmark				
Estonia				
Finland				
France	Present in the departments of Corse, Gironde and Var	Tela Botanica		
France	Present, no further details	CABI	Offered by six nurseries	www.ppp-index.de
France	Corse: present, no further details	Vassal and Mouret, 1989; CABI	Offered by nine nurseries	Florama, Jardiland, Les Botaniques du Val Douve, Pépinière de Saint Jean, Pépinières de Kerzarc'h, Pépinières Cavatore, Pépinières Eric Duval, Pépinières Saint Georges, Pépinière La Palmeraie
France	France: alien, established; Corsica: alien, unknown	DAISIE		www.ppp-index.de
Germany			Offered by six nurseries	www.ppp-index.de
Greece			Offered by three nurseries	Best Gardens, Delta-trees, Vlachos Elias

EU MS	Occurrence in wild	Reference	Area of cultivation	Reference
Hungary				
Irish Republic			Offered by one nursery	http://www.rhs.org.uk/plants/search-form
Italy	Liguria (casual), Campania (naturalised), Sardinia (casual)	Altervista, Acta Plantarum, G. Brundu personal communication		
Italy	Present, no further details	CABI, ISSG	Offered by three nurseries	www.ppp-index.de
Italy	Italy: alien, established; Sardinia: alien, not established	DAISIE	Offered by 10 nurseries	Fattoria Beretta, Florsilva, Margheriti Piante, Piante and Vivai, Vivai MGF, Vivai Nannini, Vivaio Noaro, Vivaio Piante la Fronda and Vivai Torsanlorenzo
Italy	Naturalised	Celesti-Grapow et al., 2009, 2010		
Italy				
Latvia				
Lithuania				
Luxembourg				
Malta				
Netherlands			Offered by five nurseries	www.ppp-index.de
Poland				
Portugal	2 850 ha between Pedrogão and S. Jacinto (= 12 % of the 24 000 ha coastal strip). Dense stands in the dunes and interspersed as undergrowth in *Pinus pinaster* plantations	Kull et al., 2011		
Portugal	Mainland Portugal (Trás-os-Montes, Minho, Douro Litoral, Beira Litoral, Estremadura, Ribatejo, Alto Alentejo, Baixo Alentejo, Algarve), Azores archipelago (Santa Maria island), Madeira archipelago (islands of Madeira and Porto Santo)	Invasoras, DinamisGlobe		
Portugal	Present, no further details	CABI, ISSG		

EU MS	Occurrence in wild	Reference	Area of cultivation	Reference
Portugal	Portugal, Azores, Madeira: alien, established	DAISIE		
Portugal	Mechanical and chemical control undertaken in some areas against this widespread species	Brunel et al., 2013		
Romania				
Slovakia				
Slovenia				
Spain	Present, no further details	CABI, ISSG	Offered by two nurseries	www.ppp-index.de
Spain	Present, dangerous invasive behaviour	Dana et al., 2001, 2003	Offered by five nurseries	Alberola Viveros, Comunicatión Vegetal, Viveros del Sueve, Viveros Juan Peixoto and Viveros Pla del Poule
Spain	Ponteverda, Gerona (Blanes, Figueras), Alicante (Guardamar del Segura), Galicia. Up to 100 m altitude	MAGRAMA		
Spain	Spain: alien, established; Baleares: alien/not established	DAISIE		
Sweden				
UK	Geographic distribution: British Isles included	Weber, 2003	Grown in many gardens in Cornwall	Bean, 1970
UK			Not rare in the south-west	Krüssman, 1976
UK			Findings of the psyllid *Acizzia uncatoides* have been associated with imported *A. longifolia*, also one of the more hardy species of *Acacia* and capable of growing outdoors in the UK	http://www.fera.defra.gov.uk/plants/plantHealth/pestsDiseases/documents/accizzia.pdf
UK			It can be grown outdoors in many milder areas of the country, though, even in Cornwall, it is liable to be cut back to the ground in excessively cold winters. Tasmanian provenances are the hardiest forms in British gardens	http://www.pfaf.org/user/Plant.aspx?LatinName=Acacia±longifolia
UK			Offered by three nurseries	http://www.rhs.org.uk/plants/search-form

ABBREVIATIONS

BCA	biological control agent
Cfb	warm temperate, fully humid, warm summer climate
Csa	mild with dry, hot summer climate
Csb	mild with dry, warm summer climate
EPPO	European and Mediterranean Plant Protection Organization
EU	European Union
IAS	Invasive Alien Species
MS	Member State
PLH	Plant Health