SCIENTIFIC OPINION

Scientific Opinion on the Pest Risk Analysis on *Phytophthora ramorum* prepared by the FP6 project RAPRA[1]

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

European Food Safety Authority (EFSA), Parma, Italy

ABSTRACT

The Panel on Plant Health was asked to deliver a scientific opinion on the Pest Risk Analysis on *Phytophthora ramorum* prepared by the FP6 project RAPRA, taking into account comments by Member States and additional information since RAPRA. *P. ramorum* is the oomycete causing sudden oak death in the USA and leaf and twig blight/dieback on a range of ornamental species in North America and Europe. Currently *P. ramorum* is not listed as a harmful organism in Council Directive 2000/29/EC, but the Commission adopted in 2002 provisional emergency measures to prevent introduction into and spread within the EU. Recent large-scale outbreaks in Japanese larch (*Larix kaempferi*) plantations in the UK and Ireland have worsened the potential consequences in the risk assessment area. However, the Panel concludes that the broad narrative in the RAPRA report stands and supports its conclusion that "*There is a risk of further entry (of known or new lineages and/or mating types), establishment and [...] impact*". It is advisable to avoid introductions of different lineages because of inherent phenotypic differences and the potential for sexual recombination. The Panel supports the management options proposed in the RAPRA report and adds further measures for consideration. Uncertainty remains over the extent to which the association between control measures and gradual reduction in the number of cases in nurseries is causal. The emergency measures have not prevented outbreaks occurring in the natural environment. The many other remaining uncertainties (fitness of progeny, hybridisation with other *Phytophthora* species, host range and epidemiological role of new hosts, early detection of new outbreaks, understanding of long-range dispersal, structure of plant trade networks, origin of the pathogen) call for further research on *P. ramorum* across Europe. Regulatory work should keep updated with research results on *P. ramorum* and further development of the Japanese larch outbreaks.

KEY WORDS

SUMMARY

Following a request from the European Commission, the Panel on Plant Health was asked to deliver a scientific opinion on the Pest Risk Analysis on *Phytophthora ramorum* prepared by the FP6 project RAPRA. During the evaluation of the risk assessment and the risk management options, EFSA was requested to take into account the comments on the RAPRA PRA submitted by some Member States as well as additional information which had become available after the finalisation of the RAPRA PRA, such as the report of an outbreak of this organism on Japanese larch (*Larix kaempferi*) in South West England. *Phytophthora ramorum* Werres, De Cock and Man In't Veld (Class Oomycetes; Order Pythiales; Family Pythiaceae) is the fungal-like pathogen which is the causal agent of a condition known in the USA as sudden oak death. On other host species it causes disease symptoms described by the common name ramorum blight. Following its first isolation in 1993 in Germany and the Netherlands, its presence in the EU was officially reported for the first time on 29 April 2002 in the United Kingdom, Germany and the Netherlands.

Currently *P. ramorum* is not listed as a harmful organism in Council Directive 2000/29/EC[4], but the Commission adopted on 19 September 2002 provisional emergency measures to prevent the introduction into and the spread within the EU of *P. ramorum* (Commission Decision 2002/757/EC[5]).

The scope of this Scientific Opinion is the evaluation of the RAPRA report, taking into consideration the comments of Member States as well as additional information published after finalisation of the RAPRA report, or not cited in the RAPRA report. A systematic literature search until March 2011 was carried out. It should be noted that the scientific community is concerned by the shift of *P. ramorum* to new hosts (with a special focus on the Japanese larch) and its further spread, and therefore new results of ongoing research are constantly being published.

The evaluation was conducted in line with the principles described in EFSA PLH guidance documents (EFSA, 2009a; EFSA Panel on Plant Health (PLH), 2010). When evaluating the RAPRA report, the Panel followed either of the following options in terms of ratings made: develop a new rating when it does not agree with original rating and/or based on additional information a new rating is necessary; or leave the original rating if it was agreed with and no additional information had become available which would modify the rating. Member State comments were considered one by one, together with the RAPRA response to Member State comments. Although we are aware that, at the time the Member States made their comments, the additional information on *P. ramorum* now available was not available, we note that some of this additional information on *P. ramorum* was taken into account in the RAPRA response. In some cases this additional information was used in the opinion to respond to the Member State response and we made it clear when this was the case.

In terms of the risk assessment part of the RAPRA report, the Panel concludes that:

- There are large regions across Europe with climatic suitability to *P. ramorum* and presence of susceptible and sporulating hosts;
- There is still uncertainty about the precise host range, but little doubt that the pathogen is a generalist one, with many common and charismatic species among the highly susceptible hosts (e.g. *Fagus sylvatica*), and in heathlands;
- In addition to these points, already well recognised in the RAPRA report, there is the rapid development of the outbreaks in Japanese larch plantations in the UK and Ireland which makes the potential consequences in the risk assessment area much worse than what could be

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[4] Council Directive 2000/29/EC of 8 May 2000 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. 1-148.

[5] Commission Decision 2002/757/EC of 19 September 2002 on provisional emergency phytosanitary measures to prevent the introduction into and the spread within the Community of *Phytophthora ramorum* Werres, De Cock & Man in 't Veldsp. nov. OJ L 252, 20.9.2002, p. 37-39.

assessed only a couple of years ago. Numerous new plant species are being infected under or adjacent to Japanese larch, further broadening the host range of this pathogen.

In general the Panel concludes that the broad narrative in the RAPRA report stands and agrees with its conclusion that "*There is a risk of further entry (of known or new lineages and/or mating types), establishment and […] impact*".

This conclusion has been substantiated by the recent outbreaks on Japanese larch. In addition to this, the Panel notes that there is growing evidence of differences among lineages including mating type and virulence. Such variation may enhance the level of damage and the rate of spread of *P. ramorum*. It is therefore advisable to avoid introductions of different lineages because of such inherent differences and also because of the potential of sexual recombination.

Points that need further attention include: fitness of progeny, improved early detection of new outbreaks, understanding of long-range dispersal, origin of the pathogen, hybridisation with related *Phytophthora* species, host range and epidemiological role of new hosts as demonstrated by the *L. kaempferi* infestation.

The Panel supports the risk management options identified in the RAPRA report to reduce the likelihood of introduction of *P. ramorum* in the risk assessment area in consignments from USA, Canada and third countries that represent the pathogen's area(s) of origin. Following the Guidance on a harmonised framework for pest risk assessment and the identification of pest risk management options by EFSA, the following options should be reviewed:

Detection of P. ramorum in consignments by inspection or testing. Methods are available for the detection of *P. ramorum* in consignments. However, there is a non-zero likelihood of failing to detect the pathogen where it is present, despite the progress made by new research in developing reliable molecular diagnostic tools. The likelihood is worsened by the issue of asymptomatic infection and the variety of commodities to be controlled. Markers are available to identify the three *P. ramorum* lineages (NA1, NA2 and EU1) and should be used in consignments to prevent the introduction of alien lineages into the risk assessment area.

Removal of P. ramorum from the consignments by treatment or phytosanitary procedures. Various treatments have been tested to determine efficacy in eradicating *P. ramorum* from infested plant material. No treatment can guarantee the removal of *P. ramorum* from the consignments, with the exception of heat treatments (including composting) that were considered an effective option for sanitation of *P. ramorum* plant material. However, these kinds of treatments can be applied only on non-living commodities such as wood or bark. The use of fungicides may lower infection rates but will also reduce the efficacy of detection in consignments.

Surveillance. Surveillance strategies can be based on targeted inspection of susceptible plants including *Larix spp.* in nurseries, gardens, parks, woodland, heathland and forest. However strategies independent of symptoms should be developed due to the presence of asymptomatic or cryptic infections. Targeted control of super-connected plant trade firms would optimise effectiveness of surveillance schemes.

Prevention of infestation of the commodity. In the EU, eradication of the disease in nurseries is being attempted by destroying all infected plants within a 2-m radius of a diseased plant and holding all susceptible plants within a 10-m radius plus any remaining plants from the same consignment for further assessment. These measures are reasonably consistent with published information on dispersal of *P. ramorum* by plant-to-plant spread; however, dispersal through irrigation/water films remains poorly understood. Release of these plants is allowed following two negative visual inspections during 3 months of active growth and a suspension of any treatment that could suppress symptoms. There are uncertainties over the latent period of *P ramorum* on different plant tissues, hence the 3-month rule must be treated with caution.

Establishment and maintenance of pest freedom of crop, place of production or area. Since 2002, the European Union adopted emergency measures to prevent the introduction and movement of *P. ramorum* into and within the EU territory based on specific import requirements and "plant passport" certification systems. There has been some success with eradication in nursery crops. The structure of the nursery trade in Europe and its Member States is poorly known.

No management options have been identified to reduce the likelihood of spread following introduction to areas of susceptible plant hosts in woodland, heathland and forests, other than the removal of diseased plant hosts as exemplified by the removal of infected trees in the Japanese larch outbreaks in the UK and Ireland. Management may alternatively focus on the protection of important trees, deemed worthy of conservation, by removing healthy but potentially sporulating *P. ramorum* hosts around them. Preventive chemical treatments employing phosphonates are being used in the USA and are promising, although long-term effectiveness data are still missing.

Consideration of other possible measures. A key and difficult issue to be addressed with regard to the establishment and maintenance of pest freedom for *P. ramorum* is that of disinfestation of contaminated water circulation systems in plant nurseries.

Overall conclusions and synthesis

Since 2002, the European Union adopted emergency measures to prevent the introduction and movement of *P. ramorum* into and within the EU territory based on the "plant passport" certification system. There has been some success with eradication in plant nurseries. Uncertainty remains as to whether this pattern is a mere association; implying that a decrease in *P. ramorum* occurrences in the plant trade would have happened anyway, even without control measures; or whether there is a causal relationship between control measures and decreased nursery reports, implying that without control measures the occurrence of *P. ramorum* would not have declined as it did. The emergency measures have not prevented outbreaks occurring in the natural environment.

The Panel generally supports the risk management options proposed in the RAPRA report to reduce the likelihood of introduction of *P. ramorum* into the risk assessment area by consignments from USA, Canada and the third countries that represent the pathogen's area/s of origin. The RAPRA report did not take into consideration the outbreaks on Japanese larch in the UK, given that they had not yet occurred. For the reasons detailed in this opinion, these outbreaks are a major step change in the epidemiology of *P. ramorum* and the associated risk assessment and management issues. The further development of these outbreaks should be carefully monitored, studied and considered in future regulatory work.

The US APHIS PRA states that likelihood of introduction of *P. ramorum* is determined by three factors, namely; (1) entry potential which is defined as directly proportional to the volume of plants traded; (2) establishment and spread potential, determined by availability of suitable climate and susceptible hosts; and (3) detection potential which is negatively correlated to the failure to detect the organisms. While not much can be done in terms of point (2) (although limited selective removal of hosts is ongoing in the US and in Europe), it appears clear that regulations have the power to decrease both the number of entries (e.g. by passporting plants and checking on their health status) and the number of failed detections (by improving sampling schemes and diagnostic assays), thus effectively limiting the introduction of the pathogen.

Many uncertainties regarding *P. ramorum* persist. This include (but are not limited to): (1) the lack of data related to the origin of *P. ramorum* and its occurrence in Asia; (2) the source of inoculum because of the occurrence of asymptomatic infested plants, a lack of a systematic surveillance approach, and a varying frequency of false negatives; although the detection tools have been improved in specificity and sensitivity; (3) the host range of *P. ramorum*, which is particularly worrying given the rapidity and the extension of the outbreaks on Japanese larch in the UK; and (4)

the virulence and the fitness of progeny. The many remaining uncertainties call for further research on *P. ramorum* across Europe.

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BACKGROUND AS PROVIDED BY DG SANCO EUROPEAN COMMISSION

The current common 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 EU or to be moved within the EU, the list of harmful organisms whose introduction into or spread within the EU is prohibited and the control measures to be carried out at the outer border of the EU on arrival of plants and plant products.

Phytophthora ramorum Werres, De Cock and Man In't Veld (Class Oomycetes; Order Pythiales; Familiy Pythiaceae) is the fungal-like pathogen which is the causal agent of a condition known in the USA as sudden oak death. Its presence in the EU was reported for the first time on 29 April 2002 in the United Kingdom, Germany and the Netherlands.

Currently *Phytophthora ramorum* is not listed as a harmful organism in Council Directive 2000/29/EC. However, a pest risk analysis carried out for the United Kingdom in 2002 showed that this harmful organism and its damaging effects could be of significant plant-health concern to the EU.

Consequently, the Commission adopted on 19 September 2002 provisional emergency measures to prevent the introduction into and the spread within the EU of *Phytophthora ramorum* (Commission Decision 2002/757/EC). The results of these measures have been assessed yearly, based on the surveys carried out by Member States and their notifications of the suspected occurrence or confirmed presence of this organism in their territory. In 2009 twelve Member States reported outbreaks of *Phytophthora ramorum* in their territory.

Provisional emergency measures against a plant harmful organism adopted by the Commission are meant to be, as indicated by their name, temporary measures put in place against an imminent danger of introduction into or spread within the EU of that harmful organism. Based on the experience gained from the application of these measures over a period of time a decision will be taken whether permanent measures are needed (and what type of measures). This decision needs to be based on a recent Pest Risk Analysis covering the whole territory of the EU, which takes into account the latest scientific and technical knowledge for this organism as well as its present distribution in the European Union and the experience gained from the implementation of the provisional emergency measures.

The Pest Risk Analysis (PRA) for *Phytophthora ramorum* prepared by the EU-funded Sixth Framework Programme (FP6) project entitled "Risk Assessment of *Phytophthora ramorum*, a newly recognised pathogen threat to Europe and the cause of sudden oak death in the USA" (Acronym: RAPRA; Contract Number 502672), which was published on 26 February 2009, could fulfil the above mentioned criteria. Following the presentation of this PRA at the Standing Committee on Plant Health and the comments received from Member States, it was decided to seek a scientific opinion from EFSA on this PRA before considering further steps in this matter.

TERMS OF REFERENCE AS PROVIDED BY DG SANCO EUROPEAN COMMISSION

EFSA is requested, pursuant to Article 29(1) and Article 22(5) of Regulation (EC) No 178/2002, to provide a scientific opinion on the Pest Risk Analysis for *Phytophthora ramorum* prepared by the FP6 RAPRA project. During the evaluation of the risk assessment and the risk management options, EFSA is requested to take into account the comments on the RAPRA PRA submitted by some Member States as well as additional information which has become available after the finalisation of the RAPRA PRA, such as the report of an outbreak of this organism on Japanese larch (*Larix kaempferi*) in South West England.

ASSESSMENT

1.   Introduction

1.1.   Purpose and Scope

This document presents an evaluation by the EFSA Scientific Panel on Plant Health of a pest risk analysis document (hereafter referred to as RAPRA report, available at: http://rapra.csl.gov.uk/) on *Phytophthora ramorum,* in response to a request from the European Commission.

The scope of the opinion is the evaluation of the RAPRA report, taking into consideration the comments of Member States as well as additional information that was published after finalisation of the RAPRA report (February 2009) or that was published before but, though relevant, was not cited in the RAPRA report.

The entire RAPRA report was scrutinised by the Panel, with special focus on the parts of the document commented on by Member States.

With reference to Member States comments, major and relevant comments are addressed by the Panel in its opinion. Since additional information has become available, some comments are no longer relevant and are addressed in the sections on additional information.

For the literature review, the following items were considered:

- Documents available before the finalisation of the RAPRA report, but not cited

- Additional information that became available after the finalisation of RAPRA

A systematic literature search was carried out until March 2011 recognising that the scientific community is concerned by the shift of *P. ramorum* to new hosts (with a special focus on the Japanese larch) and its spread within the EU. Therefore new results of ongoing research are constantly being published.

1.2.   The document under scrutiny

The RAPRA report presented for evaluation is a Pest Risk Analysis (PRA) for *Phytophthora ramorum* that was prepared by the EU-funded Sixth Framework Programme (FP6) project entitled "Risk Assessment of *Phytophthora ramorum*, a newly recognised pathogen threat to Europe and the cause of sudden oak death in the USA" (Acronym: RAPRA; Contract Number 502672). The PRA was prepared according to the EPPO Standard '*Guidelines on Pest Risk Analysis: Decision-support scheme for quarantine pests*' version 07-13727 (PM 5/3 (3). The document published on 26 February 2009 comprises 310 pages and includes 275 cited references. It is arranged in three parts: Stage 1 outlines the reason for preparation of a pest risk analysis as one outcome of the FP6 project mentioned above. Stage 2 documents the pest risk assessment. Stage 3 outlines the pest risk management options proposed.

During the evaluation process, the comments on the RAPRA report from Belgium, Slovenia, Spain, The Netherlands and Germany, and a response to the comments from Belgium prepared by the United Kingdom, were taken into account.

The chronology of the documents is as follows:

RAPRA published   26 February 2009
Belgium (ILVO) comments on RAPRA PRA   27 August 2009
RAPRA (UK, FERA) response to comments made by Belgium 19 February 2010
The Netherlands comments on RAPRA PRA   19 February 2010
Slovenia comments on RAPRA PRA   19 February 2010
Germany (JKI) comments on RAPRA PRA   28 June 2010
Spain comments on RAPRA PRA   not dated

1.3.   Methodology of evaluation

1.3.1.   Structure of the opinion

The individual chapters of the pest risk assessment chapter (apart from the chapter on pest categorisation) as well as the pest risk management chapter are divided into 6 main subsections.

(1) General comments of the Panel on the RAPRA report

(2) Specific comments of the Panel on the RAPRA report

(3) Summary of the Member States comments, RAPRA response to the comment at issue (if any), and reaction of the Panel to the comments.

(4) Additional information since the finalisation of the RAPRA project and additional relevant information not considered in the RAPRA report.

(5) Uncertainties associated with the specific chapter

(6) Key findings of the above subsections highlighting the most important considerations from which the conclusions have been derived.

1.3.2.   Guidance documents on evaluation and harmonised framework

The evaluation has been conducted in line with the principles described in the documents "Guidance on the evaluation of pest risk assessments and risk management options prepared to justify requests for phytosanitary measures under Council Directive 2000/29/EC[6]" (EFSA, 2009a) and Guidance on a harmonised framework for pest risk assessment and the identification and evaluation of pest risk management options" (EFSA Panel on Plant Health (PLH), 2010). In order to be in line with the latter guidance document, the Panel in its evaluation refrained from addressing economic considerations as these are outside its remit.

1.3.3.   Ratings

Given that it was prepared according to the EPPO scheme, the RAPRA report contains ratings of the individual questions of the scheme. When evaluating the RAPRA report, the Panel followed either of the following options in terms of rating:

- Develop new rating when there is disagreement with the original rating, and/or based on additional information a new rating is necessary;

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[6] Council Directive 2000/29/EC of 8 May 2000 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. 1-148.

- Leave original rating if there is agreement and no additional information has become available which would modify the rating.

1.3.4. Literature review

The Panel searched for available scientific literature on *P. ramorum* that appeared after the publication of the RAPRA report (February 2009) or had not been included even if published before its publication. Search tools such as Web of Science, Scopus, Google Scholar, and CABI Abstracts were used, applying the following keywords: ramorum, sudden oak death, tree fungal pathogen, oomycete. Lists of papers in press (online first) of relevant journals (e.g. *Canadian Journal of Forest Research, Environmental Management, European Journal of Plant Pathology, Forest Ecology and Management, Forest Science, Ecological Modelling, Ecology, Forest Pathology, Fungal Biology, Fungal Ecology, Journal of Phytopathology, Molecular Ecology, Molecular Plant Pathology, Mycologia, Nature, New Disease Reports, New Phytologist, Physiological and Molecular Plant Pathology, Phytopathology, Plant Disease, Plant Pathology*) were inspected. Papers citing older *P. ramorum* literature were sought for. Lists of references cited in new *P. ramorum* articles were scanned. Whilst the Panel cannot obviously guarantee to have retrieved all relevant literature on *P. ramorum* that appeared after the publication of the RAPRA report (given the many new studies on the topic appearing), every effort was made to find the most relevant recent information on the pathogen up to March 2011 (first review of the literature in August 2010, with update in November 2010 and in March 2011).

Personal communications are also cited in this opinion. In line with the EFSA guidance on transparency in risk assessment (EFSA, 2009b), these were cited only in specific instances where the information provided was highly pertinent to the issue in question and fulfilled scientific criteria.

1.3.5. Member States comments

Member States comments were considered one by one (apart from the minor comments of one Member State, which mainly repeated the major comments of the same Member State and were thus considered when dealing with the major comments to avoid repetition). A summary of the RAPRA response to Member States comments (this response is available for ILVO, Belgium) was included in the summary of the Member States comments. Although we are aware that at the time the Member States made their comments additional information on *P. ramorum* was not yet present, we note that some of this additional information on *P. ramorum* was taken into account by RAPRA when responding to some Member States comments. In some cases this additional information was also used in the opinion to respond to the Member States comments. We made it clear when this was the case.

2. Critical review of the document

2.1. Pest Risk Assessment

In this evaluation the Panel have carefully treated (1) the RAPRA report, (2) the Member States comments including the ILVO comments and (3) the responses to the latter, in a timely and transparent manner reflecting the additional information and scientific evidence that post dates each of these three "steps".

The RAPRA report used a draft of the European and Mediterranean Plant Protection Organisation (EPPO) Standard entitled 'Guidelines on Pest Risk Analysis: Decision making scheme for quarantine pests' (07-13727) (EPPO, 2007; Brunel and Petter, 2010) as a basis for its Pest Risk Analysis (PRA) for *P. ramorum*. The EPPO standard provides a scheme, based on a sequence of questions, for deciding whether an organism has the characteristics of a quarantine pest or not in relation to a specific risk assessment area.

The Panel did not comment on all the answers/ratings of the RAPRA report, but only on those where the Panel disagrees or is of the opinion that supplementary information is needed (at the level of knowledge when the RAPRA report was published).

As for the conclusions of the RAPRA report, the Panel's opinion, when in agreement, was also indicated.

2.1.1. Pest categorisation

The classification of an organism which is not yet present in an area as a quarantine organism, after assessing its potential impact by pest risk analysis on the basis of its effects on plants in other areas, is an accepted procedure under the IPPC and the WTO-SPS. When a new pest has already been introduced to parts of a risk assessment area and the observed impact (so far) is small, the explanation can also be, apart from the "minor impact pest" explanation, that the epidemic is still in its lag phase, or the pest has been introduced to a limited and sub-optimal host range. Such a situation is difficult to assess because some interests may push in the direction of classifying the pest as a pest of minor importance based on the limited experience with the pest in the risk assessment area, as the short term impact of phytosanitary measures in an early phase often will be higher than the damage impact caused by the pest thus far.

2.1.1.1. Identity of the pest (origin, genetic lineages)

The exotic nature of *P. ramorum* in North America and Europe was hypothesised since the discovery of its role as causal agent of sudden oak death (SOD) in 2000. A parallel hypothesis was formulated for European populations, when the identity of a new *Phytophthora* species from European nurseries and the SOD agent was ascertained (Rizzo et al., 2002). These hypotheses were based on the following observations: a) the species had never been described before in either continent (Werres et al., 2001; Rizzo et al., 2002), b) the distribution was either geographically limited (California) or clearly associated with the nursery trade (Europe), c) the very high susceptibility of hosts in California natural settings, confirmed by inoculations in controlled experiments (Rizzo et al., 2002), and the (d) segregation of different mating types in the two different continents (Brasier, 2003; Ivors et al., 2004).

Subsequent papers provided ample and convincing genetic evidence that the organism had been introduced in both continents:

- Ivors et al. (2004) showed that the genetic structure of California forest populations is extremely simplified, as expected of an organism subjected to a strong genetic bottleneck, but also indicated that California forest isolates were clearly distinct from European nursery isolates.

- A follow-up paper using microsatellites by Ivors et al. (2006) identified three genetically clearly distinct lineages in *P. ramorum*: the term lineage does not simply refer to a cluster or clade of genotypes, but to a group of genotypes that have undergone a significant independent evolutionary process. This independence has obvious potential implications as it may result in significant phenotypic diversity among lineages.

- The distribution of lineages was reported as follows (using the nomenclature suggested by Grünwald et al., 2009): NA1 in North American nurseries and forests, NA2 in North American nurseries, EU1 in European nurseries. The evidence presented in that study indicated that introduction in nature in the USA most likely had been unwittingly aided by the sale and/or movement of infected nursery plants. Moreover introduction in the nursery trade had to have occurred multiple times both in North America; as suggested by the presence of all three lineages, and in Europe; as suggested by the presence of genetically distinct clades within the single lineage present in European nurseries.

- The presence of multiple clades within the European EU1 lineage is best explained as the result of multiple introductions of distinct genotypes (Ivors et al., 2006) rather than as the result of a more "natural" diversification process of a native organism.

- Connectivity between nursery populations of the pathogen and populations present in wild (e.g. California forests) or semi-wild settings (parks in the EU) was immediately noticed (Garbelotto and Rizzo 2005). However, such correlation was not enough to determine whether escapes were from nurseries into the wild or vice versa.

- Mascheretti et al. (2008) provide the strongest evidence to date that the pathogen was introduced from nurseries into the wild, by showing that pathogen populations from nurseries are genetically ancestral to all California forest populations.

Entry in North America and EU through the nursery trade becomes thus an obvious inference from all published studies, but spread within North America and the EU is also documented to have occurred through the movement of infected ornamental plants as shown by the genetic analyses of Goss et al. (2009a) and Prospero et al. (2009) in North America. Dominance of a common genotype in Belgian nurseries (Vercauteren et al., 2010) implies a common origin, but could also be explained by plant movement. The presence of site specific genotypes (i.e. genotypes that are specific to single locations) both in Belgian nurseries (Vercauteren et al., 2010) and in California forests (Mascheretti et al., 2008, 2009) is suggestive of ongoing site-specific micro-evolutionary processes leading to local diversification, and more importantly of limited gene flow among most sites. Despite a clear nursery origin of California forest populations, it appears that plant nurseries no longer contribute genotypes to established wild populations as indicated by the lack of gene flow documented between recently established forest populations and nurseries, both in California and Oregon (Prospero et al., 2007; Mascheretti et al., 2008). Incidentally, nursery trade has been regulated in the last years in North America, potentially leading to minimal new introductions (APHIS, 2007).

The presence of genetic diversity within the pathogen *P. ramorum* warrants further consideration. Although evidence of within lineage differentiations has been presented both for wild and nursery populations (Ivors et al., 2006; Goss et al., 2009a; Mascheretti et al., 2008; Vercauteren et al., 2010) there is currently no published evidence of different phenotypes consistently associated with different genotypes within lineage. Conversely, at least two reports compare phenotypes of different lineages (Brasier and Kirk, 2004; Elliott et al., 2009b) and identify differences in mating type, growth rate, and virulence. Evidence presented at the sudden oak death COMTF meeting in 2010 in San Rafael, (Pathogen and Host Variability: How Will They Affect the Spread of SOD in Heterogeneous Environments in: http://www.suddenoakdeath.org/news-and-events/past-events/2010-annual-meeting/) indicates that in replicated experiments NA1 and NA2 display different virulence levels with NA2 being more aggressive than the widespread NA1. Considering the significant levels of genetic divergence among lineages of *P. ramorum* (Goss et al., 2009a), this difference is not surprising, and deserves consideration. In spite of clear differences among lineages on individual hosts (Appendix A.), the three known lineages are extremely similar in broad ecological and virulence traits (Elliott et al., 2009b), and there is no strong evidence suggesting the three lineages would each behave radically differently. This indicates that knowledge of ecology and biology of one lineage can be reasonably extended to other lineages.

However, sympatric presence of NA1/NA2 and EU1 lineages should be regarded as an additional potential threat because of the possibility of sexual reproduction leading to a differentiated progeny that may be selected upon (Boutet et al., 2010), and the production of hardy oospores resistant to adverse environmental conditions.

Most up to date evidence indicates lack of sexual reproduction both in North America (Ivors et al., 2006) and Europe (Vercauteren et al., 2010, 2011b). Nonetheless, Boutet et al. (2010) report that sexual reproduction is possible and that the progeny displays a range of virulence with the clear

potential for adaptive evolutionary process. Additionally, the formation of thick-walled oospores may increase the survival of the pathogen in adverse conditions as shown for *Phytophthora megasperma* (Juarez-Palacios et al., 1991). In California at least two additional foliar Phytophthoras are present, namely *P. nemorosa* and *P. pseudosyringae* (Wickland et al., 2008). These two species have overlapping host range and habitat, and cause symptoms that are undistinguishable from those produced by *P. ramorum*, but are remarkably less virulent. New evidence shows all three species to be exotic (Linzer et al., 2009), yet the less virulent *P. nemorosa* and *P. pseudosyringae* are much more broadly distributed and are found in habitats that are more extreme for temperature and water availability than *P. ramorum*. Because these species are homothallic, this suggests a role of oospores in enhanced survival in harsher habitats.

Although the available evidence points to an exotic nature of *P. ramorum* both in North America and Europe, the actual area of its origin still remains unknown. The recently reported finding of *P. lateralis* in an old-growth forest of *Chamaecyparis* in Taiwan (Brasier et al., 2010) is relevant for the issue of the geographic origin of *P. ramorum*. The two *Phytophthora* species (*P. lateralis* and *P. ramorum)* are phylogenetically related, show various common features (e.g. large chlamydospores, sporangia with short pedicels, production of stromata, a similar optimum growth temperature) and are thus likely to share a common region of origin. Brasier et al. (2010) point out that since *Chamaecyparis* is present both in Taiwan and Japan, both *Phytophthora* species could have originated from one of these two countries. The recent evidence strengthens the already settled case that *P. ramorum* is an exotic pathogen both in Europe and in North America (Hansen, 2010).

The recent report of *P. lateralis* causing root and aerial infections on *Chamaecyparis lawsoniana* hedgerow trees in Brittany, France, a region whose climate is very similar to the one of coastal Oregon, is noteworthy (Robin et al., 2011). The authors write that this is the first report of the pathogen outside nurseries apart from Oregon [and California, Kliejunas, 2010]. Given that *P. lateralis* is likely to be an aerial *Phytophthora* just as *P. ramorum*, Robin et al. (2011) argue that *P. lateralis* now poses a new plant health threat to European countries.

The comments of Member States and the PLH Panel related to the identity of the pathogen are summarised in Table 1.

Table 1: Member State comments and comments of the PLH Panel related to identity of *Phytophthora ramorum*

RAPRA page and topic	Member state comments	RAPRA response	Panel comments
48: functionality of the breeding system	*P. ramorum* progeny less aggressive than parents	Boutet et al. (2010) study shows a range in pathogenic behaviour in progeny, so selection pressures could well operate	Boutet et al. (2010) now published in Molecular Ecology
49-50: presence of *P. ramorum* in the risk assessment area	Presence of *P. ramorum* in nurseries understated	Pest is indeed present but not widely distributed, e.g. in the UK only 2/5% of nursery inspections have resulted positive. Latent presence cannot be quantified, and methodological differences among surveys in various states need also to be kept in mind	RAPRA response appears adequate; see also comment from Slovenia indicating an average of 7% of surveys in Slovenian nurseries positive for *P. ramorum* (2003/2009). Presence in 60 nurseries and almost 10 semi-wild sites reported by (Vercauteren et al., 2010)
51-52: potential economic damage through effects on plant health	Quantitative differences in behaviour of *P. ramorum* in America (major outbreak in forests) vs. Europe (some dead trees)	RAPRA stated that *P. ramorum* could cause significant damage, not that it already has caused it (although to some extent it has in Cornwall). Response also refers to the findings on Japanese larch	New development of UK *P. ramorum* outbreak in Japanese larch plantations (Brasier and Webber, 2010) makes the comments from Member State no longer relevant? Evidence provided by Elliott et al. (2009b) indicates European and North American lineages are comparable to one another in virulence and suggests history rather than biology explain differences between North America and Europe
51-52: same as above	Other *Phytophthora* species (*P. alni*, *cambivora*, *citricola*, *cinnamomi*) causing similar damage but not regulated	No requirement for RAPRA to compare *P. ramorum* with other *Phytophthora* species. There is enough information available on *P. ramorum*, which makes extrapolation from other species unnecessary	Literature on other *Phytophthora* species in relation to plant nurseries is available e.g. in Jung and Blaschke (2004) and Jung at al., (2009). Study by Harwood et al. (2009) on the benefit brought by control in nurseries in terms of reducing the country-wide spread of *P. ramorum*. Biology of *P. ramorum* requires attention as it is both a soilborne/waterborne and an airborne pathogen. The combination of these traits is new (Rizzo et al., 2005) and brings uncertainty in the potential damage to nurseries and risk of escape that warrant differential treatment. Possibility of interspecific hybridisation augmented by increasing number of *Phytophthora* species present in nurseries (Gibbs et al., 1999; Jung and Blaschke, 2004; Lilja et al., 2010)
52: same as above	Damage to nursery sector mainly due to phytosanitary measures (point backed by comment from another MS,	Norway's *P. ramorum* PRA (Oct 2009) considers that the pathogen "*is likely to have a moderate impact on the nurseries in the PRA area with current*	The RAPRA response leads to an independent opinion from Norway, and also refers below on a similar comment by a Member State to a study from Oregon, where the impact of *P. ramorum* on nursery stock and Christmas tree production was

	also in that case without data to support the statement)	*phytosanitary measures*." Without these regulations, major economic impact on nurseries is expected	quantified with and without control. From Europe, there is now the study by Harwood et al. (2009) which shows the role of control
52: same as above	Conclusion about classifying *P. ramorum* as a quarantine organism in Europe based on its effect on trees in America, before there were data on its risk for Europe. Member State states: "*If it hadn't caused high levels of mortality on specific US tree species, it would not have been made a quarantine organism*"	RAPRA here just providing summary of main elements leading to the conclusion that the pest presents a risk to the PRA area, more evidence is available in detail in previous *P. ramorum* PRA.	The classification of an organism which is not yet present in an area (and therefore, before there are data on its effects in the area), as a quarantine organism, as assessed by pest risk analysis on the basis of its effects on plants in other areas, is an accepted procedure under the IPPC and the WTO-SPS. When data for the area become available, the classification should of course be re-assessed. For the risk assessment area, studies on *P. ramorum* susceptibility of Mediterranean European tree and shrub species (by Moralejo et al., 2008; Vettraino et al., 2009) confirm RAPRA's view on the issue. Discrete but locally significant mortality of Fagaceae associated with rhododendron as a sporulating host was observed in the UK and the Netherlands (Brasier et al., 2004). Recently, after RAPRA was finished, widespread mortality of mature and juvenile trees were recorded in the outbreak of *P. ramorum* in Japanese larch plantations in the UK (Brasier and Webber, 2010).
52: same as above	The similarity of risk for European and American tree species is not supported by the data on the behaviour of *P. ramorum* in the European natural environment so far. The combination of epidemiological, climatic and host susceptibility factors limits the risk of *P. ramorum* to specific locations and habitats.	The combination of factors needed for a serious threat is provided in the relevant RAPRA section (sporulation, climate, host susceptibility).	The recent outbreak of *P. ramorum* in Japanese larch plantations in the UK contradicts the Member State statement that the risk of *P. ramorum* is limited to specific locations and habitats. Several studies present data on epidemiological, climatic and host susceptibility factors contributing to the risk of P. *ramorum* for larger areas than specific locations and habitats. Considering epidemiological and host susceptibility factors, inoculation studies (some presented in the RAPRA report) indicate that several plant species or genera widespread in the EU are sporulators or very susceptible trunk hosts (Hüberli et al., 2006). Inoculation studies by Tooley and Kyde (2007) and Tooley and Browning (2009) show that US Eastern forests, similar in many respects to Central European Forests, contain many susceptible and infectious hosts. These US hosts have close relatives in Europe, and sometimes are themselves present in Europe. Such is the case for *Robinia pseudoacacia*, identified as one of the potentially more infectious hosts known to date. Considering climatic factors, models using Climex indicate that some parts of Europe would be favourable to significant

			outbreaks (see RAPRA report). Considering the specificity of locations and habitats, not only forest areas but also heathland areas are at risk to damage by *P. ramorum*. Several ericaceous hosts such as Manzanita, *Vaccinium* and *Arbutus* species are susceptible to *P. ramorum* (Garbelotto et al., 2003) and heathland outbreaks by the similar *Phytophthora kernoviae* are known in the UK (Widmer, 2010). Tooley et al. (2004) report on variability in susceptibility within the genus *Vaccinium*, with some species at moderately high susceptibility. The RAPRA report refers to evidence that *Vaccinium myrtillus* (page 167) is infectious at levels inferior but comparable to Californian bay laurel. This combination of susceptibility and infectivity contributes to the risk of *P. ramorum* to heathland ecosystems in the risk assessment area.
52: same as above	Diversity of climate, habitat composition and fragmentation effects may slow down *P. ramorum* spread in Europe	Pathogen does have a broad host range and sporulating hosts do occur in climatically favourable areas. Moreover, new sporulating hosts are being discovered, so that fragmentation effects may be weaker than suggested by the MS	Effects of landscape fragmentation on *P. ramorum* spread have been studied in California (e.g. Ellis et al., 2010). Similar research should be undertaken in the EU. The pathogen may jump from infected habitat patches to other suitable patches due to movement of infected plants, soil or water. Evidence indicates that infested soil is inadvertently moved around by people (Davidson et al., 2005) and can be a source of infection for plants (Fichtner et al., 2009). Additionally, genetic studies combined with spatial autocorrelation analyses show that the pathogen can be effectively moved several miles from a source (Mascheretti et al., 2008).
53: potential for genotypes of the non-European lineages to enter, establish and spread in the EU.	Again on risk due to mating between two *P. ramorum* mating types, comment argues that RAPRA is speculating in an unbalanced way	RAPRA statements are balanced as they point out that sexual reproduction is unlikely, but introduction of non-European genotypes (irrespective of mating type) may end up in increasing *P. ramorum* fitness or virulence in Europe	Knowledge remains in its infancy on the issue, RAPRA appears to have provided a useful summary of the available evidence.

2.1.1.2. Occurrence in the risk assessment area (new occurrences since RAPRA)

P. ramorum findings outside of nurseries (including managed parks, gardens, public greens, woodlands, and forests) have now been reported in Europe not just from the Netherlands and the UK, but also from Belgium, Denmark, France, Germany, Ireland, Luxembourg, Norway, Serbia, Slovenia, Spain, and Switzerland (Sansford et al., 2010).

Compared with the situation in 2006, as shown on the RAPRA website at http://rapra.csl.gov.uk/objectives/wp1/2006Distribution.cfm, this means an increase of 8 European countries, of which 6 are Member States. However, already in 2004 Belgium, Slovenia and Switzerland had reported *P. ramorum* presence in the semi-natural environment: http://rapra.csl.gov.uk/objectives/wp1/2004Distribution.cfm. This raises the question of how long to wait before eradication efforts can make it possible to declare freedom from the pathogen in a certain country where *P. ramorum* has been detected outside of nurseries. This shifting baseline issue was also raised by the comments of a Member State on the RAPRA report.

Based on reports in the wild from the USA outside of California and Oregon, it appears that *P. ramorum* was reported in a drainage system in Mississippi in 2007 (COMTF 2007 Summary Report http://www.suddenoakdeath.org/pdf/2007Year-endSummary.pdf), presumably three years after it was introduced in 2004 (Garbelotto and Rizzo, 2005). This indicates that several years are needed to identify a new establishment or a re-establishment of the pathogen after the predictable bottleneck related to eradication efforts. Three years were needed in this particular case, so at a minimum three years of survey are necessary: if weather is dry that time lag will need to be prolonged significantly. The discovery of relatively advanced damage in Redwood National Park in California, in spite of ongoing surveys, indicates several years may be needed to identify a new infestation (COMTF October 2010 report; http://www.suddenoakdeath.org/news-and-events/current-newsletter/).

2.1.1.3. Regulatory status

Regulatory status in the EU

In the EU, *P. ramorum* provisional emergency measures were adopted by the Commission in September 2002. These measures aimed to prevent further introduction into and spread within the EU of *P. ramorum*. The EU *P. ramorum*-legislation was amended in April 2004 and revised again in March 2007, following changes in knowledge of host range and assessed risk. Currently, EU *P. ramorum* measures require official surveys, control of movements of the 3 most affected genera in the EU (*Rhododendron, Camellia* and *Viburnum*) and statutory action wherever *P. ramorum* is detected. A summary of the history of *P. ramorum*-regulation in Europe is provided by Hunter (2008).

Apart from the emergency measures, non-specific measures are listed in the Council Directive 2000/29/EC relating to:
- known host plants or potential hosts of *Phytophthora ramorum* originating from outside the Community;
- non-host plants for planting originating from outside the Community that might be contaminated with *Phytophthora ramorum*;
- soil and growing media as a commodity;
- foliage and cut branches of host plants of *Phytophthora ramorum* originating from outside the Community;
- susceptible isolated bark as a commodity;
- susceptible wood.

Regulatory status outside the EU

Regulations have been imposed in the USA and Canada.

In the USA all confirmed *P. ramorum* hosts are regulated, and nurseries selling these hosts are required to conduct regular surveys using a standardised sampling protocol and a combination of culturing and molecular techniques. If infected plants are found, destruction of the entire lot is required. In addition, a buffer zone of two metres around the infected lot must also be destroyed.
Once a nursery is found to be infested outside the known zone of infestation, a nursery perimeter survey using a standardised protocol is required to check for the potential escape of the pathogen in neighbouring areas. As reported elsewhere, it is often in waterways adjacent to nurseries that the pathogen is found. This indicates a close link between infestation of nursery plants and introduction of the pathogen in the water systems of production facilities. Tjosvold et al. (2009) show that use of infested water for irrigation purposes can lead to infection of plants, but a natural escape of the pathogen from waterways has yet to be proven, although it is not hard to imagine that foliar infection may occur during flooding of river banks. *P. ramorum* in fact can be easily baited from rivers, and during floods, leaves of plants can act as baits.

In all three cases, a reduction of disease incidence has been associated with regulations (Suslow 2008). Regulation allowed the identification of massive movement of the pathogen from California and Oregon into 22 states in 2005 and to identify water infestation in two states. Anecdotally since regulations started, no clear escape from nurseries into wild has been recorded in California and Oregon.

There are other regulations that may have consequences for exportation which are beyond the scope of this opinion, e.g. regulations in place in China, South Korea, New Zealand, and Mexico (Kliejunas, 2010).

2.1.1.4. Potential for establishment and spread in risk assessment area

Given the extent of the newly reported *P. ramorum* outbreaks on Japanese larch (*Larix kaempferi*) plantations in England and Wales, it is important to establish how widespread such plantations are in the various European countries (Figure 1). From the CABI Crop Protection Compendium, it appears that most European countries do have some plantations of Japanese larch. Some information is available for the following countries/regions: Ireland 3.4% of forest area (Anon, 2010a), 2,300 ha in Bavarian public forests (Staatswald) (Stimm, 2004), Norway 240 hectars (Øyen, 2006). For comparison, the area covered by Japanese larch plantations in England, Wales and Scotland is about 125,000 ha (Clark, 2010).
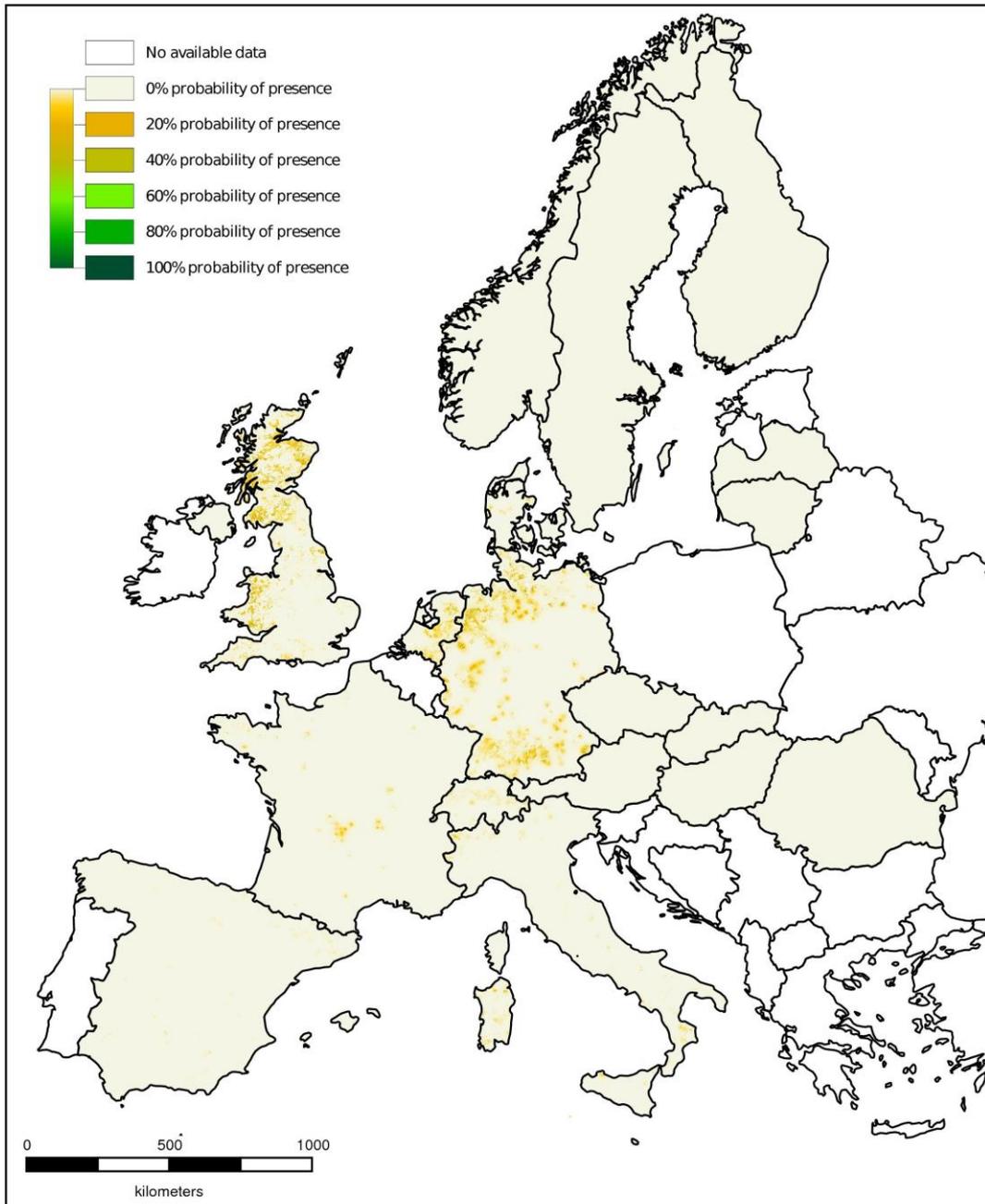
Distribution map of
Larix kaempferi Carr.
in Europe
Preliminary version 0.2 2011–03–17

Figure 1: Geographical distribution map of host species *Larix kaempferi* in Europe

The following maps (Figures 2 and 3) from UK Forest Research show not only the location of confirmed (red) and suspect (blue) *P. ramorum* outbreaks in UK Japanese larch plantations (as of September 2010 and January 2011), but also the distribution (pink) of (unaffected) Japanese larch plantations in the UK.
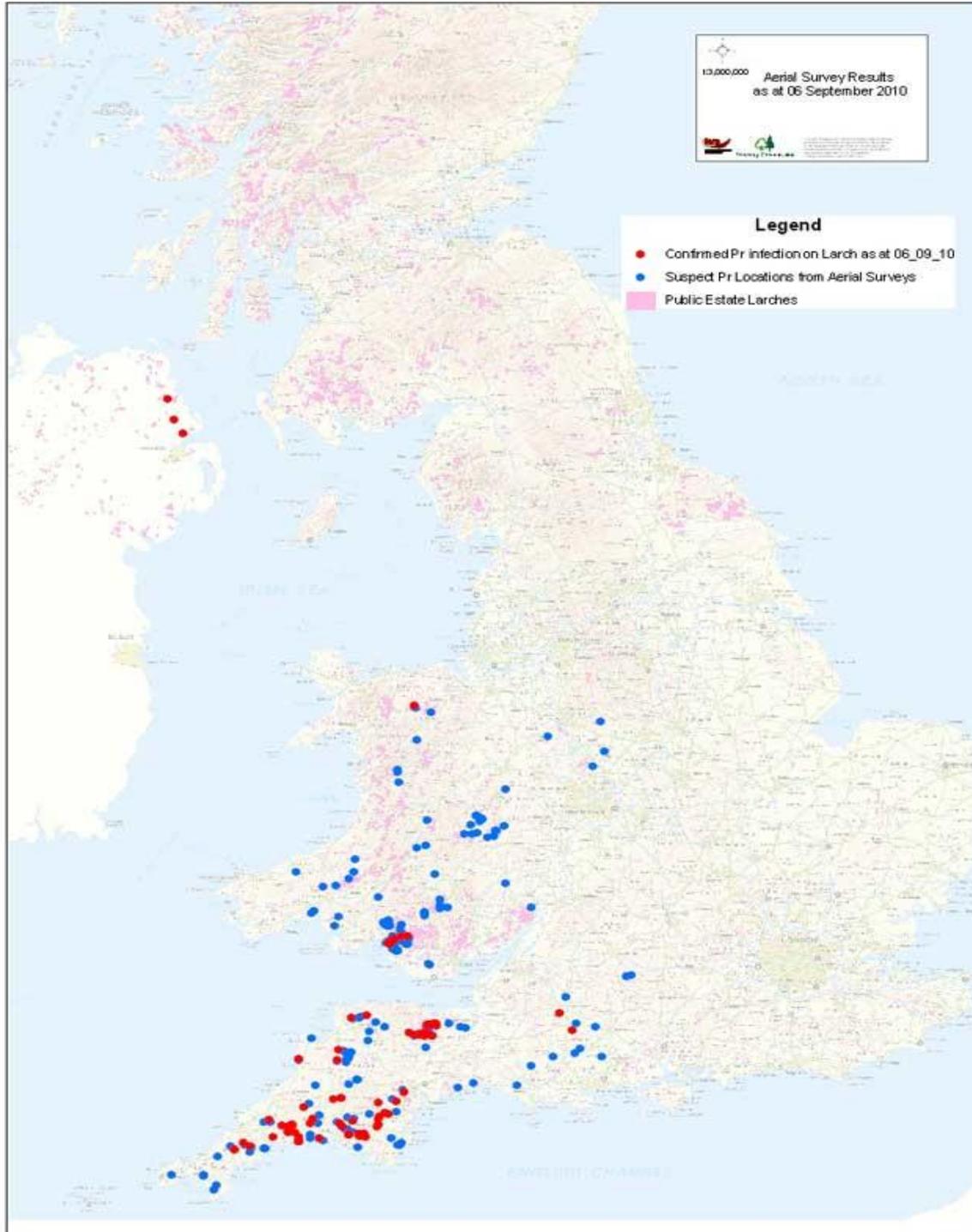
Figure 2: Location of confirmed (red) and suspect (blue) *Phytophthora ramorum* outbreaks in UK Japanese larch plantations as of September 2010 (by courtesy of the Forestry Commission).

Figure 3: Location of confirmed (red) and suspect (blue) *Phytophthora ramorum* outbreaks in UK Japanese larch plantations as of January 2011 (by courtesy of the Forestry Commission).

It is still unknown whether or not European larch (*Larix decidua*) is as susceptible to *P. ramorum* as Japanese larch has proved to be. One infected site in Wales has mixed Japanese larch, European larch and hybrid larch plots. From aerial photos *Larix decidua* had visible symptoms towards the end of 2010, so is highly suspect. But all the European larches turned colour 3 weeks early in an early autumn needle drop. These trees are due to be sampled in 2011 (Clive Brasier, personal communication, 2011). The natural distribution of European larch is mainly restricted to the Alps and the Carpathians (Figures 4 and 5). Although frost may not be expected to be a limiting factor for *P. ramorum* survival (Tooley et al., 2008), there is ample evidence both from the field and experimental settings (Davis et al., 2008) that the most favourable environmental conditions occur in the presence of rainfall and with temperatures around 20 ℃ (Davidson et al., 2005; Tooley et al., 2009). In their updated US national risk map for *P. ramorum* using climate, host and pathways data, Koch and Smith (2008) excluded areas with minimum temperature below 0 ℃ for at least 150 days (Kliejunas, 2010). Moreover, the report by DEFRA that *P. ramorum* spores are killed after 4 hours exposure to -25 ℃ (DEFRA, 2005) has been used in the mapping of *P. ramorum* risk across North America by Magarey and co-workers, obviously reducing the Northern extent of risk from the pathogen under current climate (Kliejunas, 2010). However, cold winters will probably reduce inoculum but are unlikely to eliminate it: some UK lab data suggest that about 80% of *P. ramorum* chlamydospores may survive freezing down to -10 ℃ (Clive Brasier, personal communication, 2011). It remains to be investigated whether European Larch is susceptible and whether it grows in areas that are favourable to epidemic outbreaks.
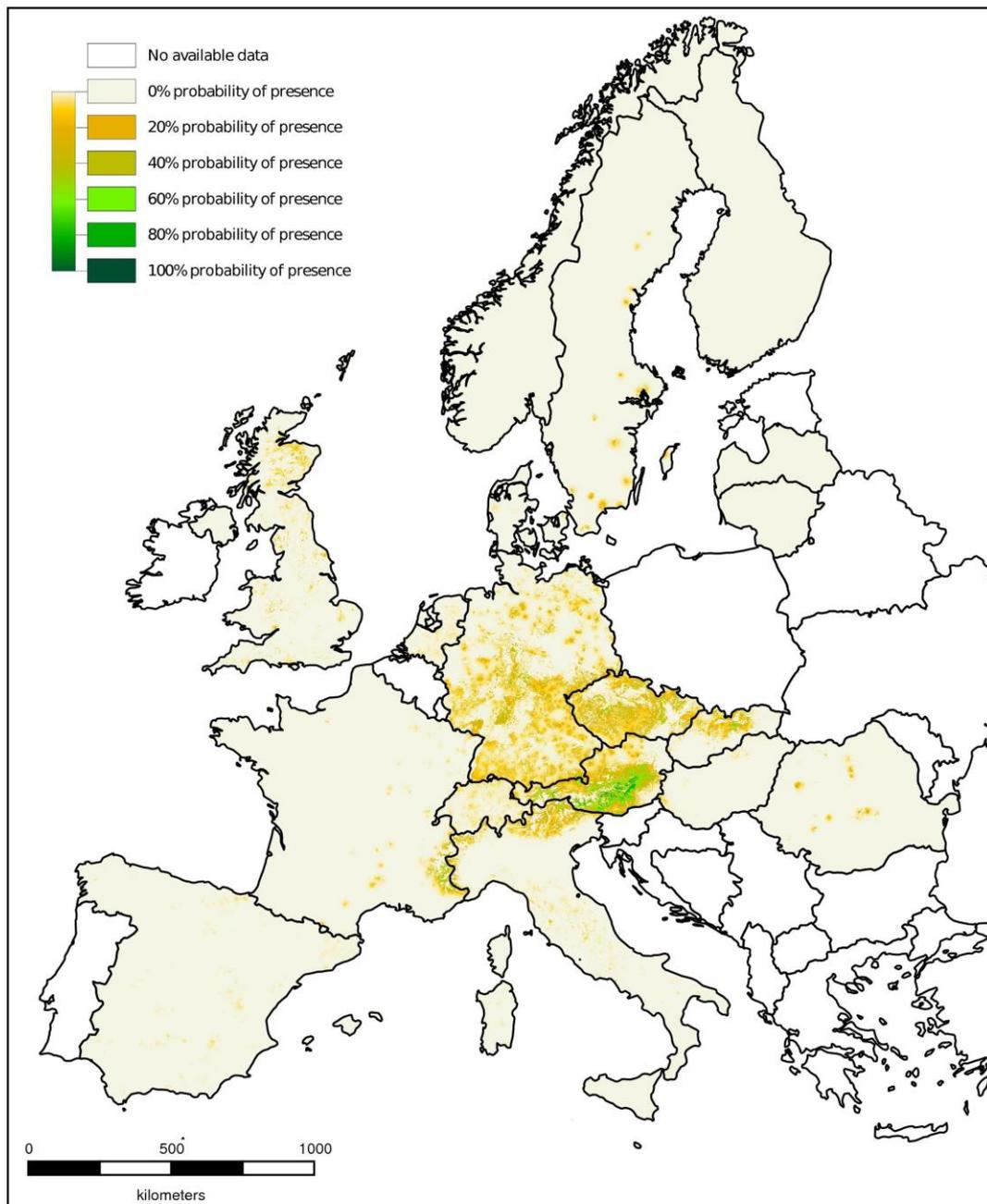
Distribution map of
Larix decidua Mill.
in Europe
Preliminary version 0.2   2011–03–17

Figure 4:   Geographical distribution map of host species *Larix decidua* in Europe.
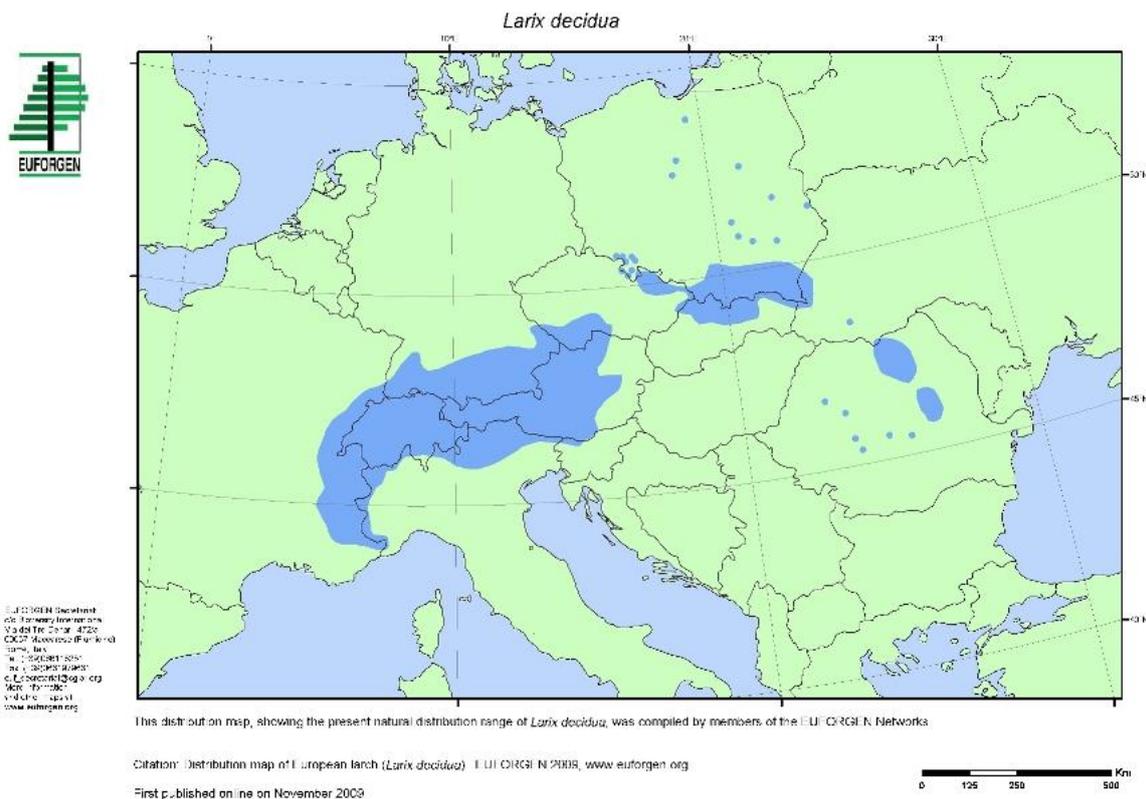
Figure 5: Geographical distribution map of host species *Larix decidua* in Europe (source: EUFORGEN).

In addition to Japanese larch, it has been observed that *P. ramorum* can indeed affect common heathland species such as bilberry (*Vaccinium myrtillus*), which had already been identified as potentially susceptible in the RAPRA report (Inman et al., 2005). Figures 6 and 7 show the widespread distribution of bilberry in the British Isles and in Europe.
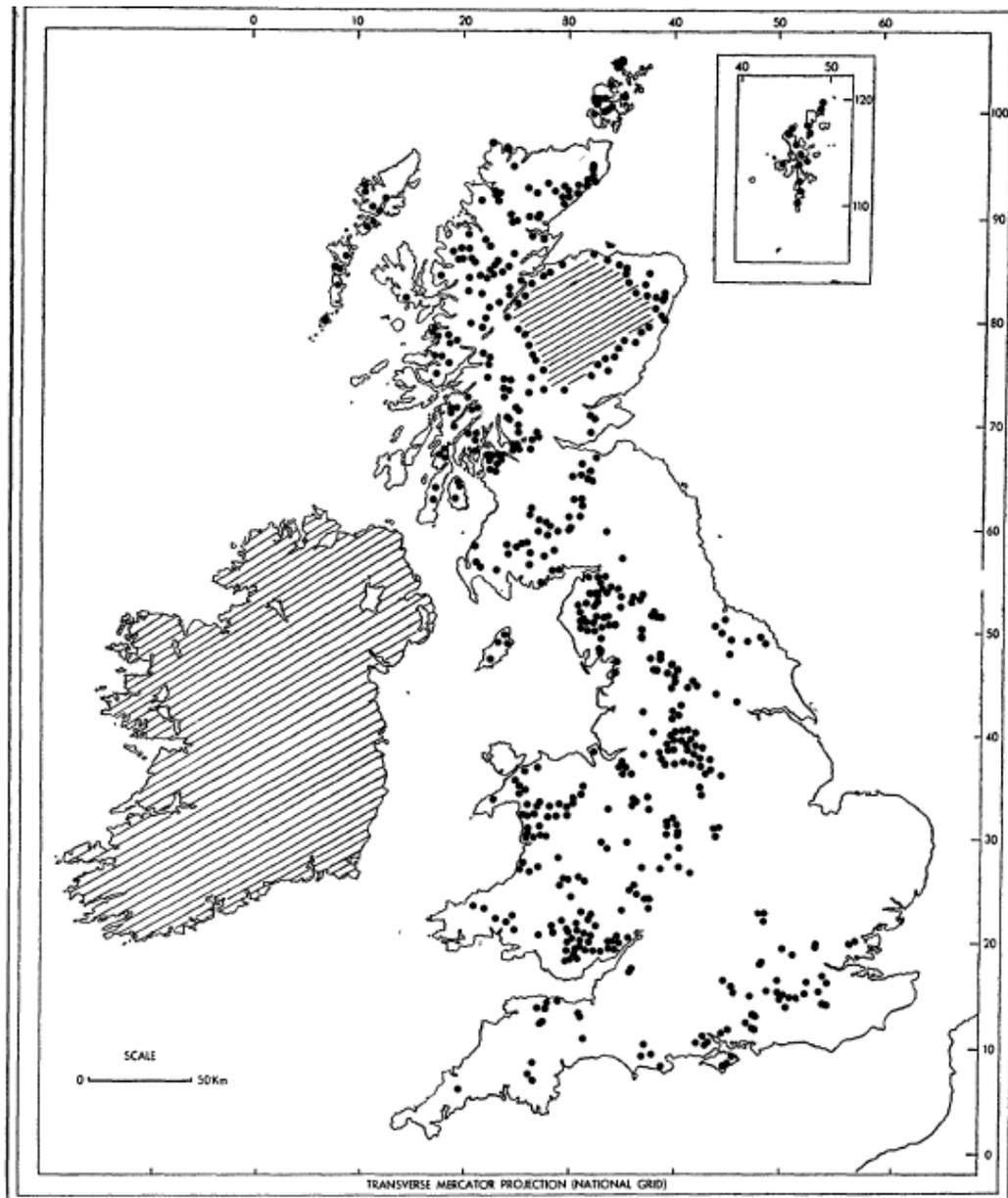
Figure 6: Geographical distribution map of host species *Vaccinium myrtillus* in the British Isles (from Ritchie, 1956, reproduced by permission from Wiley-Blackwell). Closed dots indicate single or aggregate localities and diagonal shading is used in areas where the distribution is either continuous or (as in the case of Ireland) inadequately known.
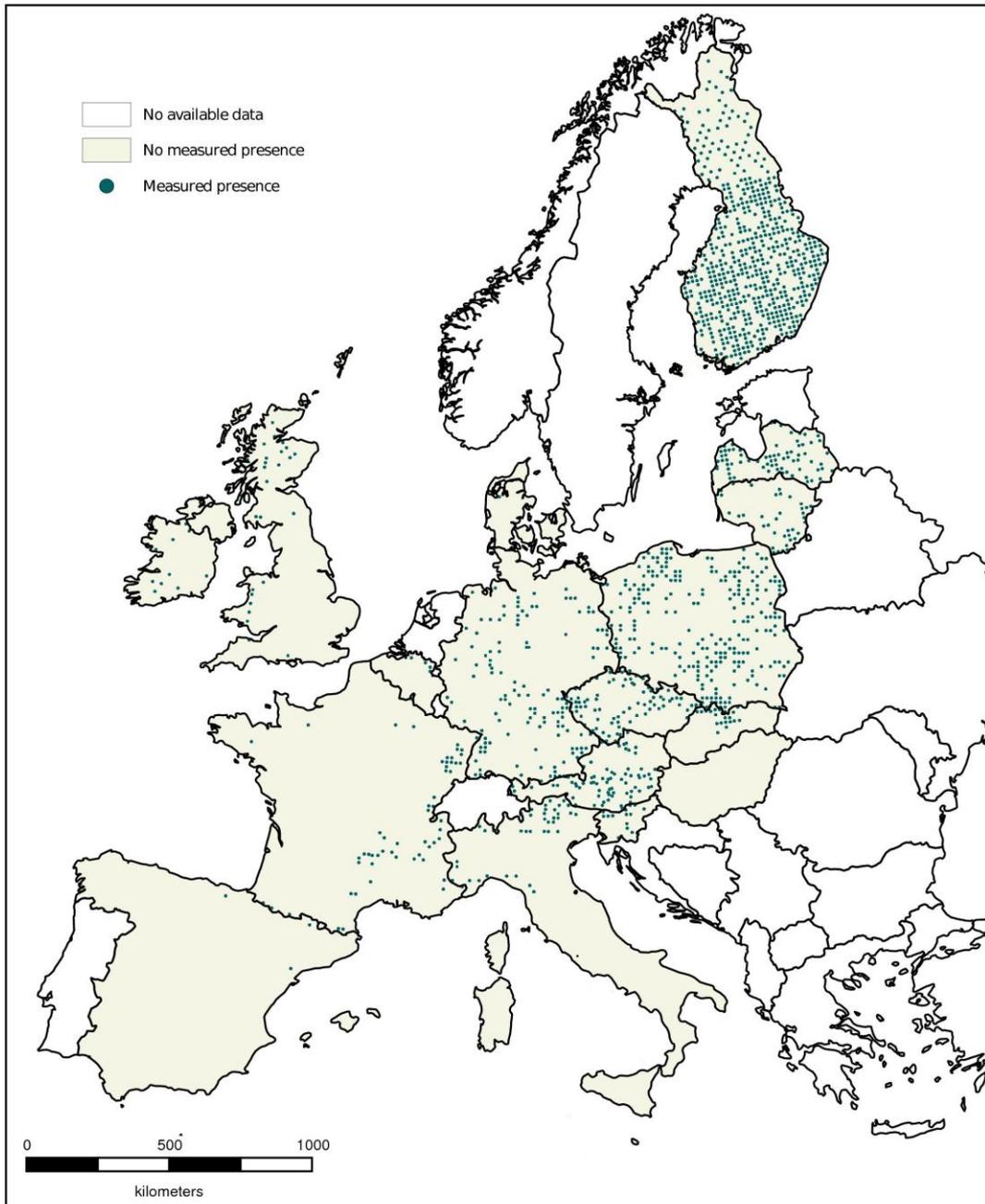
Presence of
Vaccinium myrtillus L.
in Europe
Preliminary version 0.2 2011–03–15

Figure 7:   Geographical distribution map of host species *Vaccinium myrtillus* in Europe

Although heathland cover has declined throughout Western Europe, this was once a common ecosystem along the Atlantic coasts from Spain to Norway (as shown in the map provided in RAPRA at p. 103, from the European Heathland Network of heathland around 1900). The dwindling presence of heathland in Europe makes the potential impacts of *P. ramorum* on this habitat all the more important to consider/alleviate (Figure 8).

Presence of
"Moors and heathland"
in Europe
Preliminary version 0.1   2011–03–29

Figure 8:   Presence of moors and heathland in Europe.

Ericaceous hosts are known to be susceptible hosts to *P. ramorum* in California (Garbelotto et al., 2003), and inoculation studies by Tooley et al. (2004) show several, although not all, ornamental *Vaccinium* species to be susceptible host species to *P. ramorum*. This combination of susceptibility and sporulation capacity makes ecosystems dominated by *Vaccinium* at high risk. See also the recent first report of *P. ramorum* on *V. myrtillus* in a semi-managed park (arboretum) with concurrent presence of *Rhododendron* (on which *P. ramorum* had been reported at the same location in 2005) on the SW coast of Norway, during the annual Norwegian *P. ramorum* survey in 2009 (Herrero et al., 2011). In addition, reports of *P. kernoviae* outbreaks in these ecosystems support the notion these habitats are at high risk of infection by *P. ramorum*.
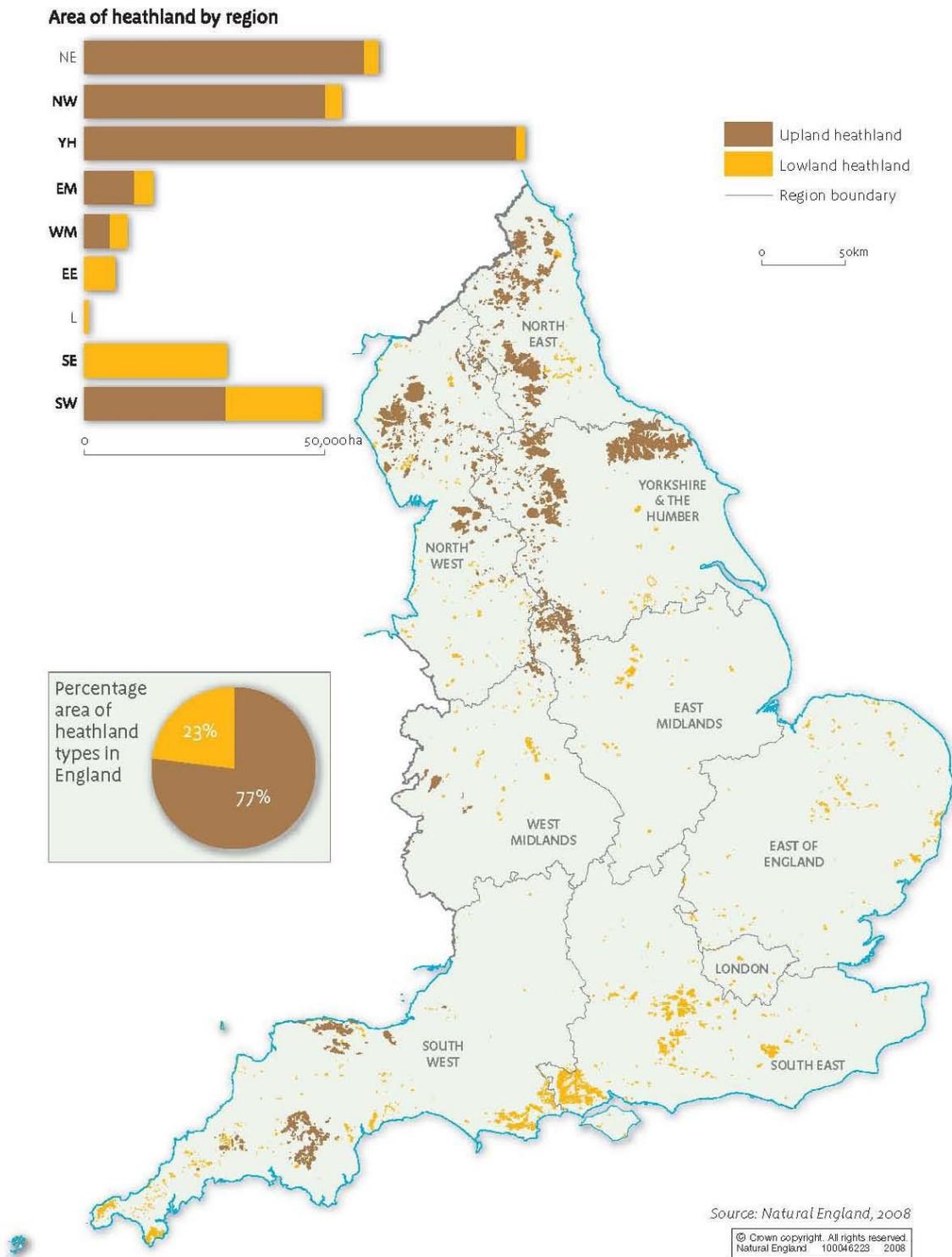
Figure 9: Extent of heathland in England (Supplied by Natural England © Crown Copyright 2008)

Britain is particularly proud of its heathland heritage, with several districts boasting the presence of lowland heathland of national significance, as shown in Figure 9.

Similar considerations apply to European beech (*Fagus sylvatica*; Figures 10 and 11), although this had already been recognised to be at risk in the RAPRA report, particularly in the presence of *Rhododendron ponticum* understory (map source: EUFORGEN). Beech is now however newly reported as a foliar host in the UK (updated August 2010 *P. ramorum* host list in the UK, http://thefera.co.uk/plants/plantHealth/documents/pRamorumHost10.pdf).
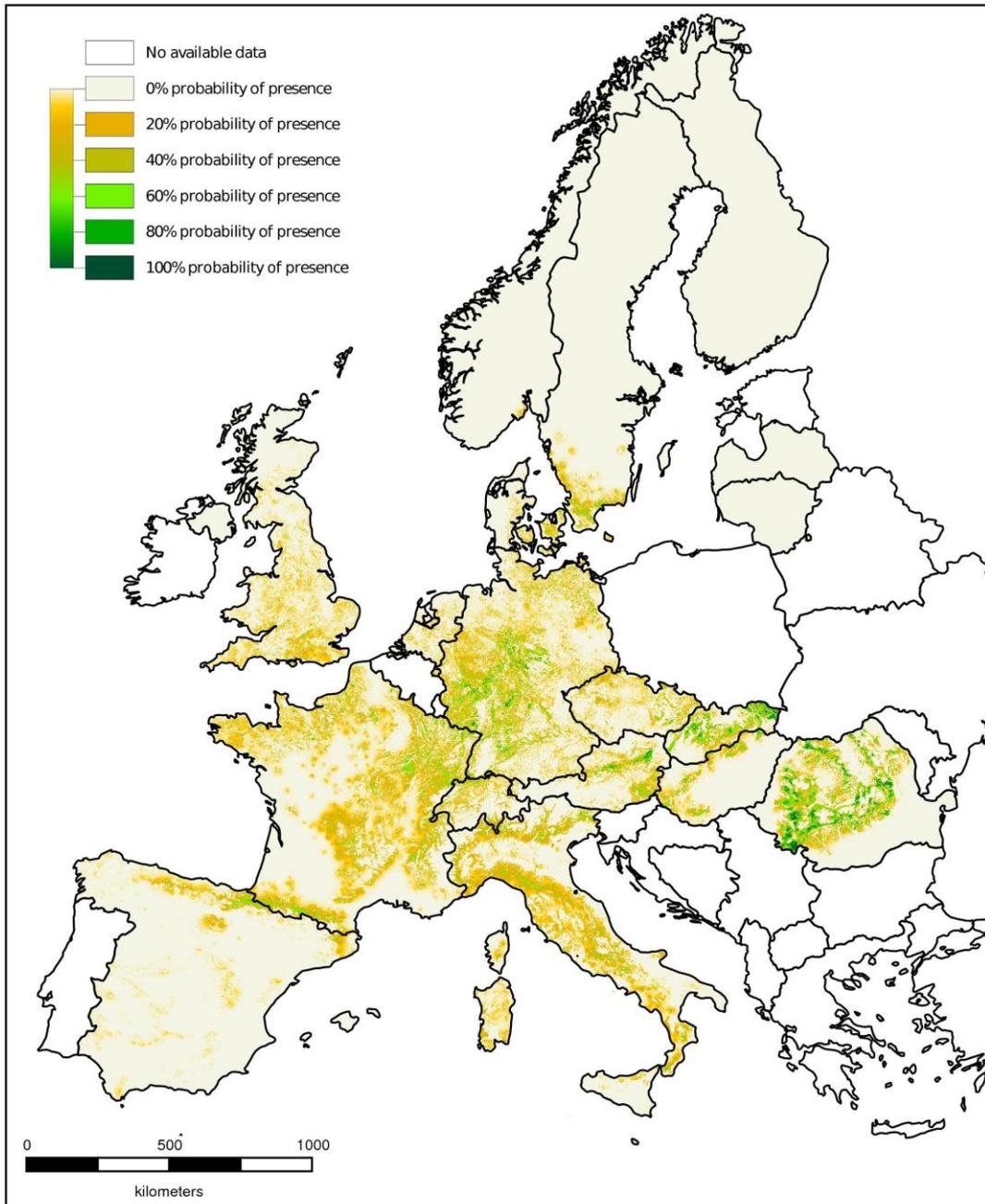
Distribution map of
Fagus sylvatica L.
in Europe
Preliminary version 0.2   2011–03–17

Figure 10: Geographical distribution map of host species *Fagus sylvatica* in Europe
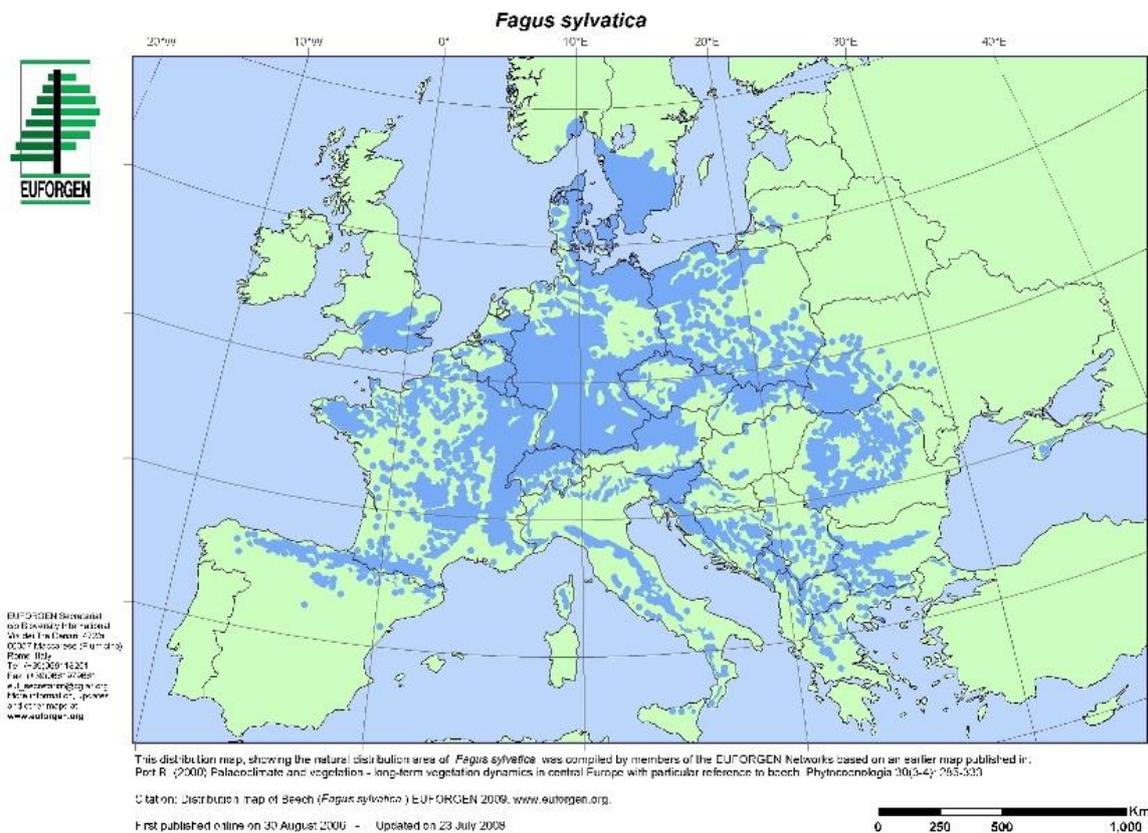
Figure 11: Geographical distribution of host species *Fagus sylvatica* in Europe (source: EUFORGEN)

Although the accuracy of EUFORGEN maps (available at: http://www.euforgen.org/distribution_maps.html) (as well as other freely available Europe-wide tree species distribution maps) varies due to differences in sampling effort among different European regions, they do provide a useful picture of the approximate natural distribution of the main European tree species, many of which are susceptible to *P. ramorum*. Of course, the presence of susceptible species is only one factor, also the occurrence of the pathogen in combination with suitable climate need to be there for disease to be expressed.

For example, sweet chestnut (*Castanea sativa*) is widely dispersed in Mediterranean countries (due to human planting since Ancient Roman times), but the following EUFORGEN map needs to be looked at for areas where climate is also favourable (based on CLIMEX there are many overlapping regions of host presence and climatic suitability to *P. ramorum*, from Galicia to Western France, from Italy to Western Turkey). Please note that in the following map (Fig. 12, first panel; distribution of sweet chestnut), the blue colour shows presence of the host species, whereas blue in the other maps denotes low climatic favourability to *P. ramorum*. Three different versions of European climate maps in relation to the potential favourability to *P. ramorum* are presented in Figure 12.
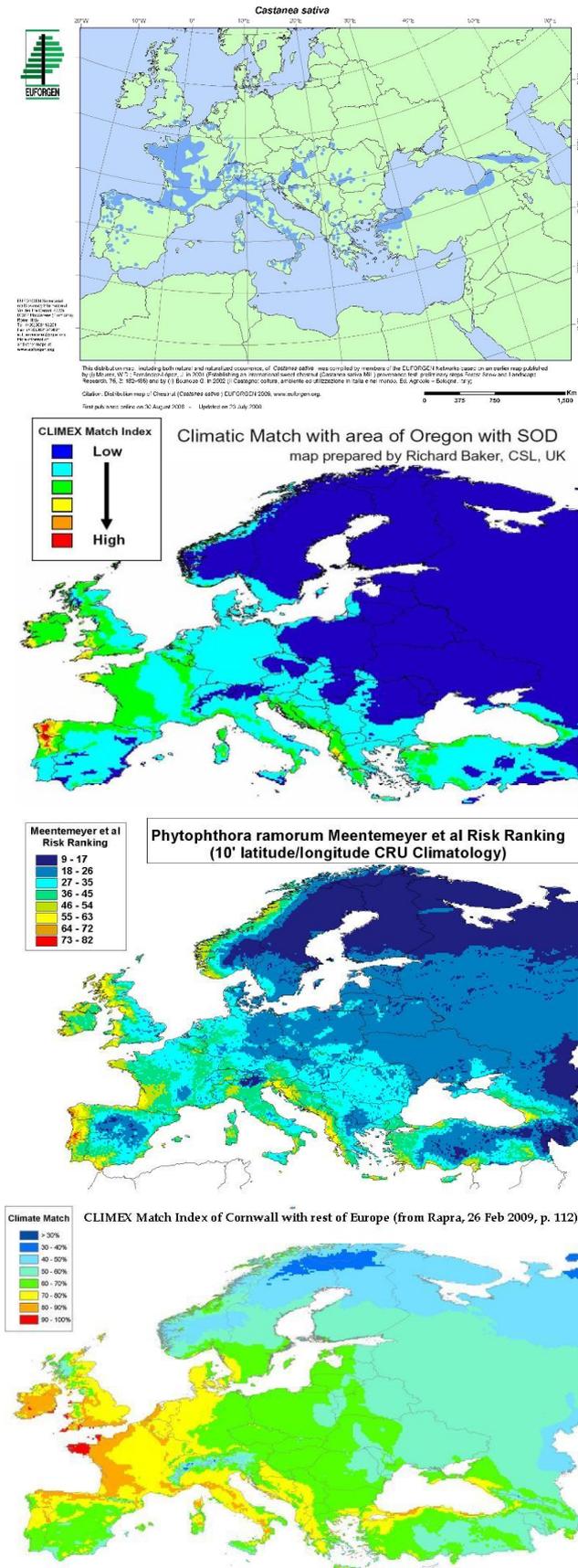
Figure 12: 1st panel: geographical distribution of host species *Castanea sativa* in Europe (EUFORGEN); 2nd panel = climatic match with Oregon; 3rd panel = abiotic variables favourable to

P. ramorum (minimum and maximum temperature, precipitation, relative humidity, see Table 15 of RAPRA); 4th panel = climatic match with Cornwall)

There is continuing work on mapping the global establishment potential of the organism (Ireland et al., 2010a).

2.1.1.5. Potential for consequences in the risk assessment area

- There are large regions across Europe with climatic suitability to *P. ramorum* and presence of susceptible and sporulating hosts.

- There is still uncertainty about the precise host range, but little doubt that the pathogen is a generalist one (Hüberli and Garbelotto, 2011), with many common and charismatic species among the highly susceptible hosts (e.g. *Fagus sylvatica*).

In addition to these points, already well recognised by RAPRA, there is the rapid development of the outbreaks in Japanese larch plantations in the UK and Ireland (Anon, 2010a; Brasier and Webber, 2010; Webber et al., 2010a, b) which makes the potential consequences in the risk assessment area potentially much worse than what could be assessed only a couple of years ago. The following hosts are being infected under or adjacent to Japanese larch (Clive Brasier, personal communication, 2011): rhododendron (*Rhododendron ponticum*), bilberry (*Vaccinium myrtillus*), European beech (*Fagus sylvatica*), Southern beech (*Nothofagus procera*), sweet chestnut (*Castanea sativa*), downy birch (*Betula pubescens*), sessile and pedunculate oak (*Quercus petraea*; *Q. robur*), Douglas fir (*Pseudotsuga menziesii*), Western hemlock (*Tsuga heterophylla*), fir (*Abies* spp.), and Lawson cypress (*Chamaecyparis lawsoniana*).

2.1.2. Potential consequences

2.1.2.1. General comments

Jung et al. (2009) provide an update on information available on *Phytophthora* diseases of trees (thus including *P. ramorum*, but not exclusively) between 2004 and 2007 (the year of the 4th IUFRO meeting on Phytophthoras in forest ecosystems). The Jung et al. (2009) review was not cited in the RAPRA assessment, as the proceedings were only published in 2009. Indeed, Jung et al. (2009) deliver an independent (albeit condensed) assessment of the RAPRA project. They write that the RAPRA project "*substantially increased our knowledge and understanding of pathogenicity, host range, distribution, and survival of P. ramorum and of the biological and climatic conditions favouring disease outbreaks.*"

2.1.2.2. Specific comments

2.1. <u>Impact on cultivated plants</u> (page 143 of RAPRA report). The Panel agrees that this is moderate for the nursery and the level of uncertainty is "*medium*". The impact on Japanese larch will be major with a low level of uncertainty, where the host occurs. The impact on overall timber production in the EU may no longer be considered minimal - at the very least, it must be minor (depending on the production of Japanese larch for timber; since the distribution of Japanese larch in many EU member states is not known, the level of uncertainty is medium). There is a potentially major impact on beech production where sporulating hosts are present.

2.2. <u>Effects on crop yield and/or quality</u> (page 148 of RAPRA report). Ratings range from minor to major depending on sector. The minor arises from the sub-section on timber production and must now be moderate, with the same qualification as in 2.1.

2.3. <u>Control without phytosanitary measures</u> (page 151 of RAPRA report). For the nurseries the ratings (with some difficulty, low uncertainty) are still valid. However the Panel argues that in managed forests and woodlands the rating should be "*with much difficulty*" with "*medium*" uncertainty (because the effectiveness of controls against Japanese larch and rhododendrons is not known).

2.4. <u>Increase in production costs (including control costs)</u> (page 152 of RAPRA report). The Panel cannot comment explicitly on economic costs, but again could certainly note that the PRA rating of "*minimal (zero)*" costs in commercial forestry, with low uncertainty, is now out of date.

2.8. - 2.9. <u>Social consequences</u> (page 161 of RAPRA report). There may be loss of recreational value in infested Japanese larch plantations.

2.10. <u>Export markets</u> (page 164 of RAPRA report). We argue that further trading restrictions are now likely rather than moderately likely, because of the timber situation, the apparent intercontinental movement of genotypes, and the unequal distribution of lineages within and between continents.

2.11. - 2.15. (page 165 of RAPRA report). It is not necessary to comment on these questions (not actually required in the original PRA).

2.16. (page 167 of RAPRA report). The Panel cannot comment explicitly on which areas are economically most at risk.

2.1.2.3.   Member State comments

Table 2:   Member State comments and comments of the PLH Panel related to the potential consequences of *P. ramorum*

RAPRA page and topic	Member State comments	RAPRA response	Panel comments
142-148: assessment of potential economic consequences	On the nursery sector not experiencing much damage from the pathogen (apart from phytosanitary measures). Member State also writes "*Within the nursery sector, P. ramorum is considered no more pathogenic than other nursery Phytophthoras such as P. cinnamomi and P. citricola.*"	MS comments unsubstantiated. Option of adopting no control measures was considered but not adopted in the UK public consultation (2008) for future management of *P. ramorum* and *P. kernoviae*. Study by Hall and Albers (2009) for the Oregon forest industry on costs associated with eradication/ control/no measures shows that the latter would be more costly by one order of magnitude	In the EU, nursery surveys have shown that less than 5% of nurseries in the single Member States are affected by the pathogen. But this proportion may have become higher in the absence of controls. In England and Wales, there have been a total of 901 outbreaks of *P. ramorum* between April 2002 and June 2009. Of the outbreaks, 261 have been in the wider environment with 85 (33%) of these now eradicated. At retail and productions sites there have been 640 outbreaks with 541 of these (85%) now eradicated (Tomlinson et al., 2009). The Member State further writes in the same comment that "*many nurseries already have been exposed to the pathogen ... this limits the new nursery area which is endangered for establishment and spread.*" This does not seem to take into account the efforts of nurseries to eradicate the pathogen from their premises when detected, and the majority of nurseries which have had no *P. ramorum* outbreak so far. As shown by the EC Standing Committee presentation (Jan, 2010), *P. ramorum* findings in nurseries are declining and represent a minority of tested nurseries, As far as the situation in North America is concerned, Mascheretti et al. (2009) show that the current distribution of *P. ramorum* in California is the result of at least eight different introductions. Multiple introductions are due to lack of regulation in the nursery trade. The study shows that allowing circulation of disease in nursery trade leads to a higher level of disease in the wild and semi-wild settings with undeniably broader impact on the environment
142-148: same as above	Costs estimated for the UK are mostly due to eradication efforts, there is no option with no regulation	Griesbach (2008) quantified the potential loss of sales of nursery stock and Christmas trees in Oregon in the case of the disease becoming endemic	Although EFSA does not deal with economic consequences, the Panel comments on this specific point raised by the Member States that no regulation may cause fewer costs to the nursery industry in the very short term, but also the costs for the semi-

		in the nursery industry. No data provided by the Member States to back up their claim	natural environment need to be considered. We also point to the recent studies on loss of house values in California due to *P. ramorum* (Kovacs et al., 2011a, b). Harwood et al. (2009) did simulate *P. ramorum* spread with and without control, and showed that control was essential to keep the epidemic under control (both in the trade and in the environment). COMTF reports indicate increasing detection of the pathogen in waterways adjacent to infested nurseries (http://www.suddenoakdeath.org/wp-content/uploads/2010/03/COMTF_Report_July_2010.pdf) thus further emphasising connectivity between nursery infestations and releases in the wild.
149: potential impact on yield/quality of ornamental species in nurseries	Only modelling (which is not surprising), no expert opinion used to justify the claim that costs would be major with low uncertainty for the nursery trade, if regulation of *P. ramorum* was lifted	Expert opinion of participants to the RAPRA project (economists, plant pathologists, experts on pest risk assessment) was used. If Member State is not surprised that rate of spread would increase in the absence of regulation, then the Member State should not be surprised at the score/level of uncertainty	Even if modelling does not lead to surprising results, it is still a useful tool. Surprising modelling is sometimes less trusted than unsurprising modelling, the important thing is whether modelling is built on reliable assumptions
153+ Costs of impacts with and without phytosanitary measures	Various comments disagreeing on economic impacts	The RAPRA response points out that Member State has either not read well the RAPRA report, or quoted some parts of it in a misleading way	One of the comments is on detection, on this issue there has been rapid progress in developing molecular tools (please see update on recent *P. ramorum* literature), hopefully these new tools will help in the control programme.

RAPRA page and topic	Member State comments	Panel Comments
52: Nursery situation	The *P. ramorum* situation in nurseries should also be compared with that of other *Phytophthora* species present in European nurseries	The RAPRA response to another Member State comment argues that this was not the purpose of RAPRA. Comparative information on *P. ramorum* vs. other *Phytophthora* species found in nurseries is available e.g. from the *Phytophthora* 2007 Symposium Proceedings, e.g. Jung et al. (2009). The number of new *Phytophthora* species detected in the last years suggests a need to keep on checking the literature related to *P. ramorum* over the coming years. The biology of *P. ramorum* is rather more complex than for other *Phytophthora* species. While a soilborne and waterborne phase remains, it is coupled with an airborne phase: this clearly singles out *P. ramorum* (Rizzo et al., 2005) as potentially different from other *Phytophthora* species already established in the

nursery trade. Because of its recent description, a differential treatment for this species seems justified.

53: - Outside nurseries – Damage	The Risk Analysis does not consider the different situations in the European countries: in UK it seems to be established outside nurseries, in NL and DE, where the pathogen has also been detected outside nurseries, there are only single detections without spread	Statement that *P. ramorum* "*seems*" to be established outside nurseries in the UK is now an understatement, given the extensive larch plantation outbreaks. These have now been reported also in Ireland (Southern and Northern), see sudden oak death Newsletter (September 2010). Japanese larch plantations are present in other European countries and thus need to be checked as a matter of priority. The Panel identifies the presence of susceptible and sporulating hosts in areas with favourable climatic conditions as conditions conducive to epidemic outbreak. It should be noted that the Oregon outbreak indicates that sporulating and susceptible hosts can actually be the same, as seems to be the case for the Larch outbreak in the UK (Note: not all susceptible hosts are also sporulating). The RAPRA report provides evidence for a range of sporulating and susceptible hosts both in Northern and Mediterranean climates. Finally, new information now convincingly shows that very low temperatures (up to -20C) do not eliminate the pathogen (Tooley et al., 2008) thus suggesting that infestations are possible in a broad geographic area.
53+: same as above	The economic damage caused by *P. ramorum* in nurseries is much less than the damage caused by the EU Decision	Please see the RAPRA response to the same comment from another MS. Although EFSA is not supposed to provide opinions on economic aspects, on this specific point raised by the MS, we note that this question will also depend on the time frame considered, and whether only damage to nurseries or also to the semi-natural environment is taken into account. Mascheretti et al. (2008, 2009) have proven that wild infestations (with all the ecological and economic damages associated with them) are directly linked to escapes from nursery settings or nursery plants. Therefore, control in the nursery trade will also result in a benefit measurable as costs avoided to protect natural environments.
63: *P. ramorum* epidemiological knowledge	Rhododendron, the plant considered to be the main host in Europe, is rarely killed by the pathogen	This Member State comment overlooks the role played by *Rhododendron* in building up *P. ramorum* inoculum, which can then lead to infection of many other native European plant species. The California forest epidemic and associated oak mortality is known to be driven by sporulation on bay laurel (Davidson et al., 2005) which does not seem to suffer significantly because of infection by the pathogen (DiLeo et al., 2009). A paper by Cobb et al. (2010) indicates that in presence of a foliar sporulating host (bay laurel) and a dead-end host (oak), *P. ramorum* actually results in an increase in the presence of the foliar host. This situation is going to lead to a progressive worsening of the epidemic. There seems to be a parallel with the Rhododendron/beech situation in Europe.

2.1.2.4. Additional information

The list of tree species with bleeding canker lesions due to *P. ramorum* in Europe (England and the Netherlands) reported in Jung et al. (2009) from the RAPRA database still does not include *Larix kaempferi* (Japanese larch). The *P. ramorum* outbreak on Japanese larch in England and Wales is a major novel finding since the RAPRA report, given the extent of the plantations of this exotic tree in Britain. An estimated 1,900 hectares of Japanese larch plantations (about 0.5 million trees) now show symptoms of *P. ramorum* infection in England and Wales (Brasier and Webber, 2010). The area covered by Japanese larch plantations in England and Wales is about 60,000 ha; there are some further 65,000 ha in Scotland (Clark, 2010).

Larix is however also not included in the APHIS regulated list of *P. ramorum* proven hosts or associated plants, updated February 2010, online at: http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/downloads/pdf_files/usdaprlist.pdf.

A study on the susceptibility of Oregon forests trees and shrubs to *Phytophthora ramorum* by Hansen et al. (2005) (not referred to in the RAPRA report, but listed in a list of hosts tested to *P. ramorum* by Woodhall and Sansford (2007)) did consider *Larix occidentalis*. The species was assessed as having (1) either resistance or low susceptibility to leaf dip in zoospore suspension, (2) resistance to whole plant dip in zoospore suspension, but (3) high susceptibility to stem wound inoculation with mycelial plugs.

Jung et al. (2009) write that up to 2007 "*stem infections have occurred only in woodlands where Rhododendron ponticum, itself an invasive species in the U.K., is a significant understorey species*," but this does not seem to be the case for the newly reported Japanese larch outbreaks, where *R. ponticum* is mostly lacking. The outbreak on Japanese larch in England and Wales in the absence of *Rhododendron* is thus an important development, as it immediately widens the susceptibility to *P. ramorum* outbreaks across (European) landscapes. It also makes the newly launched *P. ramorum* management plan in English and Welsh woodlands (Walters et al., 2010) in need of an update.

The study by Vettraino et al. (2009) concludes:

"*The presence of wide range of symptoms, even similar to those caused by other Phytophthora spp. [and] the broad host range associated with P. ramorum together with the presence of sporangia on symptomless hosts species could make difficult to detect the pathogen. For this reason potentially there is a very high risk of inadvertent introduction of P. ramorum plant through trade due to a hidden infection.*"

Together with the susceptibility studies on Iberian plant species carried out by Moralejo and colleagues (already cited in the RAPRA report, see also Moralejo et al., 2009), the Vettraino et al. (2009) study shows that there is a considerable potential for *P. ramorum* to become a major problem in Mediterranean ecosystems in case of inadvertent introduction through trade. Similar considerations apply to Australasia, where new *P. ramorum*-susceptible hosts have been reported (e.g. *Fuchsia excorticata* (foliage inoculations), *Nothofagus fusca* and *Pinus radiata* (both branch inoculations)) (Hüberli et al., 2009; see also Ireland et al., 2010b). That plant species commonly associated with laurel forests (e.g. *Castanea sativa*, *Erica scoparia, Pittosporum undulatum, Prunus lusitanica*) are potential *P. ramorum*-hosts was clearly acknowledged in the RAPRA report.

A study of tree mortality over a gradient of sudden oak death intensities (at eight sites in the greater San Francisco Bay Area - Marin, Sonoma, and Contra Costa counties) shows that coast live oaks (*Quercus agrifolia*) died between 1994 and 2004 at a rate of 4-5% per year, ten times higher than the background rate (Brown and Allen-Diaz 2009). This spike in coast live oak mortality is predicted to markedly lower its basal area in the studied forests. However other research documents a correlation

between variation among trees in date of largest lesions and variation in timing of bud burst and cambial phenology (Dodd et al., 2008).

In the same study, Dodd et al. (2008) report a genetic component in the size of lesions to coast live oaks caused by *P. ramorum* (already hinted at in a previous study, not considered in the RAPRA report, Dodd et al. (2005)). Although for *P. ramorum* in Europe and America there has not been host-pathogen co-evolution, there is still likely to be intraspecific genetic variation in susceptibility of the various host species (Nettel et al., 2009). This is an important issue, which is understudied (particularly in Europe), as it is likely to affect the long-term potential of plant hosts to adapt to *P. ramorum* (Pautasso, 2009; Hayden et al., 2010; Nagle et al., 2010, 2011). Similarly understudied are differences in virulence between isolates from infectious vs. dead-end hosts. Hüberli and Garbelotto (2011) have shown that isolates from dead-end hosts (oaks) were significantly less pathogenic than isolates from infectious hosts (bay laurel). There have been previous reports of quantitative differences in virulence of isolates from animal hosts playing different epidemiological roles, but this is a novel finding in botanical epidemiology (Hüberli and Garbelotto, 2011).

In other Californian forests affected by *P. ramorum* with dominant presence of tanoak (*Lithocarpus densiflorus*) and Californian bay laurel (*Umbellularia californica*), the high mortality experienced by tanoak is increasing the presence of bay laurel (Cobb et al., 2010), a host which is critical for further dispersal of *P. ramorum.* Bay laurel supports sporulation during the rainy season and provides a means for the pathogen to survive the dry, Mediterranean summer, but is not lethally affected by the pathogen (DiLeo et al., 2009). This shift in species composition is likely to lead to a positive feedback for the *P. ramorum* epidemic, i.e. an increased production of inoculum (Cobb et al., 2010). Research is needed to know whether such a development is likely to occur also in Europe.

In addition, new research has strengthened the potential link between sudden oak death and increased fire risk. In Californian forests where the disease is thought to have occurred since 1994, the foliar moisture content of tanoak is significantly lower in the presence of *P. ramorum* infection than without it (Kuljian and Varner, 2010). Model predictions show that this decrease in moisture content increases the risk of canopy fires (Kuljian and Varner, 2010). Crown fires can pose a major threat to human beings and ecosystem function. In addition to moisture decrease in the foliage, the increase in deadwood and fuels must be added (Metz et al., 2010, 2011). Obviously the point that the *P. ramorum* outbreak in California increases fire risk has been made before (e.g. by Hansen, 2008). It is likely that similar conclusions can be drawn for the effect on fire risk of a *P. ramorum* outbreak in the Mediterranean, where forests and shrublands are already particularly vulnerable to fires.

Researchers are starting to consider the implications of the *P. ramorum* outbreak in California for the carbon budget of forests (Madej, 2010). A comprehensive assessment of the carbon emissions caused by the introduction of this pathogen will need to take into account not only the carbon released following the death of millions of trees, but also the shift in tree species abundances and the increased likelihood of forest fires. Moreover, there is a recent confirmation that *P. ramorum* infection leads to Ambrosia beetle attacks and that these secondary infestations further reduce survival of infected trees (McPherson et al., 2010). In case of large-scale outbreaks on semi-natural vegetation in the European continent, similar mechanisms are also likely to operate.

The large-scale outbreaks of *P. ramorum* in the USA and now also in British and Irish *Larix kaempferi* plantations are of course not merely causing additional carbon emissions. In their updated pathogen profile (for time reasons, not cited in the RAPRA report), Grünwald et al. (2008) point out that

"in the USA, the economic impact of losses due to P. ramorum is estimated to be in the tens of millions of dollars due to the direct loss of nursery and ornamental crops, the decrease of property values due to dead/dying trees [see Kovacs et al. 2010], the cost of monitoring, tracking and eradicating the disease, the societal impact through loss of recreational value and cultural value, and the ecological impact through loss of food resources for native fauna."

On the issue of the wider environmental implications of *P. ramorum*, a recent study in California found that the presence of the pathogen in forests is associated with higher density of nymphal ticks. This is in turn likely to increase the risk of human exposure to Lyme disease (Swei et al., 2011). For the Eastern part of the USA, models of the invasion of *P. ramorum* in oak-dominated deciduous forests of the Southern Appalachians predict a marked decrease in the presence of *Quercus rubra*, with potential consequences to the many species depending on the masting of red oaks (Spaulding and Rieske, 2011).

2.1.2.5. Uncertainties

How high the risk of inadvertent *P. ramorum* introduction to ecosystems with a Mediterranean climate is, does not just depend on the presence and abundance of susceptible hosts in these ecosystems, but also on patterns in the European trade of plants susceptible to *P. ramorum*, the effectiveness of plant passporting and control schemes, as well as imponderable factors (e.g. awareness level of the population in the area of first introduction, number of initial infection foci, weather during the first years after introduction, resources available for eradication after detection, evolution of the international plant health regulation in the next few years (see MacLeod et al., 2010 for an update), sporulation potential of hosts in the new affected ecosystems, connectivity of the landscape from the point of view of *P. ramorum*, etc.).

Compared to North America, the impact of *P. ramorum* in the EU has so far been limited. One source of uncertainty is thus why this has been the case and whether this will continue to be so. There are various possible explanations for the lower impact of *P. ramorum* so far in the EU compared to North America:

- California and Southern Oregon provided a combination of suitable hosts that were infectious and/or susceptible, and a favourable climate, without extremes such as heat or frost. In addition, California has a different urban reality with towns and plant nurseries in a forest setting, while a more clear separation seems to occur in the EU, where nurseries are often in a more agricultural setting (as in the Netherlands) or in a more urban setting (as in the North-western part of Italy). Additionally, the 1:10 rule states that successful escapes are not very likely in the first place (e.g. Williamson and Fitter, 1996) and so it is understandable why *P. ramorum* has not escaped continuously, but once introduced as in the Japanese Larch case or in California, it can be very successful.

- Fewer susceptible hosts in Europe. Most evidence, including that provided by the RAPRA report, indicates that there are many susceptible and infectious hosts in Europe. In itself, this is enough to warrant a special consideration for this pathogen, but really what needs to happen is to determine where sporulating hosts are sympatric with susceptible hosts (sometimes the two hosts can be the same) in an area with favourable environmental factors, and to determine whether favourable environmental factors occur when phenology of the host makes them most susceptible as it has been shown for California oaks (Dodd et al., 2008). Because most inoculation tests are inevitably performed on a few individuals only, it may also be necessary to assess whether putative susceptibility occurs at the same level throughout populations of the host as has been done for coast live oaks (Dodd et al., 2005) and bay laurels (Anacker et al., 2008) in California.

- The *P. ramorum* epidemic in Europe may be still in its lag phase. It is clear now that *P. ramorum* infestations are virtually invisible for variable periods of time, depending on the affected ecosystem. If a foliar host is driving the epidemic (as in most California systems where bay laurel is a foliar host not significantly impacted by the disease, and in some European countries where Rhododendrons are driving the outbreaks) the lag phase can be very long because: a) symptoms are hard to detect (e.g small lesions on a bay leaf are not very visible, and can be easily confused with symptoms caused by other agents (Wickland et al.,

2008)), and b) in some hosts such as rhododendrons, infected plants can be asymptomatic (e.g. Denman et al., 2009).

2.1.2.6. Key findings (Potential consequences)

- In managed forests and woodlands, control without phytosanitary measures would be difficult.

- The statement that *P. ramorum* "*seems*" to be established outside nurseries in the UK is now an understatement, given the extensive Japanese larch plantation outbreaks.

- An estimated 1,900 hectares of Japanese larch plantations (about 0.5 million trees) now show symptoms of *P. ramorum* infection in England and Wales. The area covered by Japanese larch plantations in England and Wales is about 60,000 ha; there are some further 65,000 ha in Scotland.

- The outbreak on Japanese larch in England and Wales in the absence of *Rhododendron* is thus an important development, as it suddenly widens the susceptibility to *P. ramorum* outbreaks across (European) landscapes.

The *P. ramorum* epidemic in Europe may be still in its lag phase with respect to its intensity, host- and geographical range.

2.1.3.   Establishment

2.1.3.1. General comments

Pest risk assessments such as the RAPRA report, rather than providing a full review of the organism undergoing assessment, focus on achieving a state of knowledge that is sufficient to determine whether the pest qualifies as a quarantine pest in relation to the risk assessment area. Although the RAPRA report makes a very thorough and comprehensive assessment of the probability of establishment for *P. ramorum* in the EU Member States within the framework of the EPPO decision support scheme, the Panel observes that a reader may have remaining questions about details not fully answered by the questions of the EPPO scheme. This may as well be the case for the assessment of the level of uncertainty accompanied by each answer (see the specific comments for examples).

Therefore, both detailed information and descriptions about uncertainties in the risks related to *P. ramorum* may have been left out in RAPRA either intentionally or because of scheme-structural reasons, but not in order to hide important information or uncertainty.

Although all answers to the EPPO scheme questions related to establishment have been scored with low uncertainty, as in the *P. ramorum* (RAPRA) case, there can still be high uncertainty in terms of to what extent the pest will establish and the impacts it will have if introduced to the risk assessment area.

2.1.3.2.   Specific comments

1.16. *Very many* host plant species or suitable habitats in the PRA area (low uncertainty) (page 97 of RAPRA report).

RAPRA provided a detailed discussion of susceptible trees and shrubs divided by Northern/Central and Southern Europe). The document includes mention of *Aesculus hippocastanum* and *Acer*

pseudoplatanus, at higher to moderate risk, and planted conifer species such as *Abies procera*, *Abies grandis*, *Pseudotsuga menziesii* and *Tsuga heterophylla* (same RAPRA rating as *A. hippocastanum* and *A. pseudoplatanus*).

RAPRA also discussed this issue for heathland, maquis, nurseries and managed parks and gardens. The report is careful to make clear that "*although laboratory tests on bark susceptibility have been largely supported by natural records, some care should be taken when interpreting laboratory tests involving wound inoculation since field susceptibility can be affected by bark thickness*". Additional morphological features such as lenticels and fissures may play a role in the field.

1.19. *Moderately similar* climatic conditions that would affect pest establishment, in the PRA area and the current area of distribution (low uncertainty) (page 107 of RAPRA report). The level of resolution of the maps at that time was low. Conclusions drawn about their value were not fully substantiated. Further, the level of uncertainty at the local scale would be high. Indeed, RAPRA noted that "*care must be taken with the interpretation of climatic matches, although such maps can highlight other areas that could be at high risk based on climate*".

1.22. *Very likely* establishment despite natural enemies in the PRA area (low uncertainty) (page 127 of RAPRA report). The Panel agrees with a comment on endophytes, which may play a role in reducing impacts.

1.26. *Moderately likely* that, based on its biological characteristics, the pest could survive eradication programmes in the PRA area (low uncertainty) (page 130 of RAPRA report). The Panel considers that the rating should have been likely to very likely.

1.28. Relatively small populations *likely* to become established (low uncertainty) (page 135 of RAPRA report). The Panel considers that there should have been medium uncertainty whether small populations can establish.

RAPRA's conclusion on the probability of establishment: <u>significant probability that *P. ramorum* could establish in wider areas of the EU than its current distribution</u>. The Panel agrees to this conclusion.

2.1.3.3. Member State comments

Table 3:   Member State comments and comments of the PLH Panel related to the establishment potential of *P. ramorum*

RAPRA page and topic	Member State comments	Rapra response	Panel comments
119-125: climate -based risk mapping approaches	Climatic conditions suitable to *P. ramorum* need to be combined with presence of hosts and foliar hosts	These factors were certainly taken into account by RAPRA	Among other things, RAPRA mentions the study by Kluza et al. (2007), who used GARP (Genetic Algorithm for Rule-set Production) to predict the potential distribution of *P. ramorum* worldwide (and thus also in Europe). GARP takes into account host distribution
130: how successful is an attempt at eradication likely to be	Limited potential for success of *P. ramorum* eradication efforts in nurseries questions their usefulness; standard control measures in nurseries (before phytosanitary actions for *P. ramorum* were taken) already sufficient	RAPRA's choice was moderately likely eradication (with medium uncertainty), provided the pathogen is detected at an early stage. At a later stage, eradication unlikely (this explains why England and Wales have moved to a policy of containment)	Available data confirm that complete eradication is likely to be difficult. Long-term monitoring and modelling of effectiveness of *P. ramorum* eradication efforts in nurseries are needed. Comment by member state is in contradiction with experience in the UK and in the USA, where control has provided significant reduction in incidence of the pathogen (Schroder and Pfeilstetter, 2007; Xu et al., 2009). In the USA, two papers (Prospero et al., 2007; Mascheretti et al., 2008) show that currently, in the presence of regulations, there is no gene flow between nursery populations and wild populations. This is in contrast with the historical migration from nurseries into the wild documented before regulations started in the USA (Mascheretti et al., 2008, 2009)
139: likelihood of spread without human assistance	Unlikely rapid spread of *P. ramorum* in natural settings in the PRA area	RAPRA scored this as moderately likely with low uncertainty, on the basis of consensus from project consortium and available data/models. Some of the infected larch plantations have no *Rhododendron* in the understory, some are at 0.3 km from the nearest known source of *P. ramorum*	The outbreaks on UK Japanese larch plantations confirm that *P. ramorum* spread by natural means can happen rapidly also in Europe. Please also note the RAPRA response that moderately likely rapid natural spread looks now (one year after RAPRA) an underestimation. The available evidence indicates that, during the lag phase, the pathogen is undetectable. By the time the pathogen is detected, it is in rapid expansion. For all purposes, expansion after an undetectable lag phase is rapid. The recent discovery of the pathogen in Redwood National Park in California despite intensive surveys (http://www.suddenoakdeath.org/wp-content/uploads/2010/03/COMTF_Report_October_2010.pdf) supports this statement.

| 140: likelihood of spread with human assistance | Rapid spread in the nursery network does not automatically lead to rapid spread into the environment, as ornamental shrubs are not planted in woodland or heathland | RAPRA did not suggest this, the report judged that *P. ramorum* was very likely (with low uncertainty) to spread rapidly with human assistance | The study by Xu et al. (2009) on spatio-temporal patterns of *P. ramorum* outbreaks in nurseries and woodlands shows some degree of association between findings in nurseries and environment. Planting of *Rhododendron, Viburnum, Camellia* in private gardens may well lead to spread in woodlands, if these gardens are close to them. Mascheretti et al. (2009) show that the current distribution of the pathogen in California is associated with multiple introductions linked to movement of infected nursery plants. The size of the meta-populations indicates the impact of each introduction. At times, the size of a meta-population (infestation caused by a single introduction) is large highlighting the impact that individual releases in the wild may have. |

RAPRA page and topic	Member State comments	Panel comments
Various topics	MS very concerned by *P. ramorum*, due to presence of susceptible hosts and climate, however, there are doubts on moving from 3 to 74 regulated genera. At the same time, encouragement of regulating European and non-European isolates separately	Recent *P. ramorum* findings on larch plantations (these plantations are also common in Spain (e.g. Onaindia and Amezaga, 2000) make redundant the suggestion of considering separately EU and NA lineages Of course, there are trade-offs between regulatory cover of the wide range in susceptible hosts and costs incurred by plant traders/the public.
		Lineages have phenotypic differences, and on some hosts some lineages are more aggressive than others (e.g. *Camellia*). Because of uncertainty, differences on a host (out of four tested) may be significant. Additionally different lineages are associated with different mating types: mating has been reported to be less than optimal, but a recent publication (Boutet et al., 2010) shows that some pairings result in apparently viable oospores. Oospores lead to recombination and potential selection of more aggressive phenotypes, but also oospores can survive in harsher conditions. This enhanced survival rate of oospores may result in a broader distribution of the pathogen; In California, the distribution of the homothallic *Phytophthora pseudosyringae* (Wickland et al., 2008) also introduced (Linzer et al., 2009) is broader and in hotter areas, in spite of reduced virulence.
		Oospore production is likely to be key, analogously as for *P. cinnamomi* (Brasier and Hansen, 1992; Hansen, 2008).
50+ presence/ distri-bution of *P. ramorum* in Europe	There should be a clear statement whether *P. ramorum* can be considered as established in the European nurseries. Elimination of *Phytophthora* species from nurseries is	As shown by the results of the FVO survey 2010 (presentation at EC Standing Committee on Plant Health on 24-25 January 2011), *P. ramorum* findings in nurseries are declining, represent a minority of tested nurseries but are still affecting many Member States in numbers which are not negligible.

extremely difficult	Vercauteren et al. (2010) provide evidence that the pathogen is established in Belgian nurseries. Even if *P. ramorum* eradication from nurseries is difficult, inoculum reduction can still be helpful, as it is probably likely to reduce risk of escapes. The paper by Werres et al. (2007) indeed suggests that elimination is difficult in nurseries. This result is in support of stronger regulations, considering that introduction will result in long-term damage for the nursery trade.

2.1.3.4.   Additional information

It was already clear at the time of the RAPRA project that common understory species other than *R. ponticum* (e.g. *Vaccinium* spp., *Viburnum* spp., *Arbutus unedo*) can become important in case of *P. ramorum* outbreaks in the semi-natural environment of European countries (Jung et al., 2009). For example, in the IUFRO proceedings there is an assessment (using both a leaf dip inoculation method and a wounded detached stem test) of the susceptibility to *P. ramorum* for various species commonly found in Italy (Vettraino et al., 2009). *P. ramorum* caused leaf symptoms and stem lesion on the vast majority of the 65 tested plant species (35 of which had not been tested before). Based on the distribution of susceptible and sporulating hosts, about 40% of the Italian woodlands appear to be suitable to the pathogen (Vettraino et al., 2009; actually already referred to in the RAPRA report).

Given the importance of *Rhododendron* for the *P. ramorum* epidemic, De Dobbelaere et al. (2010) inoculated wounded/non-wounded detached leaves of 59 cultivars and 22 species of *Rhododendron*, with replications in three different years. They report that "*all Rhododendron species and cultivars were susceptible when using wounded leaves, but not when using non-wounded leaves, suggesting a resistance mechanism operating at the level of leaf penetration.*"

There is also the issue of the range in temperature and humidity conducive to *P. ramorum* infection for a given host. There have been early reports of the ability of *P. ramorum* to grow over a wide range of temperatures (Werres et al., 2001; Harnik et al., 2004; Swain et al., 2006). This is confirmed by an experimental study of *P. ramorum* infection (using the NA1 isolate) of Cunningham's White *Rhododendron*, with *P. ramorum* able to infect this host over a wide range of temperatures and moisture levels (Tooley et al., 2009). This result, if generalised to other host species, would make spread of the pathogen even more likely.

From North America, DiLeo et al. (2008) report that *P. ramorum* was isolated from symptomatic tissue of *Corylus cornuta* var. californica (California hazelnut), *Acer circinatum* (vine maple), *Ceanothus thyrsiflorus* (blue blossom), and *Arctostaphylos columbiana* and *A. manzanita* (two species of Manzanita*). P. ramorum* was re-isolated from symptomatic tissues of all inoculated shoots but not from control shoots. The authors point out that "*the native ranges of these new host species overlap considerably with regions that are considered to be highly favourable to P. ramorum epidemics. Although it is unknown how these newly-identified hosts affect the epidemiology of sudden oak death in California ecosystems or the spread of P. ramorum into uninfested areas, species that are very closely related to many of these are common elsewhere in North America where the climate has been estimated to be favourable to P. ramorum.*"

Similar results are obtained from a study of the susceptibility to *P. ramorum* and sporulation potential of 25 understory plant species growing in Eastern North America (Tooley and Browning 2009). Twenty-two species showed infection of more than 90% of their leaves. Sporangia and chlamydospore production varied considerably among the tested species, but were maximal for *Robinia pseudoacacia* and *Rhus typhina*, respectively. These are two species widely planted and in some cases naturalised in many European countries. *Robinia pseudoacacia* was confirmed as a potential *P. ramorum* host by a study in Serbia, where the pathogen is now reported both from a private garden and a garden centre (Bulajic et al., 2010). Bulajic et al. (2010) also experimentally showed that *Cotoneaster horizontalis* and *C. dammeri* can be infected by *P. ramorum*.

Widmer (2009) demonstrates that when assessing experimentally the susceptibility to the pathogen of potential *P. ramorum* hosts, using sporangia as the inoculum propagule may not achieve the full inoculum potential. This is because rooted rhododendron cuttings had a higher percentage of necrotic leaves per plant when inoculated with zoospores than with sporangia (Widmer, 2009). This result suggests that the epidemiological understanding of *P. ramorum* should not presume that the zoosporic stage is a weak link in the infection process. On the issue of zoospores vs. sporangia, note also the report of Shishkoff (2009) of *P. ramorum* propagules (obtained from *Syringa vulgaris* leaf tissues

infected with *P. ramorum* and placed on potting mix) including zoospores at 10 or 15 °C, but predominantly sporangia at 20 or 25 °C, thus highlighting the importance of climate for the *P. ramorum* epidemic, also in nurseries.

All evidence suggests that free water on leaf surfaces is needed to achieve maximum infection. This factor implies that most infection occurs through a zoosporic phase. Very steep slopes in curves depicting reduction in inoculum (Davidson et al., 2005) and genetic similarity (Mascheretti et al., 2008) suggest that natural spread is produced by large airborne propagules, such as sporangia, rather than small zoospores. This implies that primary infection is determined by the movement of sporangia, followed by the release of zoospores once the sporangia land on a new host.

2.1.3.5. Uncertainties

In relation to the uncertainty assessments (answers to questions in the establishment and spread sections), the Panel observes that, with one exception, the level of uncertainty in RAPRA is scored as "*low*" for all questions. This result is in contrast with the general scientific opinion on uncertainty about *P. ramorum* establishment and spread in relation to its full potential as a pest which is still significant.

Although we consider that all questions related to establishment and accompanying uncertainty assessments are answered properly in the RAPRA report, the Panel recognises that there is a possibility that uncertainty was underestimated by RAPRA.

For some of these questions, the scoring of the uncertainty appears to be related to the specific wording of the questionnaire in the EPPO scheme, rather than to the actual uncertainty present in the topics addressed. For example, RAPRA scores the question "*Estimate the number of host plant species or suitable habitats in the PRA area*" as "*Very many*" (on the scale: Very few, Few, Moderate number, Many, Very many) with low uncertainty. This could be understood as "we are certain that there are many host plants species occurring in the PRA area". On the other hand, as a relatively newly discovered organism, considerable uncertainty remains about the host range of *P. ramorum* and the susceptibility to this pathogen for known hosts.

Regarding the RAPRA assessment of the suitability of the environment, the Panel notes that no new biological information emerging from the RAPRA project was used. All modelling approaches applied, including parameter values for climate responses of *P. ramorum*, pre date the RAPRA work. This observation slightly contrasts Jung et al. (2009) who wrote that the RAPRA project "*substantially increased our knowledge and understanding of pathogenicity, host range, distribution, and survival of P. ramorum and of the biological and climatic conditions favouring disease outbreaks*". Nonetheless, RAPRA made a good attempt to compile existing information that was scattered in the literature.

The Panel recognises an interpretative challenge in that the assessment of the endangered area paragraph is placed in RAPRA in the establishment and spread section. For an untrained PRA reader it is difficult to distinguish establishment and impact, for pathogens in particular, because the area where pathogens can establish in principle can be different from the area where they may act as a pest and cause disease. The RAPRA does not treat the topic of how to classify areas where *P. ramorum* can establish but does not cause disease. On the other hand, it seems that this potential problem is a minor one in the case of *P. ramorum* as there is no evidence that there are areas with establishment without damage, i.e. areas having latent infections only. At least in the wild or semi-wild setting, the presence of *P. ramorum* equals damage.

Davis et al. (2010) have started investigating the environmental conditions associated with increased *P. ramorum* damage. Ellis et al. (2010) have assessed the relative importance of connectivity and of environmental conditions for the *P. ramorum* epidemic in California. Severity of the disease will be different among sites, but there is no evidence of pathogen presence in the wild without damage. There

are several interesting exceptions linked to presence in streams outside of that naturalized range of infection (http://www.suddenoakdeath.org/wp-content/uploads/2010/03/2009YearEndSummary.pdf): these are all cases in which the pathogen has been repeatedly found in waterways, but not on plant hosts. The water biology of this organism is not yet fully understood , but it has been shown that infested water can infect plants (Tjosvold et al., 2009), and it can be expected that water infestation will lead to plant infection, especially following flooding events of river banks.

2.1.3.6. Key findings (Establishment)

- The RAPRA report provides a thorough and comprehensive assessment of the probability of establishment of *P. ramorum* in the EU member states.

- However, some detailed information about uncertainties related to *P. ramorum* establishment may have been left out in RAPRA. This may have been because the RAPRA team strictly followed the questions of the EPPO scheme.

- Early stage impact evidence from new pest establishment events must be interpreted with care.

- Although all answers to the EPPO scheme questions related to establishment have been scored with low uncertainty, as in the *P. ramorum* (RAPRA) case, there can still be high uncertainty in terms of to what extent the pest will establish and the impacts it will have if introduced to the risk assessment area.

- The level of resolution of the maps of major potential hosts at the time of the RAPRA project was low. Therefore, some RAPRA conclusions drawn about these maps may not be fully substantiated. Problems (e.g. in uniform coverage of the various Member States, in the consistency of the methodology, in the spatial resolution) in the available host maps shown in this opinion are still present.

- The Panel supports RAPRA's view that there is a significant probability that *P. ramorum* could establish in wider areas of the EU than its current distribution.

- The outbreaks on UK Japanese larch plantations confirm that *P. ramorum* spread by natural means can be extensive also in Europe.

- *P. ramorum* lineages have phenotypic differences, and on some hosts some lineages are more aggressive than others.

- The enhanced survival rate of oospores may result in a broader distribution of the pathogen.

- The host range and range of climatic conditions which enable *P. ramorum* to establish are broader than described in the RAPRA report.

- Uncertainty regarding establishment ratings was generally underestimated by RAPRA.

Whilst the severity of the disease will be different among sites, there is no evidence of pathogen presence in the wild without damage.

2.1.4. Spread

2.1.4.1. General comments

Contrary to the assessment of the probability of entry (into the risk assessment area), the assessment of the probability of spread (within the risk assessment area) is not assessed on a per pathway basis, although a pathway is defined equally as '*any means that allows the entry or spread of a pest*' for entry and spread in the Glossary (FAO 2007). This is again a characteristic of the EPPO PRA scheme related to its main purpose namely to decide whether an organism has the characteristics of a quarantine pest in relation to a specific risk assessment area. In the case of RAPRA the risk assessment area is defined as "*The European Union Member States – a subset of the EPPO (European and Mediterranean Plant Protection Organisation) region*". Therefore, entry of *P. ramorum* from one Member State to another Member State is considered as spread and consequently not assessed in detail on a per pathway basis by RAPRA, although such assessment results could be interesting.

In the USA, regulation is in place between those States with *P. ramorum* and other States, but even within individual States there is a distinction between areas that are within and outside the zone of infestation (ZOI).

Epidemiological information on the spread of *P. ramorum* is essential to guide effective management and policy regarding this pathogen. In California, the current distribution of the pathogen can be explained by multiple human-mediated introductions followed by natural local spread of the introduced pathogen (Mascheretti et al., 2009). Although the pathogen is somewhat limited in its spread by landscape heterogeneity, there are large areas of California proven to have been colonized from a single introduction point (Mascheretti et al., 2008, 2009). In the Big Sur region the area of infestation attributed to a single introduction exceeds 100 km in radius, in Marin-Sonoma Counties it approaches 100 km, and in Humboldt county where introduction occurred after 2000, it is already approaching 30 km in spread, with a leap of almost 100 km Northward (http://www.suddenoakdeath.org/wp-content/uploads/2010/03/COMTF_Report_October_2010.pdf).
The combination of human induced introductions (Mascheretti et al., 2009), potential human-mediated spread (Cushman and Meentemeyer, 2008) and natural spread (Mascheretti et al., 2008; Davis et al., 2010) at the 10 m to 5 km scale make this a difficult pathogen to eradicate once introduced in the wild, as the Oregon experience shows (Prospero et al., 2007).

2.1.4.2. Specific comments

1.32. *P. ramorum is only moderately likely to spread rapidly by natural means* (low uncertainty) (page 138 of RAPRA report). The Panel agrees with the original rating, however the RAPRA report did not define what "*rapid*" meant. Therefore, the associated uncertainty is considered higher.

1.33. *The pathogen is very likely to spread rapidly by human-mediated means, most significantly through the commercial movement of infected plants for planting* (low uncertainty) (page 139 of RAPRA report). The Panel agrees with the original rating, however the RAPRA report did not define what "*rapid*" meant. Therefore, the associated uncertainty is considered higher.

1.34. *It is moderately likely that P. ramorum could not be contained within the PRA area, based on its biological characteristics* (low uncertainty) (page 140 of RAPRA report). The Panel agrees with the rating with a medium level of uncertainty.

RAPRA's conclusion on spread (page 141 of RAPRA report):

"*P. ramorum is very likely to spread quickly throughout the nursery network within the PRA area in the absence of statutory controls on host plants for planting. This is due to its very wide host range and the likely characteristics of the trade network itself. Spread from nurseries into the environment*

will be facilitated by the planting of infected plants; potential natural spread from nurseries in to semi-natural or natural habitats is likely to be relatively slower. Similarly, natural spread within the semi-natural or natural environment is likely to be relatively slow due to the pathogen's somewhat poor ability to disperse very long distances naturally, especially in spatially heterogeneous landscape where susceptible habitats/hosts may be fragmented.

However, in more homogenous landscapes where there is an abundance of continuous hosts, spread could be significantly more rapid. Although speed of spread is highly relevant to determining the likelihood of containment being successful or not at the spatial scale, it may not be entirely relevant to overall impact at a longer-term temporal scale."

The Panel agrees with this conclusion.

1.35. *Nurseries involved with hardy ornamental plants across the whole PRA area are likely to be favourable for P. ramorum to establish.[....]Timber plantations, especially mixed-deciduous types may be at risk where they have sporulating hosts in them as an understorey and where they fall in the climatic zones highlighted below* (page 142 of RAPRA report).

The nursery trade has proven to be a formidable pathway for dispersal (Ivors et al., 2006; Grünwald et al., 2008; Prospero et al., 2009) at the worldwide and continental levels. Notwithstanding the historical introduction in California, undeniably linked to the nursery trade (Ivors et al., 2006, Mascheretti et al., 2008), there are at least three instances of *P. ramorum* presence in US waterways in the absence of wild infestations, but close to commercial nurseries: one in California in Northern Humboldt County, one in Washington State, and one in Mississippi (http://www.suddenoakdeath.org/wp-content/uploads/2010/03/COMTF_Report_October_2010.pdf). The nursery trade is thus undeniably a link enabling the spread of the pathogen among landscape setting and between landscape and wild or semi-wild settings. Natural spread appears to be mostly through airborne inoculum (Davidson et al., 2005). Other pathways such as those represented by soil and wood are theoretically possible, but there is no evidence that *P. ramorum* may have already been spread through such pathways. Because our knowledge only goes back ten years, lack of proven dispersal through soil or wood does not mean that these are not viable pathways and should not be regulated.

It is clear that dispersal of *P. ramorum* in California does not follow a linear pattern, but really is bimodal. Short distance spread occurs on a yearly basis and is normally within a few kilometers, but long distance spread (through unknown pathways) occasionally occurs, and seems to be linked to favourable weather conditions for the pathogen.

Although it was already clear at the time of the RAPRA project that people can play a major role in the worldwide *P. ramorum* epidemic (e.g. with movements of infected plants in the plant trade), new studies have filled gaps in our knowledge e.g. of how the epidemic in the wild and in the trade are related. Goss et al. (2009b) used microsatellites to investigate 279 isolates collected from 19 US states between 2004 and 2007. The study distinguishes two eastward migration pathways, one connecting Connecticut from Oregon and Washington and the other tracing isolates from California to the remaining investigated states. Goss et al. (2009b) confirm the importance of combining genetic studies with trace-forward data of infected plant shipments.

Prospero et al. (2009) studied 576 *P. ramorum* isolates obtained between 2001 and 2005 from infected forests on the West Coast of the USA. The investigation confirms previous analyses of the *P. ramorum* epidemic in Oregon and California forests, inasmuch as the vast majority of the investigated isolates belonged to the NA1 clonal lineage. Prospero et al. (2009) also report that the high incidence of genotypes shared among populations show that there has been long-distance movement of *P. ramorum* genotypes.

However, using multilocus genotypes, Mascheretti et al. (2009) show that "*localized genetic differentiation of the pathogen is under way in California due to the lack of effective migration among*

established infestations combined with the local evolution of new genotypes". This study confirms the previously reported (Mascheretti et al., 2008, already cited in the RAPRA report) spatial genetic autocorrelation profile with an increase in relatedness from a few hundred meters to about one km, which could be related to wind-dispersal, but also to artificial movements of the pathogen.

Mascheretti et al. (2009) back up the view that the effective dispersal of this exotic pathogen over a large area in California has been made possible by a combination of human-mediated and natural spread. The importance of considering at the same time the spread in the semi-natural environment and in the plant trade is likely to apply also to Europe, as shown by spatially-explicit simulations of the *P. ramorum* epidemic in Britain integrating realistic features of the trade (Harwood et al., 2009) and by spatio-temporal analysis of the *P. ramorum* records in the semi-natural environment and in the plant trade in England and Wales from 2003 to 2006 (Xu et al., 2009).

That initial conditions (for *P. ramorum* introduction events in the plant trade) are important is confirmed by epidemiological modelling where there is a continuum of infection (infection is not just either present or absent) in directed networks of small size. This model along a continuum of infection is appropriate for the horticultural trade, where plant nurseries may contain a varying proportion of *P. ramorum*-infected plants and may not be removed from trade, unless posed under strict and continued quarantine after infection detection. Directed networks are needed to model the plant trade, given the inherent asymmetry in the contact structure of these networks, which are often of limited size (whereas most modelling of epidemics in networks has been done for large-size networks; Jeger et al., 2007). Regardless of network type (local, small-world, random and with presence of super-connected individuals) and level of connectance, there was a strong correlation at the threshold conditions between epidemic final size and the number of out-going connections of the starting node of the epidemic (Pautasso et al., 2010b).

In terms of *P. ramorum*, epidemic final size can be interpreted as the overall number of infected plants in all nurseries/retail centres or the number of nurseries/retail centres with more than a certain proportion of plants infected. In the simulations, this proportion was chosen at 1%; recent monitoring in the UK shows that positive findings on nurseries have diminished from 3 to 1% of inspected material, probably as a consequence of the emergency actions taken over the last 5 years (Walters et al., 2010). For comparison, at a plant nursery in Belgium heavily infected by *P. ramorum*, about 16% of over 1300 samples were positive for the pathogen (Heungens et al., 2010).

2.1.4.3.   Member State comments

Table 4:   Member State comments and comments of the PLH Panel related to the spread potential of *P. ramorum*

RAPRA page and topic	Member State comments	RAPRA response	Panel comments
52-53: conclusion of pest categorisation	The diversity of climate, habitat composition and fragmentation effects may slow down *P. ramorum* spread in Europe	The pathogen does have a broad host range and sporulating hosts do occur in climatically favourable areas. Moreover, the RAPRA response pointed out that new sporulating hosts are being discovered, so that fragmentation effects may be weaker than suggested by MS	We concur with the RAPRA response, although Member State is correct inasmuch as the potential effects of landscape fragmentation on *P. ramorum* spread have been studied in California. Similar research should be undertaken in the EU. We know that the pathogen may jump from infected habitat patches to other suitable patches due to movement of infected plants, soil or water, but there is still a need for a reliable dispersal kernel for *P. ramorum* in various landscape configurations and forest types (Davidson et al., 2011). The Big Sur Ecoregion is one of the worst affected areas by SOD in California (Davis et al., 2010) and genetic reconstruction suggests the pathogen may have arrived there only 20 years ago (Mascheretti et al., 2008). Despite the great heterogeneity of landscape, comprising forested valleys separated by barren dry ridges, a few genotypes of *P. ramorum* have been able to occupy the entire region (Mascheretti et al., 2009) even if disease severity may be variable depending on history and climatic conditions (Davis et al., 2010).
91: likelihood of transfer from plant trade pathways to hosts in the environment	Long-distance aerial dispersal was frequently cited in RAPRA, but is likely to be lower in Europe than in California	*P. ramorum* long-distance spread was also assessed using UK information from a DEFRA project report by Beales (2007)	There has now been a key development related to this issue, i.e. the Japanese larch outbreak, as pointed out by the RAPRA response to the Member State comment. Just recently an outbreak was reported tens of km North of the only known wild infestation in Humboldt County: genetic analysis shows the new outbreak to be linked to the pre-existing infestation in the same county but tens of km South of the pre-existing infestation (http://www.suddenoakdeath.org/wp-content/uploads/2010/03/COMTF_Report_October_2010.pdf) and shows the ability of the pathogen (whether naturally or through human mediated activities) to start new infestations at long distance from previous ones. Davidson et al. (2005) and Mascheretti et al. (2008) provide direct and indirect evidence, respectively, that airborne spread of the pathogen in a forest setting is routinely in the range of several meters. At the 2010 COMTF meeting, Garbelotto reported that sporangia were trapped at least 20 m away from any

			possible source. Reports of spread from nurseries (Heungens et al., 2010) are about one order of magnitude smaller, but conditions in forests and nurseries are drastically different and this difference should not be regarded as contradictory. In nurseries spread is often linked to water-borne spread (e.g. Garbelotto and Rizzo 2005) which is often limited to one or adjacent beds. In forests spread appears to be a function of height of infectious host, wind speed and shading provided by canopy with final distances covered by the pathogen much larger than those reported in nurseries Mascheretti et al. (2008) propose that aerial spread of a few kilometres may be linked to turbulent movement (medium-distance movement of large infectious propagules such as sporangia), only occurring in presence of winds strong enough to pick up propagules of such size. If winds are strong enough to pick up sporangia, their release would only occur at 1-5 km from the source.
139	It is unlikely that spread of *P. ramorum* in natural settings in the PRA area will be rapid	RAPRA scored this as moderately likely with low uncertainty, on the basis of consensus from project consortium and available data/models. It was further pointed out that some of the infected Japanese larch plantations have no *Rhododendron* in the understory, some are at 0.3 km from the nearest known source of *P. ramorum*	We concur with the assessment in the RAPRA response that the outbreaks on UK Japanese larch plantations confirm that *P. ramorum* spread by natural means can happen rapidly also in Europe. Please also note the RAPRA response that moderately likely rapid natural spread looks now (one year after RAPRA) an underestimation. Sporulation patterns in California (Davidson et al., 2005) show that the pathogen can build up very high inoculum loads in response to favourable climatic conditions in a fairly short period of time. This supports the concept that rapid outbreaks are actually not just possible, but very common, once the pathogen is established in a site
140: likelihood of spread with human assistance	Rapid spread in the nursery network does not automatically lead to rapid spread into the environment, as ornamental shrubs are not planted in woodland or heathland	RAPRA did not suggest this, the report judged that *P. ramorum* was very likely (with low uncertainty) to spread rapidly with human assistance	The study by Xu et al. (2009) on spatio-temporal patterns of *P. ramorum* outbreaks in nurseries and semi-natural environment in England and Wales (2003-2006) shows some degree of association between findings in nurseries and in the environment. Planting of *Rhododendron, Viburnum, Camellia* in private gardens may well lead to spread in woodlands, if these gardens are close to them. There is no doubt that the pathogen has spread within the nursery trade. Similarly, new evidence links spread in the wild to introduction from nurseries in California. Genetically distinct populations of the pathogen in California are linked to distinct introduction events for the most part undistinguishable from nursery populations of the pathogen, indicating that multiple introductions from the nursery to the wild are responsible for the current distribution of SOD in

			California (Mascheretti et al., 2008, 2009). The first two wild infestations in California, according to Mascheretti et al. (2008) occurred in two forests, one adjacent to a Rhododendron nursery in Santa Cruz County, and one adjacent to a private home in Marin County, where infected plants where planted around the residence. Additionally reports in Northern California, Washington State, Mississipi, Florida, Georgia and Alabama (http://www.suddenoakdeath.org/wp-content/uploads/2010/03/2009YearEndSummary.pdf) indicate that infested rivers in areas without forest infestations are in the vicinity of commercial nurseries that were found to be positive for *P. ramorum* one or multiple times. In the case of the Big Sur region, a single introduction caused an infestation of over 100 km in radius, in spite of spatial heterogeneity.
140: spread from plant trade into the natural environment	In countries where forests cover an important part of territory, preventing the spread of *P. ramorum* into natural habitats is of great importance.		This statement concurs with the RAPRA assessment and is endorsed given the state of the current evidence for potential *P. ramorum* spread and its likely damage to a wide range of native species in the semi-natural environments
141: conclusions on the probability of spread	On the issue of spread, one Member State declared that the risk of natural spread seems to be quite different in the countries where *P. ramorum* has been detected outside nurseries. For example, in UK this risk appears to be high, whilst in Germany the pathogen is still present at the two locations outside nurseries but there has been so far no detection of further spread during about 15 years of monitoring.		This point is legitimate, as different European countries will have different climates and host distributions, and thus different likelihoods of *P. ramorum* spread once this becomes established. However, the presence of Japanese larch plantations in various European countries (not only Britain and Ireland) makes this issue delicate and in need of further monitoring and assessment.
	The risk of spread within the PRA area is criticised for not being assessed as a pathway.	This PRA has been carried out in accordance with the EPPO scheme which restricts this approach.	

2.1.4.4.   Additional information

In the UK, key recommendations for *P. ramorum* policy and management not yet available at the time of the RAPRA project were delivered using a model of *P. ramorum* epidemic development (Harwood et al., 2009). The epidemiological model includes information on the spatial distribution of potential hosts in Britain, as well as a realistic super-imposed network of commercial plant movements. A series of simulation experiments were run, with variation in the epidemic pressure and in the connection between semi-natural vegetation and horticultural trade, with or without disease spread in commercial trade, and with or without inspections-with-eradication. The results of the simulations underline the importance of the inspection policy: epidemic final size was reduced by inspections by about 90%. However, in some simulations, escape from the horticultural trade into the semi-natural environment did occur in spite of the inspections, thus emphasising the importance of stochasticity and initial conditions in country-wide *P. ramorum* epidemic development.

The list of tree species with bleeding canker lesions due to *P. ramorum* in Europe (England and the Netherlands) reported in Jung et al. (2009) from the RAPRA database still does not include *Larix kaempferi* (Japanese larch). *Larix* as a genus is however also not included in the APHIS regulated list of *P. ramorum* proven hosts or associated plants, updated February 2010. The *P. ramorum* outbreak on Japanese larch in England and Wales is a major novel finding since the RAPRA report, given the extent of the plantations of this exotic tree in Britain. An estimated 1,900 hectares of Japanese larch plantations (about 0.5 million trees) now show symptoms of *P. ramorum* infection in England and Wales (Brasier and Webber, 2010). The area covered by Japanese larch plantations in England and Wales is about 60,000 ha; there are some further 65,000 ha in Scotland (Clark, 2010). The level of inoculum in the affected Japanese larch plantations is so high that the number of *P. ramorum* infected trees other than *Larix* (*Fagus, Castanea, Quercus*) has increased by one order of magnitude since the finding of *P. ramorum* on larch (RAPRA response to the Member States comments on RAPRA, 2010).

Evidence already available at the time RAPRA was finalised, together with additional evidence, gives insights on the spread potential of *P. ramorum*. In Oregon, new infestations have been reported 1-2 miles away from known infestations (Prospero et al., 2007). By using spatial autocorrelation analysis, it was shown that in California *P. ramorum* has a bimodal spread pattern, with a peak at 10 m from a source then decreasing sharply until 400m, and a second peak at 1-3 km from the source. This second peak was explained by the large size of sporangia, that once picked up by strong winds are deposited 1-3 km from the source (Mascheretti et al., 2008). In support of data provided by Mascheretti et al. (2008), surveys   in   California   publicly   available   on   the   web (http://nature.berkeley.edu/garbelotto/english/sodblitzresults2010.php), show new infestations at 1-5 km from established infestation sites.

This bimodal spread pattern results in potential spread at the landscape level, but in the presence of gaps of a few km (> 5) in environments favourable to the pathogen, pathogen spread should be hindered. However, research by Gilligan et al. (2007) suggests that control is difficult in the presence of coalescent infection foci at an intermediate distance, while a pathogen that moves at a larger distance normally causes discrete identifiable hotspots. For *P. ramorum*, this implies that the scale of movement by the pathogen in the wild can be considered one of the hardest to effectively control. Mascheretti et al. (2009) show that in the presence of landscape heterogeneity, migration is minimal at distances above 5 km, resulting in genetically distinct populations, while smaller gaps in habitat can easily be crossed by the pathogen resulting in homogeneous populations over large areas.

2.1.4.5.   Uncertainties

As far as the spread of *P. ramorum* in the plant trade is concerned, the modelling in small-size, directed networks suggests that heterogeneity in the contact structure of horticultural trade networks is likely to increase the risk of major epidemics occurring, if by chance a major producer with a high

number of out-links is infected. One uncertainty is here that information on the contact structure of European plant trade networks is generally lacking (Moslonka-Lefebvre et al., 2011).

The model along an infection continuum shows that the correlation among incoming and outgoing links can explain a substantial proportion of the variance in the epidemic threshold among network replicates, again irrespective of the network structure (which is largely unknown as far as plant trade in Europe as well as within European countries is concerned) (Moslonka-Lefebvre et al., 2009). Epidemiological modelling thus calls for the standardised and long-term collection of data on the contact structure of plant trade networks, i.e. not just trade volumes, but for example the frequency distribution in the number of incoming and outgoing links of trade players (producers, wholesalers, retailers) in various European regions at risk of inadvertent *P. ramorum* introduction (Pautasso et al., 2010c).

However, as it is well stated in the USDA APHIS PRA (APHIS, 2008), volume of traded plants is one of the factors affecting likelihood of introduction, but diagnostic power and presence of suitable environmental-host combinations are equally important. Uncertainty in each of these three factors, will translate into uncertainty in correct predictions. The Panel agrees that a clear understanding of the areas potentially most endangered by the pathogen is far from perfect, but also acknowledges that the available evidence indicates that improving diagnostic efficiency and reducing trade of infected plants will reduce likelihood of introduction across borders, including both areas that may be highly conducive to disease expression and areas that may be less so.

Identification of the endangered area is a potential source of uncertainty.

According to the RAPRA report, care should be taken in interpreting the results from the identification of the area endangered by *P. ramorum* in Europe:

- One reason for this is because model outputs for suitable climate differ between the various modelling approaches. There is no general scientific agreement on which modelling technique is the most appropriate out of those applied and a reliable validation is not possible using the limited data from Europe.

- Another reason why the results should be treated carefully is due to the character of the climatic data, which is summarised information from a thirty year period (1961-90) and is also interpolated from weather stations to cover wide areas. As described in RAPRA, it is therefore likely that the microclimatic factors in the western coastal fringes of Europe that appear to be particularly suitable for *P. ramorum*, e.g. woodland in a steep valley near the sea or a large water body that provides continuous high relative humidity, occur in some locations over a much wider area.

- These climate-based maps also do not account for more local microclimatic factors. For example, in the UK, the pathogen has been found in several more central or eastern areas, considered of lower risk based on climate, causing significant disease on established rhododendron: these sites have had favourable microclimates; in one case in East Yorkshire there has also been a beech tree with bleeding bark cankers.

Uncertainties still exist about the geographical distribution of suitable hosts – RAPRA points at limitations in host distribution data on the EU level.

Uncertainties are still present both regarding the host range and host suitability of *P. ramorum*. For example, a recent report of *P. ramorum* infecting mistletoe reminds us of how still little understood is the host range of this generalist pathogen (Riley and Chastagner, 2011).

2.1.4.6. Key findings (Spread)

- Epidemiological information on the spread of *P. ramorum* is essential to guide effective management and policy regarding this pathogen.

- The Panel generally agrees with the RAPRA conclusions on spread, but the RAPRA rating that rapid natural spread is moderately likely is now an underestimation, given the new information on Japanese larch.

- Potential effects of landscape fragmentation on *P. ramorum* spread should also be studied in Europe.

- Airborne spread of the pathogen in a forest setting is routinely in the range of several meters. If winds are strong enough to pick up sporangia, their release would extend some 1-5 km from the source.

- The *P. ramorum* outbreak on Japanese larch in England and Wales is a major novel finding since the RAPRA report, given the extent of the plantations of this exotic tree in Britain.

- The level of inoculum in the affected Japanese larch plantations is so high that the number of *P. ramorum* infected trees other than *Larix* (*Fagus, Castanea, Quercus*) has increased dramatically in Britain since the finding of *P. ramorum* on larch.

- The presence of Japanese larch plantations in various European countries (not only Britain and Ireland) makes the risk of spread problematic and in need of further monitoring and assessment.

- Knowledge of the structure of European plant trade networks is generally lacking.

- The available evidence indicates that improving diagnostic efficiency and reducing trade of infected plants will reduce likelihood of introduction across borders, including both areas that may be highly conducive and areas that may be less conducive.

The combination of climatic suitability and presence of (sporulating) hosts across Europe still needs investigation.

2.1.5.   Entry

2.1.5.1. General comments

There is little doubt that the magnitude in the trade of plants among (and within) countries is increasing. In a useful overview of the various PRA stages for *P. ramorum* in the UK and the EU, Sansford (2009) demonstrates *"a significant increase in imports of various ornamental plants into the U.K. between 1995 and 2005. For example ca. £6.4 million worth of trees in 1995 increased to ca. £64 million in 2005; ca. £5 million 'cuttings, slips and other young plants' increasing to ca. £16 million"*.

Sansford (2009) then goes on to argue that this increase in trade is not only confined to the UK, but has occurred around the world, and has "*led to new opportunities for plant pathogens to be moved to new areas or countries.*" A similar link of the structural changes in the international horticultural industry with a potentially increased risk of introduction of new plant pathogens is made in the

reviews by Dehnen-Schmutz et al. (2010), Webber (2010), Wingfield et al. (2010) and Stenlid et al. (2011).

2.1.5.2. Specific comments

"*In the absence of phytosanitary measures the highest risk of entry ('high') of P. ramorum into the EU is on plants for planting of host plants from the area, or areas, of origin, as well as from the USA (see Table 14). Uncertainty is highest ('medium') for plants for planting from the pathogen's origin(s) since: (a) the area/s of origin for P. ramorum are unknown; (b) the host range in the area of origin is unknown; (c) it is uncertain whether specific phytosanitary controls will be in place for host material from the area of unknown origin; (d) entry has already occurred at least once in Europe, as well as in the USA.*"(page 141 of RAPRA report)

No particular specific comments are made on entry, as *P. ramorum* is now well established within the EU territories.

The RAPRA report did not consider the entry of different lineages.

2.1.5.3. Member State comments

Table 5: Member State comments and comments of the PLH Panel related to the entry into the risk assessment area of *P. ramorum*

RAPRA page and topic	Member State comments	RAPRA response	Panel comments
69-79: pathways	RAPRA focuses on pathways of entry into PRA area, rather than within it.	RAPRA was following EPPO guidelines, and the section should thus be specifically aimed at entry into the PRA area.	Even if the RAPRA response to the Member States comment is valid, research should be undertaken to study available information (or to collect it, if missing) on trade volumes of *P. ramorum* susceptible material within vs. into Europe using a network approach and with a long-term perspective. Intra-European trade of *P. ramorum*-susceptible plant material is obviously an important issue.
91: likelihood of transfer from plant trade pathway into the environment	RAPRA overestimated the risk of *P. ramorum* introduction into the semi-natural environment (rather than into private and public gardens) from the nursery trade, because planting of *P. ramorum* susceptible species into woodland or heathland is rare.	Also the number of findings in managed gardens needs to be considered here, with reference to recent figures on the impact of *P. ramorum* (and *P. kernoviae*) on National Trust gardens. It is unknown whether transfer to forests from nurseries is rare or not.	Evidence discussed in Mascheretti et al. (2008) points to two different scenarios for escape of the pathogen, as exemplified by the discovery that two oldest infestations in California were a) in a forest around a nursery in Santa Cruz County, and b) around a landscape of planted rhododendrons in a private property. Although the need of information on frequency of planting of *P. ramorum* susceptible plants in woodland or heathland now needs to be considered with the UK *P. ramorum* outbreak on Japanese larch in mind, the study by Xu et al. (2009) on spatio-temporal patterns of positive findings in England and Wales suggests some (decreasing) co-occurrence of nursery and environment outbreaks. UK and EU-wide *P. ramorum* genetic studies as performed in California would help to know more here.
94: conclusions on the probability of entry	EU Commission measures aimed at preventing *P. ramorum* import from third countries should be kept at least as they are, if not strengthened.		Data on *P. ramorum* incidence in the various European States confirm the importance of such measures, as the number of *P. ramorum* interceptions in plant shipments among Member States appears to have declined in 2009.

Additionally, a Member State comment maintained that there is an underestimation in RAPRA of the risk of introduction for the pathway nr 5 (foliage or cut branches of susceptible hosts). The Member State agrees however with RAPRA's suggestion to declare a pest-free area for these ornamental materials.

2.1.5.4. Additional information

Goss et al. (2011) studied the genotypes of *P. ramorum* in plant nurseries in Canada compared to those found in the US and in Europe. The study confirms that plant trade networks have not only made it possible for *P. ramorum* to be introduced into North America and Europe, but are also likely to introduce new genotypes if left unregulated. In Canadian nurseries, all three of the *P. ramorum* clonal lineages were found. Surprisingly, the most common lineage in Canada was NA2, whilst NA1 (the most common one in US nurseries) was rare. In addition, the EU1 lineage was frequently detected in Canada. Goss et al. (2011) conclude that *P. ramorum* migration from Europe to North America is more likely to have occurred than migration in both directions. Evidence provided to the Panel by Matteo Garbelotto (University of California, personal communication, 2011), indicates that virulence of isolates belonging to different lineages may differ (Appendix A.), in spite of seemingly overall comparable ecological and biological traits of different lineages of this pathogen (Elliott et al., 2009b). This information strengthens the need to prevent introduction of different lineages into the area covered by the PRA.

2.1.5.5. Uncertainties

In California, the nursery that was the original source of the epidemic infested the surrounding forest. At that moment it became impossible to certify the nursery as pest-free and eventually it was closed down. This is an important source of uncertainty, because nurseries that are adjacent to susceptible habitats become hotspots for the spread that are hard to control, and it is likely (although uncertain how likely) that re-infection from the wild will occur. This implies that not all nurseries are equal: the same mitigation or clean-up policy will not yield the same results in nurseries placed in an agricultural or urban settings (where treatment is likely to be long lasting) vs. nurseries in semi-wild situations where linkage with the outside world is likely to restart infection.

One further uncertainty in relation to entry is the reliability of control and plant passporting schemes. For example, the recent first report of *P. ramorum* in Greece (in a nursery) involves a *Rhododendron* shipment from Belgium which had received a plant passport from the relevant authorities (Tsopelas et al., 2011).

2.1.5.6. Key findings (Entry)

- *P. ramorum* is now well established within the EU territories.

- The entry of different *P. ramorum* lineages has not been considered.

- Intra-European trade of *P. ramorum*-susceptible plant material is an important issue.

- UK and EU-wide *P. ramorum* genetic studies as performed in California would provide information on entry into the natural environment from the nurseries.

- Plant trade networks have not only made it possible for *P. ramorum* to be introduced into North America and Europe, but are also likely to introduce new genotypes if left unregulated.

- There is a need to prevent introduction of different lineages into the area covered by the PRA.

- There are still questions about the reliability of control and plant passporting schemes.

- The geographical origin of the pathogen is still matter of investigation.

2.1.6. Overall uncertainties

Despite the international research effort to improve our biological and epidemiological knowledge on *P. ramorum*, uncertainties still remain in e.g.:

- The origin of the pathogen;

- Host infection process and long-distance dispersal;

- The exact host range and the susceptibility in the field of each single host;

- The geographical distribution of susceptible and sporulating hosts;

- The rapidity of spread of *P. ramorum* across European landscapes;

- The reliability of plant passporting schemes;

- The structure of European networks trading plants susceptible to *P. ramorum*.

2.1.7. Conclusions of the pest risk assessment

In general the Panel concludes that the broad narrative in the RAPRA report stands and agrees with its conclusion that "*There is a risk of further entry (of known or new lineages and/or mating types), establishment and [...] impact*".

In addition to this, the Panel notes that there is growing evidence of differences among lineages. Therefore, it is advisable to avoid introductions of different lineages not only because of potential of sexual recombination, but also because of such inherent differences.

There are some points that need some attention:

- The RAPRA report concludes (p 48, 2nd paragraph) that "*the mating system may not be fully functional*". *P. ramorum* can mate easily in the lab, but not all crosses are viable. Thus, it is true that it is not fully functional, as the % of success is not close to 100%, but it definitely does mate (Appendix B.; Boutet et al., 2010).

- The conclusion that the pest is not widely distributed (p. 49 in RAPRA report) is still valid, although new outbreak reports are still being made by Member States and survey methods are not well-developed. However, we note the number of findings outside the nursery trade, in heathland and Japanese larch that have emerged since the PRA (p. 50 in RAPRA report). We also note the number of *P. ramorum* interceptions in the plant trade among Member States, as shown by Figure 13.

Figure 13: Network of *P. ramorum* interceptions in Europe (2002-2010). Arrow thickness is proportional to the number of interceptions; arrow direction shows the direction of the interception; EU data.

- Long-range spread of the pathogen appears to be cyclically linked to a) favourable weather patterns (El Nino in California, Meentemeyer et al., 2011) and/or, b) new introduction events (Kliejunas, 2010). Based on the following map (Figure 14) of the *P. ramorum* outbreak in Humboldt county (California), where there was no attempt at containment, the distribution of the pathogen has not progressed from 2002 to 2010 as quickly as can be expected for other invasive tree pathogens. This is important because it shows that efforts to slow down new introductions are worthwhile.
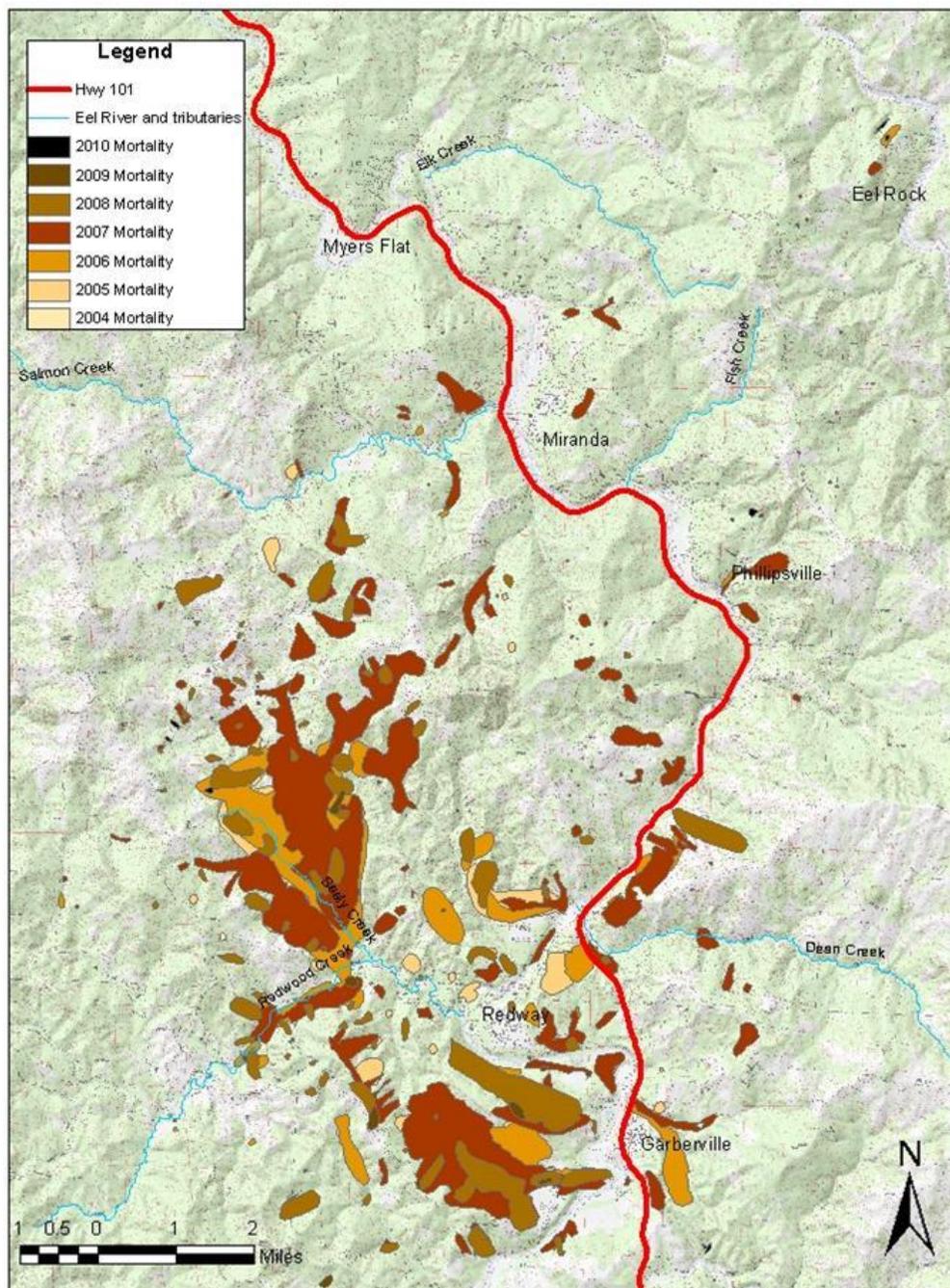
Figure 14: Reconstruction of the *P. ramorum* outbreak in Humboldt county (California), 2004-2010 (By permission from Yana Valachovic and Chris Lee (UC Cooperative Extension) with support of the USDA Forest Service Region Five Forest Health Protection.

- There is a clear potential for *P. ramorum* spread at the landscape level. Although studies on the effects of landscape fragmentation on *P. ramorum* spread have only been carried out in

California, we know that, also in Europe, if winds are strong enough to pick up sporangia, these would be released at 1 to 5 km from the source. Given (1) the level of inoculum in the affected Japanese larch plantations, (2) the extent of these plantations in Britain, and (3) their presence in various other European countries, the potential for further long-distance spread of *P. ramorum* needs further monitoring and assessment.

- There are still uncertainties on the origin of *P. ramorum* although the recently reported finding of *P. lateralis* in an old-growth forest of *Chamaecyparis* in Taiwan (Brasier et al., 2010) made it even clearer that the geographic origin of *P. ramorum* is likely to be East Asia. The two *Phytophthora* species (*P. lateralis* and *P. ramorum)* are phylogenetically related, show various common features (e.g. large chlamydospores, sporangia with short pedicels, production of stromata, a similar optimum growth temperature) and are thus likely to share a common region of origin. Brasier et al. (2010) point out that since *Chamaecyparis* is present both in Taiwan and Japan, both *Phytophthora* species could have originated from one of these two countries. This would explain why previous missions to discover the origin of *P. ramorum* in the South-East Asian mainland were not successful (Vettraino et al., 2011). In any case, the recent evidence strengthens the already settled case that *P. ramorum* is an exotic pathogen both in Europe and in North America (Hansen, 2010). The uncertainty on the origin of *P. ramorum* corroborates the importance of checking entries from undetermined third countries other than USA, Canada, Norway and Switzerland.

- Hybridisation among *Phytophthora* species has been observed (e.g. *Phytophthora alni*; Brasier et al., 2004; Stenlid et al., 2011). Many *Phytophthora* species co-occur in the nursery environment. When they are closely related and on the same host species, the risk of hybridisation is higher. Similarly, the observation of the closely related *P. ramorum* and *P. lateralis* on the same host at the same location (Clive Brasier, personal communication, 2011) suggests the potential for hybridisation.

- The list of tree species with bleeding canker lesions due to *P. ramorum* in Europe (England and the Netherlands) reported in Jung et al. (2009) from the RAPRA database still does not include *Larix kaempferi* (Japanese larch). The *P. ramorum* outbreak on Japanese larch in England and Wales is a major novel finding since the RAPRA report, given the extent of the plantations of this exotic tree in Britain, as shown by Figure 15 (Brasier and Webber, 2010).

- Infection of other hosts in the vicinity of Japanese larch demonstrates that the range of hosts is even broader than it was estimated by the RAPRA.

Figure 15: Consequences of *Phytophthora ramorum* infection in tanoak woodland (California) and Japanese larch plantations (Britain) (from Brasier and Webber, 2010; reprinted by permission from Macmillan Publishers Ltd: Nature 466, 824-825, 2010).

2.2. Pest Risk Management

2.2.1. General comments

The RAPRA report was prepared according to the EPPO Standard 'Guidelines on Pest Risk Analysis: Decision-support scheme for quarantine pests' version 07-13727 (PM5/3). Eight pathways were identified in the RAPRA report in the probability of entry section and were used to structure the pest risk management analysis:

- Plants for planting (excluding seed and fruit) of known susceptible hosts;

- Plants for planting (excluding seed and fruit) of non-host plant species accompanied by contaminated attached growing media;

- Soil/growing medium (with organic matter) as a commodity;

- Soil as a contaminant;

- Foliage or cut branches;

- Seed and fruits;

- Susceptible (isolated) bark;

- Susceptible wood.

Three types of measures were identified for each pathway:

- Measures related to consignments;

- Measures related to the crop or to the places of production;

- Other possibilities.

Risk management options were identified according to the following categories:

- Options to reduce likelihood of introduction on plant consignments;

- Options to reduce likelihood of spread;

- Options to reduce infestation and magnitude of impact in the crop.

P. ramorum outbreaks had already occurred at least in nurseries within the risk assessment area when the RAPRA report was published but only the first category of option was analysed in the RAPRA report. Options to reduce the likelihood of spread and to reduce the infestation and magnitude of impact in the nurseries should have been analysed in the nursery network. It is important to remember that *P. ramorum* findings outside of nurseries (including managed parks, gardens, public greens, woodlands, and forests) have now been reported in Europe not just from the Netherlands and the UK, but also from Belgium, Denmark, France, Germany, Ireland, Luxembourg, Norway, Serbia, Slovenia, Spain, and Switzerland (Sansford et al., 2010).

The measures recommended in the RAPRA report do not account for the pre-existing EC phytosanitary measures for the *P. ramorum* under the EC Plant Health directive (2000/29/CE).

The RAPRA report recalled the non-specific measures listed in the Council Directive 2000/29/EC. This directive was completed by a legislation specific to *P. ramorum* (Commission Decision 2002/757/EC[7]) and amended in 2004 and 2007 in order to adopt emergency measures to prevent the introduction and the movement of *P. ramorum* into and within the EU territory. When plants were imported in the EU countries from third countries such as USA and Canada, susceptible plants must have originated from a *P. ramorum*-free area or from a place of production that has been inspected by the U. S. authorities and found free of symptoms.

Within the EU territory, susceptible nursery plants (*Rhododendron, Viburnum and Camellia*) are controlled and must be found free from quarantine pest including *P. ramorum* after statutory inspections and delivery of the "plant sanitary passport" to be allowed to circulate within the EU territory. Additional measures were undertaken relating to destruction of associated growing media and plant debris and other appropriate phytosanitary measures were applied to the growing surfaces.

The EU Directives also called for all EU member states to undertake surveys for *P. ramorum* and disseminate the results. An analysis of the effect of these measures on the prevalence of *P. ramorum* during the period 2004 to 2006 showed a reduction in the percentage of inspections positive for *P. ramorum* and a reduction in the number of outbreaks at nurseries (Slawson et al., 2008). However, the continued findings of the pathogen in nurseries and in woodlands, notably on Japanese larch in UK, indicated that the phytosanitary measures have not been completely effective.

Vercauteren et al. (2010) indicate that the pathogen in Belgian nurseries includes local unique genotypes but also shares common genotypes: this finding indicates that local eradication of genotypes is hard to achieve, and/or that genotypes are still shared among nurseries through trade of infected plants.

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[7] Commission Decision 2002/757/EC of 19 September 2002 on provisional emergency phytosanitary measures to prevent the introduction into and the spread within the Community of *Phytophthora ramorum* Werres, De Cock & Man in 't Veldsp. nov. OJ L 252, 20.9.2002, p. 37-39.

2.2.2. Specific comments

The RAPRA report recommends measures for six out of the eight analysed pathways (Table 6). There is no specific measure thought to be necessary for seed and fruit of susceptible host plants. For susceptible bark, it is reported that measures are only necessary for parts of the USA where *P. ramorum* occurs in woodlands and forest. The larch outbreak in the UK may make similar measures necessary also for the states covered by the PRA. Specific comments are listed below according to the EPPO scheme when disagreement with statements of the RAPRA was noticed.

Pathway (i): Plants for planting (excluding seeds and fruit) of known susceptible hosts that are permitted entry from the USA and Canada, Norway and Switzerland (pages 178-191 of RAPRA report);

3.12. Are there any existing phytosanitary measures applied on the pathway that could prevent the introduction of the pest? If appropriate, list the measures and identify their efficacy against the pest of concern.

The current list of susceptible plants is not exhaustive. It should include *Larix kaempferi* and other newly recognized susceptible species. *P. ramorum* has been reported in the 2010 FVO report on tree species such as *Abies*, *Betula*, *Chamaecyparis*, *Ilex* and *Tsuga* and on shrubs such as *Drimys*, *Gautheria*, *Pieris* and *Sarcococca*.

3.14. Can the pest be reliably detected by testing (e.g. for pest plant, seeds in a consignment)?

Detection methods actually used are not reliable for the detection on *P. ramorum* on asymptomatic plants. Furthermore, certain detection methods do not work, e.g. PCR with some plant species.

3.22. Can infestation of the commodities be reliably prevented by growing the crop in specified conditions (e.g. protected conditions such as screened greenhouses, physical isolation, sterilized growing medium, exclusion of running water?

Multiplication of certain susceptible ornamental plants could be done by *in vitro* culture, as this prevents risk of contamination with *P. ramorum*.

3.26. Has the pest a low to medium capacity for natural spread?

The comment made by RAPRA is valid, however it should be revised because the infection pattern on Japanese larch is different to that on *Rhododendron*.

3.30. Have any measures been identified during the present analysis that will reduce the risk of introduction of the pest? List them?

Surveillance plans should be revised to cover Japanese Larch plantations in third countries.

3.34. Estimate to what extent the measures (or combination of measures) being considered interfere with international trade.

The Panel agrees with the RAPRA comments. However, it would need to update the list of host plants including Japanese larch. The use of network analysis to guide nursery control schemes would make the measures more effective.

3.41. Consider the relative importance of the pathways identified in the conclusion to the entry section of the pest risk assessment from the very low to very high.

There is no reason to consider the level of risk different in commodities coming from the USA and other infected countries such as Canada and the non-EU countries, where *P. ramorum* occurs.

Pathway (ii): Plants for planting (excluding seeds and fruit) of non- hosts plant species accompanied by contaminated, attached growing media from the USA and Canada, Norway and Switzerland (pages 191-199 of RAPRA report);

3.16. Can the pest be effectively destroyed in the consignment by treatment (chemical, thermal, irradiation, physical)?

Yakabe and MacDonald (2008) determined the effectiveness of chemicals as potential soil treatments and reported that only chloropicrin, metam sodium, iodomethane and dazomet were efficient to kill viable propagules in treated soil but the use of these compounds is banned or will soon be banned in many countries.

Pathway (iii): Soil growing medium (with organic matter) as a commodity from the USA and Canada, Norway and Switzerland (pages 199-204 of RAPRA report);

3.14. Can the pest be reliably detected by testing (e.g. for pest plant, seeds in a consignment)?

Import of soil as a commodity is prohibited according to Council Directive 2000/29/EC.

Pathway (v): Foliage or cut branches (for ornamental purposes) of susceptible foliar hosts from the USA (Norway and Switzerland – but only if foliar hosts are affected where harvesting and export to the EU occurs) (pages 207-214 of RAPRA report);

3.12. Are there any existing phytosanitary measures applied on the pathway that could prevent the introduction of the pest? If appropriate, list the measures and identify their efficacy against the pest of concern.

The current lists of *P.ramorum* susceptible plants need to be updated. They should include *Larix kaempferi* and other plant species infected in Japanese larch plantations. *P. ramorum* has been reported in the 2010 FVO report on *Abies*, *Betula*, *Chamaecyparis, Drimys*, *Gautheria*, *Ilex*, *Pieris*, *Sarcococca* and *Tsuga* (confirmed by C. Brasier, pers. comm.).

3.14. Can the pest be reliably detected by testing (e.g. for pest plant seeds in a consignment)?

Detection methods currently used are not reliable for the detection on *P. ramorum* on asymptomatic plants. Furthermore, certain detection methods do not always work, e.g. PCR with some plant species.

3.26. Has the pest a low to medium capacity for natural spread?

The point made by RAPRA is still valid but should be revised given that infection and sporulation on Japanese larch are not the same as on *Rhododendron*.

There is the additional issue of movement within the EU. For instance, if leaves are used from plants grown in EU nurseries they could carry the disease.

Pathway (vi): Susceptible (isolated) bark from the USA (pages 214-220 of RAPRA report) and **Pathway (viii):** Susceptible wood (pages 227-237 of RAPRA report).

3.16. Can the pest be effectively destroyed in the consignment by treatment (chemical, thermal, irradiation, physical)?

Composting has been considered as an effective treatment option for sanitation of *P. ramorum*-infected plant material (Garbelotto, 2003; Swain et al., 2002, 2006; Aveskamp and Wingelaar, 2005).

For susceptible bark, it is reported that measures are only necessary for parts of the USA where *P. ramorum* occurs in woodlands and forests. The larch outbreaks occurring in the UK may make similar measures necessary also for all the European states covered by the PRA.

Table 6: Recommendations of the RAPRA report per pathway and the related comments of the PLH Panel

RAPRA recommendation	Panel comment
Pathway (i) Phytosanitary Certificate (PC) and, if appropriate, Reexport Certificate (RC) Measures related to consignments: Detection of the pathogen in consignments by inspection and testing at export and/or import or Detection of the pathogen by inspection and testing during post-entry quarantine Measures related to the crop or to places of production: Pest freedom for the crop, place of production or area. Domestic certification schemes if supported by testing of symptomatic material. Other possible measures Surveillance and eradication in the importing country of the EU	• surveillance and eradication have not been entirely successful: there was some success in the nursery trade but this did not prevent the UK outbreaks in Japanese larch plantations • it can be argued that what was introduced through this pathway predated the emergency measures • detection of the pathogen by inspection: the network built on the data of EU *P. ramorum* interceptions could help in better targeting inspection • symptom-based diagnosis: it would make sense testing the plant nursery facilities, e.g. with irrigation water detection • specific certification is required by Norway for importation from nurseries in D, NL • need to continue surveys to understand whether changes in interceptions are related to policy
Pathway (ii) PC and, if appropriate, RC Measures related to consignments: Physical removal of any surplus growing media just before export. Measures related to the crop or to places of production: In areas where the pathogen occurs, treatment (sterilisation) of the growing media prior to planting and prevention of reinfestation during the growing period Pest freedom for the crop, place of production or area (i.e. non-host plants to be produced away from hostplants to avoid contamination) Other possible measures Surveillance and eradication in the importing country of the EU	• plants are imported in growing media • there is a question about whether/how to regulate the imports of plants potentially susceptible to *P. ramorum* into the EU • contaminated non-host import plants • movements into and within the EU need to be distinguished • regulation of movements within EU against a hot spot of *P. ramorum* infection (e.g. South-West of England and Wales) may be worthwhile considering
Pathway (iii)	• heat treatment effective but not practical (as

PC and, if appropriate, RC Measures related to consignments: Depending upon the volume of material heat treatment could be considered but may not be practical. Measures related to the crop or to places of production: Pest free crop, place of production or area. (For the area where soil or growing media are collected). Other possible measures Surveillance and eradication in the importing country of the EU.	pointed out by RAPRA), 50 degrees specific for soil (Linderman and Davis, 2008a) • surveillance and eradication - is this realistic/practical for this pathway?
Pathway (iv) Measures related to consignments: Cleaning and disinfection of used machinery or vehicles imported from an area where *P. ramorum* occurs. Measures related to the crop or to places of production: Not applicable Other possible measures Inspection of human travellers footwear and possible treatment at the point of entry where travellers have entered from an area where *P. ramorum* occurs	• the pathogen has been found on shoes and bicycles (tested, both in USA and UK, SOD 3rd Science Symposium; see also McNeill et al., 2011) • this pathway mostly concerns contamination into the EU (which has already taken place) • felling operation within hotspots or where large amount of soil is moved may require an additional certification
Pathway (v) PC and, if appropriate, RC Measures related to the crop or to places of production: Pest–free area for the crop, place of production or area. Other possible measures Controls on recycling for known infected material Surveillance and eradication in the importing country of the EU	• an important issue is that infection remains viable for *at least* 3 months on foliage (Swain et al., 2006) • the issues of recycling and composting need to be addressed for this pathway
Pathway (vii) PC and, if appropriate, RC Measures related to consignments: Limited end-use of known infected bark (i.e. not to be used in the nursery trade or the landscaping industry) Measures related to the crop or to places of production: Pest-free crop, place of production or area	

2.2.3. Member States comments

No comments were made by Member States on the Pest risk management part of the RAPRA report.

2.2.4. Additional information

Options to reduce likelihood of introduction on plant consignments

One problem here is that, inevitably, there is a non-zero likelihood of failing to detect the pathogen where it is present. This risk is worsened by the issue of asymptomatic infection, already referred to in the RAPRA report (see also Denman et al., 2009). However, new research is making progress in developing reliable molecular tools to find *P. ramorum* where this occurs (Bilodeau et al., 2009;

Giresse et al., 2010; Grünwald et al., 2011; Schlenzig, 2011). A comparative study of 5 detection methods concludes that "*a combination of either culturing and molecular diagnosis or of two molecular assays is the most promising approach to diagnose this pathogen*" (Vettraino et al., 2010). Nevertheless, Vettraino et al. (2010) found significant effects of season, host species and laboratory on *P. ramorum* detection accuracy. Their recommendation is that "*diagnosis should occur as much as possible during wet and warm periods favourable to the pathogen, and* [that] *proficiency tests should be performed to compare results obtained with molecular approaches in different laboratories*".

The use of a nested Taqman approach was shown to be extremely sensitive (Hayden et al., 2006), and has a zero rate of false positives in the absence of unnaturally high concentrations of DNA (Martin et al., 2009). However, in the Hayden et al. (2006) paper the following was stated: "*In the subset of 207 samples tested by both single-round and nested TaqMan methods, P. ramorum was detected in 88 of the 207 samples using the nested protocol but in only 31 using a single-round of TaqMan detection*" These data indicate that we are still far from having a fool-proof diagnostic assay available, and in fact as confirmed by Vettraino et al. (2010), the current single round Taqman assays mostly used for regulatory purposes have a significant rate of false negatives.

In the Martin et al. (2009) study cited above, 11 *P. ramorum* detection techniques were studied. However, in this case, "*with few exceptions, all assays correctly identified all isolates of P. ramorum.* [Moreover,] *low levels of false positives were observed for the mitochondrial cox spacer markers and most of the real-time assays based on nuclear markers*" (Martin et al., 2009). Nonetheless the comparison was done on pure DNA extracted from cultures and its results cannot be fully extrapolated to the real world.

It should be pointed out that the two studies above are based on the analyses of either symptomatic plant tissue (Vettraino et al., 2010) or cultures (Martin et al., 2009). However, symptoms are rarely diagnostic, because culturing is often impossible (Hayden et al., 2004) and because plants can be asymptomatic. The greater issue of how appropriately to screen a large number of plants remains unsolved, although the improved understanding of the range of symptoms that could be diagnostic (exemplified by several diagnostic guides available) and on the effects of season on the outcome is helpful. Just as helpful could be the development of reliable tools to pre-screen potential *P. ramorum* infections, as suggested by baiting of *P. ramorum* with leaves of susceptible plant such as Rhododendron and Tanoak, which has been used with success in California (Murphy et al., 2006; Aram and Rizzo, 2009) and in Oregon (Sutton et al., 2009).

Together with the need to be sure that reliable results are obtained from diagnostic tests, there is also an issue of the time involved. Some laboratories are thus focusing on developing more rapid tools for detecting the pathogen in addition to the conventional isolation and DNA extraction. Tomlinson et al. (2010) report that membranes of lateral flow devices added directly to real-time PCR can greatly reduce the time needed for the testing without affecting diagnostic sensitivity and specificity.

Tomlinson et al. (2010) also studied a pre-screening test for *Phytophthora* spp. in conjunction with the *P. ramorum*-specific test. This can be a useful way to reduce the time needed, as it will increase the prevalence of *P. ramorum* in the tested samples. If a sample does not test positive by any *Phytophthora* species, it is likely also not to test positive (and therefore not to be infected) by *P. ramorum*. In this case, the predictive value of the *P. ramorum*-specific test will depend not only on the prevalence of *P. ramorum,* but also on the presence of other *Phytophthora* species (Tomlinson et al., 2010). Tomlinson et al. (2010) argue that *Phytophthora* species other than *P. ramorum* are likely to be common for samples coming from plant nurseries, and to be rare for samples coming from confirmed *P. ramorum* outbreak sites.

Many *Phytophthora* species other than *P. ramorum* are relatively abundant in natural streams in healthy forests, even if the species present are poorly characterised, and their ecology is still largely unknown (Reeser et al., 2011). As a by-product, monitoring for the presence of *P. ramorum* is therefore helping increase our general knowledge of naturally-occurring *Phytophthora* species

(Hansen, 2008). Similar considerations apply to non-naturally occurring *Phytophthora* species, some of which have been discovered during surveys for *P. ramorum* (e.g. Linzer et al., 2009).

Options to reduce likelihood of spread

For disease management to be implemented efficiently, it is essential to have reliable knowledge on the identity, distribution and incidence of the pathogen. Boutet et al. (2010) confirm that it makes sense to avoid the introduction of NA1 and NA2 A2 isolates into Europe and further introduction of EU1 A1 isolates into North America. The available evidence so far (Ivors et al., 2004, 2006; Prospero et al., 2007; Vercauteren et al., 2010) shows that the genetic structure of the pathogen seems to be the outcome of clonal rather than sexual reproduction, although Ivors et al. (2006) could not really exclude sexual reproduction was occurring in the EU1 lineage. Although there is no evidence of ongoing sexual reproduction, Tyler et al. (2006) reported that the genome of *P. ramorum* has heterozygosity rates that are to be expected as the result of sexual reproduction, hence the potential for sexual reproduction remains (see also Martens and Van de Peer, 2010).

As well as the issue of selection on progeny, the role played by oospores in the survival of the pathogen remains debatable. As shown by comparative studies on three aerial *Phytophthora* species in California, all putative exotic species (Linzer et al., 2009), the presence of oospores is highly correlated with a much broader distribution, including areas with extreme heat and cold temperatures (Wickland et al., 2008). Notwithstanding the fact that the origin of the pathogen is still unknown, the North American nursery trade seems to harbour all three known lineages, while European nurseries only harbour one (Ivors et al., 2006). This highlights the risks of introducing different lineages by the pathogen through imports of infected plant material from North America into the EU.

In the same paper (Ivors et al., 2006) it is stated: "*The potential role of plant trade in the creation of an 'artificial' panmictic population at the continental level is highlighted by (ii) the observation that rare genotypes were found more than once within Europe, particularly in the UK, where the EU4 genotype was found multiple times in different regions, and (ii) the detection of an EU genotype within Oregon and Washington nurseries and warrants the concern about the introduction of the lineages into the EU.*" Besides different mating types, these lineages display different phenotypes such as virulence and growth rate in vitro (Ivors et al., 2006). These concerns were confirmed by the recent findings of Goss et al. (2011).

A first report of phenotypic differences among lineages has now been published (Elliott et al., 2009b). This paper shows that NA2 and EU1 lineages are more aggressive than the NA1 lineage. Data presented to the Panel by Matteo Garbelotto (University of California, personal communication, 2011) (i) support that view, (ii) suggest that NA2 is more aggressive than NA1, and (iii) provide evidence that, in general, the two lineages perform differently (Appendix A.). This difference among lineages should be taken into account: introduction of new lineages into Europe could have significant consequences both for the nursery trade and natural ecosystems.

Further progress is being made in developing markers to identify the three *P. ramorum* lineages (Elliott et al., 2009b), in examining virulence, sporulation and elicitin production of the same lineages (Manter et al., 2010) and in reconstructing the evolutionary history of the three lineages (Goss et al., 2009a; already referred to in the RAPRA report). These are thought to have been diverging for a length of time sufficient to have allowed independent evolution prior to the independent introductions to North America and Europe (Goss et al., 2009a). In addition, there is now an attempt at standardising the nomenclature for the three *P. ramorum* lineages (Grünwald et al., 2009).

In Europe, genetic studies of *P. ramorum* have focused on the presence in Belgian plant nurseries of EU1-A2 isolates (Vercauteren et al., 2010). The North-American NA1 and NA2 isolates are of A2 mating type, whereas the European EU1 isolates are of A1 mating type, with the exception of some EU1-A2 isolates collected in Belgium in 2002/2003 (Vercauteren et al., 2011a). Research is planned to compare *P. ramorum* isolates from the Japanese larch outbreaks with those from e.g.

Rhododendron, both genotypically and phenotypically (Clive Brasier, personal communication, 2011). Molecular tools can relatively rapidly provide insights for reconstructing the *P. ramorum* epidemic and in predicting its further development. It is important to remember that *P. ramorum* findings outside of nurseries (including managed parks, gardens, public greens, woodlands, and forests) have now been reported in Europe not just from the Netherlands and the UK, but also from Belgium, Denmark, France, Germany, Ireland, Luxembourg, Norway, Serbia, Slovenia, Spain, and Switzerland (Sansford et al., 2010).

However, using multilocus genotypes, Mascheretti et al. (2009) show that "*localized genetic differentiation of the pathogen is under way in California due to the lack of effective migration among established infestations combined with the local evolution of new genotypes*". This study confirms the previously reported (in Mascheretti et al., 2008 already cited in the RAPRA report) spatial genetic autocorrelation profile with an increase in relatedness from a few hundred metres to about one km, which could be related to wind-dispersal, but also to artificial movements of the pathogen.

Dodd et al. (2008) report a genetic component in the size of lesions to coast live oaks caused by *P. ramorum* (already hinted at in a previous study, not considered in the RAPRA report (Dodd et al., 2005)). Although for *P. ramorum* in Europe and America there has not been host-pathogen co-evolution, there is still likely to be intraspecific genetic variation in susceptibility of the various host species (Nettel et al., 2009). This is an important issue, which is understudied (particularly in Europe), as it is likely to affect the long-term potential of plant hosts to adapt to *P. ramorum* (Pautasso, 2009; Hayden et al., 2010; Nagle et al., 2010).

There is a recent review of sudden oak death management case studies at the local community level in California (Alexander and Lee, 2010). They write that, "*in the absence of a centrally organized and coordinated set of mandatory management actions for this disease in California's wildlands and open spaces, many local communities have initiated their own management programs*." Thus, although there was no formal experimental testing of different management strategies at the landscape level, the diversity of approaches allows the drawing of some lessons after a decade of sudden oak death management. Alexander and Lee (2010) acknowledge that: "*most of the local management efforts detailed in these case studies are not effective in the sense of eradicating P. ramorum or slowing its spread on a large scale. However, they are effective in the sense of alleviating pathogen impacts on a smaller scale; in stimulating local involvement with natural resource management; in enhancing group capacities for useful citizen science by providing testing-grounds for new management techniques; and in preparing for more effective pest outbreak responses in the future*." On this last point, see also Wright and Slawson (2010) from the point of view of British historic gardens managed by the UK National Trust, most of which have extensive collections of exotic Rhododendrons from Asia (as acknowledged also in the RAPRA report). In the United Kingdom, a best practice guide for woodlands, parks, and gardens is available from the department for Environment, Food and Rural Affairs (DEFRA) Web site (DEFRA, 2008).

Widmer (2009) demonstrate that when assessing experimentally the susceptibility to the pathogen of potential *P. ramorum* hosts, using sporangia as the inoculum propagule may not achieve the full inoculum potential. This is because rooted rhododendron cuttings had a higher percentage of necrotic leaves per plant when inoculated with zoospores than with sporangia (Widmer, 2009). This result suggests that the epidemiological understanding of *P. ramorum* should not presume that the zoosporic stage is a weak link in the infection process. On the issue of zoospores vs. sporangia, note also the report of Shishkoff (2009) of *P. ramorum* propagules (obtained from *Syringa vulgaris* leaf tissues infected with *P. ramorum* and placed on potting mix) including zoospores at 10 or 15 °C, but predominantly sporangia at 20 or 25 °C, thus highlighting the importance of climate for the *P. ramorum* epidemic, also in nurseries.

Indirect and direct evidence (Davidson et al., 2005; Mascheretti et al., 2008) indicate dispersal of the pathogen is mostly at a relatively small scale (1-10 m) as expected of a pathogen being spread by large propagules such as sporangia, however infection only occurs when water is present on plant surfaces,

indicating that zoospore release is a necessary step in the infection process. When temperatures are around 20C, this infection process appears to be optimal, as indicated in papers monitoring the infection process in wildlands in California (Garbelotto et al., 2003; Davidson et al., 2005; Hayden et al., 2008).

At the molecular level, European research has recently delivered green fluorescent protein production in *P. ramorum* strains (Riedel et al., 2009). Riedel et al. (2009) write that "*in order to prevent the spread of the pathogen with asymptomatic plants, more detailed information on the infection process, on the tissue colonization, and on the latency period [are] needed.*" The development of a reporter gene system is a tool which can help in such investigations.

One result likely to pertain also to Europe comes from modelling of the current distribution of *P. ramorum* across Californian landscapes (Václavík and Meentemeyer, 2009). The simulations show the importance of data on *P. ramorum* absences. Without including these in models, the actual distribution is over-predicted. Thus accurate large-scale *P. ramorum* surveys have a key role, even if these end up in a majority of negative reports. Figure 16 presents sites in the semi-natural environment in England and Wales where *P. ramorum* was detected between 2002 and 2010, but also surveyed sites where no infection was detected (the map does not seem to consider infected Japanese larch plantations, whose map is produced by Forest Research).
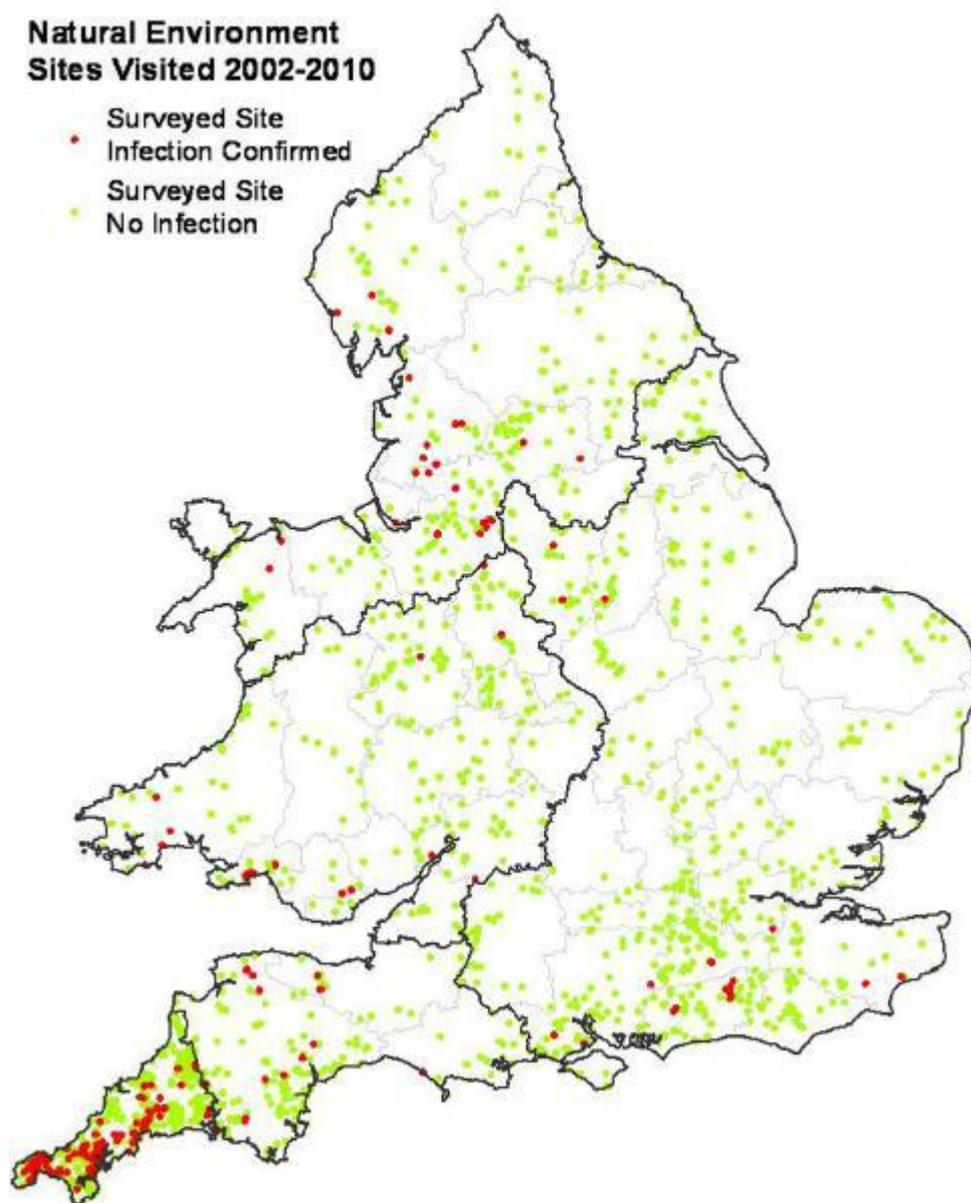
Figure 16: Distribution map of *Phytophthora ramorum* in natural environment sites, in England and Wales, 2002-2010 Supplied by The Food and Environment Research Agency, UK © Crown Copyright 2011.

Options to reduce infestation and magnitude of impact in the crop

Sanitation is important both when attempting to slow down natural epidemics and when trying to curtail the spread through human trade or movement of infected plant material. Leaves infected by *P. ramorum* can be highly infectious, and can carry the disease over long distances. It is widely accepted that the risk associated with foliar hosts is proportional to the sporulation potential on that host (both sporangia and chlamydospores are relevant). RAPRA already identifies some potential sporulating hosts, however no clear discussion is provided on how to effectively deal with infected plant material. Two papers have addressed sanitation in California: one dealt with the ability of heat and heat vacuum treatments to reduce viability of foliar infection to zero (Harnik et al., 2004), while a second paper

showed that composting following EPA guidelines successfully eliminates all traces of *P. ramorum*, even in cases where massive infection of foliage was tested (Swain et al., 2006). Risk when moving infected plant parts needs to be considered and it could be mitigated by adopting these proved techniques or investigating alternative approaches.

In heat treatments of *P. ramorum*-infested soil, the pathogen was still detectable after more than 40 days at 30 and 22 °C. However, only 3 days of soil heating above 40 °C made the pathogen no longer detectable (Yakabe and MacDonald, 2010). Linderman and Davis (2008a) also investigated fumigation treatments that effectively sterilize the soil as a means of eradicating *P. ramorum* from soil or potting media. Another study tested various plant protection products against *P. ramorum* on Rhododendron, with only phosphite and Cu hydroxide having the effect of decreasing disease development (Nechwatal et al., 2010). However work by Garbelotto et al. (2008, already cited in RAPRA) points out that not all chemicals are effective on all hosts: for instance while Cu-OH treatments worked well for the protection of the foliar host bay laurel, they were ineffective for oaks. Phosphites instead were effective on oaks, but not effective at all on bay laurels.

One important aspect deals with the efficacy of preventative (e.g. before plants are infected) vs. therapeutic (e.g. after plants are infected) treatments. All studies in the wild in California point to a lower efficacy of therapeutic compared to preventive treatments. Studies on ornamental plants have indicated that preventative chemical treatments can be effective (Linderman and Davis, 2008b), but strong evidence suggests that therapeutic treatments are only marginally effective (Tjosvold et al., 2008) even in nursery settings. Unfortunately, it is extremely hard to convince stakeholders to apply treatments preventatively, except for when risk of *P. ramorum* infection is high and imminent.

These studies raise the question of whether anti-*P. ramorum* treatments are not just effective, but also affordable by plant growers. Ndeffo Mbah and Gilligan (2010a) studied the trade-off among detection and control efforts in forests potentially affected by *P. ramorum*, thereby showing that for optimal results the resources allocated to detection and to control need to be balanced. Similar considerations apply also to *P. ramorum* in nursery settings.

In another paper, Ndeffo Mbah and Gilligan (2010b) incorporated economic factors in an epidemiological model of a pathogen able to infect two host species, with symmetric or asymmetric infection rates, a model motivated by the sudden oak death outbreaks in California. The many uncertainties about the epidemiology of *P. ramorum* make it important to consider the potential effects of uncertainty on plant epidemiological models which include economic factors (Ndeffo Mbah et al., 2010). Similar considerations have been made by Vàclavik et al. (2010) in their study about prioritising *P. ramorum* detection efforts across landscapes differing in potential susceptibility.

A study by Elliott et al. (2009a) on the biocontrol options for *P. ramorum* reports that control of *P. ramorum* on *Rhododendron* foliage with BCAs was comparable to the one obtained with some contact fungicides, although it was lower than with systemic fungicides tested in the study by Wagner and colleagues, which was cited in the RAPRA report. Nevertheless, Elliott et al. (2009a) point out that with fungicides there is an issue of resistance development, as well of course as environmental and toxicity considerations.

On this point, it is interesting that the majority of Belgian *P. ramorum* isolates have become resistant to metalaxyl (Vercauteren et al., 2010). The same authors point out that "*the extensive use of fungicides as [...] P. ramorum control measures probably had an unnoticed but similar impact on the populations of other Phytophthora species present in Rhododendron nurseries such as P. citricola and P. cactorum, and may therefore have broader implications.*"

One problem in adopting BCAs as a control strategy for *P. ramorum* may be that there was great variability among *P. ramorum* isolates in their response to the commercial Biocontrol Agents (BCAs) tested (Elliott et al., 2009a). However, there is certainly some potential as all treatments reduced *P.*

ramorum lesion size on both *Rhododendron* and *Camellia* (Elliott et al., 2009a). The study also tested various BCAs in combination but compared the combinations with the most effective treatment.

On biocontrol of *P. ramorum*, Tjosvold et al. (2009) report that root infections were detected only after pots were infested for at least 40 weeks. The authors speculate that this may be due to roots of potted Rhododendrons in nursery conditions having been generally precolonised with *P. cinnamomi* (probably not the best choice available as a BCA, but the principle is probably similar), which may have put *P. ramorum* at a competitive disadvantage for infecting roots.

A novel finding in this direction is the report for the first time of the presence of endornaviruses in *P. ramorum* (Kozlakidis et al., 2010). Kozlakidis et al. (2010) write that whether the presence of endornaviruses in *Phytophthora* species has effects on fungal growth or pathogenicity is unknown but since there are endornaviruses, which act as a hypovirulence factor, reducing virulence of the host fungus, the authors are convinced that this is an issue worth further exploration and funding. The deployment of hypovirulence, rather than the use of fungicides or the destruction of hosts, greatly helped in limiting the effects of another major plant health issue throughout much of Europe, i.e. chestnut blight (*Cryphonectria parasitica*). However, although hypovirulence worked very well as a natural biocontrol of chestnut blight in Southern Europe, this was not the case in North America.
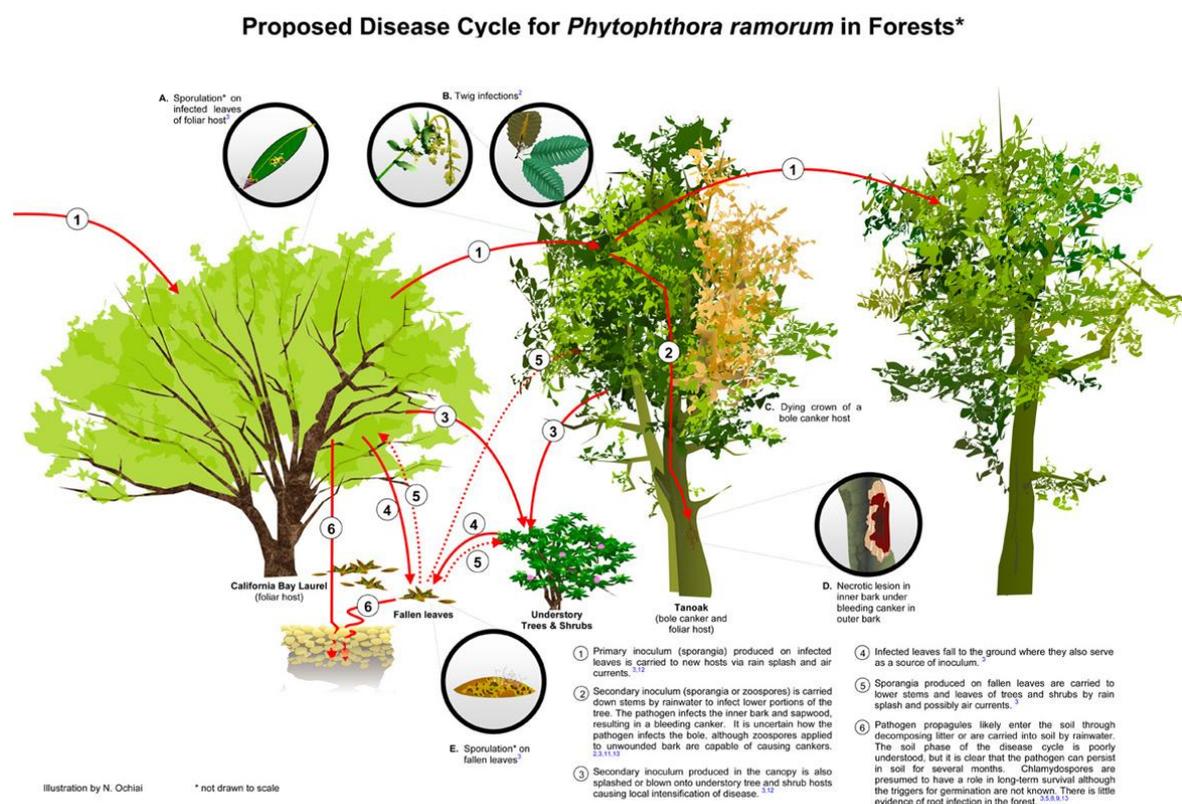

Figure 17: Proposed disease cycle for *Phytophthora ramorum* in forests. (Reproduced by permission from Parke JL and Lucas S, 2008. Sudden oak death and ramorum blight. The Plant Health Instructor. DOI: 10.1094/PHI-I-2008-0227-01).

In natural ecosystems (Figure 17), an important factor influencing the reaction of plant hosts to a *P. ramorum* outbreak will be the pre-existing health conditions of the ecosystems. Generalisation on the relationship between abiotic stress and susceptibility is difficult given the wide variety of susceptible species and ecosystems, and the potential for *P. ramorum* evolution. Nonetheless, a study by Roubtsova and Bostock (2009) found that transient abiotic stress (osmotic stress induced with salt) can be a potential contributing factor to infection severity by *P. ramorum* for two major hosts, *Rhododendron* and *Viburnum*. Moreover, symptom development in control (non-stressed) inoculated plants was delayed by one to two weeks relative to the inoculated, salt-stressed plants.

For the UK and continental Europe, there is the possibility that *P. ramorum*-susceptible ecosystems will be stressed in the coming years due to summer drought (as happened e.g. in 2003) (Desprez-Loustau et al., 2006; Tubby and Webber, 2010). Summer drought could be beneficial from the point of view of potentially reducing opportunities for splash dispersal of *P. ramorum*, which is known to be an important mechanism for infection (confirmed recently in California forests by Fichtner et al. (2009) – they also report suppression of chlamydospore production by moist redwood-associated soil – and in Belgian nurseries by Heungens et al. (2010)).

Drought however could predispose susceptible plants to infection. This link can also work in the other direction: stressed plants due to *P. ramorum* infection (which, in tanoak, affects hydraulic conductivity probably due to tylosis formation; Collins et al., 2009) may become more vulnerable to further stress due to drought. The importance of the interconnections among the various global change drivers in influencing plant health was emphasised in the review of Pautasso et al. (2010a).

Other management options are being tested. *Rhododendron*-removal in *P. ramorum* infested woodlands is being tried in the UK, but a new study shows that the pathogen can persist in roots of *Rhododendron*, so that post-clearance management of *Rhododendron* regrowth is necessary (Fichtner et al., 2011).

Recent data from California show that a forced reduction of the infectious host bay laurel obtained by removing all bay laurels in 50 x 50 m plots resulted in a significant reduction in inoculum potential both at 10 and 20 m for remaining bays (Appendix C.). Inoculum loads high enough to infect oaks were recorded 3% of the times for controls vs. 0.5% for treatments. This difference was significant at P< 0.001. This reduction is not enough to prevent infection of foliar host, but an inoculation experiment shows it is sufficient to effectively prevent infection of oaks. The selective elimination of the infectious host may thus work in Europe as well in those situations where there is a distinction between epidemiological host spreading the disease and vulnerable hosts highly susceptible to infection. This work also shows that a reduction in inoculum level, rather than an elimination of inoculum, can affect the epidemic.

Garbelotto and Schmidt (2009) report that a method consisting of an azomite soil amendment and bark lime wash was always ineffective, and did not reduce *P. ramorum* growth and infection rates. However, phosphonate treatments were shown to be effective in slowing both infection and growth rates for at least 18 months (Garbelotto and Schmidt, 2009). This study builds on the work on phosphonates by Garbelotto and colleagues already cited in the RAPRA report (there is now also a phosphonate study from Oregon, with disappointing results on seedlings (Kanaskie et al., 2010b)). Would such approach be effective in Japanese larch plantations? Would it be accepted in Europe by the local population and the many stakeholders? It should be emphasised that phosphonates act indirectly by increasing the production of secondary metabolites that have antimicrobial properties and by accelerating lignification and cell wall thickening processes and because of this mode of action they are regarded as environmentally friendly (Garbelotto et al., 2008).

2.2.5. Level of reduction of risk

Figure 18: Proposed disease cycle for *Phytophthora ramorum* in nurseries. (Reproduced by permission from Parke J L and Lucas S, 2008. Sudden oak death and ramorum blight. The Plant Health Instructor. DOI: 10.1094/PHI-I-2008-0227-01)

A sketch of the *P. ramorum* disease cycle in nurseries is illustrated above (Figure 18). Since control measures were adopted, there has been a decrease in the number of *P. ramorum* nursery reports, both in California (APHIS, 2011) and, generally, in EU states (FVO survey, 2010[8]), including the UK. Uncertainty remains about whether this pattern is a mere association (which would imply that a decrease in *P. ramorum* occurrences in the plant trade would have happened anyway, even without control measures) or whether there is a causal relationship between control measures and decreased nursery reports (which would imply that without control measures, the occurrence of *P. ramorum* would not have declined as it did). However, one can reasonably infer the positive impact of management options included in control measures adopted in the EU because the total number of occurrences of the disease have progressively decreased since 2007 (400 in 2007 versus 290 in 2010) and the number of interceptions decreased gradually, as well, from 46 in 2007 down to 17 in 2010. The specific situation observed in UK where the number of occurrences has increased was mainly caused by an epidemic development of the disease on Japanese larch (Figure 19).

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[8] Based on reports from Member States, as processed by the Food and Veterinary Office of the European Commission
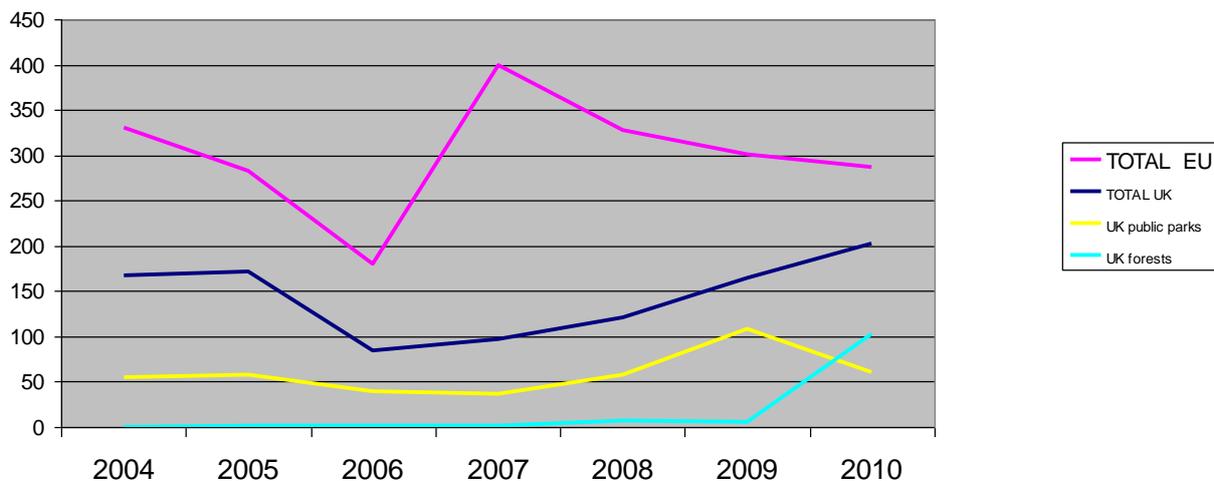
Figure 19: Number of *Phytophthora ramorum* outbreak sites since 2004 in the EU and in the UK only (FVO survey, 2010).

There is evidence that other factors may play an important role in the *P. ramorum* trend in the plant trade. For example, there has been a return of *P. ramorum* reports in nurseries from Scotland when the weather was favourable after some years without reports (Anon, 2010b). The importance of the weather pattern (in particular the amount and distribution of rain) has been underlined also by C. Brasier (pers. comm.) for the Japanese larch outbreaks and by the recent study of Davidson et al. (2011) on *P. ramorum* dispersal in different forest types in California. This implies that the level of reduction of risk is contingent on factors changing from year to year, which are difficult to predict and influence.

Moreover, inspection data from Norway show that *P. ramorum* is still present in import consignments with *P. ramorum* host plants originating in the European Union (Sundheim et al., 2009; Herrero et al., 2010). These data originate from the inspection procedure applied by the Norwegian Food Safety Authority, where import consignments showing suspicious symptoms are sampled for testing. After the peak in 2007 and 2008, there has been a sharp reduction in 2009 and 2010 (Table 6). However, this coincided with a new regulation imposed where nurseries in Germany and the Netherlands have to qualify specifically for export of *P. ramorum* host plants to Norway (http://www.mattilsynet.no/english/plant_health/regulations_on_measures_against_phytophthora_ram orum_have_been_amended_55856).

Table 7: Evolution of importation of consignments with *Phytophthora ramorum* in Norway since 2005.

year	Number of import consignments with host plants of *Camellia, Kalmia, Pieris, Rhododendron* and *Viburnum*	Number of import consignments with *P. ramorum*
2005	631	5
2006	525	6
2007	667	22
2008	242	14
2009	Not available	3
2010	Not available	1

2.2.6. Uncertainties

The critical uncertainty is that there has been no control against which the effectiveness of management measures can be assessed.

Additional uncertainties were noted in particular with regard to:

- The lack of data related to the origin of *P. ramorum* and its occurrence in Asia.

- The efficacy of detection methods because of the occurrence of asymptomatic infested plants, a lack of a systematic surveillance approach and a varying frequency of false negatives.

- The host range of *P. ramorum*, which is particularly worrying since the occurrence on Japanese larch in the U.K.

- The virulence and the fitness of progeny.

A more detailed treatment of these issues can be found in specific discussion of Member States comments in the risk assessment section or additional information in the sections above.

2.2.7. Key findings (Pest risk management)

- Options to reduce the likelihood of spread and to reduce the infestation and magnitude of impact in the nurseries were not considered in RAPRA.

- *P. ramorum* findings outside of nurseries (including managed parks, gardens, public greens, woodlands, and forests) have now been reported in Europe not just from the Netherlands and the UK, but also from Belgium, Denmark, France, Germany, Ireland, Luxembourg, Norway, Serbia, Slovenia, Spain, and Switzerland.

- An analysis of the effect of these measures on the prevalence of *P. ramorum* during the period 2004 to 2006 showed a reduction in the percentage of inspections positive for *P. ramorum* and a reduction in the number of outbreaks at nurseries.

- However, the continued findings of the pathogen in nurseries and in woodlands, notably on Japanese larch in UK, indicated that the phytosanitary measures have not been completely effective.

- An ongoing future update of the *P. ramorum* host list is needed, as the provision of new information from lab tests and field observations will continue.

- The use of network analysis to guide control schemes in the plant trade is advisable.

- There is a need to update various RAPRA statements in relation to risk management (e.g. surveillance plans, assumptions on spread, etc.) in the light of the Japanese larch outbreaks.

- Surveillance and eradication of plants for planting of known susceptible hosts (pathway i) have not been entirely successful: there was some success in the nursery trade but this did not prevent the UK outbreaks in Japanese larch plantations.

- There is a question about whether/how to regulate the imports of plants potentially susceptible (pathway ii) to *P. ramorum* into the EU.

- The regulation of movements within EU against a hot spot of *P. ramorum* infection (e.g. South-West of England and Wales) may be worthwhile considering.

- Although progress is being made in developing reliable molecular tools to detect *P. ramorum* where this occurs, there is still a non-zero likelihood of failing to detect the pathogen where this is present, which is worsened by the issue of asymptomatic infection.

- There is now published evidence for phenotypic differences among lineages. The introduction of new lineages into Europe could have significant consequences both for the nursery trade and natural ecosystems.

- The issue of intraspecific genetic variation in susceptibility of the various host species is understudied, particularly in Europe.

- Recent modelling and survey work from California shows the importance of data on true *P. ramorum* absences in order not to overpredict the potential distribution of the pathogen. Thus the importance of accurate large-scale *P. ramorum* surveys, even if these end up in a majority of negative reports.

- Nonetheless, other recent models have been developed in California to help prioritise detection efforts across regions differing in potential susceptibility. There is scope for the further development of such modelling approach also across Europe.

- In natural ecosystems, an important factor influencing the reaction of plant hosts to a *P. ramorum* outbreak will be the pre-existing health conditions of the ecosystems (which is uncertain for future decades given impending climate changes).

- Generalisation on the relationship between host stress and *P. ramorum* susceptibility is difficult given the wide variety of susceptible species and ecosystems, and the potential for *P. ramorum* evolution.

See the section on Uncertainties (2.2.6.) for further uncertain issues in the context of *P. ramorum* Pest Risk Management.

2.2.8. Conclusions of the pest risk management

The Panel supports the management options proposed in the RAPRA report to reduce the likelihood of introduction of *P. ramorum* in the risk assessment area in consignments from USA, Canada and the third countries that represent the pathogen's area/s of origin. The RAPRA report does not take into consideration the outbreaks that occurred within the risk assessment area and particularly the recent outbreak on Japanese larch in the UK. According to the Guidance on the harmonised framework for pest risk assessment and the identification of pest risk management options by EFSA Panel on Plant Health (PLH) (2010), the following options should be reviewed.

2.2.8.1. Option for consignment

- *Detection of P. ramorum in consignments by inspection or testing*

Methods are available for the detection of *P. ramorum* in consignments. However, there is a non–null risk of failing to detect the pathogen where this is present, despite the progress made by new research in developing reliable molecular tools to find *P. ramorum* where this occurs. The risk is worsened by the issue of asymptomatic infection and the varieties of commodities to be controlled. There is enough

evidence to recommend baiting with leaves of susceptible plants from soil samples collected and pooled before baiting, also baiting from water systems of facilities that are selling plants. Further progress is being made in developing markers to identify the three *P. ramorum* lineages (NA1, NA2 A2 and EU1 A1). It is essential to have a reliable knowledge on the lineages detected in consignments in order to prevent the introduction of alien lineages into the risk assessment area.

- *Removal of* P. ramorum *from the consignments by treatment or other phytosanitary procedures.*

Various treatments have been tested to determine efficacy in eradicating *P. ramorum* from infested plant material. No treatment can guarantee the removal of *P. ramorum* from the consignments, with the exception of heat treatments (including composting) that were considered an effective option for sanitation of *P. ramorum* plant material. However, these kinds of treatments can be applied only on non-living commodities such as wood or bark. The efficiency of heat treatment was not yet tested on soil. The use of fungicides will reduce the efficacy of detection in consignments.

- *Prevention of establishment by limiting the use of consignment*

No limited use of consignment was reported in the RAPRA report.

2.2.8.2. Options for the prevention or reduction of infestation and spread in the crop

Surveillance should be based on targeted inspection of susceptible plants including *Larix spp.* in nurseries, gardens, parks, woodland, heathland and forest, and surveillance strategies should be carried out independently of symptoms.

- *Prevention of infestation of the commodity*

In the EU, eradication of the disease in nurseries is being attempted by destroying all infected plants within a 2-m radius of a diseased plant and holding all susceptible plants within a 10-m radius plus any remaining plants from the same consignment as the diseased plants for further assessment. Release of these plants is allowed following two negative visual inspections during 3 months of active growth and no treatment that could suppress symptoms should be applied during the quarantine period for all susceptible plants at the place of production.

An experimental study of *P. ramorum* dispersal in a mock nursery showed that pathogen dispersal occurred mainly to neighbouring plants and that plant-to-plant contact was an important condition for dispersal to happen (Heungens et al., 2010). Moreover, there was no aerial detection of *P. ramorum* with a spore sampler, whereas there was evidence for *P. ramorum* dispersal via water films at a distance of several meters (Heungens et al., 2010). The role of splash- and irrigation water in leading to *P. ramorum* dispersal in nursery settings was also underlined by Neubauer et al. (2006) and by Tjosvold et al. (2006).

Although a preliminary study from California suggests a spread in nurseries that is within the range of what is imposed by regulations (Tjosvold, 2010), Heungens et al. (2010) reported no clear pattern of focal spread, as the same genotype was detected at more than 10 m distances in a genetic study over 2 growing seasons at a large commercial nursery in Belgium with multiple infestations of *P. ramorum*. This result may be due to coalescing foci, as suggested by a comparative analysis of different detection techniques (above- and below-ground ELISA, real-time-PCR, nested PCR and culture) in a *P. ramorum*-infested nursery growing *Camellia japonica* in California (Bulluck et al., 2006).

- *Establishment and maintenance of pest freedom of crop, place of production or area.*

Since 2002, the European Union adopted emergency measures to prevent the introduction and movement of *P. ramorum* into and within the EU territory based on the "plant passport" certification system. FVO (2010) reported some success with the eradication in nursery crops. The structure of the nursery trade in Europe and its various states is poorly known. If it has the characteristics of a scale-free network in which major nodes predominate, then control may be easier than in the absence of such super-connected nodes. By targeting nodes with many links in and out of them, regulatory controls can be more effective and efficient than random or systematic approaches. A focus on plant passporting of key traded genera susceptible to *P. ramorum* (e.g. *Rhododendron, Viburnum*) also makes sense from a network perspective. As the trade of plants susceptible to *P. ramorum* is a directed network (links from nurseries to garden centres do not imply the reverse connection), the risk of spreading the disease associated with different categories of business (producers, wholesalers, and retailers) is not the same. Simulations suggest that increasing the proportion of wholesalers (and reducing the proportion of producers and/or retailers) in plant trade networks with different structures is associated with a decrease in the epidemic threshold (Pautasso et al., 2010c). However, information on the structure of plant trade networks is mostly lacking.

No management options are identified to reduce the likelihood of spread following introduction to areas of susceptible plant hosts in woodland, heathland and forests. The epidemic on Japanese larch in UK and Ireland is indicative of that fact (although felling is currently taking place in those plantations).

- Slash and burn was attempted in Oregon to eradicate the pathogen, and may be attempted in relation to *Rhododendron ponticum* clearance, although this needs more than burning, as it regrows from stock, and reinfection coming from soil, roots or surface litter is common.

- Use of herbicides and prevention of resprouting are alternative options, but removal of *R. ponticum* at the stand/landscape level is a daunting task (the shrub is an invasive exotic and there have been previous attempts to remove it from British woodlands).

Management may instead focus on the protection of important trees, by clearing potential *P. ramorum* hosts around them. Although the Panel does not comment on economic aspects, we note that even if the epidemic may be at comparable level, when removing hosts at single locations or doing nothing, the former strategy may enable some degree of protection for individual groups of trees that are deemed worthy of preservation efforts.

- *Consideration of other possible measures*

A key and difficult issue to establish and maintain pest freedom for *P. ramorum* is the issue of water circulation systems in plant nurseries (Seipp et al., 2008).

2.2.8.3. Evaluation of risk management options

The following options are identified for reducing the magnitude of impact:

- Early detection in consignments by inspection or testing allows the halting of a spread pathway, as well as limiting local pathogen build-ups.

- Removal of infected plants and sanitation achieved by e.g. burning, composting, etc.

- Reducing density of potential sporulating hosts may change the course of the epidemic.

- Chemical control using phosphonate as preventive applications is a possible option for the future sustainable management in forested areas subject to further studies to confirm effectiveness where *P. ramorum* is established. The study by Garbelotto et al. (2008) provides data on efficacy of copper treatments for foliar hosts, but obvious ecological consequences are associated with copper.

- Chemical control in nurseries (Ufer et al., 2008).

- Irrigation water treatment in nurseries using filtration and chemical treatment.

- Biological control may be a possible option for the future sustainable management in nurseries, but no studies provide convincing support for this option.

OVERALL CONCLUSIONS

1.   Original RAPRA report

The Panel agrees with RAPRA's conclusion that "*P. ramorum fulfils the criteria of a quarantine pest*". Additionally, "*there is a risk of further entry, establishment and [...] impact*" of lineages and mating types other than those already present in the EU.

With regards to introduction of novel pathogen genotypes, the Panel notes that there is growing evidence of differences among lineages. Therefore, it is advisable to avoid introductions of different lineages not only because of potential of sexual recombination, but also because of such inherent differences. Additionally, the formation of thick-walled oospores as a result of sexual activity made possible by the introduction of the A2 mating type currently absent in the EU, may allow for broader establishment of the pathogen in the EU.

The Panel supports the view that the RAPRA project "*substantially increased our knowledge and understanding of pathogenicity, host range, distribution, and survival of P. ramorum and of the biological and climatic conditions favouring disease outbreaks*" (Jung et al., 2009).

Although RAPRA was published recently in 2009, the *P. ramorum* pathosystem is developing rapidly and much research has been performed since publication of the RAPRA report. This continuing production of new information on *P. ramorum* is not likely to stop. There is thus a need to keep updated with future research on the pathogen and to adapt measures to new key information (e.g. new major host species, effectiveness of plant passporting schemes, interactions with other plant health stressors) in an iterative way.

2.   Member States comments

Many Member State comments on the RAPRA report have been addressed by a direct RAPRA response, a summary of which is provided in this Opinion, together with the comments of the Panel on each of these issues.

Although in some cases RAPRA responded to the Member State comments in the light of additional information not available at the time of the RAPRA project or at the time the Member States made their comments, this was reasonable given that additional information on the pathogen and its outbreaks can lead to better management.

The outbreaks on UK Japanese larch plantations run counter to the view of some Member States that *P. ramorum* is unlikely to spread by natural means over large regions in Europe (although the role of the plant trade in the Japanese larch outbreaks is still unclear).

UK and EU-wide *P. ramorum* genetic studies as performed in California would provide information on entry into the natural environment from the plant trade.

Although *P. ramorum* findings in nurseries are declining and represent a minority of tested nurseries, the pathogen is still reported from plant traders of many Member States in numbers which are not negligible. Intra-European trade of *P. ramorum*-susceptible plant material is an important issue.

P. ramorum lineages have phenotypic differences, and on some hosts some lineages are more aggressive than others, so that it is important to avoid introduction of new lineages into Europe. Additionally, the presence of both mating types in the EU may lead to sexual reproduction. Sexually produced oospores may increase survival of the pathogen in harsher climatic conditions, and recombined progeny may accelerate the adaptive evolution of the pathogen. Both traits are considered as undesirable.

3.   Additional information

The outbreak on Japanese larch in the UK and Ireland in the absence of Rhododendron is an important development: it (1) suddenly widens the *P. ramorum*-suitability and connectivity of European landscapes, (2) enlarges the list of potential hosts, (3) changes the dynamics of infection and spread by indicating one plant species that can act both as infectious and lethally affected host, and (4) makes it necessary to reassess the management options for some pathways.

Modelling can help in better targeting surveys both in the plant trade (network approaches) and in the semi-natural environment (combination of climatic suitability with distribution of (sporulating) hosts).

Genetic and molecular studies are essential to get a better handle on the pathways of introduction and spread into and within Europe and to develop more rapid and reliable diagnostic tools.

4.   Synthesis

Since 2002, the European Union adopted emergency measures to prevent the introduction and movement of *P. ramorum* into and within the EU territory based on specific import requirements and the "plant passport" certification system. FVO (2010) reported some success with the eradication in plant nurseries. Uncertainty remains about whether this pattern is a mere association (which would imply that a decrease in *P. ramorum* occurrences in the plant trade would have happened anyway, even without control measures) or whether there is a causal relationship between control measures and decreased nursery reports (which would imply that without control measures, the occurrence of *P. ramorum* would not have declined as it did). This uncertainty is only slightly lowered by the same two phenomena (emergency measures in nurseries and reduction of *P. ramorum* nursery outbreaks) having also occurred in California. Additional potential evidence in favour of a reduction of spread caused by the regulatory action is provided by the reduction in interceptions in Norway since that Nation demanded that German and Dutch nurseries be certified as free of pest in order to be allowed to ship *P. ramorum*-susceptible plants to Norway. Nonetheless, the emergency measures have not prevented outbreaks occurring in the natural environment.

The Panel generally supports the management options proposed in the RAPRA report to reduce the likelihood of introduction of *P. ramorum* in the risk assessment area in consignments from USA, Canada and the third countries that may represent the pathogen's area/s of origin.

The RAPRA report did not take into consideration the outbreaks on Japanese larch in the UK, given that they had not yet occurred. For the reasons detailed in this Opinion, these outbreaks are a major

step change in the whole *P. ramorum* issue. The further development of these outbreaks should be carefully monitored, studied and considered in future regulatory work.

The US APHIS PRA (APHIS, 2008) states that likelihood of introduction of *P. ramorum* is determined by three factors, namely: (1) entry potential which is defined as directly proportional to the volume of plants traded, (2) establishment and spread potential, determined by availability of appropriate climate and susceptible hosts, and (3) detection potential which is negatively correlated to the failure to detect the organisms. While not much can be done in terms of point (2) (although limited selective removal of hosts is ongoing in the US and in Europe), it is clear that regulations have the power to decrease both the number of entries (e.g. by import regulations and checking on the health status of plants) and the number of failed detections (by improving sampling schemes and diagnostic assays), thus effectively limiting the introduction of the pathogen.

Many uncertainties regarding *P. ramorum* persist. These include (but are not limited to):

- the lack of data related to the origin of *P. ramorum* and its occurrence in Asia, and other parts of the world

- the source of inoculum because of the occurrence of asymptomatic infested plants, the lack of a systematic surveillance approach not based on symptoms, and the host- and season-dependent frequency of false negatives, although the detection tools have been improved in specificity and sensitivity

- the expanding host range of *P. ramorum*, and

- the virulence and the fitness of progeny.

The many remaining uncertainties call for further research on *P. ramorum* across Europe.

DOCUMENTATION PROVIDED TO EFSA

1. Request (Background and Terms of Reference) to provide a scientific opinion on the pest risk analysis *Phytophthora ramorum* prepared by the FP6 project RAPRA. SANCO E1/GC/svi D(2010) 510302. 27 April 2010. Submitted by European Commission, DG SANCO Health and Consumers Directorate General.

2. Summary of RAPRA *Phytophthora ramorum* PRA (Date of Report: 26 February 2009).

3. Comments on this PRA from Belgium, Slovenia, Spain, the Netherlands and Germany, and a response to the comments from Belgium prepared by the United Kingdom.

4. Briefing note from the Forestry Commission, England, on *Phytophthora ramorum* (dated 18 November 2009).

5. Overview report of the results from the 2009 and 2010 official surveys carried out by the Member States for the presence of *Phytophthora ramorum* in their territory according to Commission Decision 2002/757/EC.

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APPENDICES

A. PATHOGEN VARIABILITY, COMPARING THE VIRULENCE OF NA1 AND NA2 LINEAGES THROUGH INOCULATION EXPERIMENTS (PERSONAL COMMUNICATION BY MATTEO GARBELOTTO, 2011)

Umbellularia californica

Analysis of variance

Random Effect	Var Ratio	Var Component	Std Error	95% Lower	95% Upper	Pct of Total
Tray[Temp]andRandom	0.1464213	0.0968672	0.0472223	0.0448978	0.3407006	11.605
TreeandRandom	0.1152419	0.07624	0.0626957	0.0243263	1.0920462	9.134
Residual		0.6615654				79.26
Total		0.8346727				100

 -2 LogLikelihood = 775.32213

Source	Nparm	DF	Sum of Squares	F Ratio	Prob > F	
Temp	2	2	12.037176	9.0975	0.0011	
Lineage	1	1	7.093438	10.7222	0.0012	
Lineage*Temp	2	2	0.450442	0.3404	0.7118	
Tray[Temp]andRandom	27	24	26.18265	.	.	Shrunk
TreeandRandom	5	4	17.271706	.	.	Shrunk

Means and standard errors of transformed data – ln(area in mm^2 + 1)

Grouping	Least Sq Mean	Mean	Std Error	Tukey HSD	Back-transform LSD	back-X SE
Temp (°C)						
12	3.74	3.75	0.18	A	43.25	2.20
20	4.08	4.05	0.18	A	59.89	2.20
24	3.27	3.27	0.18	B	27.31	2.20
Lineage						
NA1	3.54	3.52	0.15	B	35.39	2.17
NA2	3.86	3.86	0.15	A	48.24	2.17
Lineage, Temp						
NA1,12	3.61	na	0.20	ABC	38.00	2.22
NA2,12	3.88	na	0.20	AB	49.25	2.22
NA1,20	3.95	na	0.20	AB	52.79	2.22
NA2,20	4.20	na	0.20	A	67.97	2.22
NA1,24	3.05	na	0.20	C	22.22	2.22
NA2,24	3.48	na	0.20	BC	33.62	2.22

Rhododendron
Analysis of variance

Random Effect	Var Ratio	Var Component	Std Error	95% Lower	95% Upper	Pct of Total
Tray[Temp]andRandom	9.52E-09	2.99E-08	0	.	.	0
TreeandRandom	8.34E-09	2.62E-08	0	.	.	0
Residual		3.1391388				100
Total		3.1391389				100

 -2 LogLikelihood =

1196.4458REML Iterations had problems. Please check iteration history. Results may be invalid.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob > F	
Temp	2	2	624.55617	99.4789	<.0001	
Lineage	1	1	13.19766	4.2042	0.0413	
Lineage*Temp	2	2	2.98554	0.4755	0.6221	
Tray[Temp]andRandom	27	24	0.00001	.	.	Shrunk
TreeandRandom	5	4	0	.	.	Shrunk

Means and standard errors of transformed data – ln(area in mm^2 + 1)

Grouping	Least Sq Mean	Mean	Std Error	Tukey HSD	back-transform LSM	back-X SE
Temp (°C)						
12	1.48	1.48	0.18	C	5.37	2.19
20	5.00	5.00	0.18	A	149.45	2.19
24	3.46	3.46	0.18	B	32.77	2.19
Lineage						
NA1	3.10	3.10	0.14	B	23.22	2.16
NA2	3.52	3.52	0.15	A	34.83	2.16
Lineage,Temp						
NA1,12	1.22	na	0.25	C	4.38	2.28
NA2,12	1.73	na	0.25	C	6.66	2.28
NA1,20	4.70	na	0.25	A	110.86	2.28
NA2,20	5.30	na	0.25	A	201.58	2.28
NA1,24	3.39	na	0.25	B	30.60	2.28
NA2,24	3.53	na	0.25	B	35.10	2.29

B. A REPORT ON GAMETANGIA FORMATION BY *PHYTOPHTHORA RAMORUM* (PERSONAL COMMUNICATION BY MATTEO GARBELOTTO, 2011)

1. INTRODUCTION

It has been suggested (Brasier, 2003) that the mating system of *Phytophthora ramorum*, may not be fully functional, however recent studies report that oospores were produced and germinated (Boutet et al., 2010). Therefore, mating tests in *P. ramorum* were carried out to study the actual level of fertility in this species.

2. Materials and Methods

Crosses of *P. ramorum* were prepared using the method developed by Brasier and Kirk (2004). Briefly, discs of 5 mm diameter cut from the margin of actively growing colonies of the twelve AFLP genotype isolates on carrot agar were mashed together with an A1 (Pr 3-74-2) and A2 (Pr 06) tester isolate on three separate CA plates per crossing . Plates were incubated in the dark at 20 °C. Oospore and oogonial diameter and antheridial width and length were measured for 30 interactions after 3 wk. Isolates that did not form gametangia with either A1 or A2 tester isolate in the first trial were retested.

Table 1: Isolates of *Phytophthora ramorum* from California and Oregon used in this study.

Isolate [a]	Source	State, county	Year	Mating type	AFLP genotype [b]	Experiments
Pr3-74-2	*Viburnum x bodnantense* 'Dawn'	Oregon, Clackamus	2003	A1[c]	European	Mating test
Pr01	*Quercus agrifolia*	California, Marin	2000	A2	7	All experiments
Pr06	*Q. agrifolia*	California, Marin	2000	A2[c]	1	Mating test
Pr27	*Q. agrifolia*	California, Marin	2000	-[d]	1	All experiments
Pr36	*Q. agrifolia*	California, Sonoma	2000	A2	3	All experiments
Pr52	*Rhododendron* sp.	California, Santa Cruz	2000	A2	5	All experiments
Pr57	*Lithocarpus densiflora*	California, Santa Clara	2001	A2	1	All experiments
Pr70	*Vaccinium ovatum*	California, Marin	2001	-	1	All experiments
Pr71	*Q. agrifolia*	California, Sonoma	2001	A2	2	All experiments
Pr75	*Q. agrifolia*	California, Monterey	2001	A2	1	All experiments
Pr102	*Q. agrifolia*	California, Marin	2001	-	1	All experiments
Pr106	*Umbellularia californica*	California, Sonoma	2001	A2	8	All experiments
Pr159	*Lithocarpus densiflora*	Oregon, Brookings	2001	-	4	All experiments
Pr345	*Sequoia sempervirens*	California, Sonoma	2002	A2	6	All experiments

3. Results

Four of 12 isolates did not form gametangia with either A1 or A2 tester isolates in two separate trials (Table 1, Figure 1). The remaining isolates, formed gametangia only with the A1 tester isolate indicating they are all A2 mating type. Gametangia were usually observed directly beneath the mashed plugs in relatively low numbers with the exception of isolate Pr01 that formed abundant gametangia of about 1 cm radius around the mashed plugs in one of two experiments. However, the capacity to form gametangia was variable as Pr01 produced oospore numbers comparable to the other isolates. Oogonia, oospores and antheridia dimensions were significantly different among isolates ($P < 0.001$).

The isolate averages for oogonia ranged from 27.4-30.1 μm diam. Oospores were always plerotic and ranged 24.4-26.6 μm diameters. In all interactions, antheridia were amphigynous and barrel shaped with dimensions of 14.0-16.7 x 15.0-16.6 μm.
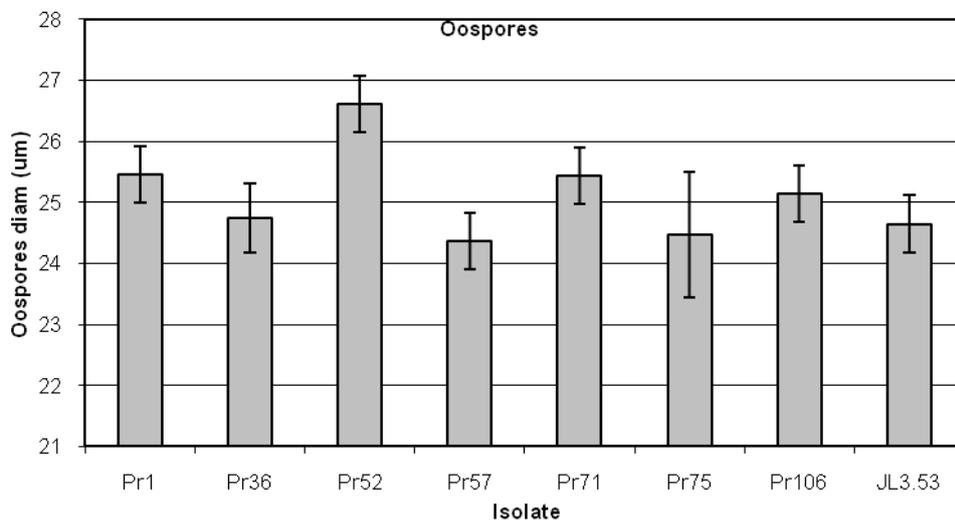

Figure 20: Mean diameter of oospores for 8 A2 isolates of *P. ramorum* belonging to the NA1 lineage. Four isolates (Pr27, Pr70, Pr102 and Pr159) did not form gametangia or oospores with either the A1 or the A2 tester strains. Numbers of oospores were comparable among isolates, with the exception of Pr75, that produced low numbers.

CONCLUSIONS

Although isolates are not completely interfertile, at least 7-8 of 12 isolates tested produced a significant and apparently viable number of oopsores. Thus, intefertility for this species can be estimated to be between 58 and 66% and should be taken as a serious possibility. We have information suggesting that lack of fertility is actually not necessarily isolate-dependent but rather depends on the physiological state of the isolate, hence the negative crosses here described may turn into positive crosses if the physiological state of the isolate were to change.

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C. DOES THE REMOVAL OF BAY LAUREL REDUCE THE INOCULUM PRESSURE OF *PHYTOPHTHORA RAMORUM* IN OAK WOODLANDS? (PERSONAL COMMUNICATION BY MATTEO GARBELOTTO, 2011)

1. INTRODUCTION

A study was initiated in 2005 and is still ongoing, to determine whether removing small patches of the infectious bay laurel will reduce the inoculum pressure by the sudden oak death (SOD) pathogen *Phytophthora ramorum*.

2. Experimental approach

Eight 50 x 50 m plots were selected in two stands at the Soquel Demonstration Forest in Santa Cruz County, South of San Francisco. Four plots were kept as untreated controls while all bays were removed in four plots.

Inoculum pressure was continuously evaluated by baiting the pathogen with 5 bay leaves placed every three weeks in 16 buckets per plot. During the dry season, each bucket was filled with at least one gallon of water carried onto the trial site. In a controlled study, it had been previously determined that infection of 4-5 leaves per bucket correspond to high inoculum levels, capable of infecting oaks, while 1-3 infected leaves out of five correspond to low and moderate levels of inoculum, that normally do not result in oak infection.

3. Results and discussion

Five years after the beginning of the trial, and three years after bay removal, number of instances of high inoculum levels was significantly lower in treatment than in control plots. In control plots, we recorded cumulatively 50 instances of high inoculum levels, while in treatment plots high inoculum levels were recorded only 8 times. Based on a Fisher's Exact Test this difference is significant at $P<0.0001$. We conclude that selective thinning of bay laurels at a small spatial scale has the potential to reduce infection levels of local oaks. Based on the scale of our experiment, removal of bay laurels should occur 10-15 m around oaks for protection to be achieved. This is the first trial showing that removal of infectious hosts has a local effect on inoculum potential.